DTU Library

New approaches for determining solubility of volatile liquid chemicals

Birch, Heidi; Trac, Ngoc Lam; Mayer, Philipp

Published in:
SETAC Europe 28th Annual Meeting - Abstract book

Publication date:
2018

Document Version
Publisher's PDF, also known as Version of record

ROME 2018

SETAC Europe 28th Annual Meeting

13–17 May 2018 | Rome, Italy

ABSTRACT BOOK

Responsible and Innovative Research for Environmental Quality

PRINT ISSN 2309-8031 - ONLINE ISSN 2310-3043 © 2018

Society of Environmental Toxicology and Chemistry Europe (SETAC Europe)

ABSTRACT BOOK
SETAC Europe 28th Annual Meeting

TABLE OF CONTENTS

This book compiles the abstracts from the platform and poster session presentations at the 28th Annual Meeting of the Society of Environmental Toxicology and Chemistry- Europe (SETAC Europe), conducted at the Rome Convention Centre La Nuvola, Rome, Italy, from 13 – 17 May 2018.
The abstracts are reproduced as submitted by the author and accepted by the Scientific Committee. They appear in order of abstract code and alphabetical order per presentation type. The poster spotlight abstracts are included in the list of poster abstracts. The presenting author of each abstract is underlined.

SETAC Europe Office
Avenue de la Toison d'Or 67
B-1060 Brussels
Belgium
T +32 2 772 72 81
F +32 2 770 53 86
setaceu@setac.org
setac.org

SOCIETY OF ENVIRONMENTAL TOXICOLOGY AND CHEMISTRY

In the 1970s, no forum existed for interdisciplinary communication among environmental scientists, biologists, chemists, toxicologists, managers, engineers or others interested in environmental issues. The Society of Environmental Toxicology and Chemistry (SETAC) was founded in North America in 1979 to fill the void, and quickly saw dynamic growth in the Society's membership, meeting attendance and publications.

A unique strength of SETAC is its commitment to balance the scientific interests of government, academia and business. The Society by-laws mandate equal representation from these three sectors for officers of the World Council and Geographic Unit Boards of Directors and Councils, and in the composition of committees and other society activities. The proportion of members from each of the three sectors has remained nearly equal over the years.

The Society is concerned about global environmental issues. Its members are committed to Environmental Quality Through Science®, to timely and effective communication of research, and to interactions among professionals so that enhanced knowledge and increased personal exchanges occur. Therefore, SETAC publishes two globally esteemed scientific journals and convenes annual meetings around the world, showcasing cutting-edge science in poster and platform presentations. Because of its multidisciplinary approach, the scope of the science of SETAC is broader in concept and application than that of many other societies.

SETAC's growth is reflected in the founding of geographic units around the world. SETAC Europe was established in 1989 as an independent organisation, followed by SETAC Asia-Pacific in 1997 and SETAC Latin America in 1999. In 2002, the four existing organisations joined together under the governance of the SETAC World Council. SETAC Africa is the most recent geographic unit, which was adopted in 2012. As evidence of international acceptance of the SETAC model and of the great interest at the local level, regional chapters and branches have emerged in a number of countries.

SETAC publishes two journals: *Environmental Toxicology and Chemistry* (ET&C) and *Integrated Environmental Assessment and Management* (IEAM). Environmental Toxicology and Chemistry is dedicated to furthering scientific knowledge and disseminating information on environmental toxicology and chemistry, including the application of these sciences to risk assessment. Integrated Environmental Assessment and Management focuses on the application of science in environmental decision-making, regulation, and management, including aspects of policy and law, and the development of scientifically sound approaches to environmental problem solving. Together, these journals provide a forum for professionals in academia, business, government, and other segments of society involved in the use, protection, and management of the environment for the enhancement of ecological health and human welfare.

SETAC books provide timely in-depth reviews and critical appraisals on scientific subjects relevant to understanding a wide range of contemporary topics pertaining to the environment. These include any aspect of environmental chemistry, toxicology, risk assessment, risk management, or environmental policy.

SETAC has two administrative offices, in Pensacola, Florida, USA, established in 1992, and in Brussels, Belgium, established in 1993.

Keynote abstracts

Keynote Sunday
Responsible Research and Innovation (RRI) - a Path towards Sustainability?
Roger Strand, University of Bergen, Centre for the Study of the Sciences and the Humanities, Norway
Responsible Research and Innovation (RRI) is a cross-cutting principle of EU's research funding programme "Horizon 2020". Indeed, in Rome 2014, scientists and policy-makers jointly produced the "Rome Declaration on RRI in Europe", that states that "excellence today is about more than ground-breaking discoveries – it includes openness, responsibility and the co-production of knowledge". The principle of RRI acknowledges that civil society is entitled to "speak back" to science and help shape the knowledge and technology of tomorrow in an ethically acceptable and sustainable direction.
What does RRI entail in practice, for researchers, innovators and policy-makers? How could RRI principles, indicators and practices help to pick up more early warnings to avoid costly late lessons from unfortunate impacts of science and technology? The lecture will present the conceptual basis of EU's RRI policy. Specifically, a full appreciation of RRI depends on a theoretical understanding not only of risk, but also of decision-making under uncertainty, ignorance and indeterminacy.

Keynote Monday
Food Safety in a Complex Changing World
Bernhard Url, EFSA, Italy
EFSA provides independent scientific advice on all matters related with food and feed with a direct or indirect impact on human, plant and animal health.
Effects on the environment are also considered as they may pose an indirect risk to food and feed. EFSA takes into account environmental risk assessment in its assessments of the application of plant protection products, the deliberate release into the environment of GMOs and the use of certain substances in food and feed (e.g. feed additives). EFSA also assesses the environmental risks related to the entry and spread of invasive alien species harmful for plant health.
EFSA is looking into the future, keeping up with a rapidly evolving and globalised world, characterised by dramatic environmental and other global changes (e.g. economic, political social, and technological) and an exponential growth and availability of data. These set new opportunities and challenges to the assessment of risks to both the environment and food safety and can drive their (re)emergence. In this context, EFSA is reviewing the methodologies for risk assessment and for the identification of emerging risks within its remit.
Predictive modelling tools based on holistic approaches for environmental risk assessment in realistic landscapes and under different scenarios of multiple stressors are being developed. Approaches considering the complex interactions and dynamics between the different food system actors, their behaviour and external drivers are proposed as tools useful for long term anticipation of emerging risks. Expert knowledge elicitation, horizon scanning, and crowdsourcing are being explored as tools to broaden participation, strengthen engagement of all relevant stakeholders and manage interconnectivity, in application of principles of resilience thinking.
Environmental quality and food safety are strongly intertwined. They need to be considered together when aiming toward the achievement of sustainable development goals. Consistent approaches for scientific assessment and data management need to be developed, integrating also societal, technological and economic drivers to effectively cope with the dramatic global changes and the data revolution we are observing.

Keynote Tuesday
Innovative Research Issues in Environmental Mutagenesis
Eugenia Dogliotti, Istituto Superiore di Sanità, Department of Environment and Health, Italy
During the 1920s, mutation research was put on a firm basis by H. J. Muller, who developed the concept of "mutation rate" and devised quantitative techniques for its measurement. These techniques allowed the discovery of the mutagenic action of ionizing radiation and paved the way for the pioneering work of C. Auerbach on chemical testing, starting with mustard gas. Since that time the recognition of the multitude of possible sources of mutagenic insults promoted the development of the science of environmental mutagenesis. Today the mechanisms by which chemicals induce mutation and the role of genetic susceptibility in the response to environmental mutagens have been largely explored. Moreover, a battery of test methods is available for regulatory purposes. What are the current challenges in environmental mutagenesis? New techniques for mutation research have been developed. The "omics" technologies such as whole genome sequencing, epigenetic profiling, transcriptomics, proteomics and metabolomics have provided a snapshot on the effects of genetic polymorphisms, gene regulation, protein synthesis and stability, metabolic pathways in the control of cell function. This presentation will describe: 1) the successful identification of the mutagenic environmental agents underlying certain types of cancer by using whole genome sequencing; ii) the evidence that epigenetic alterations mediate toxicity from environmental chemicals and, iii) the use of the exposome approach, that comprises all environmental exposures that a person experiences from conception throughout the life course, to unravel complex gene environment interactions that affect disease risk.

Keynote Wednesday
The Environmental Dimension of Antimicrobial Resistance: Assessing and Managing the Risks of Anti-infectives
Jason Snape, AstraZeneca Global Safety, Health and Environment, UK
Antibiotics are vital in the treatment of infectious disease in both livestock and human health and they are entering the environment continuously. In freshwaters antibiotics can reach concentrations up to mg/L, but more commonly they occur in the low to sub μg/L range. They selectively target bacteria and thus there is an increased likelihood for impacts on environmental bacteria populations at levels well below that for effects on aquatic vertebrates. However, current environmental risk assessment (ERA) frameworks of antibiotics, as required by the European Medicines Agency guidelines 2006, adopts the use of one species of cyanobacteria only to represent all bacterial diversity. The activated sludge respiration inhibition test (ASRIT), used to identify risk to microorganisms in sewage treatment plants has also been proven to be insensitive for antibiotics. Thus, there is concern that the ERA for antibiotics does not fully consider their potential impacts on microbial community structure, function and resilience. In addition to the risk posed to ecosystem function there is a global concern on antimicrobial resistance (AMR) development and the associated risk to human health. It has been proposed that the risk of AMR development in the natural environmental should be included in ERA but there is currently no standard experimental methodology or framework to address this. Recently, a theoretical approach that makes use of minimum inhibitory concentrations (MIC) of clinically relevant bacteria (CRB; using the European Committee on Antimicrobial Susceptibility Testing (EUCAST) database) has been proposed to predict no effect concentrations (PNEC) for AMR development (PNECR). To help define science-based protection goals for antibiotics for use in a prospective ERA frameworks and to define safe discharge concentrations for antibiotic production and patient use this presentation will review the publicly available aquatic ecotoxicity data for antibiotics to assess the following: 1) the relative sensitivity of commonly used taxa in aquatic ecotoxicity to antibiotics; 2) the value of extending the toxicity testing to a more diverse range of bacteria species and: 3) how a PNECR relates to the PNEC derived for surface waters (PNECSW) using standard ecotoxicity testing. This presentation will describe (i) the output of this analysis of protection goal data and (ii) how the wider pharmaceutical industry are addressing concerns with antibiotic residues associated with manufacturing operations.

Platform Abstracts

Modelling and monitoring of pesticides fate and exposure in a regulatory context (I)

1

The SETAC DRAW workshops - aims, approaches and progress to date

N. Mackay, FMC Corporation / Environmental Modelling; A. Alix, Dow Agrosciences / Risk Management; G. Azimonti, ICPS; A. Chapple, Bayer Crop Science AG; P. Miller, Silsoe Spray Applications Unit Ltd; K.M. Nienstedt, European Commission - DG SANCO / PPR; C. Pickl, Federal Environment Agency; T. Wolf, Agrimetrix Ltd.

In order to develop a more complete understanding of spray drift to improve the regulatory basis for representation in risk assessments, a set of SETAC workshops known as DRAW (Drift Risk Assessment Workshops) are underway to facilitate a range of efforts: Assemble and interpret a database of the spray drift trials for boom sprayers; Develop a programme of trials to more fully characterise drift influences; Use this information to develop proposals for standardized protocols for drift characterization in the field Develop an enhance role for mathematical modelling as a higher tier risk assessment option; and Expand and reinforce the toolbox of regulatory risk mitigation measures. This presentation will focus upon; Database development and study design The workshop database currently comprises 56,001 data points from a wide range of studies, providing a rich, complex basis for supporting a range of different research and regulatory efforts. Because of the variation in study designs and the consequent difficulties with interpretation there is a clear motivation to develop a more detailed and tightly defined protocol to support future research efforts. This presentation will summarise the database and efforts to develop and test a protocol to support further research efforts. Modelling Options for developing an expanded role for modelling of drift profiles have focussed upon evaluations of two models that have been used within a regulatory context in the EU; IDEFICS and the SSAU Arable Crop Spray Drift Model. This presentation will report on model assessment efforts, potential future improvements in process representation and consider options for regulatory scenario development. Flexibility in risk mitigation An earlier workshop (SETAC MAgPIE) compiled a toolbox of risk mitigation measures in use in Europe and recommended development of communication tools to support broader and more effective implementation and encourage certification and testing harmonisation. This presentation will summarise the efforts underway in SETAC DRAW to realise these objectives through the creation of a platform to support exchange on scientific, technical, professional, and legislative or regulatory aspects of the toolbox, to further develop its accuracy and effectiveness (https://www.spraydriftmitigation.info/).

2

Plant uptake in regulatory environmental exposure assessment: Refined modelling based on experimental data

C. Schriever, BASF SE; Z. Gao, Bayer AG Crop Science Division; M. Lamshoeft, Bayer CropScience AG / R&D; M. Reitz, H. Resseler, Syngenta Agro GmbH; R. Sur, Bayer AG - Crop Science Division / Environmental Safety; P. Sweeney, Syngenta; P. Volz, BASF SE; S. Webb, Syngenta Ltd; B. Zillgens, Dupont GmbH

A novel study design to determine plant uptake of chemicals for environmental fate modelling was developed and tested in a tiered approach. Ten laboratory organizations with different levels of experience with uptake testing participated in a round robin test and studied uptake of [14C]-1,2,4-triazole by wheat plants. Afterwards, uptake of ten radiolabelled chemicals with various properties by potato, tomato or wheat plants was investigated in two laboratories. The findings showed acceptable inter-laboratory variability and proved the applicability of the design to various compound/crop combinations. Experimental Transpiration Stream Concentration Factor (TSCF) values were higher than calculated values, implying that the equation of Briggs et al. (1982) underestimates plant uptake of compounds with logKow values of less than 2. Results obtained with this study type are suggested to be used for regulatory environmental exposure assessments. These schemes are usually based on a tiered approach, where modelling with refined model parameters is one of the higher tier options. One example is the plant uptake factor that is considered e.g. in the course of the leaching assessment according to FOCUS Groundwater. Consideration of plant uptake (dissolved compound mass is taken up into plants with the soil porewater) decreases compound mass in soil and can lead to more realistic predicted environmental concentrations. Recent guidance proposes a default uptake factor of 0 for modelling and suggests two refinement options: TSCF values calculated from the logKow of a substance (Briggs et al., 1982) and the substance specific TSCF value from “uptake experiments with appropriate and agreed set-up to be developed” (EFSA, 2013; EU Com, 2014). The presented study design was explicitly developed to address the need identified by EFSA and was based on suggestions from the EUregPUF workshop (York, 2013) where participants from academia, authorities and industry met to establish an up to date understanding of plant uptake science. Lessons learned from the testing and continuous exchange with academia and authorities facilitated optimisation of the study design. The current version of the study design is considered appropriate to produce reliable data on plant uptake to be used as input for refined exposure modelling. An explicit guidance, however, on how to integrate the requested study design into the regulatory process is still lacking.

3

Work of a SETAC Group to Develop the Scientific Basis for Guidance for Regulatory Groundwater Monitoring of Crop Protection Products and their Metabolites in Europe

R.L. Jones, Bayer AG Crop Science Division / Environmental Safety; A. Gimsing, The Danish Environmental Protection Agency / Pesticides and Gentechnology; J. Agert, Bayer CropScience AG / Environmental Safety; N. Baran, BRGM; A. Boivin, ANSES; F. Ferrari, AEIFORIA; R. Gibson, Health and Safety Executive; L. Hammond, Health and Safety Executive / Environmental Fate; F. Hegler, Dr. Knoell Consult; W. Koenig, UBA Umweltbundesamt; J. Kreuger, Swedish University of Agricultural Science / Centre for Chemical Pesticides; T. Van der Linden, RIVM / ENVIRONMENTAL QUALITY; D. Liss, SGS Institut Fresenius GmbH / Agro; L. Loiseau, Syngenta; A. Massey, Health and Safety Executive; B. Miles, BASF SE / Crop Protection, Environmental Fate Modelling; L. Monrozies, SCE; A. Newcombe, ARCADIS US Inc; L. Padovani, European Food Safety Authority (EFSA); A. Poot, Ctgb; G.L. Reeves, Dow AgroSciences Ltd; S. Reichenberger, DR. KNOELL CONSULT GmbH; A.E. Rosenbom, Geological Survey of Denmark and Greenland / Geochemical; H. Staudenmaier, BASF SE / Crop Protection, Environmental Fate; R. Sur, Bayer AG - Crop Science Division / Environmental Safety; A. Schwen, AGES; M. Stemmer, Austrian Agency for Health and Food Safety / Institute for Plant Protection Products; W. Tüting, German Federal Office of Consumer Protection and Food Safety; U. Ulrich, University of Kiel

Groundwater monitoring is considered a higher tier assessment in the regulatory groundwater assessment of crop protection products in Europe, but little guidance has been provided to date on study designs. The SETAC EMAG-Pest GW group (a mixture of regulatory, academic, and industry scientists) in 2015 began developing the scientific basis for guidance for use by regulators and industry scientists. Rigid study designs are not appropriate since the study design needs to be tailored to the specific study objectives and should consider environmental conditions, the properties affecting environmental behaviour of the substance being studied, and site and use conditions. To illustrate how study design can vary, the group has proposed general study designs for seven hypothetical exposure assessment options, ranging from protecting all zones of saturation below the soil surface to only groundwater used to supply drinking water. Designs include recommendations on in-field and edge of field studies, as well as studies focused on catchments and aquifers. Examples of potential designs and recommendations on the use of publically available monitoring data have also been included. Also general recommendations on well installation and sampling procedures have been provided. Methodology has also been developed for assessing the relative vulnerability of agricultural regions and the vulnerability of specific sites for use in study design and site selection. The work of SETAC EMAG-Pest GW is still in progress but the goal is to finish by mid-2018.

4

Effect of the Freundlich exponent on the finite penetration depth in a homogeneous Freundlich-SFO leaching system

J. Boesten, Wageningen Environmental Research

All models used in the EU pesticide leaching assessment since 2000 (PELMO, PEARL, PRZM and MACRO) are based on a Freundlich isotherm combined with single first-order (SFO) degradation of the pesticide concentration in total soil. Thus, this is one of the cornerstones of the EU regulatory leaching assessment. This assessment is based on the FOCUS groundwater scenarios which use weather series of tens of years and include crop development and heterogeneous soil profiles. The sensitivity of the FOCUS leaching concentration (evaluated at 1 m depth) to the parameter describing the curvature of the Freundlich isotherm (i.e. the Freundlich exponent N) as derived from simulations with these models shows a sharp decline with decreasing Freundlich exponent with the concentration going down to a submolecular level. This is counterintuitive and difficult to understand. Explanations may be found by studying a simplified version of these sophisticated models, i.e. a assuming a homogeneous soil profile with pesticide properties that are constant with depth and assuming a constant water flow rate and a constant volume fraction of water (further called ‘simplified Freundlich-SFO system). Previously it was shown that a pulse of pesticide applied at the soil surface in this simplified system has a finite leaching depth beyond which no pesticide molecule will every pass. Simulations on the effect of N for a few FOCUS groundwater scenarios were compared to this effect on the percentage leached with this simplified model and qualitatively these effects were found to be similar. Next it was shown that this finite penetration depth after infinite time in the simplified Freundlich-SFO system increases slowly when N increases from 0.5 to about 0.85; however, when N approaches 1, this finite penetration depth goes to infinity. This was expected because this finite penetration depth does only occur in a system with a Freundlich isotherm and not in a system with a linear isotherm. It was checked by inspection of a concentration profile of one of the FOCUS groundwater scenarios that these scenarios also show a finite penetration depth for low N values at the end of the simulation period.

5

Bespoke monitoring to support Tier 4 FOCUS groundwater assessment

S.L. McManus, Syngenta; S. Payvandi, Syngenta Ltd; P. Sweeney, Syngenta; L. Fish, Sygnenta Crop Protection, LLC / Environmental Safety; R.J. Andrews, D. Schofield, Ramboll Environ; J. White, ARCADIS UK; N. Jones, Syngenta Ltd; G. Langridge, CEM Analytical Services Limited; T. Oteyza, Syngenta Crop Protection AG; M. Greener, Syngenta Ltd

Data generated from this bespoke groundwater monitoring programme will offer a solution to address the non-relevance case of pinoxaden metabolites from an exposure side. Median modelled mass flux was determined using GeoPEARL 3.3.3 simulations over 20 years which represent vulnerability to leaching across the EU27 under standard conditions. These data were aggregated to a $10km^2$ level and combined with a shallow groundwater dataset and a cereal land use dataset based on wheat in CAPRI. Those grid cells in the upper 50^{th} percentile for each spatial layer (mass flux, shallow groundwater, and wheat) were considered for the site selection process. Sites identified by modelling were assessed during site walkover surveys. To justify inclusion in the programme, sites had to have a history of pinoxaden use, groundwater less than 10m bgl, no confining layers, and no influential features which may act as preferential flow pathways. In 2015, 70 sites were installed across France, Germany, Italy, Lithuania and the United Kingdom. Each site consisted of three shallow wells installed around the field perimeter. The 70 sites represent all EU FOCUS groundwater scenarios except Jokioinen. The sites have all had a minimum of two pinoxaden applications before 2016 with groundwater levels an average of 2.9m below ground level. Sampling began in 2015 from 84 down hydraulic gradient wells. Of the 871 samples collected between June 2015 and July 2017 from these 70 vulnerable sites, the average for each site never exceeded 0.1µg/L. Only minor residues of metabolites have been detected since sampling began. Monitoring is to continue until Q4 2019 to ensure a thorough assessment of groundwater vulnerability is made. The modelling approach should allow extrapolation of the modelled vulnerability to be extended to member states outside of those where the wells were installed.

6

Long-Term Trend of Aquatic Pesticide Risk

A. Paulus, UFZ - Helmholtz Centre for Environmental Research / System-Ecotoxicology; S. Knillmann, K. Foit, Helmholtz Centre for Environmental Research UFZ / System-Ecotoxicology; M. Klein, Fraunhofer Institute for Molecular Biology and Applied Ecology IME; M. Liess, UFZ Center for Environmental Research / System-Ecotoxicology

European Union member states aim at reducing ecological risks exerted by pesticides. For this, reliable trend indicators of pesticide exposure and risk are inevitable. Based on this demand we designed a long-term trend indicator of aquatic pesticide risk for Germany. It uses pesticide sale statistics, toxicity data and chemical properties as input variables. The trend indicator was designed by combining the most reliable exposure and effect models. (i) We selected the most realistic exposure model by evaluating several established exposure models. For this we tested their performance with peak concentration data monitored in small agricultural streams in central Germany. These field data comprised of event-driven samples of 46 active substances from field-campaigns performed in 1998, 1999, 2000, and 2013. The highest agreement of measured and modeled peak concentration was yielded by the risk indicator EXPOSIT/EVA (R^2: 0.38), followed by the more complex models FOCUS STEP 2 (R^2: 0.36), SYNOPS-TREND (R^2: 0.24), and GERDA (R^2: 0.24). (ii) The translation from toxic pressure to pesticide risk was implemented by applying the field based and validated exposure – response relationship $SPEAR_{pesticides}$. Based on these information and models, we calculated the trend of toxic pressure and pesticide risk in Germany from 1996 to 2016 for the 500 substances authorized in this period. The method presented here requires only few input data, is based on validated models and can be adapted to regional conditions around the world.

Hydrophobic Chemicals and Mixtures: Reliable Investigations on their Environmental Fate and Effects (I)

7

The hydrophobicity delay: symptoms and solutions

A. Celsie, Queens University; D. Mackay, Trent University / Chemistry; D. Powell, DMER Ltd.; J. Parnis, Trent University / Cemistry

The objective of this presentation is to set out the conditions under which chemicals of high hydrophobicity experience significant delays in approaching equilibrium conditions. We suggest that this delay may be misinterpreted as being caused by a change in partitioning behaviour or mechanism resulting in development of non-linear regression models describing inter-media partitioning. In reality, the partitioning is fundamentally linear but is distorted by a kinetic delay. The rate constant format commonly applied to fish biouptake from water of concentration C_W is $?C_F/?t=k_1C_W-k_2C_F$ where C_F is the fish concentration, k_1 and k_2 are the uptake and loss rate constants and k_2 is k_1/BCF where BCF is the bioconcentration factor. The characteristic time for uptake and loss τ is $L?K_{OW}/k_1$. Slower uptake and loss will occur if the partition ratio K_{OW} is large, and the fish must contact $K_{OW}\cdot L$ times its own volume to approach equilibrium. Very hydrophobic substances will experience long time delays when approaching equilibrium and correspondingly long times for loss during a depuration phase. Mackay et al. [1] modeled bioconcentration and toxicity of superhydrophobic chemicals D4, D5, and D6 using a biouptake model for fish. Due to the very high hydrophobicity (log $K_{OW}\approx 10^8$ for D5) and very low water solubilities C_W must be very low, which results in a very long equilibration time. Uptake time to equilibrium for D5 was estimated to be ~2000 days, to get C_F=2 mol/m^3 about 17 days. The study concluded that for superhydrophobic substances organisms will likely not reach toxic concentrations within the test duration which is usually 4-94h. Doucette et al. [3] reviewed foliage/air partitioning data in which the onset of a hydrophobic delay (HD) is apparent when log $K_{OA}\approx 9$. McLachlan [4] developed a model for uptake of hydrophobic chemicals by foliage. This model shows levelling off corresponding to insufficient time to achieve equilibrium. A kinetically limited regime is reached at a $K_{OA}\approx 10^9$. In our presentation we will address the HD issue that we believe is a widespread phenomenon applicable to numerous environmental systems including passive sampling and partitioning to aerosol particles. Finally, we suggest a general method for identifying the HD problem. [1] Environ Sci Technol 2015, 49(19): 11913-22. [2] Environ Toxicol Chem 2012, 31(8):1911-9. [3] Environ Toxicol Chem. 2017, Accepted. [4] Environ Sci Technol 1999 33:1799-1804.

8

Partitioning of chlorinated paraffins (CPs) to organic matter is not class specific: implications for bioaccumulation?

M. Castro, Stockholm University / ACES; M. Breitholtz, B. Yuan, Stockholm University / Department of Environmental Science and Analytical Chemistry (ACES); I. Athanassiadis, Stockholm University; L. Asplund, A. Sobek, Stockholm University / ACES

Chlorinated paraffins (CPs) belong to a group of industrial chemicals consisting of n-alkanes (from 10 to 30 carbon chain atoms) with chlorine content from 30 to 70% weight. They are widely used as high-pressure lubricants, flame retardants, and additives in plastic, rubber, and sealants, leading to high-production volumes worldwide. These chemicals are also ubiquitously found in the environment. The use of short chain chlorinated paraffins (SCCPs) in Europe has been restricted, however, medium (MCCPs) and long chain (LCCPs) chlorinated paraffins are used in Europe as substitutes for SCCPs. In some countries, all classes are still in use, leading to high production volumes (over a million tons per year globally). There is a lack of data on CP physicochemical and hazard-based properties, which is due to their inherent high complexity. CPs are hydrophobic contaminants, which complicates their aquatic toxicity testing. In this work, we validate the use of passive dosing for the study of chlorinated paraffins and demonstrate the partitioning behavior of CP technical mixtures between silicone, water and organic carbon. We used 5 different technical mixtures from three the established categories (2 SCCPs, 1 MCCP, 1 LCCP). We added *Daphnia magna* to the passive dosing system, to understand the partitioning behavior of CP technical mixtures from CP-dosed water medium to CP-free organic matter ($K_{oc\text{-}water)}$). Immobilization of *D. magna* was observed after 48 hours under different exposure concentrations. APCI-QTOF-MS was used for CP quantification. Both silicone-water and organic carbon-water partition coefficients overlap between different categories of CP technical mixtures. CP-52, labelled as a MCCP, had a similar silicone-water partitioning coefficient as a restricted SCCP – Huels 70C. We demonstrate that increasing average chlorine content of each CP mixture significantly increases the Log $K_{silicone\text{-}water}$ and Log $K_{oc\text{-}water}$. These results could have implications on the study of environmental fate of CPs: in-use CPs (MCCPs and LCCPs) might be equally or more bioaccumulative as restricted SCCPs. $K_{oc\text{-}water}$ is particularly helpful at predicting bioaccumulation of chemicals into biota. The next step is to quantify the bioaccumulation potential of CPs. With the use of the passive dosing approach, we are producing laboratory experimental data that can be used to help in the on-going regulatory discussion on MCCPs and aid their risk assessment.

9

Trophic magnification of cyclic volatile siloxane materials (D4, D5, and D6) in a freshwater lake: A Monte-Carlo analysis

K.B. Woodburn, The Dow Chemical Company / HES; R.M. Seston, Hyla Environmental Consulting, LLC / Toxicology, Environmental Research & Consulting; J. Kim, D.E. Powell, The Dow Chemical Company / Toxicology, Environmental Research & Consulting

The trophic transfer of cyclic methylsiloxane (cVMS) materials in aquatic ecosystems is an important criterion for assessing bioaccumulation and ecological risk of these compounds. Food web magnification of the cVMS materials, specifically octamethylcyclotetrasiloxane (D4), decamethylcyclopentasiloxane (D5), and dodecamethylcyclohexasiloxane (D6) was determined for the Lake Pepin, Minnesota (USA) food web. The objective of this work was to determine if cVMS materials are biomagnified in this freshwater ecosystem. To determine whether the benthic influence in the Lake Pepin aquatic food web affected the trophic magnification factor (TMF) values for the cVMS compounds, a companion study was conducted to determine the biomagnification and TMF value of a reference material, 2,2',3',4,4'5,5'-hepatachlorobiphenyl (PCB-180), in Lake Pepin. TMFs for the three cVMS materials and PCB-180 were determined using standard

methods involving feeding guild, trophic guild classifications, and the stable isotopes of nitrogen ($d^{15}N$) and carbon ($d^{13}C$) to estimate trophic position/carbon flow. The aquatic food web consisting of two benthic macroinvertebrate species and 15 fish species was evaluated for trophic magnification of cVMS materials and PCB-180. Lipid-normalized concentrations of D4, D5, and D6 were greatest in the lowest trophic levels and significantly decreased going up the food web, with the lowest concentrations being observed in the highest trophic levels. The TMFs measured for the three cVMS materials were all99% of the uncertainty for cVMS TMF values in Lake Pepin was explained by uncertainty at the base of the food web (89%) and at the top of the food web (11%). By comparison, PCB-180 had a TMF of 2.2 in the evaluated food web, indicating biomagnification. TMFs for the cVMS chemicals and PCB-180 were determined using a Monte-Carlo probability analysis technique, and the likelihood that the values exceed unity was less than 0.5% for all three cVMS compounds and >99.5% for PCB-180. This evaluation indicates that D4, D5, and D6 do not biomagnify in the benthic-dominated Lake Pepin aquatic ecosystem, a food web which does demonstrate biomagnification of the legacy contaminant, PCB-180.

10

Distribution and Bioaccumulation of Polyhalogenated Carbazoles in Aquatic Systems from the United States and China

D. Chen, Jinan University / Cooperative Wildlife Research Laboratory and Department of Zoology; Y. Wu, Southern Illinois University Carbondale / Cooperative Wildlife Research Laboratory and Department of Zoology; R. Sutton, San Francisco Estuary Institute; K. Xu, Louisiana State University / Department of Oceanography and Coastal Sciences

The present study reports the discovery of a suite of polyhalogenated carbazoles (PHCZs) in aquatic sediments collected from four watersheds located in the United States and China, including the Gulf of Mexico (USA), San Francisco Bay (USA), Lake Tai (China), and Lake Dianshan (China), and their bioaccumulation in the San Francisco Bay ecosystem. A total of 11 halogenated carbazoles, including 3-chloro, 3,6-dichloro-, 1,3,6,8-tetrachloro-, 2,3,6,7-tetrachloro-, 3-bromo-, 2,7-dibromo-, 3,6-dibromo-, 1,3,6-tribromo-, 1,3,6,8-tetrabromo-, 1-bromo-3,6-dichloro-, and 1,8-dibromo-3,6-dichloro-carbazole were screened. Halogenated carbazoles were detected in 98.7% of the sediment samples, with concentrations ranging from below method limits of quantification to 51.5 ng/g dry weight. In most of these sediment samples, PHCZ concentrations exceeded those of polybrominated flame retardants (PBDEs). The latter group of chemicals has been demonstrated to be persistent and globally distributed. PHCZs were also detected in various organisms from the San Francisco Bay, including bivalves, sport fish, harbor seal blubber and bird eggs. The median concentrations of PHCZs by species ranged from 33.7 to 164 ng/g lipid weight. Biomagnification was also observed from fish to harbor seal and was mainly driven by chlorinated carbazoles, particularly 36-CCZ. Congener compositions differed among species, suggesting that individual congeners may be subject to different bioaccumulation or metabolism in species occupying various trophic levels in the studies system. Toxic equivalent (TEQ) values of PHCZs were determined on the basis of their relative effect potencies (REP) compared to 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD). The median TEQ ranged from 4.8 to 19.5 pg TEQ/g lipid weight in biological tissues. Our data demonstrated the broad exposure of PHCZs in the studies systems and potentially in global aquatic systems. These findings raise the need of additional research to better elucidate their sources, environmental behavior, and fate in global environments.

11

Bioconcentration factors of constituents of essential oils in fish determined in an in-vivo benchmarked dietary exposure study: A case study for pine oil

C.L. Chen, Stockholm University / Department of Environmental Science and Analytical Chemistry; M. McLachlan, Stockholm University / Environmental Science and Analytical Chemistry (ACES); M. MacLeod, ITM - Stockholm University / Department of Environmental Science and Analytical Chemistry

Essential oils are fragrance materials that are registered as natural complex substances (NCS) under the European REACH legislation. One of the categories of information required in a REACH registration is information about the potential for bioaccumulation of NCS by fish. Determining the bioconcentration factor (BCF) of essential oils cannot be readily accomplished using a standard flow-through uptake/depuration experiment. Previously, we demonstrated that a single dietary exposure coupled to the benchmarking technique could be applied to an artificial mixture for measuring the in vivo BCF. Here, we report an application of our proposed BCF-determination methodology on a real essential oil – pine oil. Fish (rainbow trout) were dosed with a mixture of the pine oil and a suite of benchmark chemicals via a single dietary exposure. The depuration rate constants (k_T) in the fish soma (without GIT) for the key pine oil constituents are 0.134 d^{-1} (β-Caryophyllene, BCP) – 1.41 d^{-1} (BAc) and they were 0.0.0799 d^{-1} (HCB) – 0.517 d^{-1} (DiCB) for the reference chemicals. The test compounds depurated faster from the soma than the GIT, making estimated whole-body depuration slower (conservative) compared to the soma only. HCB was the chemical most resistant to depuration among all the test compounds. Benchmarking to HCB reduced the standard error of measured $k_{T\text{-}BM}$ from the soma for most of the chemicals, with $k_{T\text{-}BM}$ ranging from 0.001 d^{-1} (PCB52) to 2.98 d^{-1} (BAc). The apparent BCF (BCF_A) values in soma for the key components in pine oil and the reference chemicals were in the range of 98.2 L kg^{-1} (BAc) – 1030 L kg^{-1} (BCP) -.. and 267 L kg^{-1} (DiCB) – 1730 L kg^{-1} (HCB), respectively; while for the benchmarked BCF (BCF_{BM}) in soma, they are 46.3 L kg^{-1} (BAc) - 2570 L kg^{-1} (BCP), and 208 L kg^{-1} (DiCB) - 197000 L kg^{-1} (PCB52) respectively. We conclude that a single dietary exposure coupled with the benchmarking technique is a feasible experimental approach for measuring the BCF of NCS in fish.

12

ROLE OF ADIPOSE TISSUE RESPONSIBLE FOR ECHOLOCATION IN THE BIOACCUMULATION PROCESS OF LIPOPHILIC COMPOUNDS IN HARBOUR PORPOISES

I. Schaap, Utrecht University; L. Gross, The University of Queensland / School of Earth and Environmental Sciences; A. Covaci, University of Antwerp, Toxicological Center / Toxicological Centre Dep of Pharmaceutical Sciences; T. Bouveroux, OCEAMM; F. Demaret, University of La Rochelle / Observatoire Pelagis; R. Blust, University of Antwerp / Department of Biology (SPHERE Research Group); M. van den Berg, Utrecht University / Institute for Risk Assessment Sciences; L. Weijs, Griffith University / Australian Rivers Institute

Previous studies have suggested that persistent organic pollutants (POP) can lead to adverse effects in marine mammals, including harbour porpoises (*phocoena phocoena*), thereby causing illnesses. Traditionally, blubber is an ideal matrix to assess POP bioaccumulation in marine mammals. However, during times of energy deficits, blubber tissue is broken down in which POPs are redistributed in the body. Echolocating tissues melon and mandibular fat are inert lipid bodies in odontocetes and, in contrast with blubber, are less prone to release POPs, which makes them ultimate sinks for POP lifetime bioaccumulation. This study aimed to assess the lifetime bioaccumulation of POPs in harbour porpoises through 1) analysis of POPs in various tissues and/or organs of harbour porpoises, including lipid rich bodies as blubber, melon and mandibular fat, and 2) Physiologically based toxico-kinetic (PBTK) modelling of PCB 153 and PBDE 153 to compare bioaccumulation of lipophilic compounds in lipid-rich tissues with different lipid composition and purpose (echolocation versus insulation) over the whole lifespan of male harbour porpoises. Overall, POP analysis and PCB 153 modelling for male harbour porpoises reveal that despite differences in lipid composition and lipid types, lipophilic pollutants bioaccumulate patterns are similar in blubber, mandibular fat and melon with increasing age. Nevertheless, the model showed the highest levels of PCB 153 in mandibular fat, followed by melon and blubber. From these results, mandibular fat can be considered as a sink for PCB 153 and a better proxy for lifetime exposure than blubber, which can be both a sink and source of lipophilic pollutants. PBDE 153 PBTK modelling reveals that bioaccumulation differs in lipid composition and lipid type, whereby bioaccumulation predominantly occurs in echolocating tissue during juvenile stage and in blubber during adulthood.

Keywords: Echolocation, life time bioaccumulation, biomonitoring, PBTK modelling, POP

Interpretation and uncertainty - overcoming challenges of translating LCA results into reliable information

13

LCA: everything is relative and nothing is certain

J. Guinee, University of Leiden / Institute of Environmental Sciences; R. Heijungs, Leiden University / Faculty of Economics and Business Administration Department of Econometrics and Operations Research; A. Mendoza Beltran, Leiden University; P. Henriksson, Stockholm University / Stockholm Resilience Centre; E. Groen, Areto Consulting GmbH / Animal production systems group

Environmental Life Cycle Assessment (LCA) relies on data, models and knowledge from almost all environmental scientific disciplines, including related uncertainties. In addition, LCA involves making methodological choices. Over the past five years, we have published several approaches to deal with data and methodological choice uncertainties. One often heard critique is that these proposals do not account yet for ‘correlations’. We distinguish between two meanings of the term ‘correlations: correlated sampling: when applying Monte Carlo sampling for propagating uncertainty data for a comparative LCA study, the sampling can be either dependent (correlated) or independent (uncorrelated). Independent sampling implies that data of shared processes between the product alternatives compared, are sampled in different MC procedures resulting in different data sets for this shared process for both product alternatives. Dependent sampling implies that process data for the product alternatives compared are sampled based upon one and the same random drawing of parameter values resulting in identical data sets for this shared process; correlated data points: a transport process input of diesel is, for example, related to an process output of CO_2 (emission); if the process consumes more diesel for the same amount of transport, the CO2 emission will also increase. The first interpretation of ‘correlation’ has been addressed in earlier work by Henriksson et al. and recently again by Mendoza et al. The second interpretation of ‘correlation’ (between data points) has recently been addressed by Groen et al. We present an overall framework integrating the different approaches comprehensively dealing with uncertainties in LCA studies. In addition, we show the possibilities and limitations of also including data correlations into LCA uncertainty assessments. Practical application of this

framework in the daily practice of LCA practitioners needs further work, including implementation in LCA software programs and particularly data.

14

Drivers of variability and uncertainty in the chemical footprint of personal care products

M. Douziech, Radboud University Nijmegen; R. Oldenkamp, Radboud University Nijmegen / Department of Environmental Science; H. King, Unilever; R. University, J.A. Hendriks, Radboud University Nijmegen / Department of Environmental Science; A. Ficheux, A. Roudot, Universite de Bretagne Occidentale; R. Van Zelm, Radboud University / Department of Environmental Science

Chemical footprinting of products, quantifying the potential environmental impact of the product's chemicals, could be used to inform consumers choice. However, the use of chemical footprints (ChFs) for comparative purposes requires a full understanding of the uncertainty and variability sources influencing its quantification. The goal of this work was to determine the ChFs for personal care products and quantify the variability and uncertainty in the different parameters used to derive these individual ChFs. In a first phase, we focused on shampoos. The environmental impact of each ingredient was derived from an environmental load, assuming 100% discharge to the drain, determined by the ingredient's removal in activated sludge wastewater treatment plants (WWTPs) simulated using SimpleTreat, and a characterisation factor estimated with USEtox. The physico-chemical and ecotoxicological properties applied in both models were all estimated. Their reliability was derived from the prediction accuracy of the estimation models used (EPISuite, ACD Labs, ECOSAR). A Monte Carlo analysis with 1000 iterations was then performed, combining the uncertainty and variability of the different parameters, to determine the spread in ChFs. The ChFs derived by this approach spanned nearly 4 orders of magnitude (95% Confidence Interval (CI)). The wide span of the ChF's 95% CI was primarily attributable to fragrances (61%), surfactants (20%), and the amount of product used (16%). The significant contribution from fragrances and surfactants can largely be explained by the uncertainty in their environmental impacts described by the characterisation factors derived with USEtox and more precisely the estimated ecotoxicity values. These preliminary results question the use of absolute values when communicating product's chemical footprints. As long as more reliable ecotoxicological assessments are not available, identifying relative contributions to the overall environmental impacts might be more useful to target specific actions.

15

Combined uncertainty and scenario analysis within Life Cycle Assessment of waste management systems

V. Bisinella, DTU (Technical University of Denmark) / DTU Environment; K. Conradsen, DTU Technical University of Denmark / DTU Compute; T.H. Christensen, DTU Technical University of Denmark / DTU Environment; T. Astrup, Technical University of Denmark / Department of Environmental Engineering

Life Cycle Assessment (LCA) is being increasingly used for decision support in the waste management field. LCAs are subject to uncertainty regarding both the input values for the LCA model (or parametrical uncertainty) and its modelling choices (or epistemic uncertainty). Parametrical uncertainty can be systematically addressed with parametrical uncertainty analysis, while epistemic uncertainty can be addressed with scenario analysis. However, the communicability and usefulness of parametrical uncertainty and scenario analyses can be hindered by the fact that such analyses are carried out separately, dividing the interpretation of the results in two separate channels and potentially limiting the value of LCA as a decision support tool. This study presents a novel method that aims at combining uncertainty and scenario analysis, illustrated on a case study on three hypothetical waste management options for treatment of residual household waste in the municipality of Copenhagen in 2025. The waste management solutions were provided with uncertainty for the model input values (parametrical uncertainty) and were assessed within four different hypothetical background conditions (scenario analysis). Within each impact category, the results of the parametrical uncertainty analysis were used to identify the most *robust* waste management option, *i.e.* the waste management option obtaining the highest average probability measure of providing the best environmental performance across the considered background scenarios. The method allowed obtaining various levels of analyses for the interpretation of the LCIA results: parametrical uncertainty analysis for each of the assessed waste management options, with identification of the parameters mostly contributing to the uncertainty around the results, within each of the assessed scenarios for the background conditions. The parametrical uncertainty analyses, which were necessary for carrying out discernibility analyses across background conditions, allowed obtaining useful insights on the changes in sensitive parameters induced by the change in background conditions. The discernibility analysis results allowed obtaining a clear quantification of the probability measure of each waste management option to provide a better environmental performance than another, for each of the assessed impact categories and investigated background conditions, and in a manner simply conveyable to the users and final receivers of the LCA.

16

Which impact categories are relevant for LCA results interpretation?

A. Esnouf, Institut National de la Recherche Agronomique, Narbonne-France / UR Laboratoire de Biotechnologie de lEnvironnement; E. Latrille, J. Steyer, INRA LBE; A. Hélias, Montpellier SupAgro / LBE ELSA

LCA is intrinsically a multicriteria approach comparing (almost) all the potential environmental impacts of human activities. However, multicriteria decisions pose challenges as a wide range of environmental impacts results may lead to unclear conclusions. Based on their relevance, a choice among the impact categories may be necessary. It can be carried out by examining how the information given in the Life Cycle Inventories (LCIs) is used by the impact categories. The Representativeness Index (RI) proposed by Esnouf et al. was initially used to compare the adequacy of Life Cycle Impact Assessment (LCIA) methods regarding LCIs. Here, the RI is used to explore the impact categories belonging to a given LCIA method. Thus, the present study focuses on how the interpretation of the LCA results can be undertaken given the choice of relevant impact categories. With a geometrical standpoint, LCIs of the ecoinvent database and impact categories of the ILCD method are standardized and localized within the same R^n vector space. This vector space is generated by all the dimensions (i.e. elementary flows) from which the LCIs of the database are described. The RI is a proximity measurement between standardized LCI vectors and standardized impact category vectors, corresponding to the cosine of the angle between two vectors. This measurement does not assess the relevance of the environmental model behind impact categories, but rather translates the main elementary flows from an LCI based on how they are represented by the impact categories of an LCIA method. Two inventories referred to the production of 1 kWh of high voltage electricity mix are used in this study as an illustrative example (areas analysed: NPCC, North-eastern North America, U.S. only, and Germany). Results show that comparing the NPCC and the German electricity mixes is more relevant based on the ionising radiation impact categories. The freshwater eutrophication, the climate change and the ozone depletion are the three other impact categories that focus on the main environmental issues that best represent those two LCIs in regard of the whole database. This analysis provides additional information for characterizing the impact categories towards LCI representativeness within the global context of a given database. While performing a LCA study, practitioners could benefit of the developed methodology to select impact categories to focus the results interpretation on relevant environmental issues.

17

Reduce the uncertainty of LCA results by prioritizing the regionalization effort: a sectorial meta-analysis

L. Patouillard, CIRAIG - École Polytechnique de Montréal; P. Collet, IFP Energies nouvelles; P. Lesage, CIRAIG / CIRAIG; P. Tirado Seco, CIRAIG; C. Bulle, CIRAIG - ESG - UQAM / Strategy & corporate social responsibility; M. Margni, CIRAIG - École Polytechnique de Montréal / Mathematical and Industrial engineering

Uncertainty in Life Cycle Assessment (LCA) can limit the results interpretation. Regionalization is one of the ways to reduce the uncertainty due to spatial variability. Life Cycle Inventory (LCI) regionalization deals with increasing the geographic representativeness modelled in LCI. Life Cycle Impact Assessment (LCIA) regionalization deals with regionalized impact characterization that accounts for the spatial variability of the receiving environment. Regionalized characterization factors (CF) apply to spatialized elementary flows (EF), called LCI spatialization. However, integrating regionalization requires additional effort on data collection and treatment for LCA practitioners and database developers. Thus, prioritizing the regionalization effort on the most sensitive data (input data with uncertainty having the highest influence on the resulting uncertainty) would ensure an optimal use of resources to reduce LCA results uncertainty. This research work proposes a procedure to prioritize regionalization efforts based on global sensitivity analysis (GSA) to reduce the spatial uncertainty of LCA results. We applied this procedure to all the activities of two economic sectors (biofuel production and passenger land transport) defined in the ecoinvent database v3. The regionalized impact methodology IMPACT World+ is used to assess environmental impacts. Statistical tests are then used to derive sectorial recommendations regarding the impact categories (IC) and LCA phases (LCI or LCIA) that should be regionalized in priority. Those recommendations are meant to help LCA practitioners and LCI database developers to define their strategy for regional data collection to lower the LCA results uncertainty. Results show that contrasting IC ranking depending on the economic sector. For the biofuel production sector, land transformation encompasses almost all the uncertainty, whereas it is distributed among several impacts (global warming and marine acidification) on the land passenger transport sector. For LCA phases ranking, it confirms that inventory should be spatialized in priority for regionalized impact categories. This methodology allows providing different recommendations specific a sector to refine data collection in order to reduce uncertainty and enhance results interpretation. To our knowledge, this is the first time that an uncertainty analysis discriminating IC and LCA phase ranking is performed.

18

Poster spotlight: MO387, MO388, MO389

Environmental fate of emerging contaminants in the water cycle: analytical challenges and engineered solutions (I)

19

Unravelling longitudinal pollution patterns in freshwaters by non-target screening and cluster analysis

L. Beckers, Helmholtz-Zentrum für Umweltforschung GmbH - UFZ / Effect-Directed Analysis; W. Brack, E. Müller, T. Schulze, M. Krauss, Helmholtz centre for environmental research - UFZ / Effect-Directed Analysis

Pollution of aquatic ecosystems with *emerging organic contaminants (EOCs)* has been intensively studied over the past decades. The vast number of *EOCs* and their occurrence in complex and variable mixtures is a major challenge for monitoring, risk assessment and management and is beyond the scope of target screening. Thus, novel approaches are needed to characterize these mixtures and identify unknown *EOCs* including transformation products and natural background. In this study, we propose a novel workflow for unravelling pollution patterns along a river course identifying longitudinal dynamics of pollutant groups, entry pathways and the fate of *EOCs* along the river course using *non-target screening* by *LC-HRMS* and *cluster analysis*. Sixteen grab samples were taken along the 42 km- long course of the Holtemme River (Saxony-Anhalt, Germany), whereas the first sampling in the national park marked a reference point for pristine conditions. Chemical screening was performed on an UltiMate 3000 LC system (Thermo Scientific) coupled to a hybrid quadrupole - Orbitrap MS (QExactive^{TM} Plus, Thermo Scientific) with a heated electrospray ionization source. MS/MS analysis was performed in a full scan experiment (100-1000 m/z) at a nominal resolving power of 140,000 at *m/z* 200. Peak extraction including peak picking, gap filling, componentization and target annotation was implemented in R. *Cluster analysis* was performed using the R package 'kmL'. Four clusters were suggested for the data set representing A: *EOCs* from treated wastewater input of the two wastewater treatment plants (WWTP), B: *EOCs* specific for first WWTP due to specific local emissions, C: *EOCs* from diffuse (i.e., agricultural and urban surface run-off) and small point-source input (e.g., rain sewers and creeks) and D: low continous background signals. The identified patterns gave insights into the spatial dynamics of complex chemical mixtures along a river course, highlighting differences in point-sources and areas governed by diffuse input and identifying points of complex mixtures of mixtures (e.g., first WWTP). Deeper investigation including structure elucidation will resolve the origin of non-target signals in these clusters. The proposed workflow proved to be a fast method for unravelling pollution patterns in non-target HRMS data and may also applied to study other longtidunal data such as temporal dynamics in pollution at hotspots and comparision of treatment and transformation processes.

20

Tracing sewage-derived contaminants from mainland towards the ocean by high resolution mass spectrometry

P. Lara-Martin, University of Cadiz / Physical Chemistry; A. Chiaia-Hernandez, Eawag - Swiss federal Institute of Aquatic Science and Technology / Environmental Chemistry; M. Biel, R. Baena-Nogueras, University of Cadiz / Department of Physical Chemistry; J. Hollender, Eawag / Environmental Chemistry

The ocean is the ultimate sink of most of the organic synthetic compounds produced and consumed by humans. Among the different pollution sources affecting this environment, discharge of treated and untreated sewage from mainland is of high relevance due to its continuous input, high volume and poor efficiency of conventional wastewater treatment plants (WWTP) to remove many potentially harmful substances. Even after dilution, some of these contaminants may still be detected at low concentrations (ppt-ppb level), especially in coastal waters, and their effects over marine biota are still widely unknown. This work focused on identifying a wide range of polar and semipolar chemicals that can be detected in both WWTP influents and effluents, as well as in the receiving waters (rivers and estuaries) and even in the open ocean. In order to do this, we carried out several monitoring campaigns in the Gulf of Cadiz (Atlantic Ocean, SW Spain), sampling wastewater from one of the biggest local WWTPs in the area (Jerez de la Frontera, 250 000 inhabitants), adjacent surface river and coastal waters, and oceanic waters at different depths (down to 400 m) taken up to 50 km away from the coastline. Solid phase extraction followed by liquid-chromatography high resolution mass spectrometry were used in combination with statistical tools (e.g., principal component and cluster analyses), specific vendor and open-access software, and online library searches (mzCloud) to tentatively identify more than 300 sewage features persistent enough to be also detected in oceanic waters. These compounds included different classes of surfactants (e.g., linear alkylbenzene sulfonates) and their byproducts (e.g., DATS) and metabolites (e.g., NPEC), polymers (PEG, PPG and many ethoxylated derivatives), pharmaceuticals (e.g., valsartan, diclofenac, carbamazepine, etc.), personal care products (UV stabilizers) and food additives (e.g., sucralose), some of them (e.g., sulfurol) identified in the environment for the first time. The list of compounds reflected here not only shows many of the substances that can potentially escape from wastewater treatment but also constitutes a first step towards a more detailed characterization of the chemical exposome in the marine environment.

21

Pharmaceuticals, personal care products (PPCPs), and artificial sweeteners (ASWs) in river and groundwater from the Ganges River Basin, India

B.M. Sharma, Research Centre for Toxic Compounds in the Environment Faculty of Science Masaryk University / Faculty of Sciene; J. Bečanová, Graduate School of Oceanography, University of Rhode Island, Narragansett, Rhode Island 02882 / Chemical Oceanography; M. Scheringer, ETH Zurich / Institute for Chemical and Bioengineering; L. Nizzetto, NIVA

Pharmaceuticals and personal care products (PPCPs) and artificial sweeteners (ASWs) are environmental contaminants of emerging concern. In this study, we investigated the occurrence and distribution of 15 pharmaceuticals and personal care products (PPCPs) and five artificial sweeteners (ASWs) in surface and groundwater of the Ganges River Basin in India. The Ganges River Basin is the largest river basin in India and home of about 7% of the total global population. PPCPs and ASWs were ubiquitously present in the river and groundwater. Most frequently detected compounds were caffeine, DEET, ketoprofen, cyclamate and sucralose. Except caffeine and DEET, concentrations of other PPCPs and ASWs in river water were found to be higher in densely populated areas. Concentrations of PPCPs and ASWs in the groundwater were lower but on same order as detected in the river water. Similar to river water, elevated concentrations of PPCPs and ASWs in groundwater were detected in middle and lower reaches along the Ganges River. PPCPs and ASWs concentrations were lower than those in developed countries, still, their instantaneous loads in the Ganges River were comparable to those in rivers from developed countries. The presence of PPCPs and ASWs in the surface and groundwater can be interpreted as a consequence of inefficient wastewater management in the basin, which pose a concern for human exposure.

22

Data-dependent fragment ion search for detection of sartans and related compounds in wastewater and surface water

B. Zonja, IDAEA-CSIC / Environmental Chemistry; M. López de Alda, Institute of Environmental Assessment and Water Research IDAEA-CSIC / Department of Environmental Chemistry; D. Barcelo, IIQAB-CSIC / Department of Environmental Chemistry

Presence of polar contaminants like angiotensin II receptor antagonist pharmaceuticals (sartans) in the aquatic system is directly linked to human impact. Like other xenobiotics, they can be metabolised in the body with enzymes such as cytochrome P450 (CYP), UDP-glucuronosyltransferase (UGT), and glutathione S-transferases (GST) which are present in the human liver at high abundance. Due to biological and/or abiotic processes that the contaminants undergo from the discharge site to the ground or surface water where they are detected, they can be transformed to transformation products (TPs). These TPs are usually detected and identified first at lab-scale in order to evaluate the degradability of a compound. This is typically followed by a targeted method development and it is not up until the compounds have been identified (and in some cases isolated) they are actually searched for in real aquatic samples to report their presence. Here, we propose an alternative approach, based on data-dependent fragment ion search, where real-world samples are initially screened for plausible TPs, metabolites or related compounds. The starting point here was a suspect screening of a list of all marketed sartans in wastewater effluent and surface water samples, which were extracted with a generic solid-phase extraction method using four cartridges with different chemistries. Out of the compounds detected, five of them had an identical core structure, and it was postulated that this sub-structure would fragment identically in all compounds. Following a series of experiments with different MS parameters modified, a list of hit compounds was obtained using fragment ion search. After all of the compounds investigated, available human metabolites and internal standards were purchased, a set of biodegradation experiments using activated sludge was performed in order to "source" the detected m/z and compare the possible TPs fragmentation to the one obtained in the bio-reactors. In parallel, a literature search for reported human metabolites was used to complement the identification of compounds detected in cases where no such compound was found to bio-form from the parent compound. Finally, a targeted method was developed for the quantification of these compounds in wastewater and surface water. This work was possible due to support from EU-FP7 programme (Solutions project), Merck (LC columns) and Biotage (SPE cartridges).

23

HR-MS non-target analysis for transformation products of emerging organic contaminants in waste water fractions pre-screened by ELISA

R.J. Schneider, H. Hoffmann, BAM Federal Institute Materials Research and Testing / Department of Analytical Chemistry Reference Materials

High-Resolution Mass Spectrometry has its benefits but still wastewater samples challenge the analyst on the quest for "unknowns", metabolites and transformation products of emerging organic contaminants (EOCs). Their detection requires non-target analysis which involves not only costly instrumentation but also scientists with the time to plough through the enormous amount of data collected. An approach is presented using antibodies as selectors to pre-screen fractions of an

HPLC run for "binding" in order to detect hitherto unknown but structurally related compounds. Carbamazepine (CBZ), an anti-convulsant and anti-depressant, sulfamethoxazole (SMX), an antimicrobial for humans, and estrone (E1), a hormone and estradiol metabolite have been studied by this LC-ELISA approach. Immunoassays had been developed for all compounds but overestimations of wastewater concentrations were frequent, with CBZ even at a constant level (+ 30 %) that did not result from the considerable cross-reactivity to CBZ-10,11-epoxide (ca. 70 %) or 2-hydroxy-CBZ (14 %). Fractions from HPLC runs of pre-concentrated wastewater samples were collected into a 96-well glass plate in small aliquots, with fractions adapted to the desired resolution along the run. One plate is sufficient to collect a 30 – 40 min. run. The fractions were evaporated to dryness under a gentle stream of nitrogen. Before ELISA analysis, all fractions were reconstituted in a few hundred µL of water. For CBZ, UPLC-QTOF-ESI-MS applied on a specifically "positive" fraction revealed an exact mass of m/z = 389.168 and a chlorine pattern. The compound is cetirizine, an antihistaminic. It appeared in our samples from spring on and was responsible for 20 % of the overestimation we found initially with the CBZ antibody. The LC-ELISA for SMX displayed a series of unidentified peaks in the ELISAgram. Careful analysis of the fractions led to the identification of N4-acetylsulfamethoxazole, an SMX metabolite which is present in the samples. With estrone, interferences by polar matrix compounds eluting early could be identified.

24

Designing a risk based monitoring program for groundwater sources for drinking water production – based on target and suspect screening combined with clustering techniques

R. Sjerps, KWR Watercycle Research Institute / Chemical Water Quality and Health; A. Brunner, B. Bajema, Vitens; P. Bauerlein, KWR / Analytical and Environmental Chemistry; M. de Jonge, Vitens; Y. Fujita, M. Schriks, KWR Watercycle Research Institute; A. van Wezel, KWR Watercycle Research Institute / Chemical Water Quality and Health

Drinking water utilities heavily invest in monitoring occurrence of chemicals in drinking water sources and produced drinking water. Worldwide, drinking water regulation prescribes drinking water limits for a limited number of chemicals, the EU Drinking Water Directive (EU DWD) for example lists drinking water limits for 26 organic and anorganic chemical parameters. However, most drinking water utilities monitor a broad set of parent chemicals and their transformation products, using both target, non-target and bioanalytical methods. The EU DWD stimulates that drinking water monitoring is performed in a more flexible way, provided protection of public health is ensured. Compared to surface water, groundwater is less intensively studied and monitored. However, groundwater can be highly influenced, by anthropogenic activities related to the land-use above the groundwater, by infiltrating surface water, by historical contamination as well as by activities in the sub-soil. The susceptibility of the groundwater aquifers to these pressures depends on soil type and groundwater hydrology. Chemical properties such as persistence and mobility and their retardation during groundwater flow are reflected in the spatio-temporal patterns of the chemicals. Treatment technology applied, such as filtration and sorption techniques, determines removal efficiencies during drinking water production for specific compounds. Water utility Vitens services drinking water in a large area in the Netherlands, mostly using groundwater as a source. Their set of chemical parameters in the monitoring program tripled in the last decade. The water utility aims to prioritize their measured chemicals and develop a tailored risk-based monitoring program. We present here is the development of a risk-based monitoring program for all 113 supply zones involved, mostly consisting of groundwater. We use both target and non-target/suspect monitoring data and well characteristics. We use clustering techniques combined with prioritization techniques including substance properties and *in vivo* as well as *in vitro* toxicity information. We analyse full scale removal efficiencies by the treatment technologies applied. Finally we propose a risk based monitoring program.

Wildlife ecotoxicology: laboratory dosing studies to field population assessments (I)

25

An interspecies correlation model to predict acute dermal toxicity of plant protection products to terrestrial life stages of amphibians

L. Weltje, P. Janz, BASF SE, Crop Protection - Ecotoxicology; P. Sowig, Bayer CropScience / Ecotoxicology

In this presentation, a model to predict acute dermal toxicity of plant protection products to terrestrial amphibian life stages from (regulatory) fish data will be presented. By combining existing concepts, including interspecies correlation estimation (ICE), allometric relations, lethal body burden (LBB) and bioconcentration modelling, an equation was derived that predicts the amphibian median lethal dermal dose (LD_{50}) from standard acute toxicity values (96-h LC_{50}) for fish and bioconcentration factors (BCF) in fish. Where possible, fish BCF values were corrected to 5% lipid, and to parent compound. Then, BCF values were adjusted to an exposure duration of 96 h, in case steady state took longer to be achieved. The derived correlation equation is based on 32 LD_{50} values from acute dermal toxicity experiments with 15 different species of anuran amphibians, comprising 15 different plant protection products. The developed ICE model can be used in a screening approach to estimate the acute risk to amphibian terrestrial life stages from dermal exposures to plant protection products with organic active substances. Applying this method has the potential to reduce unnecessary testing of vertebrates.

26

Overview of the EFSA Scientific Opinion on the state of the science on pesticide risk assessment for amphibians and reptiles

S. Pieper, German Federal Environment Agency (UBA) / Plant Protection Products; P.I. Adriaanse, Alterra Wageningen University and Research Centre; A. Aldrich, Agroscope / Ecotoxicology; C. Berg, Uppsala universitet, Dept. of Environmental Toxicology / Dept of Environmental Toxicology; P. Berny, VETAGRO-SUP / Toxicology; K. Machera, Benaki Phytopathological Institute / Department of Pesticides Control & Phytopharmacy, Athens, Greece; M. Ortiz Santaliestra, Institute for Game and Wildlife Research (IREC) UCLM-CSIC-JCCM; C. Topping, University of Aarhus; S.M. Weir, Queens University of Charlotte / Biology; F. Streissl, EFSA / Pesticides Unit; R. Smith, School of Applied Sciences, University of Huddersfield

Concerns have been raised that the current risk assessment schemes may not sufficiently cover the risk for amphibians and reptiles exposed to intended uses of plant protection products (PPP). To address these concerns, the European Food Safety Authority (EFSA) has published a Scientific Opinion on the state of the science on pesticide risk assessment for amphibian and reptiles [1]. Specific protection goal (SPG) options for amphibians and reptiles were developed to address the general protection goals for non-target organisms and biodiversity in Regulation EC 1107/2009 [2]. When proposing SPG options, the endangered status of a great proportion of amphibian and reptilian species was taken into account, as well as the importance of amphibians and reptiles as drivers of valuable ecosystem services in agricultural landscapes. The analysis of literature data reveals that amphibians and reptiles occur in agricultural landscapes, where they may be exposed to PPP in the in- and off-field areas, and that unacceptable, adverse effects on their populations may occur. It was concluded that the current risk assessment scheme does not fully cover all relevant life stages and thus might not protect the persistence of amphibian and reptilian populations in the long term. The EFSA working group proposes a general risk assessment framework based on a tiered approach and adapted to assess local and landscape-level risks for amphibian and reptilian populations. Identified knowledge gaps regarding amphibians and reptiles concern on the one side the exposure assessment, with e.g. a lack in data about size and location of ponds inhabited by amphibians. On the other side, central information is missing for the effect assessment, e.g. on the impact of oral and dermal exposure routes. Informative field studies are very difficult to perform with these organisms and risk mitigation options – if available - could usefully be included in risk assessment as an alternative to full-scale field studies. Practical mitigation options would need to be developed and adapted for local environments to be most effective. [1]EFSA PPR. 2017 Scientific Opinion on the state of the science on pesticide risk assessment for amphibians and reptiles. EFSA Journal 2017; 329 pp. doi:10.2903/j.efsa.20YY. [2]EC. 2009. Regulation (EC) No 1107/2009 of the European Parliament and of the Council of 21 October 2009 concerning the placing of plant protection products on the market. OJ L 309/1

27

Ecotoxicological assessment of Caretta caretta (Linnaeus, 1758) in the Mediterranean Sea using an integrated non-invasive protocol

S. Casini, University of Siena / Scienze Fisiche della Terra e dellAmbiente; I. Caliani, M. Giannetti, L. Marsili, S. Maltese, D. Coppola, N. Bianchi, T. Campani, University of Siena / Department of Physical, Earth and Environmental Sciences; S. Ancora, University of Siena / Physical sciences, Earth and environment; M. Fossi, University of Siena / Department of Physical Sciences, Earth and Environment

Caretta caretta is the most common sea turtle in the Mediterranean Sea. The IUCN assessment for this carnivorous long-lived reptile underlines the lack of information regarding pollution and pathogens and indicates as a priority efforts to investigate and reduce the impacts of these threats. Up to now very few studies were conducted investigating biological endpoint potentially influenced by contamination on *C. caretta*. The aim of our study was to conduct the first ecotoxicological assessment of this species in the Mediterranean sea using a non-invasive integrated methodology. We set up and applied a monitoring protocol which also includes endpoints, such as CYP1A, Lipid peroxidation, ENA assay, B esterases, never investigated before in this species. Seventy-five loggerhead turtles were sampled in a non-invasive way in Italian Sea Turtle Rescue Centers or free-ranging along the Spanish coasts. Blood, skin and carapace samples were used to test biomarker responses and contaminant (OCs, PAHs, Pb, Cd, Hg) levels. We measured biomarkers of exposure to lipophilic contaminants (CYP1A in skin biopsies), biomarkers of indirect and direct effects investigating neurotoxic (esterases inhibition) and estrogenic (vitellogenin) effects, oxidative stress (lipid peroxidation), genotoxicity (Comet and ENA assays) and liver damage (Gamma Glutamyl Transferase). Elaboration of experimental results was carried out taking also into consideration different age classes of the specimens. Among the main results obtained we should underline the statistically significant correlation between carcinogenic PAHs in blood and DNA fragmentation as well as between Cd in carapace and GGT in plasma. We measured a very sharp band with a molecular weight of 59 kDa in skin sample that can be attributed to CYP1A, never investigated earlier in this species. We also evidenced as the youngest animals showed significantly higher DNA fragmentations, BChE inhibition and increase of GGT, these alterations can be potentially related to their coastal habits. Older specimens showed the highest levels of erythrocyte nuclear abnormalities which may indicate a long term toxicological stress. This study contributed to expand the knowledge about the ecotoxicology of *C. caretta* in the Mediterranean, the non-invasive protocol could also be applied to other marine ecosystems and other sea turtle species, and implemented with new endpoints in the near future.

28

Sucking clams or hunting seals - consequences to walrus health

H. Routti, The Norwegian Polar Institute; S. Bourgeon, University of Tromsø / Department of Arctic Marine Biology; B. Diot, UiT The Arctic University of Norway; N. Duale, Norwegian Institute of Public Health; A.T. Fisk, University of Windsor / Great Lakes Institute for Environmental Research; M. Fossi, University of Siena / Department of Physical Sciences, Earth and Environment; L. Hanssen, M. Harju, NILU Norwegian Institute for Air Research; K.M. Kovacs, C. Lydersen, Norwegian Polar Institute; I. Nymo, Norwegian Veterinary Institute; C. Panti, University of Siena / Department of Physical Sciences, Earth and Environment; S. Scotter, M. Tryland, UiT The Arctic University of Norway; G.D. Villanger, Norwegian Institute of Public Health

The walrus (*Odobenus rosmarus*) is an ice-associated marine mammal with distinct feeding habits. Concentrations of the main chlorinated pollutants, namely polychlorinated biphenyls (PCBs) and chlordanes, in walruses that likely feed on seals are very high - similar to levels observed in polar bears, whereas pollutants concentrations in walruses feeding on benthos are lower. Although multiple studies have associated contaminant exposure to adverse health effects in polar bears and other marine mammals with similar contaminant exposure, there are, to our knowledge, no studies to date investigating effects of pollutants in walruses. The goal of our study was to investigate contaminant and pathogen exposure and endocrine disruption in walruses feeding at different trophic levels. Samples from adult male walruses (n=39) were collected from at Svalbard, Norway. Stable isotope values determined in seven body compartments indicated that all of the walruses in this study fed at a low trophic level. However, concentrations of blubber lipopohilic compounds showed very high individual variation. Concentrations of chlorinated compounds have decreased since they were last studied in walruses sampled, in the same area as the current study, during 2002-2004. Plasma PFAS concentrations varied less betweeen individuals. δ^{15}N values in red blood cells and in mid parts of vibrissae were positively related to concentrations of lipophilic compounds, but not to PFASs. Antibodies against *Brucella* spp. and *Toxoplasma gondii* were detected in 26 % and 15 % of the walrus plasma samples, respectively. Presence of *Brucella* spp. and *Toxoplasma gondii* were not related to contaminant exposure or stable isotope values. Among the 5 thyroid hormone concentrations analyzed in plasma, concentrations of TT4 were negatively related to concentrations of lipophilic compounds. Finally, we analyzed transcript levels of 21 target genes in blood cells and 7 target genes in blubber related to endocrine and immune functions by real-time quantitative PCR. The preliminary results indicate few relationships between transcript levels of genes involved in endocrine functions and pollutant exposure.

29

Triclosan-induced embryotoxicity in the yellow-legged gull

C.D. Possenti, Università degli Studi di Milano; G. Poma, S. Defossé, University of Antwerp Toxicological Center; N. Saino, University of Milano; A. Covaci, University of Antwerp, Toxicological Center / Toxicological Centre Dep of Pharmaceutical Sciences; M. Parolini, University of Milan / Department of Environmental Science and Policy

Triclosan (TCS) is a chemical compound extensively used as synthetic and antimicrobial agent in a wide range of personal care products. Because of its hydrophobic nature and its discharge in the sewer system, TCS accumulates in settled sewage sludge and surface water, contaminating aquatic and terrestrial ecosystems. However, information on the presence and toxicity of TCS towards wildlife species is very scarce. Seabirds are highly exposed to environmental contamination because of their ecological habits, high trophic position in the food webs and relative long life-span. Their eggs are a useful tool to monitor the levels of environmental pollutants and their potential adverse effects because these chemicals can be maternally-transferred to the offspring. However, such information on TCS is lacking. The aim of this study was to explore, through *in ovo* injection, the potential embryotoxicity of TCS in the yellow-legged gull (*Larus michahellis*). In a within-clutch experimental design, 150 ng/g egg weight of TCS were injected into the egg yolk and the effects on embryo morphology, oxidative stress and genetic damage in embryo liver were investigated. Specifically, we assessed effects on embryo body mass, tarsus length and head size, as well as liver and brain mass. The amount of oxidant species (i.e. ROS), enzymatic activities (SOD, CAT, GST) and the levels of lipid peroxidation (LPO) were measured as biomarkers of oxidative stress, while levels of DNA fragmentation were measured as genetic damage endpoint. To check for the reliability of the injection method, we quantified TCS concentration in the yolk of unincubated eggs, while to assess its transfer to the embryo, we measured TCS in residual yolk and in the liver and brain. TCS concentrations in yolk from unincubated eggs were similar to the nominal ones (158.9±35.3 ng/g wet weight), while lower concentrations were found in residual yolk soon before hatching (2.9±1.1 ng/g wet weight). TCS was also detected in the liver (2.3±1.1 ng/g wet weight) and limitedly in the brain (0.2±0.1 ng/g wet weight). TCS treatment did not significantly affect embryo morphological traits. However, TCS significantly increased ROS levels and promoted GST activity, leading to a marginally non-significant increase of both oxidative and genetic damage. Thus, these findings demonstrated, for the first time in a wild bird species, that TCS may affect offspring phenotype and may represent a potential threat for coastal ecosystems.

30

Egg overspray with herbicides and fungicides reduces chick survival in red-legged partridges

M. Ortiz Santaliestra, Institute for Game and Wildlife Research (IREC) UCLM-CSIC-JCCM; V. Alcaide, IRIAF / Centro de Investigación Agroambiental El Chaparrillo; R. Mateo, IREC-CSIC- UCLM / Grupo de Toxicología de Fauna Silvestre; F. Mougeot, IREC

Toxicity characterization in pesticide risk assessment for birds is derived from oral exposure of adults. However, for ground-nesting species, a temporal and spatial overlap of egg laying and incubation with pesticide applications may result in direct exposure of the eggs. Using formulations commonly applied to cereal crops in spring, we conducted two experiments to analyse the effects of 2,4-D and tebuconazole on embryonic development and post-hatching survival of a common farmland bird, the red-legged partridge (*Alectoris rufa*). The first experiment simulated egg overspray with pesticides and the second one the incubation of eggs upon a soil that had been previously sprayed. For both experiments we used an application rate, corresponding to a 30% of the labelled application rate of each product (i.e. assuming a 70% interception by crop), and a control consisting of water application in stead of pesticide formulations. Eggs were incubated at 37ºC and 45% humidity until hatching (23-26 days). Sixteen eggs per treatment were removed from the experiment at different incubation times to analyse pesticide uptake (ongoing analyses, results will be presented at the meeting), and a minimum N=20 per treatment was monitored for embryonic development and post-hatching survival. Chicks were weighted and measured (tarsus length), and body condition calculated, at hatching and at days 8, 16, 24 and 32 post-hatching. Egg overspray with pesticides significantly increased chick mortality (Wald's $X^2 = 29.909$, 14 d.f., $p = 0.008$). Although pesticides did not affect survivorship at hatching time, *in ovo* exposure to both 2,4-D and tebuconazole caused increased mortality of chicks afterwards, resulting in a reduction of productivity 32 days after hatching of 30.6 and 25.9%, respectively. Incubation of eggs in pesticide applied soils did not significantly affect chick survival (Wald's $X^2 = 15.603$, 14 d.f., $p = 0.338$), and nestling growth was not affected in either experiment by embryonic exposure to pesticides. These results suggest that reduction of embryonic and chick survival because of egg overspray with pesticides can be a potential way by which these products may affect reproduction in birds. Likewise, potential time lapses between the exposure period and effect occurence need to be considered when monitoring pesticide impacts on avian reprodutcive output. Financed by the Spanish Ministry of Economy and Competitiveness (Project ref. CGL2016-75278-R)

Biocides and Veterinary Medicines: latest developments in regulatory risk assessment, research and monitoring

31

Regulatory improvement in the assessment of environmental risks from veterinary medicines; a European Perspective

J. Weeks, Joint Nature Conservation Committee

This presentation will focus on the developments in the environmental risk assessment of veterinary medicines from a European regulatory perspective. There is a need to continuously develop and react to changing requirements to perform more sophisticated, quantitative or robust environmental risk assessments for veterinary medicine products. The centralised approval of new veterinary medicines within the EU, is the responsibility of the European Medicines Agency (EMA); however, developed best practices are shared by all member states. The EMA also aids in coordinating other European procedures with several member states involved. EMA frequently authorises and takes advice from specialist working parties aligned to the specific provision or modification of regulatory guidelines or procedures within the committee for veterinary medicine products (CVMP). One such group focuses on the improvement of the guidance to industry and other stakeholders on environmental risk assessment alongside the provision of reflection documents that aid understanding or address specific areas for clarification in regulatory procedures. The overview will highlight some of these recent developments, in improved regulatory advice for current or future procedures. It will summarily cover issues around assessing and limiting veterinary medicines in groundwater, the use of higher tier testing of dung fauna, the use of higher tier plant testing, improvements in PBT assessments, future plans around developing improved guidance for the assessment of risks from aquaculture

medicines and the role of veterinary medicines in driving AMR in the environment and its potential consequences and mitigation. The session will feature successes, current issues and developments in improving the guidance on the assessment of veterinary medicines in the environment; and will reflect on the future challenges and difficulties faced by the regulators and industry alike. This paper will acknowledge the significant continuing contribution made by the Environmental Risk Assessment Working Party (ERA-WP) of the CVMP.

32

Risk of veterinary medicines to plants: Reflections for an updated approach.

R. Carapeto García, Spanish Medicines Agency / Veterinary medicines; A. Haro Castuera, Spanish Medicines Agency / Veterinary Medicines Department; G. Cortés Ruíz, C. Rubio Montejano, Spanish Medicines Agency / Department of Veterinary Medicines

In an Environmental Risk Assessment (ERA) the General Protections Goals need to be translated into Operational Protection Goals in order to achieve efficient and robust ERAs. Not doing so hinders the process of Risk Management in those cases where a risk is identified. In the current regulatory framework of ERA of Veterinary Medicinal Products (VMP) the General Protection Goal ("Protection of ecosystems") is not translated into Operational Protection Goals. Hence, when risks are found it is complicated to manage or mitigate such risks. In the taxonomic level of "terrestrial plants" some VMPs have shown different levels of risks. From the Risk Assessor perspective it is difficult to deal with these risks, partly due to the lack of guidance on Operational Protection Goals. Here we analyze a proposal of using, in the VMPs framework, two Operational Protection Goals in the risk assessment of terrestrial plants: "Protection of Human Interest" and "Protection of Environmental Interests".

33

Innovative environmental assessment of a veterinary medicinal product: watershed-level impacts of trenbolone acetate and 17β-estradiol

J.P. Staveley, Q. Ma, J. Ma, Exponent; C. Celly, Intervet Inc. d/b/a Merck Animal Health; G. Scheef, MSD Animal Health Innovation GmbH / Preclinical Development

Environmental assessments of pharmaceuticals are required by regulatory authorities as part of the drug approval process. Revalor-XR is an extended-release implant for use in cattle fed in confinement (steers and heifers) that contains trenbolone acetate and 17β-estradiol (17β-E2) as active pharmaceutical ingredients (APIs). Both APIs are metabolized *in situ* resulting in the excretion of 17β-trenbolone (17β-TB), 17α-trenbolone (17α-TB), trendione (TBO), 17β-E2, 17α-estradiol (17α-E2), and estrone (E1). The similarity in chemical structures and many of the environmental fate properties among 17β-TB, 17α-TB, and TBO, and that among 17β -E2, 17α-E2, and E1 promote the use of surrogate compounds to represent the trenbolone compounds and the estradiol compounds in the environmental assessment. Data on the individual compounds were collected from various laboratory studies and literature sources, aggregated to generate representative values for the surrogate compounds to characterize their environmental fate, and used for exposure assessments at feedlot-, field- and watershed-scales. Nine exposure pathways were evaluated at the feedlot and field scale, allowing for elimination of insignificant pathways for the watershed-scale modeling, which considered the major exposure pathways and was conducted for two representative watersheds, one in Texas and one in Iowa, using the U.S. EPA's BASINS/HSPF model. The outputs of the modeling efforts resulted in Predicted Environmental Concentrations (PECs) for the surrogate compounds for individual as well as aggregated exposure pathways. The effects assessment was focused on potential reproductive impacts to fish from chronic exposure, which is the most sensitive ecological endpoint for these compounds, and generated Predicted No-Effect Concentrations (PNECs) for 17α-TB, 17β-TB, 17α-E2, and 17β-E2. Risk characterization involved comparison of the PECs for the surrogate compounds to the PNECs of the individual compounds. The assessment at the watershed scale demonstrated that it is highly unlikely that the compounds associated with Revalor-XR would have any significant environmental impacts when used according to the Revalor-XR label. The environmental assessment supported a Finding of No Significant Impact by the U.S. Food and Drug Administration from the use of Revalor-XR in beef steers and heifers in the US.

34

How can mesocosm studies increase realism in risk assessment of biocides and veterinary medicines?

L. Dören, ERM / Product Stewardship; U. Hommen, Fraunhofer IME; P. Ebke, MESOCOSM GmbH Institut für Gewässerschutz; R. Duering, Justus Liebig University Giessen / Research Centre for BioSystems, Land Use, and Nutrition (IFZ), Institute of Soil Science and Soil Conservation

Mesocosm studies can be used to assess the environmental impact of potential stressors based on model-ecosystems under realistic environmental conditions. They are an important link from laboratory to field. Mesocosms provide the assessment of a broad range of different species of different ecological groups forming food webs with complex interactions. Therefore mesocosm studies can support a better understanding of the environmental impact of stressors on population level as well as on ecosystem level (e.g. direct and indirect effects on community structure and ecosystem functions as primary production). In addition, mesocosm studies provide data on the fate of test substances under realistic outdoor conditions, which can be used to test the prediction based on laboratory studies. While for the risk assessment of Plant Protection Products (PPP) mesocosm studies are an established higher tier approach and are considered as the surrogate reference tier, the use of mesocosm studies for risk assessment of biocides, veterinary medicines and chemicals under REACH is rare, although mesocosms are recommended in the *Guidance on information requirements and chemical safety assessment - Chapter R.10: Characterisation of dose [concentration]-response for environment* and in the *Guidance on the Biocidal Products Regulation - Volume IV Environment - Assessment and Evaluation* both provided by ECHA. One reason for this might be, that mesocosm studies have the reputation to be very complex and difficult to evaluate by regulators. This presentation intends to take some fears of contact with mesocosms. It will explain the most important aspects to validate the quality of a mesocosm study and the relevance of the results. Further, it will give some insights to the use of (aquatic) mesocosm studies in the context of PPP risk assessment and will provide important aspects for planning a mesocosm study for biocides, veterinary medicines and chemicals in the context of REACH.

35

Emission estimation of insecticides in mink farms

R.G. Ovesen, Danish Environmental Protection Age; H. Bækgaard, Kopenhagen Fur

Biocides are regulated in EU by the BPR [1]. To evaluate if an active substance (a.i.) or product may be authorised, an assessment of the environmental exposure is required. For insecticides used in stables an Emission Scenario Document (ESD) [2] is used covering application methods and a range of animal categories. The ESD does not cover biocides used in mink farms. A scenario has therefore been developed, where emission of a.i. from mink farms is calculated based on either amount applied or measured concentration in straw. Default values have been estimated from regulation and general practice in mink production in the Nordic countries, where Denmark has the highest production of mink in Europe [3]. Each breeding animal is kept individually in one cage to be treated at the start of the season. Mother and cubs stay together in one cage and are separated into pairs after lactation, where all cages are retreated. Each mother will bear 5.55 cubs/year according to Danish regulation [4]. The number of "breeding females" (BF) is 1 mother+5.55 cubs. The number of nest boxes that is treated/BF may be calculated as follows: 1 animal/nest box before separation and 6.55/2 animals (=3.275)/nest boxes after separation. In Europe it is prohibited to discharge waste from stables to public sewer. Emission is therefore only expected to be to agricultural land. Emission of manure/straw may be from up to 50 BF per hectare (ha) per year based on regulation in the Nordic countries. Emission according to application pattern: $Y = Q_{prod} \times F_{conc} \times (N_{app\ before\ sep} + 3.275 \times N_{app\ after\ sep}) \times B \times 10^{-3}$(Eq. 1) Where Y is emission of a.i. in kg/(ha x year), Q_{prod} is amount of product/nest box in g, F_{conc} is concentration of a.i. in the product in g/kg, $N_{app\ before\ sep}$ is number of treatments before separation of adults and cubs, $N_{app\ after\ sep}$ is number of treatments after separation of adults and cubs and B is amount of straw/manure that may be applied to land in number of BF/ha (B = 50). Emission based on amount of straw/manure applied to the field: Y = Concentration of a.i. in straw/manure x 750 kg straw per BF per ha(eq. 2) Where amount of straw used per BF is 10-15 kg/year according to Kopenhagen Fur. The emission based on Nordic countries regulations and information from Kopenhagen Fur on amount of straw used per BF is 50 BF per ha x 15 kg straw per BF=750 kg straw per BF per ha. Predicted Environmental Concentration in soil may be calculated according to Volume IV Part B [5].

36

Biocidal active substances in municipal wastewater - what product groups are the sources?

S. Wieck, Leuphana University of Lüneburg / Institute for Sustainable and Environmental Chemistry; O. Olsson, Leuphana University of Lueneburg / Institute for Sustainable and Environmental Chemistry; K. Kümmerer, Leuphana University of Lüneburg / Institute of Sustainable and Environmental Chemistry

The emission sources of biocidal active substances in households have been under discussion since these substances have been detected frequently in municipal wastewater. The emissions from the outsides of houses, e.g. facades, have already been discussed in detail. However, until now the specific sources from the interior of these households remained unknown. To investigate the products responsible for these emission to wastewater, we analysed the wastewater of one neighbourhood for a set of biocidal active substances and compared these results with household product inventories. Time-proportional sampling of daily samples was conducted during one year for one week of each season. The 14 substances analysed with an LC-MS/MS method were BIT, C12-benzalkonium chloride, carbendazim, CMIT, DCOIT, DEET, diuron, icaridine, OIT, piperonyl butoxide (PBO), triclosan, tebuconazole, terbutryn and tetramethrin. In comparison with data available from household product inventories of this neighbourhood, we investigated the product groups possibly being responsible for the emissions to the wastewater. Except for CMIT, DCOIT, PBO and tetramethrin, all substances have been detected in at least 10 % of the samples. Highest concentrations were measured for C12-benzalkonium chloride with an average concentration of 6.6 µg/L. Besides C12-benzalkonium chloride, BIT, DEET and icaridine were measured in all samples. The results show that washing and cleaning agents are important sources for preservatives such as BIT and OIT, while triclosan was apparently mainly emitted through personal care products. The mosquito repelling substances DEET and icaridine were found throughout the whole year, with highest emissions in summer and autumn. C12-benzalkonium chloride concentrations were associated with the inventoried disinfectants. Material preservatives such as terbutryn, diuron, tebuconazole or carbendazim were also detected. As these were not listed on the inventoried products, emission via treated materials such as paint, render, seals or textiles seems likely. We were able to show that biocidal active substances are emitted from the inside of households in considerable concentrations. Those emissions are not only due to biocidal products but also washing and cleaning agents, personal care products and preserved materials. For this reason, measures should not only tackle biocidal products when it comes to the reduction of biocidal active substances in wastewater.

The environment as a reactor determining fate and toxicity of nanomaterials (I)

37

Comparative multi-generation study of long-term effects of pristine and wastewater-borne silver and titanium dioxide nanoparticles on reproduction in Daphnia magna

S. Hartmann, University of Siegen, Institute of Biology / Department of Chemistry and Biology; R. Louch, University of Manchester; R. Zeumer, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Bioaccumulation and Animal Metabolism; C. Schlechtriem, Fraunhofer IME / Department Bioaccumulation and Animal Metabolism; K. Witte, University of Siegen / Department of Chemistry and Biology

Manufactured nanomaterials (MNMs) and especially Ag- and $Ti0_2$-NPs are processed in daily used products such as cosmetics, clothing and in medical supplies. After passing wastewater treatment plants these MNMs reach the aquatic environment and can accumulate in the aquatic ecosystem and cause toxicity to aquatic organisms. To assess the risk potential of these NPs to aquatic invertebrates under more realistic circumstances, we investigated and compared possible effects of pristine Ag-NPs and Ti02 NPs with those after passing a model wastewater treatment plant on the reproductive success (number of offspring), mortality and body size of adult daphnia as endpoints in up to six generations. We exposed daphnia to: (i) pristine Ag-NPs (NM300K) and TiO_2-NPs (NM105) or (ii) wastewater borne Ag- and TiO_2-nanoparticles from effluent from the model WWTP. The first generation of daphnia was exposed to four concentrations of Ag-NPs (nominal: 1.25 µg/L, 2.5 µg/L, 5.0 µg/L and 10.0 µg/L), to solvent control (NM300K DIS), or to three concentrations of TiO_2-NPs (nominal: 25 µg/L, 50 µg/L, 100 µg/L) in line with the OECD guideline No. 211. Each generation was exposed for 21 days and started with the third brood from the previous one. In all six generations the exposure with pristine Ag-NPs (NM300K) for 21 days caused a significant reduction in the mean number of offspring in daphnia compared to the control. However, wastewater-borne Ag-NPs had no effects on reproduction in any generation. In the pristine Ag-NPs treatment the mean body length of daphnia was significantly larger at 5 µg/L in generation F2 and at 2.5 µg/L in generation F3 compared to the control. In the wastewater-borne Ag-NP treatment the adults´ body length was significantly larger at 2.5 µg/L. Thus, adult´s body length showed no consistent pattern towards both scenarios. When passing WWTPs most Ag-NPs might be transformed and enter the aquatic environment as silver sulfide. That may be the reason for the lower toxicity than compared to other forms of Ag-NPs. Our results provide a first, direct comparison between the toxicity of pristine Ag-NPs and TiO_2-NPs with those from WWTP. To our knowledge, the present study is the first one showing that Ag-NPs from a wastewater treatment plant had a minor and no chronic toxicity to *Daphnia magna*. The used experimental approach allows a more realistic risk assessment of Ag-NPs and TiO2-NPs for the aquatic environment. The experiment with TiO2-NPs are in progress.

38

Development of a rapid screen to assess bioaccumulation potential: from ex vivo to in vivo using pristine and aged nanomaterials in fish

N. Clark, School of Biological Sciences, Plymouth University; D. Boyle, Plymouth University; R. Handy, University of Plymouth

Bioaccumulation is one of the key triggers of concern for environmental risk assessment that has had little consideration for engineered nanomaterials (ENMs). Given that ENMs undergo surface chemical reactions, agglomerate and sediment, the likely exposure route to higher trophic organisms (e.g., fish) is through the diet. Here, we address the suitability of an *ex vivo* gut sac technique to rapidly screen the bioaccumulation potential of pristine (Ag NPs) and environmentally aged (Ag_2S NPs) materials. Additionally, we assess whether the results of the gut sac experiment can predict *in vivo* chronic dietary exposure. The gut sacs were prepared by removing the entire gastrointestinal tract and separating it into the oesophagus, stomach, anterior, mid and hind intestine compartments. Compartments were exposed by filling the lumen with one of four solutions: physiological gut saline or saline spiked with 1 mg/L Ag as $AgNO_3$, Ag NP or Ag_2S NP. Following a 4 h exposure, tissues were cut open and the mucosa was separated from the underlying muscularis, through scraping via a microscope slide. For the *in vivo* chronic dietary exposure, fish (n = 350) were graded into tanks (n = 3 tank/treatment). Fish were fed either a control (no added Ag), 100 mg/kg as $AgNO_3$, Ag NPs or Ag_2S NPs. Fish were sampled each week (1, 2, 3 and 4; n = 2 fish/tank/time point). Following this, all tanks were placed on the control diet for another two weeks to measure Ag elimination. During sampling, the mid and hind intestine, liver, gallbladder, kidney, spleen, gill, brain and carcass were dissected. Tissues from both experiments were analysed for total Ag using ICP-MS. The gut sac experiment demonstrates the uptake of Ag is associated with the mid and hind intestine. There was significantly less Ag in the muscularis of the mid and hind intestine after exposure to Ag NP and Ag_2S NP compared to $AgNO_3$, but no difference between ENM treatments. The *in vivo* experiment demonstrated significantly more Ag in the mid and hind intestine of Ag NP and $AgNO_3$ treatments compared to Ag_2S NPs. Silver from all the exposures were able to pass the gut epithelium and cause total concentrations in the liver to rise, despite the form being unknown. In conclusion, the *ex vivo* gut sac method can be used to rapidly screen the bioavailability of Ag NPs and Ag_2S NPs. However, if the data are ranked in the mid and hind intestine by total Ag accumulation, the gut sac does not directly predict *in vivo* accumulation.

39

Fate and Effect of Wastewater Borne Manufactured Nanomaterials on the Aquatic Food Chain

R. Zeumer, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Bioaccumulation and Animal Metabolism; B. Knopf, Fraunhofer Institute for Molecular Biology and Applied Ecology IME / Environmental Specimen Bank and Elemental Analysis; B. Meisterjahn, Fraunhofer Institute for Molecular Biology and Applied Ecology IME / Ecological chemistry; I. Lopes, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro; V. Galhano, Department of Biology & CESAM - University of Aveiro / Biology (dBio); M. Monteiro, Aveiro University / Biology; S. Loureiro, Universidade de Aveiro / Biology; C. Schlechtriem, Fraunhofer IME / Department Bioaccumulation and Animal Metabolism

Manufactured nanomaterials (MNMs) are widely used in various applications and commercial products, e.g. textiles, sunscreens, paints, cosmetics etc. Even though MNMs are mostly removed during wastewater treatment, the remaining and mostly transformed MNMs in the effluents are significant and may show an increased toxicity for aquatic organisms due to their modification during the WWT. The impact of wastewater-borne MNMs (TiO_2 and Ag MNMs) on different trophic levels and their bioaccumulation within a relevant food chain (algae-*Daphnia*-fish) have been investigated with innovative analytical and experimental approaches.Several model WWTPs were conducted according to OECD Guideline 303A. The collected effluents were used to perform acute and chronic tests with *Daphnia magna* and *Onchorhynchus mykiss* according to the OECD guidelines 202, 211, 215 and 305. Animals were exposed to (i) effluent from model WWTPs contaminated with MNMs, (ii) uncontaminated effluent, manually spiked with MNMs and (iii) dilution water enriched with pristine MNMs. Tissue samples of the different test organisms were analyzed for changes in the levels of several biochemical markers [lipid peroxidation; activities of acetylcholinesterase (AChE), lactate dehydrogenase (LDH), superoxide dismutase (SOD), catalase (CAT) and glutathione S-transferase (GST)]. Furthermore, uptake and elimination kinetics of the MNMs were investigated by quantitative ICP-MS and ICP-OES analysis.No chronic effects were found in *D. magna* after exposure to effluents with transformed AgNPs. However, when supplemented into uncontaminated effluents or dilution water, the amount of offspring per adult daphnid decreased with increasing AgNP concentration. For nano-TiO_2, no effects on the reproduction of *D. magna* could be shown at environmentally relevant concentrations. Only after chronic exposure to the very high concentrations of 5 mg/L and 10 mg/L significant effects could be shown.Neither nano-Ag nor nano-TiO_2 showed an effect on the growth of juvenile rainbow trouts after 28 days of exposure.The analysis of biochemical markers showed that several effects induced by chronic exposure to MNMs were observed. However, no general effect pattern could be identified.Total MNM levels were measured in several tissue samples in *D. magna* and *O. mykiss* following exposure via the water or food. Pristine nanomaterials showed a significantly higher uptake into the test organisms compared to supplemented and treated WWTP effluents.

40

Uptake and elimination kinetics of pristine and aged silver nanoparticles in freshwater benthic organisms

P. Silva, Universidade de Aveiro; C. van Gestel, Vrije Universiteit Amsterdam / Ecological Science; R.A. Verweij, Vrije Universiteit Amsterdam / Department of Ecological Science; S. Loureiro, Universidade de Aveiro / Biology

Nanomaterials (NMs) can undergo changes in their properties and behaviour during application and disposal. Once in the environment, different forms of NMs can be taken up by organisms and suffer biologically-driven alterations. Toxicokinetic modelling can provide important information about ways of uptake, internal processing and elimination of NMs. Freshwater systems are important sinks for NMs, especially considering the sediment phase, where benthic organisms can be exposed through both water and sediment. Considering this, the aim of the present study was to determine the uptake and elimination constant rates of pristine and (simulated) aged silver nanoparticles (Ag-NPs) in freshwater benthic organisms. In this study the pulmonate snail *Physa acuta*, the non-biting midge *Chironomus riparius* and the planarian *Dugesia tigrina* were used as tests species. Pristine Ag-NPs of different sizes (3-8nm, 50nm and 60nm), a 27nm silver sulphide (Ag_2S-NPs) simulating aging, and their ionic counterpart as silver nitrate ($AgNO_3$) were tested. Bioaccumulation tests consisted of an uptake phase, where organisms were exposed in glass beakers with sediment and contaminated aqueous medium in a concentration of 10 µg Ag.L^{-1}, and an elimination phase where organisms were transferred to clean medium. Animals were sampled during the tests and total body Ag concentration was analysed by graphite furnace atomic absorption spectrometry. Kinetics of Ag-NPs and ionic Ag were described by one-compartment models. In this work, uptake and elimination kinetics of the different Ag forms showed to differ between organisms. In general, chironomids presented higher k1 values which can be related to their higher exposure to settled Ag in the sediment. Larvae exposed to Ag 50nm showed the highest uptake rate constant (k1) and the highest elimination rate constant (k2), suggesting that Ag 50nm was easily taken up and eliminated from the body. Ag_2S-NPs displayed a k2 close to zero, indicating that not only uptake was less in the larvae but also that they were less eliminated. Snails showed faster uptake and elimination of Ag_2S-NP from the body compared to other Ag forms. For planarians, results revealed very similar k1 values, with the highest k1 for animals exposed to ionic Ag and the lowest for Ag 60nm exposure. Analysis of Ag in the sediment will be soon available to elucidate the behaviour of Ag, especially at the water-sediment interface, where most benthic organisms are exposed.

41

Transformation of silver nanomaterials by ubiquitous zinc finger peptides

K. Wheeler, Z. Amaris, G. Park, M. Eiken, K. Baumgartner, J. Marckwordt, Santa Clara University / Chemistry & Biochemistry

In biological systems, chemical and physical transformations of engineered silver nanoparticles (AgENMs) are mediated, in part, by proteins and other biomolecules. Given the high affinity of thiolate ligands for silver, metalloproteins are key targets to evaluate the role of biomolecules in AgENM transformations. In turn, metalloprotein interactions with AgENMs are also central in mechanistic studies of cellular impacts of AgENMs, including toxicity, antimicrobial, and resistance mechanisms. Despite the shared preference of both silver and zinc for thiolate and amine coordination, the interactions of zinc finger domains with AgENMs is not well studied. Zinc fingers constitute a large class of metalloproteins, ubiquitous in eukaryotes, that use a combination of cysteine and histidine residues that bind Zn(II) as a structural element. Zinc finger domains within proteins typically serve as interactors and can binding DNA, RNA, proteins or small molecules to mediate cell processes like transcription or translation. Using a small library of zinc finger peptides, we have evaluated the impact of Zn fingers on AgENMs aggregation and dissolution. Zinc finger peptides drive AgENM dissolution resulting in release of Ag(I)(*aq)* at orders of magnitude higher rates than other model proteins, including a few metalloproteins. The release of Ag(I)(*aq*) is central to mechanisms of cellular response and toxicity of AgENMs. Indeed, Cu(I) binds to both the apopeptides and the Co(II)-substituted peptides; the stoichiometry of Ag(I) binding is dependent on the peptide primary sequence. Additional studies using fluorescence spectroscopy to monitor Ag(I) binding to the Zn finger peptide indicate that the Ag(I) effectively completes with Zn(II) at the metal binding site, despite the high affinity of Zn(II) for the peptide. Circular dichroism spectroscopy used to assess changes in the peptide secondary structure demonstrate that the addition of either form of silver alters peptide structure and structural perturbations are again dependent upon the peptide sequence. These results show that Zn finger peptides can mediate AgENM transformations within eukaryotic cells. In turn, for the Zn finger peptides studied here, Ag(I) is the thermodynamically favored metal despite the known high Zn(II) affinity of zinc finger domains. This works suggests that Ag(I)-substituted zinc finger domains might be relevant in the context of both silver toxicity mechanisms and silver-responsive transcription factors.

42

Fate and effects of transformed Ag and TiO2 nanoparticles aged through a lab-scale wastewater treatment system

A. Georgantzopoulou, Norwegian Institute for Water Research NIVA; C. Vogelsang, NIVA Norwegian Institute for Water Research; C. Coutris, Norwegian Institute of Bioeconomy Research NIBIO / Dpt for Soil Quality and Climate; F. Polesel, Technical University of Denmark (DTU) / DTU Environment; B. Plósz, University of Bath; K. Ndungu, NIVA Norwegian Institute for Water Research; P.A. Carvalho, SINTEF Materials and Chemistry; A. Macken, NIVA / marine pollution

The increase in production and use of Ag and TiO_2 nanomaterials has led to their release in wastewater streams and subsequently in the environment. Nanoparticles (NPs) can undergo transformations in environmental media such as wastewaters leading to an alteration in behavior, bioavailability and toxicity that may differ from their pristine counterparts and make predictions challenging. In this context, the overall goal of the study was to elucidate (i) the behavior and transformation of Ag and TiO_2 NPs in realistic matrices such as wastewater effluents and activated sludge and (ii) the subsequent effects of transformed particles in comparison to their pristine counterparts. In this study, a laboratory-scale wastewater treatment system was established and combined with a battery of ecotoxicological assays and characterization techniques. The system contained activated sludge and was operated as a pre-denitrification system fed with synthetic wastewater spiked daily with 10 µg Ag NPs/L (PVP coated, 25 nm, nanoComposix) and 100 µg TiO_2 NPs/L (nominal primary size of 5 nm, NM-101, JRC) over a period of 5 weeks. During that period the effluents were collected weekly and the excess sludge was stored for the evaluation of terrestrial toxicity. Samples from all reactors and effluents were collected weekly and analyzed by sequential filtration and ICP-MS to determine the partitioning of NPs and their transformation products. Transmission electron microscopy and sp-ICP-MS were performed on selected samples. The effects of aged particles were assessed using a battery of bioassays including freshwater and marine algae (growth inhibition and reactive oxygen species -ROS- formation), crustaceans, as well as *in vitro* models of relevance for NP toxicity assessement (RTgill-W1 cell line, effects on metabolic activity, epithelial integrity, ROS formation, gene expression). The extent of the observed effects was dependent on the organism exposed, with bottom feeding organisms and algae being more sensitive, while the *in vitro* model was a good tool for environmental samples. Furthermore, the biosolids generated from the lab-scale continuous system were used in terrestrial microcosm experiments, giving insight into the fate and potential accumulation in a model terrestrial system. Experimental data generated from the continuous-flow operation of the activated sludge system and the targeted batch experiments will be used to model the fate and the removal of NPs.

Advances in environmental risk assessment of oil spills and offshore oil & gas operations (I)

43

Optimization of Oil Spill Response Planning and Preparedness Using Spill Mitigation Impact Assessment (SIMA)

R. Wenning, Ramboll Environ / Ecology & Sediment Management; E. Bizzotto, M.J. Bock, H. Robinson, H.R. Diogo, Ramboll / Ecological Services

Oil spill emergency response plans (OSRP) are required as part of permitting for offshore operations. An OSRP typically includes risk assessment and detailed plans for responding to different types of oil spill accidents involving shipping, pipelines, platforms, and/or subsea well heads. The owner/operator must demonstrate to state and federal regulatory authorities that the company's OSRP for offshore exploration and production operations conform to all applicable regulations and international standards and practices, and further demonstrate that the necessary equipment and trained personnel are in place to respond quickly and effectively to an oil spill accident. In the event of an oil spill accident, the priorities for oil spill response (OSR) are to protect people, prevent or mitigate environmental damages, and prevent impact to affected communities. Spill Impact Mitigation Assessment (SIMA) is a science-based framework evolved from Net Environmental Benefits Analysis (NEBA) to broaden the focus from consideration of mitigation of ecological impact to include mitigation of socioeconomic and cultural impacts, as well. SIMA is a method for identifying and comparing the socio -environmental and -economic benefits of alternative OSR options, with the goal of selecting options that best mitigate the consequences of spilled oil and impose the lowest additional negative consequences on environmental and social resources. SIMA is both site- and spill- specific, and is particularly useful during the initial planning and preparedness period for oil and gas exploration and when new technologies and best practices emerge that need to be adopted into safety, health and environmental operational programs. This paper describes a spill impact mitigation assessment framework using recent examples of OSRP work conducted for the Arctic Oil Spill Response Technology Joint Industry Project, American Petroleum Institute Gulf of Mexico Deepwater Project, and several companies working in tropical marine environments. The applicability of SIMA to marine resources and habitats is also discussed. The SIMA framework typically includes (a) assessment of initial impacts and potential consequences in the marine environment, and (b) effectiveness and consequences of deploying different spill response strategies.

44

Adapting the SIMA Process to Assess Offshore Decommissioning Options

T. Coolbaugh, ExxonMobil Research & Engineering; A. Aziz, ExxonMobil Upstream Research Company; P. Taylor, Petronia Consulting Limited; G. Coelho, Sponson Group Inc.

For several decades, the oil and gas industry has used the Net Environmental Benefit Analysis (NEBA) approach for oil spill response contingency planning. Recently, IPIECA-API-IOGP published guidelines on the implementation of NEBA, using a novel process known as Spill Impact Mitigation Assessment (SIMA). When applied to oil spills, the objective of SIMA is to conduct an evaluation that will enable decision-makers to choose response options that will result in the best overall recovery of the ecological, socio-economic and cultural resources. One of the key advantages to the SIMA process is its transparency – it clearly shows and documents the assumptions and decisions that were used to arrive at the conclusions. In most spill scenarios, no single response option is likely to be completely effective; oftentimes, the best approach to minimize environmental impacts is to employ multiple response options. This will require consensus between key stakeholders and the various decision-makers on the benefits and drawbacks of each option, thereby developing response strategy. Similarly, oil and gas operators are faced with complex options for decommissioning offshore installations as part of their decommissioning plans. There is a need to evaluate these options in a scientifically-defensible and consistent manner, while adequately assessing risks that can be challenging to asses. An adapted SIMA process could be a valuable tool for fostering collaboration between operators, stakeholders and regulators, thereby ensuring a transparent review of engineering studies and available decommissioning options in a meaningful way. This paper evaluates the use of the SIMA process in a hypothetical decommissioning of an offshore platform to determine if this process lends itself to this purpose. It describes the challenges encountered when trying to conduct this comparative risk assessment, originally intended for oil spill response, to the task of decommissioning. Finally, it proposes adaptations to the SIMA process that might enable its use in the future as a credible tool in assessing environmental risks for oil and gas decommissioning.

45

Detection and quantification of oil contamination in vegetated areas using hyperspectral remote sensing

G. Lassalle, ONERA; S. Fabre, ONERA / Optics and Associated Technics; A. Credoz, R. Hédacq, TOTAL SA / Environment; P. Borderies, ONERA / DEMR; G. Bertoni, INRA / Dynafor; D. Dubucq, TOTAL SA; A. Elger, University of Toulouse / EcoLab

In the field of oil and gas production, there is a constant challenge in developing new techniques of oil detection for prospection (natural seeps) and environmental monitoring purposes (pipeline leaks, production mud pits). Among recent techniques proposed, remote sensing, and especially hyperspectral spectroscopy, has provided promising results for the detection of oil in vegetated areas. It is achieved by extracting information from the spectral signature of vegetation, which corresponds to its reflectance measured by a sensor over multiple, narrow and contiguous wavelengths. Vegetation reflectance is driven by leaf structure, pigments and water content, which can be affected by oil. As a result, the spectral signature of vegetation is modified so it is possible to detect and quantify oil exposure. The final objective of this rapid and non-destructive approach is to be applied on airborne hyperspectral images at high spatial resolution (Rubus fruticosus L.) exposed for 32 days to 6 to 25 $g.kg^{-1}$ total petroleum hydrocarbons (TPH) from crude oil and mud pits under controlled conditions. Spectral signatures were measured at different scales (leaf, plant and canopy) with a portable spectroradiometer, using a leaf-clip or fixing the sensor above the plant. After 18 days, the signature of TPH-exposed plants was strongly modified. Compared to controls, their reflectance increased in all wavelengths at leaf scale, up to 0.15 greater. The low ground coverage of TPH-exposed plants induced an opposite response in the near- and short-wave infrared (750-2500 nm) at plant and canopy scales. Vegetation indices (VI), computed by reflectance ratio at different wavelengths, were able to discriminate among treatments, and remained robust from leaf to canopy scale. Plant pigments, chlorophyll fluorescence and stomatal conductance were also affected by TPH. The following step was to study the spectral response of the species in situ, in an oil and gas brownfield with the same contamination, ranging from 16 to 58 $g.kg^{-1}$ TPH. Results indicated that the same VI were highly correlated to TPH ($r > 0.7$). Finally, VI allowed identifying the brownfield from an airborne hyperspectral image at high spatial resolution, and thus confirmed the potential of this technique for assessing environmental risks deriving from oil and gas production in vegetated areas.\n\n

46

A tool for tracking complex ecotoxicological effect data after large pollution events with use of the Deepwater Horizon oil spill as a case study

J. Beyer, NIVA - Norwegian Institute for Water Research; H.C. Trannum, T. Bakke, Norwegian Institute for Water Research; P.V. Hodson, Queens University / School of Environmental Studies; T.K. Collier, Delta Independent Science Board Ninth Street Suite Sacramento CA

The Deepwater Horizon Oil Spill (DWHOS) in 2010 is the largest and most studied accidental marine oil spill in history. More than 100 new research studies concerning the effects of the DWHOS have been published each year since 2011. Key issues investigated include the behaviour and fate of oil in deep spills, the effect of oil dispersant applications, microbial oil degradation, oil-associated marine snow formation, oil impacts on deep water corals, seafood quality and safety, oil contaminants effects on fish, birds and marine mammals, effects of combined stressors on species and habitats, and habitat and ecosystem recovery processes. To keep order in this flow of new knowledge is an important albeit challenging task. It is essential that the lessons of DWHOS are applied globally to improve pre-spill and post-spill measures to minimize the adverse ecological impacts of the next big marine oil spill, wherever it may happen.In this presentation, we demonstrate a convenient tool for keeping track of the large amount of ecotoxicological data and knowledge that typically emerges from research and monitoring after marine pollution disasters, using the DWHOS as a case study. In addition, we provide a summary of the new insights about oil spill effects on marine ecosystems that have been gained from the DWHOS research, and identify some key knowledge gaps still remaining. The presentation will update a comprehensive review about the environmental effects of DWHOS that we recently published in Marine Pollution Bulletin.

47

Oil spill combat and effects in the Arctic coastal environment; self-cleaning potential and in situ burning

S. Wegeberg, Aarhus University / Department of Bioscience; J. Fritt-Rasmussen, Aarhus University / Department of Bioscience - Arctic Environment; O. Geertz-Hansen, M.B. Larsen, Greenland Institute of Natural Resources; K. Gustavson, Aarhus University / Department of Bioscience - Arctic Environment

What is the environmental effects of a beaching oil spill in the Arctic, how well will the shoreline potentially be able to self-clean and will combating the oil by in situ burning at the coast just do more harm to the communities in the tidal zone? To answer these questions, several studies have been performed at the west coast of Greenland in 2016 and 2017. One aim of the studies was then to support net environmental benefit analysis, NEBA, related to oil spill in Arctic waters. A NEBA is often performed to achieve the optimal environmental effect with respect to choice of oil spill combat methodology and biology at risk. Hence, a synthesis will be presented of following studies: 1) removal rate and ecotoxicological effects of oil smother on seaweed (*Fucus distichus*), an important organism of the communities in the coastal tidal zone; 2) self-cleaning potential of a coast line, including natural removal by seawater wash and physical degradation; and 3) effects on the tidal communities after combat of a beaching oil spill by in situ burning. Effects of oil smothering of the macroalgae *Fucus distichus*, which inhabit the intertidal zone of the coasts in the Arctic, as well as its self-cleaning potential by wash in sea were studied over a period of 2 weeks in a field test at Disko Island on the Greenland west coast. The rate of natural removal and degradation of stranded oil on rocky surface were investigated at three locations of low Arctic, middle Arctic and high Arctic climatic regimes along the west coast of Greenland. The study included experiments of natural removal of a crude oil and a heavy fuel oil from tiles mimicking rocky shore substratum and was run in the period from May-September 2017. The tiles were placed in different height levels of the tidal zone, and hence natural removal and degradation rates in correlation to different water cover regimes and air exposure times were obtained. The oil remains on the tiles were analysed for chemical compositions. A pilot scale costal in situ burning operation was performed during summer in a bay in western Greenland with a crude oil for testing burning efficiency and environmental exposure and effects. The studies were funded by the European Commission Horizon 2020 programme and the Government of Greenland.

48

How stable are our indices? - differentiating between sources in a weathering environment

S.M. Mudge, NILU - Norwegian Institute for Air Research / IMPACT

Crude oil contains many hundreds of compounds and some of these are widely used to differentiate between different oils and products, especially in spill scenarios. Traditionally, we have developed chemical fingerprints based on a suite of compounds such as the steranes and terpanes although the concept of a “fingerprint” suggests this is static in time. However, it is also well known that these compounds have differential degradation rates in the environment and the pattern changes with period of exposure. It would be more appropriate to use a “signature” analogy when comparing oils by this approach. The weathering processes change the chemical signature and old oil may have a different chemical composition to the original source oil. When we analyses such samples, we may need to ask if this is the same oil as the proposed source, or a different oil with a different signature that is also present. The steranes and terpanes contain several homologues and analysis of the chemical signature during the Deepwater Horizon Response clearly indicated that several of these compounds were not behaving conservatively and were degrading at a faster rate than anticipated given the exposure time. Comparisons with the actual oil released clearly identifies the compounds most likely to alter and the environments where they degrade. In this case, the Louisiana marshes were clearly a site where biodegradation was significantly faster than expected. This was also true of the alkylated PAHs which had been used as source identifiers in previous spills such as the Exxon Valdez. The triaromatic steranes were also degrading at a significant rate while the oil was at sea and the exposure to UV light may have led to a relatively rapid abiotic transformation. When it comes to distinguishing between sources, less may be more! We need to select the compounds we include in our analyses with care since each question may need a different approach: if we want to know if the oil is weathering, we use a suite of compounds with differential properties appropriate to the environment of the spill. If we want to conduct source apportionment, we may need to choose the most recalcitrant of the compounds rather than all of them.

Fish model species in human and environmental toxicology (I)

49

Exposure to bisphenol S alters microRNA expression in male zebrafish (Danio rerio)

j. Lee, K. Ji, Yongin University

In response to the restriction of bisphenol A (BPA), bisphenol S (BPS) has been widely used in the manufacturing of polycarbonate plastics and epoxy resins as an alternative compound. BPS has been found to affect reproduction, development, and immune system. Although microRNAs (miRNAs) play a crucial role in many metabolic activities, whether and how they are involved in the process of BPS-induced toxicity is largely unknown. BPS-induced changes in miRNAs and target gene expression in male zebrafish (*Danio rerio*) gonad, and the potential mechanism was investigated. Male zebrafish were exposed to 0, 5, and 50 µg/L BPS for 21 d. miRNA was isolated from the gonad pool and the expression profiles of 255 known zebrafish miRNAs were analyzed using Affymetrix microarrays. Quantitative real-time polymerase chain reaction was used to validate the expression of several miRNAs in the microarray data. The GO term analysis revealed that miRNAs significantly affected by BPS exposure were involved in hematopoiesis, lymphoid organ development, and immune system development. Among 14 miRNAs that were significantly regulated after exposure to 5 and 50 µg/L BPS, six miRNAs targeted *cyp19a1b* gene, suggesting the role of BPS-induced toxicity via the interference with the aromatization process. The results of this study will provide novel insight into the epigenetic regulatory mechanisms of BPS-induced toxicity in male zebrafish.

50

Zebrafish as a model to investigate mechanisms of adverse metabolic and cardiovascular outcomes associated with elevated dietary selenium exposure

D.M. Janz, University of Saskatchewan / Toxicology Centre; C. Pettem, University of Saskatchewan - Toxicology Centre / Toxicology; J. Thomas, University of Saskatchewan   Toxicology Centre; L.P. Weber, University of Saskatchewan / Veterinary Biomedical Sciences

A variety of anthropogenic activities cause increased loading of the essential trace element selenium into aquatic ecosystems, where it poses an extreme toxicological hazard to fishes due to the narrow range between essentiality and toxicity. Although several studies have reported developmental toxicities in early life stages of fishes, fewer studies have investigated sublethal toxicological effects that may occur following dietary selenium exposure in adult fishes. Adult zebrafish were exposed to dietary selenomethionine (SeMet) at Se-normal levels (1.1 - 1.3 µg Se/g food, dry mass) and environmentally relevant supraphysiological levels (3.4 – 28.8 µg/g) for 90 days. Swimming performance, O_2 consumption and metabolic rates were determined using a swim tunnel respirometer. Cardiac function was assessed using high resolution (30 µm) ultrasound biomicroscopy. Whole-body energy stores (triglycerides and glycogen) and mRNA transcript abundance of selected genes of interest were determined. Compared to controls, adult zebrafish exposed to elevated dietary SeMet exhibited impaired swimming performance (lower fatigue velocity or Ucrit). This was associated with elevated basal metabolic rate and reduced aerobic scope, indicating impaired aerobic capacity. Triglycerides (the primary fuel for aerobic swimming) were elevated in a dose-dependent manner, which was associated with altered transcript abundance of several genes involved in lipid homeostasis. Ultrasonography revealed decreased cardiac output, which was associated with increased echodensity at the atrial-ventricular junction and reduced mRNA expression of the collagenase, MMP2. These results suggest significant ecophysiological effects that may impair the fitness of fishes exposed to elevated dietary Se in contaminated ecosystems. From a comparative biomedical viewpoint, these results highlight the utility of zebrafish as a model to investigate mechanisms of metabolic, energetic, and cardiovascular toxicities caused by excess dietary Se exposure, since similar responses following selenium over-supplementation have been reported in the human clinical toxicology literature. A proposed adverse outcome pathway (AOP) based on this study will be presented that links changes in gene expression to key events leading to adverse outcomes at the individual, and potentially population, levels of biological organization.

51

Toxicity and neurotoxicity profiling of sediments from Gulf of Bothnia with Danio rerio embryos

R. Massei, Helmholtz Centre for Environmental Research   UFZ; H. Hollert, RWTH Aachen University / Institute for Environmental Research; M. Krauss, T. Schulze, Helmholtz centre for environmental research - UFZ / Effect-Directed Analysis; C. Weidauer, Helmholtz Centre for Environmental Research   UFZ; P. Haglund, C. Galampois, Umea University; M. Tysklind, Umea University / Department of Chemistry; W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis

Sediments are a well-known sink for a large variety of organic pollutants that may cause distress to benthic and pelagic species in case of their remobilization to the water-phase. Risk assessment of complex mixtures may involve component-based approaches applying chemical analysis together with measured or predicted toxicity data of individual components and mixture risk modeling or whole mixture approaches using bioassays. Both are complementary and an integration of both approaches using chemical and bio- analytical tools for characterization for example of sediment contamination is expected to provide a more comprehensive picture of toxic risks to aquatic organisms. One of the most promising organisms for diagnostic *in vivo* testing of sediment may be zebrafish embryos (*Danio rerio*) being a versatile *in vivo* model suitable for high-throughput analysis while keeping several advantages of *in vitro* approaches (i.e. low-cost, sensitivity, short duration of the test). The fish embryo test (FET) with *Danio rerio* has been considered as a good surrogate for the acute toxicity fish test and was successfully used in several studies for the detection of toxicity and neurotoxicity in sediments samples. One of the major advantages of the FET with *Danio rerio* is the possibility to monitor several toxic endpoints including the modification of biochemical and molecular processes, which can be related to the exposure of specific pollutants . The present study provides a first attempt to integrate a diagnostic whole mixture assessment workflow based on *in vivo* toxicological profiling of *Danio rerio* after direct exposure to sediments from Gulf of Bothnia (Sweden) for 4 days. The objectives of the present study was were 1) to validate a screening approach for sediment of samples 2) to offer a first *in vivo* toxicological profile of sediment from three different polluted sites from Gulf of Bothnia.

52

Proteomics based screening tool to detect molecular responses following aromatase inhibition

S.U. Ayobahan, IME Fraunhofer / Department of Aquatic Ecotoxicology; E. Eilebrecht, M. Teigeler, Fraunhofer IME / Ecotoxicology; M. Kotthoff, Fraunhofer IME / Environmental and Food Analysis; S. Kalkhof, University of Applied Sciences Coburg / Department of Bioanalytics; C. Schaefers, Fraunhofer-Institut / Ecotoxicology; H. Hollert, RWTH Aachen University / Institute for Environmental Research

Chemical exposure to endocrine disruptors can have adverse outcomes on organism health and function; however, the current reliance on end-points such as egg number, plasma VTG content and morphological changes to determine effects of endocrine disrupting chemicals has given rise to series of questions related to chemicals exhibiting similar effects but different mode-of-action (MoA). Mechanistic identification of biological responses preceding to apical endpoints has become crucial for analyzing, accessing and determining chemical effects. Proteomics, therefore, show appreciable promise as a molecular screening tool for identifying specific alterations between exposures and controls, which is therefore imperative in discriminating endocrine disruptors from substances with a non-endocrine MoA. Such tool waives the need for elongated higher-tier testing. The main aim of this study is to identify alteration in molecular-toxicity pathways that are specific to chemical-induced apical responses in zebrafish. The study focused on fadrozole, a known inhibitor of cytochrome P450 aromatase. Thus an excellent model substance to evaluate and validate proteomic methods with the integration of organ-specific effects. Spawning adult zebrafish groups (5 males, 5 females) maintained at 25-26°C on a 16:8 h light/dark cycle; were exposed for 21 days to fadrozole (0, 0.1, 1, 10 µg/L) and analysed for plasma vitellogenin content, egg numbers and organ histopathology. Livers and gonads were isolated for shotgun proteomics and qPCR to characterize substance induced specific molecular toxicity pathways. Proteins involved in steroid hormone secretion and estrogen stimulus such as vtg1, vtg3, vtg6 and lman1, were significantly deregulated. Several of the prominently affected pathways involved regulation of xenobiotic stimulus, lipid metabolism, metabolic processes, TCA metabolism and calcium signalling. Our study demonstrated that the downstream induced-estrogen receptor suppression by aromatase inhibition triggered the downregulation of estrogen synthesis, which was assumed to induce the observed decrease in egg numbers and oocyte atresia with membrane folding in the ovary. We anticipate that this improvement leads to the identification of reliable biomarkers to determine chemical-induced adverse outcomes of ecological relevance in order to avoid unnecessary extensive testing.

53

Zebrafish embryos are able to conduct complex biotransformation processes and activation of chemicals

E. Küster, Helmholtz Centre for Environmental Research, Dept.Bioanalytical Ecotoxicology / Department Bioanalytical Ecotoxicology; B. Seiwert, Helmholtz Centre for Environmental Research- UFZ / Department Analytical Chemistry; S. Speer, Helmholtz centre for environmental research - UFZ / Dept. Bioanalytical Ecotoxicology; S. Brox, Helmholtz centre for environmental research - UFZ / Department Analytical Chemistry; T. Reemtsma, Helmholtz Centre for Environmental Research / Department Analytical Chemistry; S. Scholz, Helmholtz Centre for Environmental Research / Department Bioanalytical Ecotoxicology

Zebrafish embryos are proposed and partially already used as replacement for tests with (adult) animals conducted for human and environmental hazard assessment. There has been concern that they may exhibit a limited biotransformation which could interfere with prediction of adverse effects for safety assessment. The concern was provoked by anecdotal observation of the lack of activation for some compounds leading to a weak acute toxicity in fish embryos. However, internal concentration profiles of parent compounds suggest that fish embryos are principally capable for metabolic transformation of chemicals (Kühnert et al., 2013, Environ. Toxicol. Chem. 32, 1819–1827). In order to assess the biotransformation more systematically we studied the biotransformation of two pharmaceuticals (clofibrate, celecoxib). Overall similar transformation products could be observed in zebrafish embryo as known from human studies. Interestingly, the ratios of the different products in fish embryos seemed to be different from the ratio in humans. Biotransformation is of particular relevance for compounds that require (metabolic) activation to elicit their intended biological effect or where activation "accidently" produces more toxic substances. Organophosphates (OP) can serve as a model compounds for a proof of concept for activation in fish embryos as in many cases activation is mediated by cytochrome P450 oxidation. Therefore, we studied the activation of organophosphates by comparing the inhibition of acetylcholinesterase (AChE) activity for the parent compound and the oxon-metabolite in homogenates and intact embryos. For homogenates only the oxon metabolites were able to provoke AChE inhibition in a concentration dependent manner. In intact embryos inhibition was also found for the parent compounds. The inhibition EC50 was gradually reduced in later stages of embryos, if 24h exposure intervals for different stages were compared. The findings suggest that fish embryos are principally able to activate organophosphates and potentially also other compounds and that later embryonic stages may exhibit advanced biotransformation capacity.

54
Differing PM2.5 Filter Extraction Methods: Impact on Chemical and Toxicological Analyses
C. Roper, Oregon State University / Department of Environmental and Molecular Toxicology; S. Simonich, Oregon State University / Depts of Chemistry and Environmental Molecular Toxicology; R.L. Tanguay, Oregon State University / Sinnhuber Aquatic Research Laboratory and the Environmental Molecular Toxicology
Toxicology research is essential to improve the understanding of the global public health burden of fine particulate matter ($PM_{2.5}$) exposures. However, research groups use differing filter extraction methods to prepare $PM_{2.5}$ and the potential toxicity bias from different extraction methods is rarely considered, possibly eliminating inter-laboratory comparisons and misrepresenting the toxic responses to $PM_{2.5}$ constituents. To determine the impact of filter extraction methods on chemical constituent recovery and toxicity outcomes we took equal portions of a single hi-volume $PM_{2.5}$ filter sample collected in Riverside, CA. Each filter portion underwent a different extraction method (n=6) and recovered $PM_{2.5}$ was then prepared for developmental toxicity testing by collecting the soluble fraction from DMSO extraction. Zebrafish (n=32/treatment) were treated with controls (DMSO, blank filter portions) and treatments ($PM_{2.5}$ filter portions undergoing filter extraction) starting at 6 hours post fertilization. Aliquots of these $PM_{2.5}$ solutions were used for chemical constituent analysis of polycyclic aromatic hydrocarbons (PAHs, n=120) and elements (n=20). Significant increases in mortality were observed for $PM_{2.5}$ from 5 of the 6 filter extraction methods when compared to both the DMSO and blank filter controls. Combined mortality and morphological changes were significantly increased following $PM_{2.5}$ treatment in all extraction methods compared to DMSO controls. Importantly, two of the methods showed significant mortality and morphological changes with blank filters when compared to DMSO controls. Chemical analysis is underway and differences in $PM_{2.5}$ solutions between extraction methods will be investigated. Correlations between chemical components and developmental toxicity outcomes will identify components that are driving toxicity and potentially altered during specific extraction procedures. This research highlights the toxicity bias due to $PM_{2.5}$ filter extraction methods that must be considered when conducting research with complex ambient mixtures. Ultimately, this work identifies extraction procedures for use in this cost-effective surrogate to compare the inherent toxicity differences of $PM_{2.5}$, and provides a path that will ultimately promote improved understanding of $PM_{2.5}$-associated health effects.

Safeguard and Conservation of Cultural Heritage: the contribution of chemistry

55
Cultural Heritage and Climate Change: impact and adaptation
C. Sabbioni, CNR-Istituto di Scienze dellAtmosfera e del Clima
Cultural heritage, which is a non-renewable resource, is a sector extremely complex for the diversity of materials, structures and systems. The access to citizens and visitors need to be favoured, but at the same time, it is our responsibility to transmit this heritage we received from the past to the future generations. It is urgent to include cultural heritage in the value chain of sustainable development: the priority that faces the world today. Research on the threats that climate change will have on cultural heritage has been very limited until now and it has not yet generated policies designed to mitigate the impact and to develop preventive adaptation strategies. The presentation will be focused on future scenario on the effects of climate variables on the vulnerability of cultural heritage at European level. Recommendations on the inclusion of cultural heritage in the national adaptation strategies and plans to climate change will also be discussed.

56
Nanotechnologies for the conservation and connected risks
M.J. Mosquera, University of Cadiz
Most products commonly employed in the restoration and conservation of cultural heritage materials have not been specifically developed to preserve such elements. In addition, they are plagued by limited performance and structural drawbacks such as low adhesion, poor penetration, and cracking. Another disadvantage is the requirement for most products to be dissolved in volatile organic compounds (VOCs), which produce environmental and human health risks in their use. In this lecture, I will review the most meaningful achievements of my group in this field. We have developed an innovative sol-gel route for preserving Cultural Heritage building materials. Specifically, a surfactant-assisted sol-gel synthesis to produce, in-situ on the building, crack-free nanomaterials to be used as long-term consolidants. Additionally, hydrophobic, water-repellent, self-cleaning, and biocidal properties can be incorporated into the product by innovative chemical modifications of the proposed synthesis route. Finally, I will summarize the future challengers of our group related to conservation of historic concrete in the framework of the Horizon 2020 project “InnovaConcrete”.

57
Towards the European Research Infrastructure in Heritage Science: E-RIHS
L. Pezzati, CNR-Istituto Nazionale di Ottica
The European Research Infrastructure for Heritage Science (E-RIHS) entered the European strategic roadmap for research infrastructures (ESFRI Roadmap) in 2016, as one of the six new projects. E-RIHS support research on heritage interpretation, preservation, documentation and management. Both cultural and natural heritage are addressed: collections, buildings, archaeological sites, digital and intangible heritage. E-RIHS is a distributed research infrastructure with a multi-level star-structure: facilities from many Countries will be organized in national networks, coordinated by separate National Hubs, and the E-RIHS Headquarters will provide the unique access point to all E-RIHS services, by coordinating the net of National Hubs.

58
Discussion & Conclusions

Modelling and monitoring of pesticides fate and exposure in a regulatory context (II)

59
Scenario Development for Off-field Soil Exposure and Risk Assessment
M. Wang, WSC Scientific GmbH / Dept Efate Modelling; J. Kleinmann, WSC Scientific GmbH; T. Schad, Bayer Ag / Environmental Modelling; K. Hammel, Bayer AG, Crop Science Division / Environmental Safety; G. Ernst, Bayer Ag / Ecotoxicology; G. Goerlitz, Bayer CropScience AG / Environmental Safety; P. Neumann, Bayer Ag; S. Bub, Tier3 Solutions GmbH
In its Scientific Opinion on risk assessment for in-soil organisms EFSA proposes a preliminary approach for off-field soil exposure by adding up entries from the different major exposure routes. EFSA indicates the worst-case character of its scenario "In the absence of appropriate off-field exposure scenarios…", and hence, emphasises the necessity for model and scenario development. The present work aims to undertake first steps (i) to develop a model approach for off-field/off-crop soil exposure due to runoff, erosion, and drift, (ii) to develop exemplary schematic and real-world scenarios, (iii) which allow to gain insights in off-field soil exposure and risk using case studies. The developments are based on the Specific Protection Goals options. A tiered modelling approach is presented which allows to build exposure scenarios ranging from simple schematic and conservative to more realistic landscape-scale tiers, which can be easily linked to effect modelling (toxicological, population, community). Results are intended to support the design of off-field soil exposure and risk characterisation scenarios and the development of assessment endpoints relevant to address SPGs.

60
Biogenic residues formation from pesticides - an overview
K. Nowak, TU Berlin / Institute for Environmental Research (Biology V); A. Miltner, Helmholtz Centre for Environmental Research UFZ / Department of Environmental Biotechnoloy; M. Kästner, Helmholtz centre for environmental research - UFZ / Department of Environmental Biotechnology
Anthropogenic organic chemicals are deliberately (e.g. pesticides) released in major amounts to nearly all compartments of the environment. Soils and sediments as complex matrices provide a wide variety of binding sites and are the major sinks for these compounds. Pesticides entering these complex systems may undergo various turnover processes. They can be degraded chemically (e.g. photolysis), biologically by microorganisms, volatilised leached to the groundwater or taken up by living organisms or immobilised in the form of non-extractable residues (NER). Microorganisms can use C and N from a pesticide to synthesize their biomass compounds, e.g. amino acids (AA) and fatty acids (FA). The extraction of known microbial biomarkers from soil or sediment after addition of C and N isotope tracer allows an estimation of microbial activity in the transformation of pesticide. We investigated the turnover of different pesticides (2,4-D, glyphosate, metamitron, bentazone, bromoxynil and clodinafop-propargyl) with the particular focus on the metabolic incorporation of the isotope label into AA, FA and their fate over time. An agricultural soil and water-sediment were incubated with stable isotope labelled respective herbicide in the dark and at constant temperature (20°C). Soil and sediment samples at the respective sampling date were analysed for the amount and the isotopic composition of AA, FA, CO_2, solvent-extractable parent compound and metabolites and total NER. The presented data indicated that easily biodegradable herbicides e.g. glyphosate, 2,4-D or metamitron were utilized as a carbon (and

nitrogen) source and the NER were mainly biogenic. The major formation of biogenic residues is supposed to be relevant for easily biodegradable contaminants under significant CO_2 formation. However, in the case of more recalcitrant pesticides like bentazone, the incorporation of C into microbial biomass, although reported to be very low, cannot be completely excluded.

61

Derivation of a foliar wash-off factor for FOCUS modelling based on literature research

S. Sittig, DR. KNOELL CONSULT GmbH / E-Fate Modelling; C. Wollmann, Dr Knoell Consult GmbH; G. Reinken, Bayer AG, Research & Development, Crop Science / Environmental Safety

After foliar application, plant protection products (PPP) undergo several routes of dissipation of which one is precipitation-induced wash-off from the canopy. This process is accounted for in the European exposure assessment framework for PPP authorization and included in the corresponding numerical models, e.g. FOCUS PEARL, PELMO, PRZM and MACRO. A numerical wash-off factor for modelling is applied, quantifying the wash-off from plant surfaces by a given amount of precipitation. Consequently, this factor is relevant for the calculation of predicted environmental concentrations (PEC) for the compartments soil, groundwater, and surface water. In case a measured wash-off factor is not available, a default value is to be applied. An increase of this default value from 0.5 cm^{-1} to 1 cm^{-1} has been proposed by EFSA, which results in more exhaustive wash-off from the plant surface. Generally, the extent of rainfall-induced displaced substance depends on several factors. An ECPA working group recommended a harmonized experimental approach to derive wash-off factors in the greenhouse: a 24h time interval between pesticide spraying and 10 to 20 mm of artificial rain, followed by an extraction of the plant material with an acetonitril/water mixture of 80:20 (v/v). This standardized experimental procedure has been defined in order to derive a reliable numerical wash-off factor as input for FOCUS modelling. In this study, the pertinent literature was reviewed for the availability of data suited for the calculation of a wash-off factor, reflecting a variety of different investigation types in terms of time of (artificial) rainfall after application, rainfall amount and intensity, formulation, crops under investigation, etc. Published experimental wash-off studies are usually not conducted according to the standardized experimental procedures. Thus, only a limited number of the published studies are suitable to derive a wash-off factor for modelling. The outcome of the literature review presented herein suggests that a meaningful default wash-off factor should be well below 1 cm^{-1}. Keeping the existing default value of 0.5 cm^{-1} retains a sufficient protection level while at the same time avoids a large number of unnecessary refinement studies.

62

Application of a dynamic aquatic food web model for FOCUS exposure assessment

L. Padilla, Stone Environmental, Inc.; A. Del Signore, D. Sprenger, L. Weltje, BASF SE / Crop Protection Ecotoxicology; M.F. Winchell, Stone Environmental, Inc. / Environmental Systems Modeling

In a regulatory setting, the potential for bioaccumulation and biomagnification of plant protection product active substances in aquatic organisms is evaluated with simple screens on the basis of a substance's log K_{OW}, where typically a value greater than or equal to 3 indicates concern. However, this criterion may lead to false positive identification, because it does not account for biotransformation of the substance in aquatic organisms or bioavailability in the environment. Dynamic aquatic food web models are more refined tools for determining bioaccumulation and biomagnification potential, because they can account for chemical bioavailability and temporal and spatial variability in exposure concentrations due to seasonal and regional differences in weather and agricultural practices. The aim of this work is to demonstrate a modelling approach that couples standard FOCUS landscape and water body models with a dynamic aquatic food web model to assess whether a hydrophobic insecticide with logK_{ow} above the screening threshold of 3 will bioaccumulate/biomagnify. The Simon Fraser University (SFU) aquatic food web model, which predicts chemical concentrations in biota at six different trophic levels within an aquatic ecosystem, was selected based on the availability of data for relatively few input parameters and its demonstrated capability to predict observed chemical concentrations for a wide range of species, chemicals, and aquatic environments. To maximize relevance for agricultural systems in Europe, the food web model was adapted to accept environmental concentration time series input from the established TOXSWA model used in EU pesticide registration procedures. The modelling approach leveraged the transient form of the aquatic food web model to account for time-varying pesticide loadings characteristic of agricultural settings. Modelling results included both the daily time series predictions of organism concentrations for the six trophic levels and the uptake and elimination rate constants calculated from organism sub-models. In total, nine FOCUS scenarios were simulated and compared (five drainage scenarios with MACRO and four runoff scenarios with PRZM) and dominant organism uptake pathways were identified.The approach may be used to refine log(Kow)-based screening bioaccumulation and biomagnification evaluations for regulatory purposes.

63

Improved assessment of pesticide peak exposure in cultivated mountain watersheds

M. Morselli, University of Insubria / Department of Science and High Technology; E. Terzaghi, University of Insubria (Como) / Department of Science and High Technology, Como; A. Di Guardo, University of Insubria / Department of Science and High Technology

Agricultural activities can involve the use of plant protection products (PPPs) and the use of such chemicals can occur near surface waters bodies, thus creating a potential for adverse effects on aquatic ecosystems. Due to the spatio-temporal variability of chemical applications and of the processes regulating their fate and transport to surface waters, ecosystems are often exposed to pulses of contaminants. In certain environmental scenarios, such as small mountain watersheds, where runoff fluxes are particularly rapid due to side slopes, exposure peaks can be shorter but much higher. Monitoring campaigns are often inadequate or too expensive to be carried out and modelling tools are therefore vital for exposure assessment and their use is encouraged by current legislation. However, currently adopted models and scenarios (e.g., FOCUS for PPPs) are often too conservative and/or "static" to accurately capture exposure variability, and the need for more realistic and dynamic tools is now one of the major challenges for risk assessment. In a previous work, the new fate model DynAPlus was developed to improve pesticide fate predictions in cultivated mountain basins. The model was successfully evaluated against chlorpyrifos water concentrations measured in the Novella River (Non Valley, Northern Italy), where more than 1000 ha of apple orchards surround the river and its tributaries. However, the need for some model improvements and application to other chemicals and scenarios was highlighted. In this work, the DynAPlus model was improved to increase realism, by including vegetation to both terrestrial and aquatic environments, dissolved organic carbon (DOC) in water, and a soil-erosion module to compute particle-mediated chemical transport to surface waters. The improved DynANet was first applied to the Novella River case study and the new predicted chlorpyrifos concentrations were compared to measurements, suggesting an improvement of model performance, particularly due to the inclusion of the soil-erosion module and DOC. The model was then parameterized to simulate another cultivated mountain basin located in Northern Italy and applied to simulate the fate of three pesticides with different physical-chemical properties and persistence. The resulting peak exposure profiles were discussed to highlight the added value of such a dynamic modelling approach in providing information on exposure which could not emerge from the application of current approaches.

64

Implementation of mitigation measures and assessment of its impact under field-specific environmental conditions in the risk indicator SYNOPS-WEB for Norway

A. Dominic, Julius Kuehn Institute / Institute for Strategies and Technology Assessment; O. Eklo, Norwegian Institute of Bioeconomy Research NIBIO / Department of Pesticides and Natural Products Chemistry; M. Stenrød, Norwegian Institute of Bioeconomy Research (NIBIO); E. Solbakken, R. Lågbu, Norwegian Institute of Bioeconomy Research; P. Horney, Julius Kühn Institut; D. Daehmlow, Julius Kühn-Institute / Institute for Strategies and Technology Assessment; B. Golla, Julius Kuehn Institute / Institute for Strategies and Technology Assessment; J. Strassemeyer, ral Research Centre for Cultivated Plants / Institute for Strategies and Technology Assessment

In response to the implementation of the EU-directive on sustainable pesticide use by Norway, the project SMARTCROP (funded by the Research Council of Norway), was started to address the challenges of developing and providing farmers with the necessary IPM tools. Towards this objective, SYNOPS, a risk indicator developed by the Julius-Kühn Institut, Germany, was adapted for Norway and provided with a graphical user interface such that a farmer or non-expert could perform risk assessments for field-specific pesticide applications. US-EPA PRZM5 and VFSMOD have been incorporated in SYNOPS for a more realistic modelling of the runoff/erosion modules and the functioning of the vegetated filter strip. The Norwegian tool, *SYNOPS-WEB, Norway* is available in both English and Norwegian. It uses Norwegian land-use, surface water and soil data, plant protection products registered for use in Norway, modified crop data for Norwegian conditions and station-based daily weather data. Risk assessments are carried out for specific field and application scenarios. Another important new feature is the implementation of various mitigation measures such as vegetated filter strips, hedges, tillage/mulch, and cover crops. Risk assessments can be performed for a combination of mitigation measures in order to select the optimal application strategy under specific field conditions. In this presentation, we describe and discuss the mitigation measures implemented in *SYNOPS-WEB, Norway* and the corresponding adjustments to the model input parameters. We provide example scenarios based on realistic application patterns, without and with mitigation measures. Aquatic and terrestrial risk indices are presented to the user as Exposure Toxicity Ratios (ETR) in the form of colour-coded tables for an easy visual appraisal of the environmental risk under different conditions. In addition the predicted environmental concentrations can be viewed on a daily basis for the selected time period.

65

Acute Toxicity of Pyrene Associated with Dissolved Organic Matter of Various Molecular Weights to Daphnia magna

H. Lin, X. Xia, S. Bi, X. Jiang, H. Wang, W. Wen, School of Environment, Beijing Normal University

Dissolved organic matter (DOM) is a key environmental factor for the toxicity of hydrophobic organic compounds (HOCs) in natural waters. However, the toxicity of DOM-associated HOCs is still not clear. In this research, pyrene was selected as a model HOC and its freely dissolved concentration (C_{free}) was maintained by passive dosing systems. The immobilization and enzymatic activities of Daphnia magna were examined to analyze the toxicity of DOM-associated pyrene. The results indicated that the immobilization of Daphnia magna in the systems containing various molecular weight DOM and pyrene was ordered as middle molecular weight (MMW, 5-10K Da) DOM > higher molecular weight (HMW, > 10K Da) DOM > lower molecular weight (LMW, < 1K Da, 1-3K Da, and 3-5K Da) DOM. Furthermore, the superoxide dismutase (SOD), catalase (CAT), and peroxidase (POD) activity of Daphnia magna decreased gradually with the increasing C_{free} in the systems of MMW and HMW DOM, whereas increased when C_{free} was at a low level and then decreased when C_{free} was at a higher level in the control group with pyrene only and the system of LMW DOM. The influencing mechanisms of DOM molecular weight on the toxicity of DOM-associated pyrene to Daphnia magna were related with the amount of pyrene sorbed on DOM, the uptake routes of DOM by Daphnia magna, and the desorption of pyrene from DOM in the gut of Daphnia magna. The findings obtained in this research suggest that the toxicity of DOM-associated HOCs should be taken into account for the eco-environmental risk assessment of HOCs in water systems.

66

Passive dosing for constant concentration and defined composition of hydrophobic organic mixtures

R. Hammershøj, Technical University of Denmark / Department of Environmental Engineering; H. Birch, DTU Environment / Department of Environmental Engineering; K. Knudsmark Sjøholm, University of Copenhagen / Dept. of Plant and Enviornmental Sciences; P. Mayer, Technical University of Denmark / Department of Environmental Engineering

There is regulatory and scientific attention on the fate, exposure and effects of chemical mixtures including complex mixtures of hydrophobic chemicals such as petroleum substances and essential oils. These mixtures have recently been categorized as multicomponent substances and substances of unknown or variable composition, complex reaction products or biological materials (UVCBs). The dosing of such mixtures in environmental experiments and tests can be challenging and there is an urgent need for new methods to provide stable concentrations and defined composition of these mixtures in aquatic fate and toxicity testing. Passive dosing from a silicone rod has successfully been used in biodegradation and toxicity testing of hydrophobic chemicals covering a broad chemical space in terms of K_{ow} and K_{aw}. This study aims to extend the applicability of the novel passive dosing method to hydrophobic multicomponent substances and UVCBs (i.e. complex mixtures). The method is straightforward: a silicone rod is loaded by direct addition of the mixture and subsequently equilibrated with ultrapure water to create constant and defined concentrations of each mixture constituent and thus also a constant and defined mixture composition. The aqueous concentration level can be controlled by the amount of mixture added to the rod. Early results show a good performance of the method with very fast dosing kinetics, aqueous concentrations increasing linearly with loading level and good reproducibility of the passive dosing for a petroleum substance and an essential oil. The presentation will focus on 1) the fast and reproducible loading of selected UVCB mixtures, 2) reproducibility and optimized passive dosing kinetics for one petroleum substance and one essential oil and 3) the performance and characteristics of the passive dosing method compared with more traditional dosing methods.

67

Biodegradation of volatile substances in soil - Challenges and optimization of test setups (OECD 307)

P. Shrestha, Fraunhofer Institute for Molecular Biology and Applied Ecology IME; B. Meisterjahn, Fraunhofer Institute for Molecular Biology and Applied Ecology IME / Ecological chemistry; C. Hughes, Shell Health / Risk Science Team; P. Mayer, Technical University of Denmark / Department of Environmental Engineering; H. Birch, DTU Environment / Department of Environmental Engineering; M. Klein, Fraunhofer Institute for Molecular Biology and Applied Ecology IME; E. Vaiopoulou, European Petroleum Refiners Association; D. Hennecke, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Ecological chemistry

Higher tier biodegradation laboratory tests in soil, sediment and/or surface water systems are conducted using standard OECD guidelines. As stated in these guidelines, they are not suitable for testing volatile chemicals, however a threshold based on Henry´s law is not defined, except in OECD 309. In the actual setups, incomplete mass balance is a major problem while testing volatile chemicals. Optionally, OECD 307 and 308 allow biometer-type incubation setups but it does not require any data to prove if the systems remains aerobic. In addition, the degradation kinetics in a closed test system can largely be influenced by air-water partitioning as described by Birch et al. 2017. Our objective was to design a closed incubation test set up where maintaining and measuring of aerobic conditions was possible without the loss of test chemical. Additionally, a full scale OECD 307 with two model chemicals was performed to check the reproducibility of data in terms of mass balance and to better understand the obtained degradation data. The test setup consisted of 100 mL flask with 50g soil, CO_2-trap and a Tenax tube completely closed using a stainless steel lock system. Oxygen saturation in the headspace was measured in a reference sample using optical measurements without the need to open the vessel. If the oxygen saturation was < 15%, the samples was aerated with oxygen-rich air. Applying this setup, degradation of ^{14}C-labelled Tetralin and Decane was conducted on four soils with different texture, organic carbon and microbial activity. At sampling dates the headspace air of the samples was stripped off through the Tenax tube using a vacuum pump. The Tenax tube and the CO_2-trap were taken for analysis and the soil was taken for extraction using appropriate methods. The solid extraction residue was subject to combustion analysis to determine the non-extractable residues (NER). The average overall recovery of 99.29% (N=90) for Decane and 104.34% (N=90) for Tetralin with a variation (between the individual replicates) < 11% for both studies suggests that obtaining a complete mass balance with the new test setup was reproducible. It was observed that radioactivity adsorbed on Tenax (100% parent), was higher in the soils with lower OC content suggesting that sorption of the chemical affecting its volatilization and hence degradation. Thus, how to deal with the volatilized parent fraction while calculating degradation kinetics is still a part of ongoing research.

68

Untangling the biodegradation of hydrophobic chemicals in OECD and novel tests using a unified modelling approach

F. Polesel, Technical University of Denmark (DTU) / DTU Environment; A. Brock, DTU Environment / DTU Environment; F. Stibany, K. Smith, RWTH Aachen University / Institute for Environmental Research; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics; M. Kästner, Helmholtz centre for environmental research - UFZ / Department of Environmental Biotechnology; S. Trapp, Technical University of Denmark DTU / DTU Environment

Persistence assessment using standardized (e.g., OECD) tests is one of the main challenges for hydrophobic organic chemicals (HOCs). While the use of radiolabeled chemicals brought major advances, mechanistic models can still provide deeper insights in experimental results and underlying processes. In this context, the key objective of this study was to test the applicability of a unified modelling approach across the spectrum of OECD degradation tests and newly developed experimental tests for HOCs. We specifically aimed at (i) elucidating biodegradation kinetics and improving their estimation by including a new method for microbial yield calculation; (ii) determining ^{14}C fractions (mineralized, incorporated in biomass and non-extractable residue NER) at the end of tests as persistence indicators. The unified model for sorption and biodegradation (in combination with the Microbial Turnover to Biomass growth yield estimation method) was used to predict mineralization to CO_2, growth of degrading microorganisms and NER formation in aerobic degradation tests with selected ^{14}C-labeled HOCs (triclosan, pentachlorophenol—PCP, propargite and pyriproxyfen). Model predictions were fit to experimental results obtained elsewhere in conventional degradation tests activated sludge or soil or in novel passive dosing setups, in the presence of a single degrader strain. Overall, good agreement between model predictions and empirical data was shown by adjusting only the ratio v_{max} / K_s, which describes biodegradation kinetics according to the Michaelis Menten equation. Overall, a high range of v_{max} / K_s values was shown for the selected substances(0—55 $m^3 g^{-1} d^{-1}$), indicating that both limited bioavailability and intrinsic recalcitrance can explain HOC persistence. This study represents a first attempt of using a unified modelling approach for predicting biodegradation of HOCs across a variety of tests, showing promising results towards persistence prediction of organic chemicals during regulatory screening. Ongoing research is focusing on extending the model applicability by (i) including the formation of intermediate transformation products; (ii) determining (de)sorption limitation based on dedicated experiments; and (iii) using uncertainty-based approaches to support decision makers within REACH.

69

History of polychlorinated biphenyl deposition to snow and ice from the Lomonosovfonna glacier, Svalbard.

M.H. Hermanson, Hermanson & Associates LLC / Environmental Chemistry; O. Garmash, University of Helsinki; E. Isaksson, Norwegian Polar Institute; C. Teixeira, D.C. Muir, Environment and Climate Change Canada / Aquatic Contaminants Research Division

Polychlorinated biphenyl (PCB) deposition to the north polar regions has long been recognized. Here we investigate quantitative and qualitative PCB inputs to the Lomonosovfonna glacier on Svalbard from an ice core drilled in 2009, and a snow pit from spring 2010. Lomonosovfonna is the highest-elevation ice on Svalbard at 1250 masl. It is above the tropospheric boundary layer at all times of year (maximum ~1000 masl), so all of the contaminant inputs have sources from long

distances, and do not include any local PCB sources on Svalbard. Total PCB deposition history to the core and surface snow shows that amounts have not declined in net amount since the mid 1950s. The peak flux in the surface snow and to ice core layer is 18.5 - 19 pg cm-2 yr-1. Average 5-day air mass back trajectories from peak flux periods beginning in 1998 show more frequent 5-day air mass trajectories from Russia, and from Europe south of 60o N latitude, particularly extending into the U. K., relative to 1989-1998, which had 5-day trajectories ending in the far north of Russia and Norway. The surface snow PCB congener profile is dominated by PCB 110, 70+74, 101, 95, 11. Combined, these five congeners represent ~27% of ?PCB. The upper-most ice core sample is dominated by PCB 95, 52, 101, 110, 70+74, which represent ~42% of ?PCB. The most apparent difference between the two profiles is the lower amount of dichloro- and trichloro- congeners in the ice core sample in Figure 1B. The indication is that the more volatile congeners in the dichoro- and trichloro- homologues are deposited to surface snow shown in Figure 1A, but are volatilized back to the atmosphere during periods of higher summer air temperature. The percent of PCB-11 throughout the samples ranges from 0.90-3.4% and is present in all samples dating from 1957. It is the dominant congener among mono- and di-chloro PCBs. This PCB congener has very low or no presence in Aroclor products, and apparently is not found in other PCB parent mixtures. Its source is often considered to dairylide yellow pigment or products containing it. Its presence in the environment is sometimes associated with disposal of products containing this pigment. The source of PCB-11 to Lomonosovfonna is uncertain, but could be waste incineration facilities in Europe where it has been found in flue gas.

70

Environmental occurrence and distribution of organic UV stabilizers in the sediment of the Bohai and Yellow Seas

C. Apel, Helmholtz-Zentrum Geesthacht; J. Tang, Yantai Institute of Coastal Zone Research, CAS; R. Ebinghaus, Helmholtz-Zentrum Geesthacht / Department for Environmental Chemistry

Organic UV stabilizers are of emerging environmental concern due to their large production volumes and potential endocrine disrupting properties. UV stabilizers are widely used in plastic products, paints and coatings to improve the product stability against UV light. Furthermore, some UV stabilizers are approved as ingredients in personal care products like cosmetics and sunscreens. The pathways into the marine environment are either indirect by wastewater treatment plant discharges or direct by recreational activities like bathing and swimming. Four benzotriazole UV stabilizers are classified as SVHC (Substances of Very High Concern) under the EU legislation REACH. Numerous others are currently listed under the European community rolling action plan (CoRAP) to be (re-)evaluated in the next years. Due to their chemical properties, most UV stabilizers accumulate in sediment (logKow > 3) and have potential for persistence or pseudo-persistence. Environmental data for the coastal and marine environment are sparse. For this study 74 surface sediment samples of the Bohai and Yellow Seas were analysed for 19 commonly used organic UV stabilizers. The sample pretreatment and analysis was carried out as following: First, the samples were homogenized with sodium sulphate. Afterwards, extraction and clean-up was performed using an accelerated solvent extraction (ASE-350, DIONEX, Germany) method. For this, 22 mL stainless steel ASE cells were filled with 3 g of 10% deactivated silica and approximately 5 g sediment that was spiked with appropriate isotopically labelled standards. The cells were extracted using dichloromethane for three 15 min-cycles at 100 °C. The extracts were solvent-changed to methanol, reduced in volume to 150 µL. The instrumental analysis was performed on a LC-MS/MS system (1290 Infinity coupled to 6490 triple quadrupole LC/MS, Agilent Technologies, Germany) equipped with an APPI-source and a C18 column (Eclipse Plus RRHD 1.8 µm, 2.1 x 150 mm, Agilent Technologies, Germany). This study shows levels of contamination and distribution of organic UV stabilizers in surface sediments of the Bohai and Yellow Seas for the first time. 14 substances have been positively detected in concentrations in the low ng/g dw range. Characteristic pollution profiles and distribution pattern have been identified, which indicate to different indirect sources of UV stabilizers into the study area.

LCIA method developments in a global perspective: Status and outlook (I)

71

Implications of spatial differentiation on LCA-based decision-making: a case study of biochar systems in Indonesia

M. Owsianiak, Technical University of Denmark; G. Cornelissen, S. Hale, Norwegian Geotechnical Institute; H. Lindhjem, Menon Economics; M. Sparrevik, NTNU

The development of spatially differentiated life cycle impact assessment (LCIA) methods and their use in regionalized life cycle assessment (LCA) has intensified in the past few years. However, it is less investigated whether spatial differentiation leads to more correct decisions based on the LCA, in addition to just more accurate and realistic LCIA results. The aim of this work was therefore to assess the implications of spatial differentiation on the interpretation phase of a comparative LCA. Biochar production from biomass residues and its use as soil conditioner in Indonesia was used as case study. Comparisons were made between 4 villages, 3 biochar production techniques, and 2 fertilization strategies. Results showed that (i) regionalized impact scores for individual impact categories either increased or decreased compared with site-generic scores, depending on the impact category (by up to 1 order of magnitude); (ii) total damages to human health were approximately 3 to 5 times higher when compared to site-generic scores and (iv) irrespective of the geographic locations, regionalized total damages to biodiversity were close to site-generic scores. This is mainly because of trade-offs between categories, where increase in impact scores for some categories was compensated by decrease in others. Overall, irrespective of the approach to spatial differentiation in LCIA, biochar production and use in agriculture is generally expected to bring environmental benefits. When parameter and inventory uncertainties were considered, there was no influence of spatial differentiation on identification of best performing villages in terms of total damage to human health and ecosystems, although village performing worst with regard to total damage to human health changed. There was a general tendency that biochar production using both Kon Tiki and Adam retort kilns performed better than earth-mound kiln, and furthermore biochars brought largest benefits where no-biochar agricultural production systems were based on inorganic fertilizers. This rather consistent ranking was mainly due to relatively large geographic differences in life cycle inventories between villages, which were often larger than geographic differences in characterization factors between site-specific and site-generic approaches. Thus, although spatial differentiation improved accuracy and realism of environmental impacts in this comparative case study, it did not necessarily contribute to more correct decisions.

72

Considering space debris related impacts within the LCIA framework

T. Maury, University of Bordeaux / ISM-CyVi; P. Loubet, CyVi-ISM / ISM CyVi; A. Gallice, ArianeGoup / Design for Environment; G. Sonnemann, University of Bordeaux / ISM CyVi

The space sector is a new area of development for LCA studies. The European Space Agency (ESA) has been working since 2012 on environmental issues for space activities through its *Clean Space Initiative*. ArianeGroup, which is the prime contractor for the development of the new Ariane 6, is currently performing an LCA of this launcher in exploitation phase (ESA's contractual requirement). However, the current studies adopt a *Cradle-to-Launch pad* approach due to lack of relevant modelling for use and disposal phase. In addition, a rising sustainability concern is occurring in the space sector particularly regarding impact of space debris: 29,000 human-made objects, larger than 10cm, are orbiting the Earth but only 6% are operational spacecraft, being today a significant and constant danger for all space missions. Consequently, considering end-of-life management during the design of space missions becomes necessary to ensure a sustainable use of space orbits. Given this situation, there is an opportunity to make the link between space debris concern and eco-design of spacecraft (satellites & launchers) using the LCA methodology. A focus should be put on the comparison of several missions & post-mission disposal scenarios to study potential trade-offs between typical impact categories (e.g. toxicity and climate change), but also with regard to the growing issue of space debris. Hence, our challenge is to integrate, *via* a dedicated additional indicator, space debris related impacts within the LCIA to broaden the scope of LCA for space systems. The Area-of-Protection Resources has been identified to reflect the depletion of available orbits by the potential generation of space debris. Considering generation of debris in operating orbits with a resource depletion perspective allows us to address the environmental footprint of the spacraft's disposal. Volume occupation by debris and dead spacecraft leads to a decrease of the orbital resource availability enhancing the risk of collision/break-up and then propagation of new clouds of debris. As a consequence, the lack of access to the orbital resource in the future (scarcity) could be handled as environmental and socio-economic impacts. The presentation will prove the relevancy of this approach by presenting the impact pathway linking the occupation of operational orbits and environmental impacts. Environmental mechanisms and impacts (midpoints) will be exposed with associated specific characterization factors.

73

Implementing ozone formation effects due to poplar plantations for biomass production in Europe in life cycle impact assessment

P. Vercoulen, Radboud University; R. Kranenburg, C. Hendriks, TNO; R. Van Zelm, Radboud University / Department of Environmental Science

Poplar trees are known to emit volatile organic compounds, among them isoprene, which enhances tropospheric ozone formation. Ozone exposure, in turn, causes adverse effects to human health and ecosystems. In the context of an energy transition, it has been proposed to use poplar biomass for the generation of electricity. The goal of this research was to determine country specific characterization factors (CFs) for ecosystem damage due to ozone formation from isoprene emissions caused by poplar tree plantations in Europe. CFs were defined as the change in potentially affected fraction of plant species (PAF) due to a change in the country-specific poplar plantation area (in km^2_{veg}?yr/km^2_{poplar}) and consists of a fate factor and an effect factor. To determine the fate factor, changes in Accumulated Ozone over a Threshold of 40 ppb (AOT40) in all grid cells connected to isoprene emissions resulting from additional poplar plantations on 1% of agricultural land in each country were estimated with chemistry transport model

LOTOS-EUROS on a 0.5x0.25° scale. The effect factor was based on a lognormal relationship between the PAF of plant and grassland species and ground-level AOT40. To test our developed CFs, a case study was performed of electricity generation in a coal power plant for varying fuel mixtures of hard coal and poplar biomass. The functional unit was 1 kWh of electricity generated. ReCiPe2016 was used for the impact assessment of other impact categories. Largest CFs were shown in southern and central parts of Europe, which is mainly caused by larger effect factors in these areas. Largest impact was obtained for Slovenia, i.e. $7.6 \cdot 10^{-2} km^2_{veg}$?yr/$km^2_{poplar}$. The area-weighted European average CF was $2.9 \cdot 10^{-2}$ km^2_{veg}?yr/km^2_{poplar}. Case study results show that ozone formation caused by cultivation of poplar plantations can contribute significantly to overall ecosystem impacts due to electricity generation from poplar biomass. When fully using biomass for electricity production, cultivation of poplar plantations in Italy even contributed 20% to the total adverse effects to ecosystems. With our work ozone formation impacts due to poplar tree plantations for biomass production can be quantified. We showed that contributions to ecosystem damage from these emissions in an electricity generation case study were not negligible. Therefore, we recommend to include the CFs in future LCA case-studies where poplar plantations are included.

74

Relative potency approach for using in vitro information for calculating human effect factors in LCIA

B. Salieri, EMPA / Technology and Society Lab; M. Rösslein, J. Kaiser, Empa Swiss Federal Laboratories for Materials Science and Technology / Particles-Biology Interactions Laboratory; R. Hischier, EMPA / Technology and Society Lab; B. Nowack, Empa Swiss Federal Laboratories for Materials Science and Technology / Technology and Society Lab; P. Wick, Empa Swiss Federal Laboratories for Materials Science and Technology / Particles-Biology Interactions Laboratory

Although manufactured nanomaterials (MNMs) offer several advantages compared to their bulk form, several concerns have been raised on their environmental and human risk. The LCA methodology is a valid tool to assess the environmental and human impact of nanoproducts. But overall, LCA studies of MNMs and nanotechnology are currently affected by a gap of knowledge regarding the exposure and toxicity of MNM releases into the environment during different life cycle stages. Within the LCA methodology, the human toxic effect (EF) is evaluated based on ED_{50} extrapolated by *in vivo* studies or human studies. In vision of the "Toxicity Testing in 21st Century", the *in vivo* tests are expected to be replaced by *in vitro* tests. Also, for emerging materials such as nanomaterials, still a scarce number of *in vivo* data are available in literature. Given the expansion of *in vitro* testing, there is probably a potential to use outputs from such in vitro testing in order to derive ED_{50} values for the use in LCA studies. Here, we propose to integrate *in vitro* data in the assessment of the human toxic potential using a relative potency (RP) approach. The RP approach has been widely used by toxicologist for ranking chemical, to calculate the equivalent dose of one chemical that produce the same response as another at a specific dose or to define toxic equivalency factor. *In vitro* toxicity test for nano-CuO, nano-Ag and nano-ZnO have been performed on THP-1 cell line, CaCo-2 cells and Hep-G2 cell line. Based on the EC_{50} values obtained by the *in vitro* test a RP factor has been calculated and used to calculate the ED_{50} for the above mentioned nanoparticle.Therefore, we present a first attempt for the derivation of EF values for their implementation into LCIA factors for human toxicity, by using in vitro data values for nano-CuO, nano-Ag and nano-ZnO in combination with a relative potency (RP) approach. Until more comprehensive toxicity data (i.e. ED_{50}) as well as a more sophisticated method to convert *in vitro* to *in vivo* data become available, we can consider the in this study applied procedure as the good approximation in order to make use of the already large and continuously increasing body of *in vitro* toxicological studies on nanoparticles and like this allow their use in the field of LCA.

75

Integrating endocrine disruption into life cycle impact assessment

Y.A. Emara, A. Lehmann, M. Siegert, Technische Universitaet Berlin / Chair of Sustainable Engineering; M. Finkbeiner, Technische Universität Berlin / Chair of Sustainable Engineering

Converging lines of evidence suggest that exposure to 'endocrine-disrupting chemicals (EDCs)', i.e. chemicals with the ability to interfere with and alter functions of the endocrine system, is linked to multiple adverse effects on humans and wildlife (e.g. diabetes and reproductive dysfunctions). Currently, life cycle impact assessment (LCIA) models targeted at characterizing toxicity-related impacts of chemical emissions do not model endocrine toxicity. This study proposes a new approach to include endocrine disruption (ED) as a new impact pathway within LCIA and establish two new impact categories (Human ED and wildlife ED), thereby capturing adverse endocrine-mediated effects on humans and ecosystems separate from other toxicity-related impacts. Relying on the USEtox model, the calculation of fate and exposure factors remains unchanged, while the effect factor is determined using effect data on several ED-sensitive toxicological endpoints, thus reflecting the spectrum of endocrine mechanisms by which an EDC is known to act (e.g. estrogen receptor antagonism or interference with thyroid pathways) and the resulting mosaic toxic effects. To overcome potential data constraints in finding sufficient toxicological effect data for the thousands of chemicals suspected to exhibit endocrine-disrupting characteristics, data provided by the United States Environmental Protection Agency on the basis of in-vitro high-throughput screening assays for the endocrine bioactivity of more than 1,000 chemicals is examined for its suitability to be used in the proposed approach. New characterization factors for a set of known EDCs will be provided to directly enable characterization of EDCs and their adverse effects within future life cycle assessment studies.

76

Poster spotlight: MO090, MO091, MO100

Environmental fate of emerging contaminants in the water cycle: analytical challenges and engineered solutions (II)

77

Review on removal and reactions of micropollutants in biofilms under growth and non-growth conditions

K. Bester, Aarhus University / Environmental Science; M. Escolà Casas, Aarhus University / Department of Environmental Sciences; U. Bollmann, Aarhus University / Environmental Science; P. Carvalho, Aarhus University, Department of Environmental Science / Department of Environmental Science; E. Torresi, Kruger A/S; H. EI-taliawy, Aarhus University / Department of Environmental Science; L. Zhang, Aarhus University / Department of Bioscience; G. Ooi, K. Tang, DTU Environment; H.R. Andersen, Technical University of Denmark / Department of Environmental Engineering; M. Christensson, Anox Kaldnes

Pharmaceuticals and other compounds need to be removed from wastewater. This contribution will give an overview on the possibilities of removing micropollutants with biofilms. Biofilms occur in nature, but are also increasingly used in technical installations to remove micropollutants from water (wastewater and drinking water), porous media biofilm systems (sandfilters), moving bed biofilm systems (MBBRs). Biofilms can remove considerably better than suspended bacteria: diclofenac is recalcitrant in sludge systems, while it can be degraded with half-lives of 2 h in biofilm systems. In this contribution it is highlighted which ecological conditions (aerated versus denitrifying; high and low BOD loads) have been found to favor degradations. It was found that BOD supply has massive impact on the removal: On the one hand high BOD loads led to enhanced growth of biofilm, resulting in high turnover and reaction rates also of the respective micropollutants. On the other hand the biomass related reaction rate constants are considerably higher with low BOD loads. The highest reaction rate constants and biomass related reaction rate constants were found for systems with intermittent BOD loading. The switch from aerated to denitrifying conditions, however only gives effects for a chosen few compounds like ibuprofen, while reaction rates usually are similar. So the redox conditions as such are obviously not critical. Biofilms have often proven to have unique degradation pathways leading to hitherto strange metabolites. On the other hand compounds that were hitherto believed to be recalcitrant (like diclofenac) could easily be degraded in relatively short time periods. For single compounds degradation pathways for biofilm systems are discussed and compared to other systems. – While oxidation pathways are relatively common it seems like biofilms often perform a combination of oxidation and sulfatation pathways. Interestingly enough, it was possible to reach high removal rates for otherwise persistent ozonation-by products such as macrolide N oxides while avoiding back reactions to the parents with a moving bed biofilm reactor (MBBR). This also holds for most of the ozonation products of diclofenac.

78

Biodegradation of emerging organic contaminants using an enzyme-mediator system and study of the resulting transformation products

I. Caraene, Cutin University / chemical department; C. Joll, Curtin University / chemical department; K. Linge, Y. Gruchlik, Curtin University; A. Paparini, Murdoch University

Due to improvements in analytical screening methods, a large number of emerging organic contaminants (pharmaceutically active compounds, personal care products, pesticides, surfactants, plasticizers, corrosion inhibitors, flame retardants, artificial sweeteners andothers) have been identified in the aquatic environment. Biodegradation is one of the processes that can remove potentially hazardous emerging organic contaminants from different environments, with the help of microorganisms (e.g. algae, bacteria or fungi) and their extracellular products, in both aerobic and anaerobic conditions. The objective of this study was to investigate the biodegradation of a series of antibiotics and one anticonvulsant using laccase enzyme, extracted from a white-rot fungi *Trametes Versicolor*, in the presence of 2,2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS), as a mediator. The experiments were conducted in MilliQ water at different pH values, with different ratios of laccase and mediator, under aerobic and static conditions. The degradation was monitored by measuring the concentration of the remaining antibiotic over 168 hours, using a high performance liquid chromatograph with UV detection. The formation of new peaks was also monitored

and high resolution mass spectrometry (HRMS, LTQ Orbitrap) was used to identify potential transformation products. Furthermore, the microbial activity of the antibiotics and their transformation products was assessed, using an *E. coli* culture and microbial disks. Results showed that 89% degradation of sulfamethoxazole can be achieved at pH=5, with an enzyme activity ranging between 110-120 U/L and an initial mediator concentration of 200 µM, while trimethoprim only degraded by 43% under the same conditions. The results of similar biodegradation experiments on other antibiotics will be discussed in this conference presentation. The identification of transformation products of the antibiotics using high resolution mass spectrometry, and the microbial activity of the transformation products, will also be presented. This study provides a better understanding of the biodegradation of emerging organic contaminants and their transformation products. Further work can assist in assessing the possible health and environmental risks associated with the reuse of treated wastewater, for applications such as irrigation and groundwater replenishment.

79

Evaluation of macrolide antibiotic transformation in model biodegradation and ozonation experiments using target and non-target analyses and ecotoxicological bioassays

S. Terzic, Rudjer Boskovic Institute / Division for Marine and Environmental Research; P. Kostanjevecki, I. Krizman-Matasić, I. Senta, Rudjer Boskovic Institute; T. Jurina, Faculty for Food Technology and Biotechnology; N. Udikovic-Kolic, Rudjer Boskovic Institute; J. Ćurko, Faculty of Food Technology and Biotechnology; M. Matosic, Faculty for Food Technology and Biotechnology; J. Lončar, I. Mihaljević, T. Smital, Rudjer Boskovic Institute; M. Ahel, Rudjer Boskovic Institute / Division for Marine and Environmental Research

The aim of the present study was to investigate the transformation of three prominent representatives of macrolide antibiotics (azithromycin - AZI, clarithromycin - CLA and erythromycin - ERY) in model biodegradation and ozonation experiments. The study included determination of the dissipation kinetics of the parent compounds, identification of transformation products and ecotoxicological evaluation of transformation processes using two different end-points. The biodegradation efficiency was studied using the sludge culture enriched in the presence of AZI (10 mg/L) over a period of 4 months while the ozonation experiments were performed in different matrices by applying selected pH conditions and ozone concentrations. The dissipation kinetics of parent compounds as well as the formation of transformation products (TPs) were followed by ultra-performance liquid chromatography/quadrupole-time-of-flight mass spectrometry. Antibiotic activity test was based on the inhibition of bacterial growth (*Bacillus subtilis*), while toxicity test was performed with the freshwater green algae *Desmodesmus subspicatus.* At the applied experimental conditions, both biodegradation and ozonation experiments resulted in nearly full elimination of the tested parent compounds. The biotic and abiotic removal of all parent compounds was associated with the formation of different TPs, some of which were rather abundant and persistent to further degradation. The highest number of detected TPs was associated with the elimination of AZI, while the number of CLA and ERY TPs was comparatively much lower either under biotic and abiotic conditions. The environmental relevance of the identified biotransformation products, some of which included previously unknown linearized TPs, was proven by screening the municipal wastewater extracts for their presence. The effect-driven evaluation of the studied transformation processes, based on toxicity to algae and residual antibiotic activity, indicated a significant reduction of harmful effects, however formation of numerous stable transformation products, warrants further ecotoxicological assessment.

80

DI-SPME - On-fiber Derivatization - GC-MS. An innovative green and cost-effective approach to determine CECs and TPs from a novel anoxic-aerobic photobioreactor

R. López-Serna, University of Valladolid / Chemical Engineering and Environmental Technology; E. Posadas, P.A. García-Encina, R. Muñoz, University of Valladolid

The demand of multicomponent methods for the analysis of compounds of emerging concern (CECs) in environmental matrices is a reality today. However, conventional techniques based on Solid Phase Extraction (SPE) coupled to Liquid Chromatography Mass Spectrometry (LC-MS) are very often only available in high-tech laboratories. The cost-competitive methodology presented here, successfully developed and validated, intends to fill the existing gap between current environmental needs and analytical capacities. It consists of an innovative method for the analysis of 12 CECs, including 3 Transformation Products (TPs), in sewage and sludge using a fully automatized online DI-SPME – On-fiber Derivatization – GC-MS. The validated method was proven to be reliable, thanks to the combination of two quantification approaches, i.e., matrix-matched and internal standard, as well as sensitive (LODs below 20 ng L^{-1} for most of the target compounds in sewage and 30 ng g^{-1} in sludge), versatile and green. The method was successfully applied to real samples from a novel pilot scale anoxic-aerobic photobioreactor, where the influence of the organic load on the removal efficiencies (REs) of the CECs was evaluated. The three operational stages, at three different concentrations of chemical oxygen demand (COD) (669±6 mg L^{-1}, 493±11 mg L^{-1} and 434± 11 mg L^{-1}), were maintained for 40 d (≈4 times the SRT) to achieve representative steady states. The maximum REs of ibuprofen, naproxen, salicylic acid, triclosan and propylparaben were 91±1%, 28±7%, 83±5%, 85±0% and 85±15%, respectively. COD concentration only affected clearly ibuprofen and naproxen REs. This pointed out oxidation as an important removal mechanism for those cases. In contrast, salicylic acid and triclosan REs slightly increased at lower COD loads. For propylparaben, high elimination rates (above 80%) were observed regardless the COD concentration. Oxidation, biodegradation, sorption, volatilization and photodegradation were discussed as the possible removal mechanisms of the tested contaminants. This constituted the first evaluation of CECs removal by a synergetic interaction between algae and bacteria depending on the organic carbon load.

81

Abatement of amoxicillin, ampicillin and chloramphenicol from aqueous solutions using activated carbons prepared from grape slurry

R. Chitongo, Cape Peninsula University of Technology Cape Town South Africa / Chemistry; B. Opeolu, Cape Peninsula University of Technology / Faculty of Applied Sciences; O.S. Olatunji, Cape Peninsula University of Technology / Chemistry

There has been an increase in the use of pharmaceutical compounds for promotion of human and animal health, and the prevention of diseases over the past few decades. The sources of water and environmental contamination from these compounds include effluent discharges from household and several industrial activities. The capability of the sewage treatment processes in removing pharmaceuticals from wastewaters in African countries is also not fully known. There is scarcity of information concerning the utilization of grape slurry waste as a precursor of carbon based adsorbents, as well as its application for the removal of amoxicillin (AMX), ampicillin (AMP) or chloramphenicol (CHLR). This study therefore aimed at monitoring of the three antibiotic residues in selected surface waters. Activated carbons produced from grape slurry were also characterized and explored for abatement of the antibiotics' residues from aqueous solutions. An UHPLC-UV-DAD was optimized for the separation, detection and quantification of antibiotics in aqueous matrix. Solid Phase Extraction (SPE) procedure was optimized for recovery studies. Surface water samples were collected along the mainstream transects drainage stretch of the Diep River at different sampling points over two seasons. The removal of antibiotics from aqueous solutions using activated carbons produced from grape slurry was also studied. Activated carbons were characterized using FTIR, SEM and EDX in order to understand the removal mechanisms of the contaminants by activated carbons. The three antibiotics studied were detected in environmental water samples. Attempts were made to remediate these compounds using activated carbons produced from grape slurry waste. The sorption data indicated that all the operating conditions employed in this study were crucial for the control of antibiotics adsorption. The percentage sorption was enhanced with increasing adsorbent dose, contact time and pH, while increasing initial antibiotic concentration and temperature did not favour the sorption of the antibiotics. The equilibrium data fitted satisfactorily into the three isotherms studied. The pseudo-second-order kinetic model better described the adsorption of the antibiotics onto activated carbons used. Thermodynamic evaluation showed that the sorption was exothermic, feasible but non-spontaneous with increased in temperature.

82

Biodegradation of organic micropollutants in constructed wetlands: comparison of design and operational parameters

P. Carvalho, Aarhus University, Department of Environmental Science / Department of Environmental Science; Y. Zhang, Southern University of Science and Technology / School of Environmental Science and Engineering; L. Tao, Nottingham Trent University / School of Animal, Rural and Environmental Sciences; L. Zhang, C.A. Arias, Aarhus University / Department of Bioscience; K. Bester, Aarhus University / Department of Environmental Science; H. Brix, Aarhus University / Department of Bioscience

Wastewater has been considered a major source of contaminants of emerging concern to the environment, as conventional treatment systems do not completely remove them. Constructed wetland systems (CWs) have, however, been shown to be able to degrade some of these emerging contaminants, namely organic micropollutants. But the processes at the core of the removal of these compounds in CWs are yet unknown. Research being developed at Aarhus University aims at understanding the removal processes and fate of organic micropollutants in different types of CW systems. Five different experiments have been conducted in the past four years. Two experiments have been performed under hydroponic conditions in a growth chamber aiming to understand the uptake, translocation and metabolisation of the organic pollutants. Two other experiments compared the effects of season, plant presence and plant species, initial concentration, hydraulic loading rate and CW design in different pesticides and phamarceutical compounds. A sixth setup targeted the impact of support matrix in compounds sorption and biofilm microbial community function. The plants *Typha latifolia* and *Phragmites autralis* were the most efficient plant species in removing ibuprofen and iohexol. Phragmites was the most efficient species to remove the pesticides tebuconazole and imazalil. Uptake, translocation and degradation of chiral pesticides inside the

plant tissue was documented. Formation of transformation products was assessed, but the mass balances were not closed. Organic micropollutants sorption to support matrix was low. Removal of different compounds was higher in summer than in the winter. Planted reactors showed higher efficiency than unplanted reactors, stressing the synergies between the plant and the microbial community. Unsaturated systems tended to be more efficient. Removal correlated with the nitrification activity and with the biofilm activity, suggesting that bacterial processes play an active role in the micropollutants biodegradation. The removal of the organic micropollutants in CWs is affected by several design and operational parameters. Plant uptake does occur but phytoaccumulation is low as the compounds can be degraded inside the plant tissues. Due to overlying effect of the plants, the extent of microbial degradation could not be quantified. Further studies on transformation products in this type of technical systems are needed.

Wildlife ecotoxicology: laboratory dosing studies to field population assessments (II)

83

Effects of PAH exposure on fuelling ability in a long distance migratory shorebird

K. Bianchini, University of Saskatchewan - Toxicology Centre / Toxicology; C.A. Morrissey, University of Saskatchewan / Biology

Many shorebirds are long distance migrants that stop to refuel along the journey where they can be exposed to pollutants that may impede fuelling for migration. Exposure to organic pollutants can cause potential effects on migration success, speed and subsequent population parameters since pre-migratory fuelling is correlated with reproductive performance upon reaching the northern breeding grounds. The polycyclic aromatic hydrocarbons (PAHs) found in marine oil pollution have the potential to interfere with pre-migratory fuelling physiology in shorebirds. However, a link between PAH exposure and pre-migratory fuelling has yet to be established. Our objective was to determine if PAHs or associated contaminants can affect condition and fuelling rates in a captive shorebird, the Sanderling and in the field at major shorebird stopovers. In this study, a captive population of 49 Sanderling (*Calidris alba*) was orally dosed with a commercial PAH mixture for 21 days at ecologically relevant concentrations (0, 12.6, 126, and 1260 µg PAH/kg body weight/day). We found that EROD activity was significantly elevated in the high dose group relative to controls and fuelling rates and condition were also lower in dosed birds. Higher PAH exposures were associated with reduced serum bile acid concentrations, elevated serum creatine kinase concentrations, and with high serum lipase concentrations (in females). These results suggest that PAH exposure can interfere with lipid transport and metabolism and can cause muscle damage leading to poorer condition. We also captured Sanderling from the Gulf of Mexico, which is subject to recurring oil spills and from Chaplin Lake, Saskatchewan, a relatively uncontaminated site. We measured each bird's body condition, fuelling status, and plasma PAH levels and attached miniature radio transmitters to a subset of birds (n=75). Motus radio telemetry array technology was used to determine the arrival and departure timing and stopover duration. We found that mean stopover durations in the Gulf of Mexico were longer than in Chaplin Lake (27 versus 15 days), and that stopover duration was associated with body condition and fuelling status at capture. We also measured higher plasma PAH concentrations in birds at certain Gulf sites, suggesting that PAH exposure is associated with lower pre-migratory fuelling rates. This work will inform shorebird conservation by providing valuable insight into a potential cause of migratory shorebird declines.

84

PFAAs levels, oxidative status and reproductive success in great tits (Parus major) inhabiting a contamination hot-spot.

A. López Antia, Universiteit Antwerpen / Biology; T. Groffen, Systemic Physiological and Ecotoxicological Research (SPHERE), University of Antwerp / Biology; L. Bervoets, Universiteit Antwerpen; R. Lasters, E. Prinsen, H. Abd Elgawad, University Antwerp; M. Eens, University of Antwerp / Department of Biology

Perfluoroalkyl acids (PFAAs) are substances which have been produced for more than five decades. Their unique properties of repelling both water and oil, make them suitable for many industrial and consumer applications such as water and dirt repellents for clothes and carpets, active components in firefighting foams or precursors in Teflon® production [1]. Its extensive use, together with their high persistence, has resulted in global contamination of the environment, wildlife and even humans [2,3]. This ubiquity contrasts sharply with the limited amount of available information about their effects on organisms. We report here PFAAs egg and plasma levels in wild populations of great tits (Parus major) settled along an established pollution gradient starting from a fluorochemical plant in Antwerp (Belgium). Using two generations of great tits we have obtained important results in some poorly known issues such as the differences between sexes, maternal transfer of compounds or possible effects on the oxidative status or the reproductive success. The levels we detected in eggs and plasma, demonstrate that Antwerp is one of the major hot-spots in the world for perfluorinated compounds pollution. With regard to the possible effects, negative correlations were observed between PFAAs levels in the eggs and reproductive parameters, including the total hatching success, eggshell thickness or the total breeding success. PFAAs levels in blood correlated with protein damage in adult birds while in chicks they correlated with higher activity of antioxidant enzymes (GPX and CAT). The obtained data represent an important step towards the understanding of the behaviour, effects and consequences of PFAAs in wild bird populations. [1] Buck RC, Franklin J, Berger U, Conder JM, Cousins IT, de Voogt P, Jensen A, Kannan K, Mabury S, Van Leeuwen SP (2011). Perfluoroalkyl and polyfluoroalkyl substances in the environment: terminology, classification, and origins. Integr Environ Asses 7: 513-541. [2] Giesy JP and Kannan K (2001). Global distribution of perfluorooctane sulfonate in wildlife. Environ Sci Technol 35: 1339-1342. [3] Giesy JP and Kannan K (2002). Peer-reviewed: perfluorochemical surfactants in the environment. Environ Sci Technol 36: 146-152.

85

Active and passive monitoring of lead poisoning in birds of prey in Spain

R. Mateo, IREC-CSIC- UCLM / Grupo de Toxicología de Fauna Silvestre; E. Descalzo-Sanchez, Instituto de Investigación en Recursos Cinegéticos IREC CSICUCLMJCCM; P.R. Camarero, Instituto de Investigación en Recursos Cinegéticos, CSIC-UCLM-JCCM / Grupo de Toxicología de Fauna Silvestre; I.S. Sanchez-Barbudo, UCLM-CSIC / Grupo de Toxicología de Fauna Silvestre

The ingestion of lead ammunition is the most important source of exposure to this metal in birds of prey, and consequences on their health are well-know. The objective of the present study is to improve our knowledge on the exposure to Pb in birds of prey in Spain by means the integration of active and passive monitoring programs based on Pb analysis in blood and liver of raptors and by the evaluation of the effects on their health by using non-destructive blood biomarkers. We have performed a passive monitoring by measuring blood (n=27) and liver (n=685) lead levels in birds of prey of 16 species found dead or sick in Spain between 2004 and 2017, but also an active monitoring by measuring blood lead levels in birds (n=196) of 9 species trapped alive in the field. Adverse effects of lead exposure on heme biosynthesis, P/Ca metabolism, oxidative stress and immune function were also evaluated in the active monitoring by means non-destructive biomarkers. The active monitoring showed that some individuals of bearded vulture (1/3), Eurasian griffon vulture (87/118), Spanish imperial Eagle (1/6) and red kite (1/18) presented abnormal blood Pb exposure levels (>200 ng/ml). Passive monitoring revealed that the species with lead levels in liver associated with clinical poisoning (18-30 µg/g d.w.) were cinereous vulture (1/39), Eurasian griffon vulture (2/228) and western marsh-harrier (1/32); and the species with clinical severe poisoning (>30 µg/g d.w. of Pb in liver) were Eurasian griffon vulture (19/228), red kite (1/129) and golden eagle (3/36). The study of biomarkers reveals a negative relationship between δ-ALAD activity in blood and blood Pb concentration. Ca/P homeostasis was also affected by Pb exposure, because elevated blood Pb levels were associated with lower Pb levels and higher Ca:P ratio in plasma of birds. Carotenoid levels in plasma were also increased in birds with higher blood Pb levels, indicating a possible allocation of antioxidants in plasma to cope with adverse effects of Pb. The integration of active and passive monitoring permits to have a more complete perspective of the risk that Pb represents for raptors. Here we confirm with the active monitoring the elevated blood Pb levels (73.7% with >200 ng/ml) in field-trapped Eurasian griffons as found in previous studies, but also report a significant mortality (8.3% with >30 µg/g d.w.) in Eurasian griffons and golden eagles with the passive monitoring.

86

Persistence of elevated p,p'-DDE levels and HCB-related protoporphyrin IX decrease in eggs of common kestrels from Tenerife (Canary Islands, Spain)

A. Buck, Instituto de Investigación en Recursos Cinegéticos IREC CSICUCLMJCCM / Wildlife Toxicology; J. Carillo, University of La Laguna; P. Camarero, IRECInstituto de Investigación en Recursos Cinegéticos; R. Mateo, IREC-CSIC- UCLM / Grupo de Toxicología de Fauna Silvestre

Persistent organochlorine (OC) pesticides, including p,p'-DDT, have been banned in many parts of the world for more than 30 years, but they are still present in the top predators of terrestrial and aquatic food webs. The Canary Island were one of the Spanish regions with the highest use of OC pesticides due to the intensity of its agriculture. A previous study performed between 1988 and 1994 with 14 unhatched eggs of West Canarian common kestrel (*Falco tinnunculus canariensis*) from Tenerife Island showed elevated concentrations of p,p'-DDE (17.9 µg/g dw; equivalent to 4.9 µg/g ww). Here, we have monitored the levels of OC compounds (pesticides and polychlorinated biphenyls) in 40 unhatched eggs of West Canarian common kestrels from Tenerife Island collected between 2009 and 2016. We have also measured the porphyrin composition of the eggshells to explore the use of these pigments as biomarkers of organochlorine pollution in birds. Biometry, status of embryo development and eggshell thickness were recorded from each egg and information about habitat characteristics were recorded for each nest. Because the eggs were at different degrees of desiccation, the content was lyophilised in order to measure OC concentrations in dry and lipid weight of content. OC analysis was performed by extraction with n-hexane:dichloromethane (4:1), evaporation (for lipid weight calculation) and resuspension in n-hexane, followed by four clean-ups with sulfuric acid and determination by GC-ECD. For porphyrin determination, eggshells were homogenized and extracted with acetonitrile:HCl 3N (2:1) and then analysed by HPLC-DAD. Egg content showed the following OC levels (mean ± SE; µg/g dw): p,p'-DDE, 15.2 ±1.7; p,p'-DDT, 0.118 ± 0.020; PCBs, 0.459 ± 0.121; HCHs (hexachlorocyclohexane isomers), 0.021 ± 0.003; and HCB (hexachlorobenzene), 0.0042 ± 0.0004. p,p'-DDE levels have remained elevated for more than 20 years and these levels were statistically associated in generalized linear models with the surface of active and abandoned cropland in a 200 m-radio around the nest (+), distance from nest to urban areas and greenhouses (-), altitude (-) and year (highest in 2011). PCB levels were associated with distance from nest to roads (-) and altitude (+). The shell index was not affected by p,p'-DDE levels, but decreased with embryo development. Protoporphyrin IX was the only pigment in eggshells and its concentration was negatively affected by HCB levels in egg content.

87
Long-term increase in secondary exposure to anticoagulant rodenticides in European polecats in Britain
K.A. Sainsbury, University of Exeter / Environment and Sustainability Institute; R. Shore, Centre for Ecology & Hydrology (NERC); H. Schofield, L. Croose, The Vincent Wildlife Trust; M.G. Pereira, Centre for Ecology & Hydrology / Lancaster; D. Sleep, NERC Centre for Ecology & Hydrology; A.C. Kitchener, G. Hantke, National Museums Scotland; R. McDonald, University of Exeter / Environment and Sustainability Institute
As a result of legal protection and population recovery in Great Britain, European polecats (*Mustela putorius*) are expanding into areas associated with greater usage of second-generation anticoagulant rodenticides (SGARs). We analysed livers from polecats found dead (mostly road casualties) from 2013-2016 for residues of five SGARs. We related variation in residues to polecat traits (age, sex, provenance), to potential exposure pathways by analysing stable isotopes of carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) in whiskers, and to data collected from polecats in the period 1992-99. In all, 54 of 68 (79%) polecats from 2013-16 had detectable liver residues of at least one SGAR. Bromadiolone (71%) was the most commonly detected compound, followed by difenacoum (53%) and brodifacoum (35%). Liver SGAR residues did not vary with sex or with the season in which the polecat died. We found a positive association between occurrence of liver SGAR residues and $\delta^{15}N$ values. Polecats in Britain feed predominantly on rats and rabbits and our findings are consistent with the concept that individuals feeding on rats (higher trophic level than rabbits) are more likely to be exposed to SGARs. Total SGAR liver concentrations were higher in polecats from arable than pastoral habitats, consistent with more intensive SGAR use on arable farms, and higher in western than eastern regions although the reason for this is unclear. Both number of compounds and total SGAR concentrations were positively associated with age, presumably due to multiple sub-lethal exposures during an animal's lifetime; older animals may thus be at most risk from poisoning due to progressive accumulation of liver residues. When we compared data for polecats from 2013-16 with those for polecats that died in 1992-99 and accounted for differences between studies in detection limits, we found that the rate of detection of SGARs in polecats in Britain increased 1.7 fold over the 25 year period. This increase was not restricted to newly recolonised areas and suggests an increase in the risk to polecats from SGARs throughout their range.

88
Poster spotlight: MO035, MO036, MO083

Environmental risk assessment in time and space - new approaches to deal with ecological complexity

89
The threshold option, the recovery option and landscape modelling
P. Thorbek, Syngenta / Environmetal Safety; N. Galic, Syngenta / Environmental Safety; V. Forbes, University of Minnesota / Ecology, Evolution & Behavior
Landscapes provide a multitude of ecosystem services, but the relationships between the populations of the organisms providing them, stressors and the delivered ecosystem services are dynamic and often nonlinear. In order to understand how pesticides may affect ecosystem services and biodiversity at the landscape level, it is necessary to understand both exposure and effects at the organism level, but also how life history, movement patterns and farming activities such as tillage and harvest affect population dynamics. If this has to be done for all species in all landscapes in Europe the modelling task quickly becomes unmanageably complex, and the interpretation of the modelling outputs will be challenging. Here we present a tiered system for model design to aid managing the complexity. We outline what model design features are necessary for modelling based on species mobility and whether the ecological threshold option (ETO) or the ecological recovery option (ERO) is chosen. Ecological production functions quantitatively link the service-providing units to the services delivered and are therefore essential for understanding how attributes of population and ecosystems (e.g. biomass or functioning) contribute to the final ecosystem services enjoyed by the recipients. Such understanding can be used to set the protection goals for different service-providing units for both ETO and ERO. The attributes which link to service delivery can be difficult to measure at the landscape level, but by combining ecological models and ecological production functions, thresholds can be set for lower tiers of the risk assessments, which may be easier to measure. In some cases the ecological production functions are quite simple if a population directly delivers the service (e.g. for angling). However, in other cases, the link is far from straightforward and such ecological production functions have largely been ignored in pesticide risk assessment. This should be a priority area for future research.

90
Understanding risk - a better approach to reduce uncertainty
M. Wang, WSC Scientific GmbH / Dept Efate Modelling; M. Foudoulakis, Dow Agrosciences / RSRA ERS
For many compounds the intrinsic toxicity as determined in toxicity studies does not reflect toxicity and risk adequately. Rather other mechanisms determine which species are most at risk (focal species) and how large the risk posed to these species is. These include for example elimination rates and feeding behaviour, which are not considered in the first tier. In the present presentation results from two case-studies are given which demonstrate how uncertainty in the risk assessment can be reduced by trying to understand mechanisms that lead to toxicity and mechanism determining the actual and long-term risk of mammals and birds in the field. Field data help to verify the obtained knowledge and to determine an empirical margin of safety. Finally, population modelling is used to answer what-if questions and to answer questions on the relevance of effects when considering specific worst-case assumptions. In both example compounds metabolism and excretion together with feeding behaviour mainly determined the acute and long-term risk. All of these mechanisms are not considered in the first tier risk assessment and without these it would not be possible to understand the risk of the compounds shown here. This understanding significantly reduced the uncertainty of the risk assessment, because with the gained knowledge it is possible to identify critical scenarios.

91
Developing spatio-temporally realistic representations of agricultural landscapes for assessing the impacts of pesticides on non-target organisms
E. Ziółkowska, Jagiellonian Univeristy / Institute of Environmental Sciences; C.J. Topping, Aarhus University / Department of Bioscience; A. Bednarska, Polish Academy of Sciences / Institute of Nature Conservation; R. Laskowski, Jagiellonian University / Ecotoxicology & Stress Ecology Group
Species richness and population sizes in agro-ecosystems have decreased dramatically during last few decades. The current scheme of agricultural intensification resulting in landscape simplification is considered one of the main causes of this biodiversity loss, along with widespread use of pesticides. As the management of landscape heterogeneity seems to be crucial for maintaining vital populations in agro-systems, it is necessary to include the landscape component in ERA and as the important mitigation strategy. We present a methodological framework for modelling the spatio-temporal heterogeneity in agricultural landscapes. The framework has been implemented within the ALMaSS simulation system allowing to investigate the effects of changes in landscape structure and management on the population size and distribution of animals. We describe spatial landscape heterogeneity through a detailed land cover map, in which farmed areas are represented as accurate maps of fields grouped into farm units of different types (e.g., cattle or arable farms). The temporal component of agricultural landscape heterogeneity refers to both crop management throughout a year, described through individually tailored management plans for each crop, and the cropping system understood as a pluriannual crop rotation. Crop management plans consist of combinations of farm activities (including pesticide treatments), as well as time windows and probabilities of carrying out activities. The temporal component also includes weather conditions and vegetation growth models for all vegetation types and crops. Such approach gives a highly realistic, updated on a daily basis, dynamic landscape with vegetation growing in response to the weather, and the pattern of farming activities related to each specific crop, farm, and field. Our methodological framework, supported with semi-automated procedures for spatial data management, makes creation of highly-realistic model agricultural landscapes feasible and usable for landscape-scale risk assessment. More importantly, the presented tools allow for testing *in silico* various scenarios of agricultural practices, including pesticide use, in differently structured landscapes. This seems at the moment the most promising strategy for elaborating sustainable agricultural practices that would allow for high productivity, whilst still protecting the agrobiodiversity. This study was supported by the National Science Centre, Poland (2015/19/B/NZ8/01939).

92
Where are the Springtails? A vertical distribution model for Collembolans
V. Roeben, RWTH Aachen University, Institute for Environmental Research / Institute for Environmental Research BioV; L.S. Tzschoppe, RWTH Aachen University / Institute for Environmental Research BioV; T. Preuss, Bayer Ag / Environmental Safety; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics; M. Ross-Nickoll, RWTH Aachen

University, Institute for Environmental Research / Institute for Environmental Research

With respect to environmental risk assessment it is crucial to know where and when to protect an organism but still little is known on the dispersal of collembolan communities in agricultural landscapes. Especially for the environmental risk assessment of plant protection products vertical movements can be relevant for exposure assessment of in-soil organisms. Thus, ecological modeling offers a powerful tool to link exposure and effect. We will present the individual-based model of the soil-dwelling collembolan *Folsomia candida* FOLCAS (Folsomia candida simulation). FOLCAS is a vertical distribution model simulating an agricultural soil column, which can be applied to demonstrate the effect of variations in environmental parameters on the population and its dispersal. In addition, the model features the option to evaluate the effect of a pesticide application. The model consists of two submodels: the lifecyle and the movement submodel. The movement of the individuals in FOLCAS is influenced by temperature, pore space, pH and the organic matter as a proxy for food availability. In order to assess the importance of food availability as a main trigger for movement a vertical distribution experiment was designed. In this experiment we assessed the vertical dispersal of *F.candida* in OECD soil in relation to food location and time. Transparent PVC columns were filled with 350 g OECD soil up to 20 cm column height and 86 individuals of *F.candida* of different age classes were added. Each column was divided in 6 compartments from top to bottom: 0-1cm, 1-2.5cm, 2.5-5cm, 5-10cm, 10-15cm and 15-20cm. The location of feeding was varied by four different regimes while all other parameters were kept constant (20'C, no light, soil moisture of 50% of WHCmax). The columns were provided either with food at the top (1st), the middle (4th), the ground (6th) or at all three compartments simultaneously. The vertical distribution study will show that the dispersal of *F.candida* within a soil column is influenced by the location and availability of food. The study will give insights not only about the population dispersal in relation to food as a single stressor, but also on the population composition. This data will be incorporated into the movement submodel and simulation results of the vertical dispersal of collembolans will be presented. A case study will be used to elucidate the importance of the vertical dispersal of non-target arthropods in effect assessment.

93

A practical application of an individual-based stickleback model in the ERA of PPPs

K. Mintram, University of Exeter / Biosciences; C. Tyler, University of Exeter / Biosciences College of Life and Environmental Sciences; S.K. Maynard, AstraZeneca / Safety Health and the Environment; A. Brown, Exeter University / Biosciences; C. Liu, Syngenta / Environmental Safety; S. Parker, Cefas Weymouth Laboratory; P. Thorbek, Syngenta / Environmetal Safety

Population models are employed in the environmental risk assessment (ERA) of chemicals, including plant protection products (PPPs), to extrapolate from individual-level effects to predictions of effects on whole populations. Individual-based models (IBMs) allow for the incorporation of individual variability, population-level interactions, and specific behaviours. IBMs can therefore be used to extrapolate from a large number of individual-level endpoints and simulate potential effects on populations under realistic environmental conditions. We present an IBM of the three-spined stickleback (*Gasterosteous aculeatus*) developed for the purpose of predicting population-level effects for exposure to chemicals. The IBM was developed from a series of sub-models parameterised from empirical data obtained from the published literature. Modelled population dynamics (e.g. size/ age class distributions and annual fluctuations in population abundance) emerge from the adaptive traits, behaviours and interactions between individuals and their environment (including toxicant exposure). Here, we describe the development and validation of a stickleback IBM and demonstrate its practical application in ERA. Empirical data quantifying the reproductive effects and subsequent recovery, following exposure to a fungicide, were input into the IBM as a sub-model. Various exposure and recovery scenarios were simulated to predict population-level effects over time. The modelled outputs demonstrated that exposure duration and individual recovery post-exposure can influence the overall effects of chemical exposure on population abundance. We suggest that using IBMs to incorporate realistic exposure and recovery scenarios may improve current ERA and result in more realistic protection standards for wild populations.

94

Using the Bayesian network relative risk model to integrate molecular effects, ecological context and ecosytem services to estimate risk over space and time

W.G. Landis, Western Washington University / Institute of Environmental Toxicology; J.D. Stark, Washington State University / Dept of Entomology; K. von Stackelberg, NEK Associates LTD / Department of Environmental Health; C. Mitchell, Washington State University / School of the Environment; V. Chu, Western Washington University / Environmental Science; M. Harris, Whatcom Conservation District / Institute of Environmental Toxicology; L. Wallis, Western Washington University / Institute of Environmental Toxicology

An ongoing dilemma in risk assessment is the perceived difficulty in successfully integrating scales that range from the molecular to ecological, timeframes from days to decades, and endpoints that can be species specific to a host of ecosystem services. Starting In the late 2000s to now there has been an interest in defining ecosystem services and in the calculation of risk to these properties. It has been suggested that ecosystem services are a method to encourage a systems approach to sustainability. Human well-being has become part of the lexicon to included endpoints such as a sense of place, education, employment, public safety and traditional activities. In a recent publication (Harris et al. 2017) it was demonstrated that it is possible to estimate risk in a contaminated site to ecological endpoints, human health and ecosystem services using a clearly defined causal pathways and Bayesian networks. Now we are extending the integration of ecological endpoints, ecosystem services and human well-being from the scale of a contaminated site to that of the Salish Sea. The Salish Sea is a term applied to both the Puget Sound and its watersheds in the United States and the Straits of Georgia in Canada. Vancouver, Seattle, Tacoma, major ports, numerous refineries, paper mills, and high tech industries. The same area is also noted for intense agricultural use, outdoor recreation and the harvest of marine resources. The region is also home to more than 30 recognized Tribes in the U. S. segment and First Nations in Canada. We will use three watersheds in this region, the Skagit, the Nooksack and the Cedar as case studies. Time frames will be from current conditions to 2070 and will include climate change projections for water temperature and precipitation. We will demonstrate the application of the Bayesian-network relative risk model to integrate pesticide effects at the molecular level and the alteration of watersheds to calculate risk to the ecological endpoint Chinook Salmon, the specific economic ecosystem services provide by the endpoint and the watersheds, and finally demonstrate the risks to human well-being as defined from a variety of cultural perspectives.

The environment as a reactor determining fate and toxicity of nanomaterials (II)

95

Mobilisation of silver sulphide nanoparticles in soil column by earthworms´ bioturbation

M. Baccaro, Wageningen University / Toxicology Department; H.H. van den Berg, Wageningen University / Dept of Toxicology; D. Hermans, L. Sloot, Wageningen University and Research; N. van den Brink, Wageningen University / Dept of Toxicology

Ag_2S-NP is the main product of transformation of Ag-NP in the waste water treatment plant (WWTP). In many European countries, sewage sludge containing Ag_2S-NPs is applied on the surface of the soils as soil amendments. Earthworms are important ecological engineers in the soil ecosystem, which, on one hand, may be affected by Ag released from the amendments and, on the other hand, may influence the distribution of metals. Therefore, the aim of this study was to determine effects of Ag_2S-NP application on an important earthworm driven process, i.e. bioturbation and the effect of the earthworm activity on the vertical distribution of Ag_2S-NP in the top soil. Their interplay was assessed in two experiments, in presence or absence of artificial rain fall. Around 2 cm of soil treated with 10 mg Ag Kg^{-1} dry weight soil of Ag_2S-NP (28.0±20.4 nm) was applied on top of natural soil columns (10 cm) mimicking an application of 200 Mg sludge ha^{-1} dry weight. For the first experiment, columns were prepared with and without Lumbricus rubellus and with and without Ag_2S-NPs. Every week for 28 days earthworms and four different layers of the soil columns (0-2, 2-4, 6-8, 10-12 cm depth) were sampled. In the same way a second experiment was performed with daily application of 2 mm of artificial rain water, allowing collection of leakage samples from the bottom of the columns. Total Ag content was measured in all samples by ICP-MS following acid digestion and nano-Ag in leakage samples by spICP-MS. Results of the first experiment show that mobility of Ag along the soil column is significantly higher in the columns with earthworms overtime. Ag reached the bottom layer of the columns where worms were present while no Ag was found at the bottom layer of the column without worms. This indicates that earthworms do not avoid the contaminated top layer. Ag content in earthworms was relatively constant overtime with an average value of 1.06±0.32 mg Ag kg^{-1} dry weight. The first study shows that uptake of Ag_2S-NPs in earthworms occurred regardless of the partial exposure and points towards the crucial role of earthworm bioturbation in the mobilisation of metal nanoparticles in the top soil. The second experiment of the study is currently being performed, results will be presented at the meeting.

96

Short- and long-term approaches to determine the fate of silver nanoparticles in soil

M. Hoppe, BGR / Groundwater and Soil; D. Rückamp, E. Wargenau, A. Lamparter, C. Stange, S. Kaufhold, German Federal Institute for Geosciences and Natural Resources; E. Fries, Federal Institute for Geosciences and Natural Resources; P. Königer, German Federal Institute for Geosciences and Natural Resources; K. Schlich, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology; K. Hund-Rinke, Fraunhofer IME / Department of Ecotoxicology; M. Kraas, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Ecotoxicology; R. Mikutta, Leibniz Universität Hannover; J. Utermann, Federal Environment Agency; G. Guggenberger, Leibniz Universität Hannover

Engineered silver nanoparticles (Ag ENP) are present in many consumer products. Hence, the ENP enter into sewers and wastewater treatment plants with a high predicted removal into the sludge. If the sludge is applied to agricultural soils, decompostation might result in resuspension of the ENP. The fate and impact of Ag ENP in soils is still unclear. Short- and long-term column remobilization experiments with disturbed soils, short-term column percolation experiments with undisturbed soils, and long-term field lysimeter experiments were conducted. All experiments were performed with sterically stabilized Ag ENP (AgNM-300k), and a slightly loamy Cambisol (RefeSol 01A). Additionally, a medium clayey silt (Luvisol) was used for the column experiments. The **column remobilization** of the Ag concentration after digestion (Ag_{total}) was on a very low level in all percolation steps in both soils. The first percolation step after three days of the Cambisol incubation showed the highest remobilization of Ag which was below 1 % of the Ag_{total} concentrations in the soil columns. The correlation between remobilized Ag_{total} and Al_{total} concentrations suggests that the remobilized amount of Ag was associated to soil colloids. The breakthrough of Ag ENP in the **column percolation experiments** was high but incomplete in the Cambisol and the Luvisol. Particularly, columns with preferential flow pathways showed low Ag ENP retention. In the unsaturated experiments, a nearly complete retention was found for the Luvisol that showed a clearly smaller pore size structure than the Cambisol. The horizontal displacement of Ag_{total} in the **lysimeter experiments** was low and very likely related to soil tillage as well as bioturbation. A low Ag_{total} release to the percolate water (t= 480 d, control= 24 ng l^{-1}, Lysimeter (7 mg kg^{-1}) = 56 ng l^{-1}, DIN 38402-11) was obtained for the lysimeter with the highest Ag ENP application. This lysimeter induced a steady inhibition of the soil microflora that was not detected in the lysimeter with the lower Ag ENP concentration.All roots (wheat, canola, barley) showed a low uptake of Ag_{total}. All approaches showed a more or less high retention of Ag ENP in soils why soils are a sink for Ag ENP. However, the demobilization in the lysimeter was incomplete because of root uptake and inhibition of the soil microflora. Thus, the impact of a repeated sludge application to the soil microorganism and the root uptake (e.g. beet) needs further long-term investigations.

97

Determination of attachment efficiency (α) for ENPs in different types of soils by saturated column experiments

K. Norrfors, SLU Uppsala / Soil and environment; G. Cornelis, Swedish University of Agricultural Sciences / Soil and environment

The attachment efficiency (α) has been suggested as the most appropriate fate descriptor for transport of engineered nanoparticles (ENPs) in soils and saturated column experiments as the most accurate method to obtain α. Due to the complexity of the soil composition and texture, a small change in performance of the column protocol may affect the resulting attachment efficiency obtained from the results. The aim of this work is to study the effect of soil composition, flow velocity, initial ENP concentration and the size of ENPs on the calculated attachment efficiency for the specific ENP-soil systems. The α values for nominally 20 and 80 nm citrate coated gold ENPs (Au ENPs), as well as 30 nm sulphonated silver ENPs (Ag_2S ENPs) were determined from saturated packed column experiments in different soils sampled in the UK. Artificial rainwater was used as the eluent. 10 mM $NaNO_3$ was used as a concervative tracer to estimate the effective porosity and dispersivity. All columns were packed with an excess of rainwater to limit the amount of air present in the saturated soil. α was either calculated from breakthrough curves of Au/Ag or from the irreversible attachment rate modelled using Hydrus 1D or the relative mass recovery of the ENPs in the break through curves. Preliminary results show no significant differences in α values for 80 nm and 20 nm Au ENPs. However, the Au ENP breakthrough curves appeared dependent on the flow rate. Even though the shape of the break through curves changes with flow rate, this can be compensated during modeling arriving at consistent α values between the systems with varying flow rates. Furthermore, presence of air in the column affects the distribution of ENPs in the packed columns. Finally, an increase in initial ENP concentration give higher α values and cannot be accounted for in the equations used for estimating the attachment efficiency. In conclusion, when varying the initial ENPs concentration into the columns, the α value is significantly affected. Hence, low NP concentrations need to be used in the column experiments to maximize reproducibility of calculated α values. Moreover, inclusion of air in the systems appears to induce artefacts that complicate determination of α for specific NP-soil combinations.

98

The transformation of copper and zinc (-nanoparticles) during sewage sludge combustion

J.J. Wielinski, ETH Zürich/Eawag / Process Engineering; A. Gogos, Eawag, Swiss Federal Institute of Aquatic Science and Technology / Process Engineering, Particle Lab; A. Voegelin, Eawag   Swiss federal Institute of Aquatic Science and Technology; E. Morgenroth, Eawag   Swiss federal Institute of Aquatic Science and Technology / Process Engineering; R. Kaegi, Eawag - Swiss federal Institute of Aquatic Science and Technology

Engineered nanoparticles in wastewater streams are effectively retained by wastewater treatment plants and accumulate in sewage sludge. Digested sludge is subsequently combusted for further volume reduction to allow for phosphorous recovery at a later stage. This study focuses on two metals Cu and Zn, as both are present in digested sludge but are also used as nanomaterials. We investigated (i) the transformation of ZnO and CuO-NP during anaerobic digestion, (ii) the subsequent transformation of Cu and Zn during sewage sludge combustion, and (iii) whether the form of Cu and Zn affects the fate during anaerobic digestion and combustion. We spiked CuO-NP, ZnO-NP, Cu^{2+} and Zn^{2+} to four aliquots of digested sewage sludge and kept them under anaerobic and mesophilic conditions for 24h. One aliquot was kept as control. Thereafter, sludge was combusted in a pilot fluidized bed reactor and ashes were collected. Sludge and ashes were prepared for Cu - and Zn K-edge X-ray absorption spectroscopy (XAS) measurements. The speciation of the metals was obtained through linear combination fitting (LCF) of X-ray absorption near edges spectra (XANES) and extended X-ray absorption fine structure (EXAFS) data. LCF fits of experimental XANES and EXAFS data suggest a high degree of sulfidation of both Cu and Zn during anaerobic digestion, irrespective of the spiked form of Cu and Zn. For control sludge and sludge spiked with Zn^{2+}, LCF results from EXAFS data suggest that ~90% of the Zn was present as sulphides, the remaining fraction was best described by a ZnO reference spectrum. For sludge spiked with ZnO-NP, a lower degree of sulfidation (~83% ZnS) with a concomitantly high fraction of ZnO (17%) was calculated. After combustion, EXAFS spectra of Zn were best described by a spectra of Zn that was co-precipitated with Ferrihydrite. All LCF fits of Cu in the digested sludge indicated complete sulfidation of Cu. After combustion, LCF fits of the experimental EXAFS spectra revealed the presence of ~30% Chalcopyrite, indicating that Cu was not completely oxidised during the combustion. Comparable fractions of CuO and $CuSO_4$ were returned from LCF analyses. All Cu spectra of the sludge and the ashes were very comparable and independent of the added form of Cu. For Zn, however, the addition of ZnO-NP resulted in a slightly lower degree of sulfidation compared to the control sludge and to the sludge that was spiked with dissolved Zn^{2+}. All Zn spectra of the ashes were comparable.

99

Soil ecotoxicity and dissolution of a marketed nanosilver product - a direct comparison with ionic silver

J. Mertens, Precious Metals and Rhenium Consortium c/o EPMF; K. Arijs, ARCHE; E. Smolders, Katholieke Universiteit Leuven; D. Leverett, wca; K. Oorts, ARCHE

As part of the REACH Substance Evaluation for silver, new data was generated to further justify read-across from ionic silver to silver nanoforms. Therefore, the soil ecotoxicity and dissolution of ionic silver vs nanosilver were tested. The smallest silver nanoform with the highest specific surface area registered under REACH was used for testing (aqueous suspension containing approximately 37% nanoparticles, spheroidal-like shape, mean primary particle size 9.4 nm). Silver nitrate was tested as source of ionic silver. Soil nitrification was tested according to the internationally standardised and accepted assay for testing toxicity to soil microorganisms (OECD Test Guideline No. 216). Three soils were selected falling within the P10-P90 interval of European agricultural soils for pH, organic carbon content and cation exchange capacity. Total silver in soil, and total dissolved (0.45 µm membrane filtered) and 'truly' dissolved silver (1 kDa centrifuge filtered) in porewater were measured (ICP-MS). Toxicity of nanosilver on soil nitrification was similar to or less than silver nitrate when expressed on the basis of total Ag in soil. Total and truly dissolved Ag in porewater decreased over time after silver nitrate spiking, suggesting ageing processes. Concentrations were always higher or equal to corresponding values after nanosilver spiking. For nanosilver spiking, total and truly dissolved Ag in porewater initially increased, suggesting dissolution processes. From day 4-7 after spiking onwards, concentrations decreased over time suggesting that ageing becomes more important than dissolution. Truly dissolved Ag in porewater qualitatively explained observed toxicity of silver nitrate compared to nanosilver. The data show that soil ecotoxicity data for ionic silver are conservative for soil ecotoxicity of nanosilver.

100

Tackling nanoparticle fate assessment in surface waters - heteroaggregation as a key process

H. Walch, University of Vienna, Dep. of Environmental Geosciences / Environmental Geosciences; A. Praetorius, F. von der Kammer, T. Hofmann, University of Vienna / Department of Environmental Geosciences

The increasing use of engineered nanoparticles (ENPs) inevitably entails emissions to the environment, raising calls for nano-specific environmental risk assessment approaches and regulations. As surface waters are the major receiving compartment, assessing risks requires understanding the aquatic fate of ENPs which, unless soluble, is determined by aggregation, including homo- and heteroaggregation with natural suspended particulate matter (SPM), or stabilisation by natural organic matter (NOM). Due to the omnipresence and larger size of SPM, heteroaggregation is much more likely than homoaggregation. However, integration of this process into fate models and exposure assessment requrires parametrisation and is still limited by the lack of simple, yet environmentally relevant experimetal protocols. Such could be developed along the lines of the recently adopted OECD testing guideline 318 on ENP dispersion stability, currently accounting only for homoaggregation. The principles of homo- & heteroaggregation are basically the same: the probability of particle attachment is controlled by the intrinsic particle properties and modified by the hydrochemical conditions (pH, electrolytes, NOM). Distinct from homoaggregation is the complexity added to the system by SPM in the case of heteroaggregation. In this contribution we therefore propose an approach to develop a heteroaggregation testing protocol based on the OECD TG 318, with a focus on tackling SPM analogue selection. The development of such a protocol requires (1) selecting SPM analogues and hydrochemical conditions complex enough to represent relevant environmental characteristics, and simple enough for routine testing, (2) an easy-to-handle experimental setup to estimate a heteroaggregation parameter, and (3) an accurate experimental method to validate the latter. Point (1) requires informed simplifications based on a profound understanding of the system. Relevant hydrochemical testing conditions have been established for homoaggregation in the OECD TG 318 and will also apply for heteroaggregation. However, suitable analogues for natural SPM still need to be selected. We therefore reviewed literature for typical compositions of riverine SPM and carried out screening tests aiming at the creation of complex analogues representing relevant characteristics. Comparisons with simple SPM analogues revealed distinct aggregation behaviour, indicating the importance of complex SPM analogues for heteroaggregation.

Advances in environmental risk assessment of oil spills and offshore oil & gas operations (II)

101

MC-252 biomarkers as indicators of oil exposure and pollutant concentration in sediments of the northern Gulf of Mexico

L.M. Basirico, Louisiana State Univeristy; R.J. Portier, Louisiana State University / Environmental Sciences

Different types of crude oil can be identified by the arrangement of constituents, or their chemical fingerprint. In addition, chemical fingerprinting can be used to associate contaminated sediments with specific spill events like the *Deepwater Horizon* disaster of 2010. Mississippi Canyon-252 (MC-252) source oil, the type of crude oil specific to the *Deepwater Horizon* event, has been fully characterized by researchers at LSU, including pattern identification of the ion 217 and 218 hopane/sterane biomarker families. The presence of biomarkers in the sediments allows for the identification of MC-252 crude oil intrusion into sampled areas, as the arrangement of the ions delineates the source of the quantified PAHs. From 2012 to 2014, five inshore and three offshore transects representing the major estuarine and shoaling regions of the Mississippi River delta were sampled to measure sediment concentrations of polycyclic aromatic hydrocarbons (PAHs) and MC-252 biomarkers. Pattern A, pattern B or NO pattern was assigned to each sample based on a visual assessment of the chromatographic profiles. Additionally, a main effects-model was implemented in order to determine the impact of environmental variables, including the presence and pattern of MC-252 biomarkers, on the sediment concentrations of ten PAHs and three toxicity indicators. Ninety-three percent of all sampled sediments (N=1,032) did not contain MC-252 biomarkers. Of the sediment samples containing detectable crude oil biomarkers, 5 percent displayed pattern A and 2 percent, pattern B. Most of the samples containing the families of biomarkers were located at the southern end of the Barataria Bay transect, a region that experienced moderate to heavy oiling during the *Deepwater Horizon* oil spill event. Additionally, MC-252 biomarkers did not account for any of the variability in the concentrations of the measured pollutants according to the main effects-model used in the current study. The lack of MC-252 biomarkers in the vast majority of the sampled sediments, indicated that the PAH contamination in the current study was not from the Deepwater Horizon oil spill event. There is need for better markers of the origination of PAHs in freshwater and marine sediments. Furthermore, crude oil is not the only or even the best indicator of potential toxicity of these sediments.

102

Downregulation of hsp90 and increased intermoult duration in the blue crab, Callinectes sapidus, in response to oil exposure

S. Chiasson, Loyola University / EEB; S.M. Giltz, C.M. Taylor, Tulane University / Ecology & Evolutionary Biology

The 2010 Deepwater Horizon (DWH) oil spill in the northern Gulf of Mexico (NGOM) resulted in over 780 million liters of crude oil spilling into Gulf waters. In an effort to disperse the oil, nearly 7.6 million liters of dispersant was applied. Many commercially and recreationally important species reside in or near the area of the spill. The blue crab, Callinectes sapidus, is common in the NGOM and is both economically and ecologically important in this region. In this study, after exposing juvenile blue crabs to oil or a mixture of oil and dispersant we tested for relative expression of heat shock protein 90 (*hsp90*) by measuring the corresponding mRNA expression. Expression of *hsp90* is normally upregulated in response to thermal or environmental stress, and it also plays an important role in the regulation of estrogen dependent cell signaling. We also monitored crabs over two molts to test for effects on growth after exposing crabs to oil. Expression of *hsp90* was significantly downregulated in juvenile crabs exposed to dispersed oil but not oil alone. This suggests that dispersed oil interferes with either the pre-mRNA transcription of *hsp90* or potentially causes alternative splicing of pre-mRNA. *hsp90* expression in crabs exposed to oil alone was slightly elevated, although not significantly. However, the intermoult duration of crabs exposed to oil increased, meaning that exposure to oil results in delayed molting and therefore slower growth.

103

Physiological and molecular impacts of crude oil and/or dispersant-contaminated seawater and sediments on the sponge Halichondria panicea (phylum Porifera).

J. Vad, Heriot-Watt University / School of Energy, Geosciences, Infrastructure and Society; J.M. Roberts, The University of Edinburgh / Grant Institute; T.B. Henry, Heriot-Watt University / The School of Energy, Geoscience, Infrastructure and Society

Sponges (phylum Porifera) are a diverse group of filter-feeder organisms present in most aquatic environments. In the marine environment, sponges perform a wide range of ecological functions including cycling of nutrients such as carbon, nitrogen and silica; and, in areas where they are present at high densities (sponge grounds), they provide a habitat for a diverse range of benthic organisms. Because of their importance within marine ecosystems, the impacts of anthropogenic activities such as hydrocarbon exploration and production on marine sponges must be assessed. The objectives of this study were to:(1) determine the physiological impact of crude oil and/or dispersant contaminated seawater and sediments in model sponge *Halichondria panicea*; and (2) characterise the effects of crude oil and/or dispersant contaminated seawater exposure on the transcriptome of *H. panicea*. A series of 48-h experiments were conducted to investigate effects of exposure to seawater or sediments contaminated with Schiehallion crude oil and/or Slickgone NS dispersant in *H. panacea.* Sponge respiration rate and filtration rate (by clearance rate) were measured throughout exposures, and tissue samples were collected for evaluation of the transcriptome. Throughout the exposure experiments, respiration rate displayed a high inter-individual variability, consistent with scientific literature. A decreasing trend in respiration rate was observed when sponges were exposed to contaminated seawater or sediments. Filtration rate was significantly decreased in sponges exposed to contaminated seawater or sediments, and filtration rate did not recover for 48h after the end of the exposure to contaminated seawater. The transcriptome has been sequenced and analysis is underway to detect changes in gene expression patterns associated with treatments. Overall, results indicate that sponges respond to short-term exposure to crude oil and/or dispersants by cessation of their filtration behaviour. These initial results and our ongoing investigations will contribute to better understand the sensitivity of marine sponges to oil production activities.

104

Advances in the effects of UV on oil toxicity in aquatic organisms

A.P. Roberts, K. Bridges, University of North Texas / Advanced Environmental Research Institute; J. Morris, Abt Associates; B.J. Venables, University of North Texas / Advanced Environmental Research Institute; M.O. Krasnec, Abt Associates; M.L. Gielazyn, NOAA / co USEPA Region IV

Polycyclic aromatic hydrocarbons (PAHs) are a class of organic contaminants composed of two or more fused carbon rings and are a major constituent of crude oil. Exposure to ultraviolet radiation (UV) can exponentially increase the toxicity of photodynamic PAHs to biota, leading to adverse outcomes well below the threshold of other mechanisms of toxicity. This phenomenon is known as photo-induced toxicity and is well documented in a wide range of aquatic organisms. Consequently, laboratory tests investigating effects of PAH on aquatic biota which fail to account for potentiation by UV may significantly underestimate toxicity. The intensity of UV exposure to biota is highly variable within aquatic ecosystems, due to a number of factors intrinsic to the water column, and extrinsic factors (e.g. cloud cover, time of day, seasonal variations). Tissue repair mechanisms may be sufficient to counteract some effects of photo-induced toxicity during periods of relief from UV exposure. Here, we report the results of experiments in which larval red drum *(Sciaenops ocellatus)* and zooplankton (*Daphnia magna*) were exposed to either a single PAH (fluoranthene) or a complex PAH mixture prepared from weathered crude oil with varying PAH and UV exposure scenarios. Red drum tests were conducted as a single pulse exposure, and daphnia tests were conducted as static renewals. Toxicity (LC_{50}) was UV and PAH dependent in both species. In red drum tests, shorter PAH pre-exposure times resulted in LC_{50}s that were considerably lower than LC_{50}s associated with longer pre-exposure periods. This is likely due to lag time in the initiation of physiological metabolism/clearance mechanisms in the organism and loss of PAH from the test chamber. A similar pattern was observed in photoperiod testing for both species. Significant latent mortality was observed in daphnia for several days following the conclusion of the UV and PAH exposures. We also report the effects of various UV-modified photoproducts on marine fishes. Taken together, these data suggest that even short-term, transient exposure to low concentrations of PAHs (common during a spill event) results in acute toxicity in aquatic organisms, and those effects may be manifested outside of standard bioassay testing durations.

105

Photoenhanced Toxicity of Petroleum to Aquatic Invertebrates and Fish: Review of the Science

M.G. Barron, U.S. EPA / Gulf Ecology Division
Photoenhanced toxicity is a distinct mechanism of petroleum toxicity that is mediated by the interaction of solar radiation with specific polycyclic aromatic compounds (PACs) in oil. Phototoxicity is observed as a 2 to greater than 1000-fold increase in chemical toxicity to aquatic organisms that have also been exposed to light sources containing sufficient quantity and quality of ultraviolet radiation (UV). When tested under natural sunlight or laboratory sources of UV, fresh and weathered middle distillates, crudes and heavy oils can exhibit photoenhanced toxicity. These same products do not exhibit phototoxicity in standard test protocols because of low UV irradiance in laboratory lighting. Fresh water, estuarine and marine waters have been shown to have sufficient solar radiation exposure to elicit photoehanced toxicity, and a diversity of aquatic invertebrate and fish species can exhibit photoenhanced toxicity when exposed to combinations of oil and UV. Risks of photoenhanced toxicity will be greatest to early life stages of aquatic organisms that are translucent to UV and that inhabit the photic zone of the water column and intertidal areas exposed to oil.

106
Pilot microcosm study to assess the fate and toxicity of diluted bitumen in an outdoor aquatic environment.
J.M. Blais, University of Ottawa / Biology; M.L. Hanson, University of Manitoba / Environment and Geography; D. Orihel, Queens University; B. Hollebone, Environment Canada / Emergencies Science & Technology; V. Palace, M. Paterson, IISD-Experimental Lakes Area; J. Rodriguez Gil, University of Ottawa / Department of Biology
Pipelines are the safest mode of transporting Canada's oil to markets, but they are a concern for the public, especially the potential effects of diluted bitumen (dilbit) spills on the environment. We added diluted bitumen (dilbit) to two land-based microcosms (2 m diameter) containing water and sediment from a nearby lake at the IISD-Experimental Lakes Area in Northwestern Ontario for a span of 11 days, and compared our results to a control enclosure with no added dilbit. Microcosms were treated with 0 (Control), 0.15, or 1.5 liters of Cold Lake Winter Blend dilbit (CLB-W), representing dilutions of 1:10,000 and 1:1000 (oil:water, v/v), which spans the range of historical dilbit spills to water. Samples of water, sediment, air and oil were collected through the study in order to determine the fate, weathering, and behaviour of the dilbit. Total petroleum hydrocarbons in the high treatment microcosm gradually increased from under 100 mg/L in the first 24 hours to over 1200 mg/L by day 11, with no evidence of reaching equilibrium over this duration. Although a decrease in total phytoplankton biomass was observed in all microcosms over the study, the biomass in the high microcosm was about one-half or less than that in the control microcosm for the first week. Thereafter, the rate of biomass loss in the dilbit-treated microcosms slowed down, which could indicate recovery of the primary producers as the oil slick sank to the sediments. This study is among the first to examine the behaviour of dilbit in an outdoor setting under natural conditions of sunlight, wind and rain, and provides a case study that will inform future dilbit studies in natural (outdoor) environmental settings.

Fish model species in human and environmental toxicology (II)

107
Life-stage, and species-specific effects of dietary methylmercury exposure
K. Bridges, University of North Texas / Advanced Environmental Research Institute; Y. Zhang, University of North Texas Health Science Center; T. Curran, J.T. Magnuson, University of North Texas / Biology; M. Allen, University of North Texas Health Science Center; B.J. Venables, A.P. Roberts, University of North Texas / Advanced Environmental Research Institute
Mercury is a globally distributed contaminant, found even in remote aquatic ecosystems. Once deposition occurs, it can be microbially transformed into organic forms, such as methylmercury (MeHg) [1]. MeHg is highly bioavailable, and it bioaccumulates and biomagnifies in biota leading to potentially toxic body burdens in long-lived organisms at high trophic levels. MeHg can be actively transferred from mother to offspring, through large amino acid transporters with the potential to cause severe, irreversible effects on developing organisms. Here, we describe the developmental effects of exposure to maternally-transferred dietary MeHg on a model fish species *(Pimephales promelas)*. Exposure to environmentally relevant concentrations of MeHg during development led to alterations in the dopaminergic system, metabolome, gene expression, behavior, hatch time, size, and embryo-larval survival. Similarly, effects on the dopaminergic system in specific regions of the adult *P. promelas* brain were observed after a 30-day dietary exposure. Recently, a functional link between gut microbiota and dopamine production in teleosts has been established. The bidirectional communication between the gut and the central nervous system (CNS) is referred to as the gut-brain axis, which plays an important role in behavior, brain function, neurodevelopment, and the progression of neurodegenerative disorders. Therefore, we characterized MeHg-mediated changes to the gut microbiome composition in *P. promelas* adults. Because the dopaminergic system is highly conserved among taxa, we sought to confirm the altered dopamine concentrations in *P. promelas* brainin a higher vertebrate species. Metabolomics was performed on the mid-brains of male mice *(Mus musculus CD-1)* exposed to similar concentrations of dietary MeHg for 30-days. Changes in dopamine concentrations of the teleost brain were mirrored in the mid-brains of male mice, and several other significant changes to the mouse mid-brain metabolome were detected. Collectively, these results suggest current environmental exposure scenarios to MeHg are sufficient to induce a number of molecular-level changes that are associated with costs to whole organism fitness, with consequences for multiple life stages, and species. Due to the similar changes detected in mice, there is increasing evidence to suggest teleosts as a surrogate model species for studies assessing effects of MeHg on highly conserved systems in higher vertebrates.

108
Characterization of molecular toxicity pathways of Fluoxetine in rainbow trout and white sturgeon using RNA-Seq whole transcriptome analyses
A. Alcaraz, University of Saskatchewan - Toxicology Centre / Toxicology Centre; B.K. Eisner, University of Saskatchewan / Toxicology Centre; S. Tang, Chinese Center for Disease Control and Prevention; M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre
The increasing number of emerging chemical contaminants(ECC) and their unknown effects to aquatic ecosystems serves as an impetus to develop advanced environmental risk assessment(ERA) approaches to improve regulatory decision-making. This is because current ERA rely on live animal testing that are expensive, time consuming and of ethical concern. Furthermore, the use of model organisms does not assure protection towards native species because of the limited understanding of their relative sensitivities and vulnerability. Hence, there is a need to establish an unbiased approach to characterize toxicity pathways that allow probing of an entire biological system without a priori knowledge of the mechanisms of toxicity. Advances in 'omics technologies can improve current testing strategies as they offer high-throughput and cost-effective approaches to examine patterns of mechanistic toxicity which could guide endpoint selection across species in predictive ERA. The objective of this study was to develop toxicity pathway models to predict outcomes of regulatory relevance for the selective serotonin reuptake inhibitor, fluoxetine(FLX), in 2 fish species of concern in Canada. Juvenile rainbow trout(RBT) and white sturgeon(WS) were exposed to 125 µg/L FLX in 96h static-renewal system, and sequence-by-synthesis whole transcriptome analysis was used to determine global differential gene expression in fish livers and brains. A 0.05 cut-off false discovery rate identified differentially expressed contigs between FLX and control groups. A total of 406 and 429 contigs were significantly altered in RBT livers and brains, respectively. Of these, 238(59%) and 236(55%) matched unique gene names. In WS, 252 and 192 contigs were significantly altered in livers and brains, respectively, with 145(58%) matched unique genes in livers and 126(66%) matched unique genes in brains. Pathway analysis using ontologies based on zebrafish in KEGG and GO Consortium showed a total of 101 affected pathways. Over half(58%) of the affected pathways were involved in biological processes while others were involved in cellular components (13%), molecular function (18%), and genetic information processing (11%). The results of this study will be compared to apical outcomes assessed in a parallel chronic study, and which will allow the assembly of toxicity pathways across multiple levels of organization with the end goal of identifying molecular markers that are indicative of adverse effects.

109
Transgenerational effects of early life stage exposure to endocrine disruptors across biological scales in a euryhaline model fish
S.M. Brander, Oregon State University / Environmental and Molecular Toxicology; B. DeCourten, J. Forbes, University of North Carolina Wilmington / Biology and Marine Biology; N.P. Burns, University of North Carolina Wilmington / Department of Biology and Marine Biology; H. Roark, Hunter Roark / Biology and Marine Biology; J.W. White, Oregon State University / Department of Fisheries and Wildlife; A.C. Mehinto, Southern California Coastal Water Research Project / Toxicology; M.L. Settles, University of California Davis / Genome Center; R.E. Connon, University of California, Davis / School of Veterinary Medicine
Emerging research demonstrates that EDCs, which agonize, antagonize, and / or synergize the effects of endogenous hormones, can cause deleterious effects in adulthood as a result of early-life exposure, as well as transgenerational effects. A paucity of studies exist in non-model fish species, such as *Menidia beryllina*, a euryhaline fish with short generation time that is found throughout North America and is demonstrated to be sensitive to contaminants. As such, we exposed *Menidia beryllina* embryos (8 hpf) until 21 dph to environmentally relevant of an androgenic or estrogenic EDC of emerging concern: levonorgestrel (Levo) (10 ng/L), bifenthrin (Bif) (5 ng/L), respectively, and coupled this exposure with testing of an established androgenic or estrogenic EDC: trenbolone (TB) (10 ng/L), and ethinylestradiol (EE2) (5 ng/L). We are now evaluating the potential for transgenerational EDC effects across three generations, with EDC exposure isolated to the parental generation (to 21 dph) only, across biological scales. This study is examining changes in gene expression, DNA methylation, histological analysis of reproductive organs, as well as altered fecundity, sex ratio, morphology, and immune response in the F0, F1, and F2. We are also sequencing the *M. beryllina* genome. F0 results show that early-life exposure to EE2 significantly skewed adult sex ratios (feminized) relative to controls. Findings from the F0 and

F1 generations demonstrate that exposure to EDCs increased growth in the parental larvae, and that androgenic treatment groups (Levo, TB) maintain this growth through the subsequent F1 generation. In the F0 adults, differences in immune response are apparent between bifenthrin and levonorgestrel, and this pattern is stronger in F1 adults, with significantly greater T-cell proliferation in bifenthrin-exposed individuals relative to controls. Bifenthrin-exposed parental females have increased atretic follicles, and developmental defects are more pronounced in F1 embryos and larvae relative to parents. Future data gathered on gonadal histology, gene expression and DNA methylation will allow us to further hone in on the mechanisms causing higher order downstream effects. Elucidation of the mechanisms contributing to these higher order downstream effects will inform adverse outcome pathways, as well as allow for the quantification and comparison of responses to established and emerging endocrine disruptors across multiple biological scales.

110
Integrated OMICS and imaging for a better understanding of ecotoxicological mechanisms - PAH developmental toxicity as an example
E. Vehniäinen, C. Rigaud, A.N. Eriksson, University of Jyvaskyla / Department of Biological and Environmental Science; A. Krasnov, NOFIMA; M. Keinänen, University of Eastern Finland; A. Rokka, S. Saraei, T. Suomi, Turku Centre for Biotechnology; A. Laiho, University of Turku and Åbo Akademi University; J. Lihavainen, University of Helsinki; J. Haverinen, M. Vornanen, University of Eastern Finland; J.V. Kukkonen, University of Jyvaskyla / Biological and Environmental Science
Polycyclic aromatic hydrocarbons (PAHs) are ubiquitous contaminants in the environment. Many of them cause developmental defects in fish, and cardiovascular tissue seems to be the most sensitive tissue. The mechanisms of toxicity remain largely unresolved for many PAHs, though partial adverse outcome pathways (AOPs) exist for those that are aryl hydrocarbon receptor (AhR) agonists. Rainbow trout (*Oncorhynchus mykiss*) yolk sac larvae were exposed to sublethal concentrations of PAHs that act via different mechanisms of toxicity: Retene, an AhR agonist causing dioxin-like toxicity; pyrene and phenanthrene, weak AhR agonists causing toxicity independently of AhR; and fluoranthene, a CYP1A inhibitor interfering with PAH metabolism. Also the effects of a mixture of retene and fluoranthene were studied. Information was gained at multiple levels of biological organization to reveal the mechanisms of toxic action. Changes in cardiac transcriptome, proteome and metabolome were explored over time. Physiology and function of the heart were also studied. At the whole organism level, growth, yolk consumption, and developmental defects and abnormalities were monitored. Each PAH caused a unique pattern in OMICS analyses, and the mixture of retene and fluoranthene caused a different transcriptomic profile from that of each of the single compounds. Retene differentially regulated genes involved e.g. in muscle contraction and ion metabolism (ion channels). Retene and phenanthrene impaired cardiac function in larval rainbow trout. Both caused bradycardia, and phenanthrene caused also arrhythmias. Phenanthrene affected cardiomyocyte electrical characteristics. As cardiovascular development is modulated by the beating heart and blood flow, alterations in cardiac function during development may have long-lasting impacts in cardiovascular tissues. Different PAHs clearly have different mechanisms of toxicity. The transcriptomic changes can at least partly account for the cardiotoxicity of retene, but the cardiotoxicity of phenanthrene seems to involve a direct effect on cardiac ion channels.

111
Physiological / Reproductive Status of Native Fish Exposed to a Complex Chemical Mixture in the BíoBio River, Central Chile
M. Quiroz-Jara, Universidad de concepcion / Biomarcadores; S. Casini, University of Siena / Scienze Fisiche della Terra e dellAmbiente; M. Fossi, University of Siena / Department of Physical Sciences, Earth and Environment; R.O. Barra, Universidad de concepcion / Aquatic systems; J. Gavilán, Universidad de concepcion / Celular Biology, Faculty of Biological Science
The BioBío River (Central Chile) is the third most important basin in Chile and is considered a Biodiversity Hot Spot with 17 native species. It presents a high degree of intervention due to the use of water resources for various industrial, agricultural and urban activities, with many focal places of point and diffuse contamination along its main axis, in addition to a high degree of modification of land use associated with the basin. The high degree of intervention and fragmentation of this river has affected the biota and water quality mainly in its lower third, due to the convergence of complex chemical mixture and anthropic interventions in the main course. The objective of this study is to determine how the development associated with this river impacts the physiological/reproductive state of the native species *Percilia irwini* ($n = 66$) *in situ*. Different sublethal responses were evaluated through biomarkers at different levels of biological organization (Biochemical, cellular, and individual) and environmental parameters (pH, temperature, conductivity and total dissolved solids). The results obtained indicate an increase in the Hepatic EROD activity (ethoxyresorufin-O-deethylase) towards the lower third of the river. The Gonadosomatic Index (100 * (Total weight / (Total weight - Gonadal weight))) shows an increase towards the lower third, however, the gonadal histology indicates a protoplasmic growth and reduction of the diameter of the different stages of development of the oocytes determined for this species ($n = 2332$, $p < 0.05$). On the other hand, the collected specimens show a difference in weight and length, presenting specimens of less frequency of length in the lower third with respect to those present in areas with less intervention. These responses are associated with the increase of the values of environmental parameters towards this zone. The results of this study indicate a gradient of adverse biological effects by the convergence of point and diffuse contamination of complex chemical mixture and establishes the possible relationships between the physiological/reproductive alterations observed and the high degree of intervention of this river.

112
Poster spotlight: MO248, MO249, MO256

Sustainable Development Goals: the global context defining the agenda for government, business and academia

113
Can the Agenda 2030 and the Sustainable Development Goals be the drivers to change the world?
E. Giovannini, ASviS

114
How the SDGs are being addressed in Horizon 2020
M. Tamborra, European Commission - DG Research and Innovation

115
Examples of EU projects related to SDGs
M. Recchioni, European Commission - EASME

116
Why SDGs are relevant for a large enterprise
A. Valcalda, ENEL

117
Conclusions
E. Tonda, UN Environment / Division of Technology, Industry and Economics (DTIE)

118
Questions and answers

Mercury Biogeosciences - Fate, Effects and Policy

119
Rethinking Atmospheric Mercury Chemistry
M. Gustin, University of Nevada, Reno / Natural Resources and Environmental Science
Mercury (Hg) is considered a global pollutant. This is because it has a long atmospheric residence time. Because of the continued and increasing emissions of this pollutant to the atmosphere associated with anthropogenic activities, and the fact that once released from a geologic repository an atom of Hg may be potentially bioavailable for thousands of years, the Minamata Convention was developed and has come into force. This global treaty focuses on protecting human health and the environment from the adverse effects of mercury. There are 3 general forms in the atmosphere- gaseous elemental Hg, gaseous oxidized Hg (Hg(I) or Hg(II) compounds), and that bound to particles. Gaseous elemental Hg can be transformed to gaseous oxidized Hg (GOM) by a variety of atmospheric oxidants. Once generated, GOM is readily deposited to ecosystems. Understanding the chemistry of GOM is important for predicting deposition velocities, availability in ecosystems, and potential for conversion to methylmercury. Methylmercury is a subtle neurotoxin and is bio-accumulated in ecosystems. Recent work using cation exchange membranes in the University of Nevada Reno –Reactive Mercury Active System (UNR-RMAS), and an air Hg calibrator system (Utah State University) have demonstrated that the standard measurement method for GOM –collection on a KCl denuder- results in underestimation of GOM concentrations by 2-to -13 times. In addition, thermal desorption profiles of GOM compounds collected using nylon membranes indicate that different chemical forms exist in the atmosphere. Data collected in urban areas, in the marine boundary layer, and at high elevation indicate that GOM compounds present are influenced by oxidants present in the air.

Different oxidized forms are produced in the free troposphere, marine boundary layer, and due to local oxidants in urban areas. Understanding atmospheric chemistry of GOM is important for developing instruments that will accurately measure GOM, and helping guide policymakers in developing solutions for reducing Hg emissions and contamination of ecosystems.

120
Evaluating spatial dynamics and species variation on mercury and selenium molar ratios in Northeast Atlantic marine fish communities
A.M. Azad, NIFES / Contaminants and biohazards; S. Frantzen, B.M. Nilsen, A. Duinker, National Institute of Nutrition and Seafood Research / Contaminants and biohazards; L. Madsen, National Institute of Nutrition and Seafood Research / Seafood in modelsystems; M.S. Bank, Institute of Marine Research / Contaminants and biohazards; A. Maage, NIFES / Monitoring Programme
Seafood is the main dietary source of methylmercury (MeHg) exposure for humans and MeHg is a primary contaminant of concern for seafood consumption advisories. Co-occurrence of the Selenium (Se) and mercury (Hg) in seafood directly affect their bioavailability and toxicity. The protective and antagonistic effects of Se against Hg have been addressed in several studies although consensus on this issue is still widely debated. Here we evaluate Se:Hg molar ratios across a latitudinal gradient in the Northeast Atlantic Ocean (NEAO) fish communities to assess species variation and spatial trends. In this study, the concentrations of total Hg and Se in 17 teleost fish species were measured using ICP-MS following microwave digestion(Julshamn, 2007 #72) and the Se:Hg molar ratios were calculated. Marine fish samples (n = 8525) were collected from the Barents Sea, Norwegian Sea, North Sea, Skagerrak, North Atlantic and Norwegian fjords and coastal areas between 2006-2015. The mean Se levels ranged from 0.27 to 0.78 mg kg^{-1} wet weight (ww) in Atlantic cod (*Gadus morhua*) and wolffishes (*Anarhichas* spp.), respectively. The mean Hg levels ranged from 0.04 to 0.77 mg kg^{-1} ww with the lowest level in Atlantic mackerel (*Scomber scombrus*) and the highest in blue ling (*Molva dipterygia*), leading to variation in the mean Se:Hg molar ratios from 1.9 in blue ling to 43.2 in Atlantic mackerel with Hg levels contributing most to the variation. In general, pelagic species had the lowest Hg levels and the highest Se:Hg ratios whereas deepwater demersal species had the highest Hg levels and the lowest Se:Hg ratios. Most species had a large portion (more than 50%) of specimens with a Se:Hg ratio exceeding 5 except for tusk (*Brosme brosme*) (4% less than 1, 53% between 1 and 5) and blue ling (19% less than 1, 80% between 1 and 5). Se and Hg levels showed weak positive correlation (R from 0.24 to 0.70) in 13 of 17 species. The Se:Hg ratio was negatively correlated to fish length and Hg levels. Mean Se:Hg molar ratios varied across offshore areas for all species and a gradual decreasing trend was found for all species from north to south due to increasing Hg concentration. Considering the EU maximum level of Hg and a Se:Hg molar ratio above one, we emphasize that fish from NEAO are generally safe regarding Hg contamination. Less than 1% of the total analyzed specimens had Se:Hg molar ratio less than 1 and the surplus Se may ameliorate the toxic effect of Hg to some extent.

121
The interaction of mercury and selenium across environmental media
J.R. Gerson, Duke University; L. Naslund, Duke University / Biology; H. Hsu-Kim, Duke University / Department of Civil Environmental Engineering; E. Bernhardt, Duke University / Biology
Both mercury (Hg) and selenium (Se) can bioaccumulate within aquatic ecosystems and are toxic to organisms when found at high concentrations. Individually, high concentrations of either Hg or Se in water result in harmful impacts to biota, but the presence of both trace metals has been suggested to have antagonistic effects. Due to this relationship, many studies propose that increased environmental concentrations and consumption of Se is a pathway to reduce Hg toxicity in organisms. Yet, despite this important link, little is understood about the biogeochemical processes that promote this antagonistic relationship. In fact, only two published studies have simultaneously examined the interaction of Hg and Se in both multicellular organisms and environmental media containing microbes (sediment, water). In this study, we seek to better understand the uptake of Hg and Se into biota, as well as the biogeochemical conditions that promote this pathway and evidence for antagonistic effects. We use samples collected from the mountaintop mined region of West Virginia, USA, where high concentrations of contaminants have previously been found in these watersheds. To answer this research question, we analyze total Hg (THg), MeHg, total Se (TSe), and Se speciation in water, sediment, biofilm, stream macroinvertebrate, and spider samples. Our results show that Hg, MeHg, and Se are bioaccumulating in the food chain, with the highest concentrations found in macroinvertebrates. We also find that there is an aquatic-terrestrial subsidy of mercury and selenium, with spiders containing elevated concentrations of these two contaminants. Our data provide evidence of a wedge-shaped cluster for the relationship between Se and percent MeHg in bulk sediment and biofilm, suggesting that at high Se concentrations, percent MeHg in bulk sediment and biofilm are reduced. In craneflies and spiders, we find a negative correlation between Se concentration and both absolute MeHg concentrations and percent MeHg. These results suggest that Se inhibition of MeHg accumulation might occur both at the microbial and macroinvertebrate levels.

122
Constraining Uncertainties in the Global Mass Balance of Mercury Using Observations and a Bayesian approach
S. Mustala, IIT Hyderabad; A. Qureshi, IIT Hyderabad / Civil Engineering
Uncertainties in global mass balance of mercury are constrained in this work using all the currently available observations of mercury species in the global environment, and a previously published multimedia model for mercury, Unit-WorM3. Reducing uncertainties help in estimating mercury concentrations and mass balances with greater confidence. Ten key input parameters that were identified to be significantly contributing to the output uncertainties in previous studies. These included: emissions of mercury to the atmosphere, reduction and oxidation of mercury in surface and sub-surface oceans, and partition coefficients of mercury species groups [Hg(0), Hg(II), Hg-p] in surface and sub-surface oceans. Then, a survey of literature on observations of mercury in the global environment is made. As these observations (for example, concentration of total mercury in air) are also key model outputs, we can update model inputs by comparing model simulated outputs to the actual observations. For this updating, a Markov chain Monte Carlo (MCMC) technique called Metropolis Hastings which is based on the Bayes rule is adopted. The observed concentrations of Hg(0) in atmosphere, dissolved gaseous mercury and total mercury in surface ocean are collected from published literature and used to obtain a likelihood function. Input parameters and their confidence range are revised. A revised mass balance is obtained through a forward Monte Carlo analysis using updated inputs. It is found that the uncertainties in key input parameters (such as partitioning of reducible divalent mercury between suspended solids and water in surface oceans) have been constrained to a considerable extent, from factor 100 to about a factor 30. As a result of updating of input uncertainties, uncertainties in key output results, such as evasion of mercury from ocean to atmosphere, are also reduced. From factor 650 to factor 50, and in net reduction of Hg(II) to Hg(0) in atmosphere from factor 36 to 10. Analysis of contribution to variances of inputs to output variacnes suggests that, still, parameters describing oceanic processes such as partitioning between suspended solids and water, and mercury redox reactions in oceans, contribute more to variances in key model outputs, compared to parameters such as global mercury emissions. Therefore, more significant effort must be made in understanding mercury process in oceans rather than in conducting emission inventory exercises.

123
Effects of probable nutrient limitation on the relationship between mercury and marine microorganisms in seawater
I. Zivkovic, V. Fajon, J. Kotnik, Jozef Stefan Institute; M. Solic, J. Lusic, G. Kuspilic, Institute of Oceanography and Fisheries; M. Ordulj, University of Split; F. Matic, B. Grbec, N. Bojanic, Z. Nincevic Gladan, Institute of Oceanography and Fisheries; M. Horvat, Jozef Stefan Institute
Microbial transformations of monomethylmercury (MMHg) and dissolved gaseous mercury (DGM) at the lower marine trophic levels are still not well understood. This is especially important in oligotrophic and nutrient-limited seas, where microbial food web and microbial loop dominate over classical (herbivorous) food web. Our research focused on the examination of the effects of probable phosphorus limitation (^{P}P-limitation) on relations between different mercury fractions (total methylated mercury – MeHg, and DGM) and autotrophic and heterotrophic microorganisms. We determined total mercury (THg), MeHg and DGM, alongside with relevant microbiological and chemical parameters in the Central Adriatic Sea. Using statistical analysis (non-metric multi-dimensional scaling, principal component analysis, Pearson’s product-moment correlations), we assessed the microbial effects on Hg transformations and bioaccumulation. Only in the absence of ^{P}P-limitation conditions ($^{NO-P}$P-limitation), we found that MeHg was significantly related to most chemical and microbial parameters, which is an indication of metabolism-dependent Hg transformations. The activity of heterotrophic low nucleic acid bacteria seems responsible for most of Hg methylation in seawater under $^{NO-P}$P-limitation. Under $^{NO-P}$P-limitation conditions, DGM shows strong correlation with microbial fractions and chlorophyll *a*, which confirms previous research about biological DGM production. Contrary to MeHg, DGM transformations are probably not metabolically dependent, as most of these correlations can also be observed under ^{P}P-limitation. MMHg biomagnification from microseston to mesozooplankton was observed through an increased bioaccumulation factor between these fractions. Mercury biotransformation and uptake are probably enhanced under $^{NO-P}$P-limitation, which emphasizes its impact on Hg transfer to higher trophic levels. In order to test our nutrient-limitation hypothesis, we have performed statistical analysis on previously published data from the Southern Atlantic Ocean. We found similar correlations between MeHg (DGM) and physico-chemical characteristics of seawater under probable nitrogen limitation compared to those found under ^{P}P-limitation in our study. These results indicate that mercury methylation is impeded in seawater under probable nutrient limitations.

124
Poster spotlight: MO333, MO334, MO335

Bioavailability and realistic risk assessment of organic

125
Anisotropic exchange kinetics of organic contaminants with passive samplers in stagnant sediment: is multiple-thickness passive sampling the better alternative?
D. Gilbert, NGI / Environmental Technology; A.M. Oen, Norwegian Geotechnical Inst. / Environmental Technology; N. Berrojalbiz, Norwegian Geotechnical Institute / Environmental Technology; H. Arp, NGI / Environmental Technology
Passive sampling with thin polymer sheets is increasingly recognized as a superior monitoring tool for obtaining realistic risk assessments of nonpolar organic chemicals in sediment porewater. For deducing true freely dissolved concentrations in the aqueous phase from measured polymer concentrations, the compounds are required to reach thermodynamic equilibrium between the polymer and the water phase. However, for in-situ deployment in stagnant sediment equilibration times are beyond practical time scales. The spiking of passive samplers with performance reference compounds (PRCs) has therefore been introduced as a way to deduce equilibrium concentrations from the release of PRCs over the deployment period. This approach relies on the assumption of isotropic exchange kinetics between the uptake of the native compounds into the polymer and the release of spiked PRCs from the polymer. Our aim was to test whether this assumption is valid in stagnant sediments in in-situ and ex-situ conditions, considering different types of sediment and PRC spiked concentrations. For the field study, we immersed low-density polyethylene (PE) and silicone thin sheet passive samplers of multiple thicknesses and spiked with PRCs for 5 months into contaminated sediment in the Oslo harbour. For the ex-situ study, Oslo harbour sediment and Horten harbour sediment was incubated with PRC-spiked PE under stagnant conditions at room temperature in the laboratory and samples were taken at various time points. From the PRC depletion field data, sediment porewater concentrations were modelled using the Fernandez-one-dimensional-diffusion model. In addition, the field equilibrium concentrations were modelled using a multiple thickness one-dimensional diffusion model. The ex-situ uptake and release data were modelled with a one-dimensional diffusion model (uptake) and a simple exponential one-compartment model (release). The results showed for both the in-situ and ex-situ data that uptake and release kinetics were not identical. In addition, the ex-situ experiment revealed that PRC release kinetics is also dependent on the initial PRC spiking concentration. In conclusion, the data question the usefulness of PRCs for passive sampling in sediment, as the use of polymers of multiple thicknesses can produce results that are free from biases caused by anisotropic exchange kinetics.

126
Sediment toxicity of chlorpyrifos: whole sediment bioassay vs. silicon disc passive dosing
K. Walker, University of Amsterdam / IBED-ELD; N. Wieringa, University of Amsterdam/IBED Institute / FAME; M. de Baat, M. Kraak, University of Amsterdam / IBED-FAME; J. Parsons, University of Amsterdam / IBED-ELD; S. Droge, University of Amsterdam/IBED Institute / IBED
Realistic risk assessment of sediments polluted with organic contaminants is much more complex than tests with water samples. Physical and biological processes as well as contaminant bioavailability, may strongly influence the adverse effects on test organisms in whole sediment testing. The current work is the first of a series of studies in our group on whether sediment-equilibrated silicon rubber (ESR) could allow for a major simplification of the assessment of the overal impact of organic contaminants in sediment. The aim of the current study was to demonstrate that ESR can transfer the chemical activity of the insecticide chlorpyrifos from spiked sediment to aquatic bioassay with ESR as a passive dosing material. The effect level of chlorpyrifos in a 28d whole sediment bioassay was compared to effect levels observed in a 4d ESR passive dosing test using first instar larvae of the midge *Chironomus riparius*. Additional sampling with polyacrylate solid phase microextraction (SPME) fibers in both sediment and ESR dosed water was used to align the bioavailable concentrations in both tests designs. The ESR samplers accumulated chlorpyrifos up to 12% of the total spiked chlorpyrifos amount within 1 month. SPME samplers in sediment had 1-3x lower concentrations than SPME equilibrated with ESR. Thus, the chemical activity in sediment as well as that released from the ESR in water were comparable within a factor of 3. The insecticide chlorpyrifos showed only slightly more toxic effect levels in a 28d whole sediment test than in the 4d ESR dosing assay. Bioavailable concentrations in SPME samples in both assays indicated lethally toxic freely dissolved concentrations in the range of 0.02 - 0.1 µg/L. This study suggests that the 4d ESR dosing assay with sensitive first instar midge larvae provides valuable and realistic insight in the toxic potency of insecticide contaminated sediment comparable to much more elaborate 28d whole sediment tests. Also, at lowest tested toxic insecticide levels, concentrations in SPME extracts were close to detection limits, so accurate measurements of safe bioavailable chlorpyrifos concentrations via SPME becomes problematic. This suggests that ESR dosing assays and chronic whole sediment studies could be used more effectively to demonstrate specific pollutant toxicity than chemical analysis of realistic sediment exposure levels.

127
Implementing desorption extraction methods into bioavailability-oriented bioremediation
R. Posada, IRNAS CSIC / Agroquimica y Conservacion del Suelo; J. Garcia, Instituto de Recursos Naturales y Agrobiologia de Sevilla CSIC; M. Cantos, IRNAS CSIC; J. Ortega-Calvo, Instituto de Recursos Naturales y Agrobiologia / Agroquimica y Conservacion del Suelo
Regulators are starting to consider bioavailability within retrospective risk assessment frameworks for organic chemicals, however, implementation is not straightforward because the developments of bioavailability science have not always been translated into ready-to-use approaches for regulators. Possible pathways for translating bioavailability science into regulation of organic chemicals have recently been identified (*Environ. Sci. Technol*. 49:10255-10264, 2015). A simplified approach was proposed in which the assessments of soil/sediment and the target chemicals should be based on two measurable values: the total extractable concentration and the bioavailable concentration as measured with robust and reproducible chemical and/or biological methods. One of the chemical methods which has been proposed to measure bioavailability of hydrophobic chemicals (HOCs) such as PAHs is the desorption extraction with Tenax during 20 h (ISO 17402) (Environ. Toxicol. Chem. 20:706–711, 2001; Integr Environ Assess Manag, 11:208–220, 2015). Understanding the role of bioavailability in the biodegradation of chemicals is relevant not only for retrospective contaminated land management but also prospective risk assessment applied in the approval and regulation of organic chemicals. With the aim of providing pathways for implementation into regulatory contexts, we carrying out desorption extraction measurements with Tenax in a greenhouse experiment in which different strategies (use of microorganisms, plants and (bio)surfactants) in a PAHs contaminated soil, oriented to decrease the fraction of bioavailable pollutants. The most relevant result in this study was that bioavailability increases in planted soils receiving rhamnolipids, as evidenced by Tenax extraction and it was accompanied by an increased biodegradation in soil slurries. In conclusion, tenax extraction during 20 h resulted a reliable and robust method to determine bioavailable concentrations in a wide set of operational conditions ranging from a different time scale to dissimilar treatments (planting, biosurfactant application, etc.).

128
Prediction of very slow biodegradation of PAHs in soil and validation in a pilot of 25 years
R. Rietra, Alterra and Wageningen University / sustainable soil management; J. Harmsen, Wageningen Environmental Research / CALM
Biodegradation of polycyclic aromatic hydrocarbons (PAHs) and mineral has been followed during periods up to 25 years. Biodegradation took place on real scale fields (six landfarms and one depot) within the Netherlands. On these fields dredged sediments were ripened and PAHs were measured in the original sediment and resulting soil. PAHs were present in concentrations up to 550 mg/kg d.m (Dutch list). The objective of the investigation was to find experimental prove on the existence of long term biodegradation in field conditions. The measured data showed continuation of PAH degradation and this could be distinguished in 1) fast degradation in the first year 2) slow degradation in the following 6 years and 3) very slow degradation of PAHs from 6 years until at least 25 years. Knowing the long time necessary for biodegradation, it will be necessary to supply regulators with data and prediction to convince them that biodegradation will be a safe option to remediate the contaminated soil or sediment Bioavailability as measured with Tenax can be used to explain and predict the rate of biodegradation of PAHs. Three desorbing fractions can be measured. Tenax applied at 20ºC gives the fast desorbing fractions, applied at 60ºC the slow desorbing fraction is measured. The amount in the residue represents the very slow desorbing fraction. These are the same fractions as considered in the approach of Ortega-Calvo et al., (2015). In the soil, desorption makes the PAHs bioavailable and if conditions allow biodegradation (sufficient oxygen and water), this will occur. Using results measured in stored original sediment the different bioavailable fractions were measured and using a model with three first order derations (fast, slow and very slow) the really observed degradation curve could be predicted. Moreover, the fractions measured in present soils, shows that biodegradation will continue, however with a very small slow rate. Experiments applied in the nineties of last century had already shown that risks measured with bioassays were already not measurable after 6 years of landfarming. After 25 years the PAHs concentrations were 10 mg/kg d.m. or lower which made the soil reusable within the Dutch legislation.

129
Linking bioavailability of complex mixture to toxicity changes to assess recovery of contaminated soils
S. Cipullo, School of Water, Energy and Environment, Cranfield Water Science Institute / School of Water, Energy and Environment, Cranfield Water Science Institute; I.G. Negrin, L. Claveau, Cranfield University / School of Water Energy and Environment Cranfield Water Science Institute; B. Snapir, Cranfield University / School of Water Energy and Environment Cranfield Agrifood; P. Campo Moreno, Cranfield Univeristy / Cranfield Water Science Institute; F. Coulon, Cranfield University / School of Water Energy and Environment Cranfield Water Science Institute
A six-month laboratory scale experiment was carried out to assess the effect of biochar and compost amendment on the behaviour and toxicity of tar mixtures in

contaminated soils collected from two gasworks sites in the UK. Site 1 had a total polycyclic aromatic hydrocarbons (PAHs) concentration of 3500 mg/kg, and total aliphatic concentration of 1500 mg/kg. Site 2 had a lower concentration of both aromatic and aliphatic hydrocarbons 250 mg/kg and 350 mg/kg respectively. Heavy metals (HMs) concentrations in both soils ranged between 600 and 1200 mg/kg and the main elements were Zn, Pb and Cu. Both soils were amended with either 5% biochar or 15% compost in order to achieve metal stabilization and enhance tar oil degradation. The total and bioavailable concentrations of the organic fractions as well as HMs were determined after 0, 30, 90 and 180 days. Further to this microbial biomass, soil respiration, phospholipid analysis, Microtox toxicity, earthworm's lethality and seeds germination were carried out to assess how ecological health changed as the soils underwent remediation treatments. The study showed that, for both soils, microcosms amended with compost showed the most significant reductions in toxicity. The microbial number and respiration activity increased by two orders of magnitude after compost addition. Conversely, there was no significant difference between non-treated and biochar amended soils. At the onset of the experiment, no seed germination was observed in Soil 1 (non-treated) whereas an increase in seed germination was observed after 90 days for mustard and rye grass, and after 30 days for peas. The compost treatments had the highest percentage germination for both soils across all seeds types. Similarly, the earthworms assay showed there was significantly greater survival rate at the end of the experiment compared to the onset. The biochar treatment resulted in a lower survival counts compared with compost treatment, with the non-treated samples having the lowest results. Preliminary results suggest that addition of compost and biochar accelerated the degradation rate of hydrocarbons compound and contributed in reducing toxicity of the soils. The degradation of PAHs and re-distribution of HMs will be evaluated with a multivariate analysis; data will be explored to highlight associations between contaminant's concentration (reduction) and its influence on biological properties and toxicological responses in mixed contaminated soils.

130

Impact of Biochar Additions to Soil on Contaminant Sorption and Plant Bioavailability

W.J. Doucette, Utah State University / Utah Water Research Laboratory; J. Flashinski, D. McAvoy, Utah State University

Reclaimed water is increasingly used in arid and semi-arid regions for irrigation. Contaminants in the reclaimed water, (e.g. pharmaceuticals and personal care products (PPCPs) and metals) could accumulate in exposed crops. Biochar is a potentially cost-effective soil amendment and contaminant sorbent that could reduce the plant bioavailability of reclaimed water associated contaminants. Biochar is biological material (often plant material) that has partly or fully undergone pyrolysis (decomposition at high temperatures with no oxygen). The main objective of this study is to investigate the impact of wood biochar on the crop bioavailability of selected PPCPs often found in reclaimed water. A secondary objective is to quantify the contaminant sorption-desorption characteristics in the amended soils and to determine if there is a relationship wirh plant bioavailability. PPCPs were selected as target contaminants because of their widespread occurrence in reclaimed water and their potential impact on animals feeding on the irrigated crops. Target PPCPs were selected based on chemical properties, widespread use, frequent detection in WWTP effluent, and potential risk to the environment. The target PPCPs represent a range of therapeutic uses including antibiotics linked to antibiotic resistance in bacteria (sulfamethoxazole (SMZ) and triclosan (TRI)), an anticonvulsant that prevents seizures and relieves nerve pain (carbamazepine), an antidepressant (fluoxetine (FLX)), and an antihyperlipidemic (gemfibrozil (GBZ)). Atrazine (ATZ) was also selected because it is an herbicide commonly used on corn and has been used in a number of sorption studies with biochar. Pinyon Juniper, Russian Olive, and Lodgepole Pine derived biochars were chosen because they were produced from tree species that often require removal because they are considered invasive or due to insect infestations.. Corn was used as the test plant because of its commercial value and has been grown with reclaimed water in the past. After the 28 day growing period, it was found that there was no negative impact of the biochars on corn growth. Once the plant tissue analysis and sorption studies are completed, the impact of biochars on contaminant uptake will be evaluated along with its relationship to PPCP sorption/desorption behavior. Extaction and analysis of the plant tissue is being conducted along with the sorption/desorption experiments. Final results expected by December 2017.

LCIA method developments in a global perspective: Status and outlook (II)

131

A novel framework for a new generation of water consumption indicators in LCA and footprint studies

M. Núñez, TU Berlin / Sustainable Engineering; R.K. Rosenbaum, National Research Institute of Science and Technology for Environment and Agriculture - Irstea / UMR ITAP; S. Karimpour, CIRAIG; A. Boulay, CIRAIG - École Polytechnique de Montréal / Chemical engineering department; M.J. Lathuilliere, University of British Columbia / IRES; M. Margni, CIRAIG - École Polytechnique de Montréal / Mathematical and Industrial engineering; L. Scherer, Institute of Environmental Sciences (CML); F. Verones, NTNU / Department of Energy and Process Engineering; S. Pfister, ETH Zurich / Institute of Environmental Engineering

Available freshwater consumption indicators for use in LCA and water footprint studies implemented in LCA software are based on either a volumetric approach of the water consumed (what we called first generation of indicators) or on stress indices (what we called second generation of indicators). The first generation only performs an inventory and thus a partial footprint assessment according to ISO 14046, while the second generation shows limitations in describing the consequences of a lack of water at the endpoint. To date, no LCA method comprehensively distinguishes water masses (e.g. surface water and ground water) and their transport flows (e.g. from river to atmosphere) within the boundaries of a watershed (e.g. evaporation) and beyond (e.g. groundwater and air advection), thus overlooking details in hydrological processes that affect environmental relevance of the assessment. In addition, a structured LCIA framework is currently lacking, as can be observed by the scattered and often incompatible developments of water fate and impact assessment models published in recent years. These models are all valuable contributions in themselves, but impossible to combine to an integrated, global characterization model that makes such developments operational in LCA. The challenge of improving environmental relevance of current water consumption indicators has been tacked by the Water Use in LCA (WULCA) working group within the UN Environment Life Cycle Initiative by developing consensus-based guidelines for the third generation of water consumption indicators, with a focus on consequences of water consumption on ecosystem quality. Unlike currently available approaches, the new generation of indicators will include, for the first time, fate and transport of water flows in a global regionalized multimedia model. In the framework, we provided spatial and temporal specifications for the LCIA and specified data requirements for the LCI. To demonstrate how to apply principles and recommendations of the guidelines, we developed an illustrative example. The operationalisation of the guidelines has the potential to harmonize current and future methods under a unique framework and to enhance the environmental relevance of the water use impact category in LCA.

132

A midpoint indicator for freshwater resources

C. Pradinaud, IRSTEA Montpellier / ITAP ELSA; G. Junqua, Ecole des Mines dAlès / LGEI; P. Roux, Irstea / ITAP ELSA-PACT; A. Hélias, Montpellier SupAgro / LBE ELSA; Y. Penru, SUEZ groupe / CIRSEE; R.K. Rosenbaum, IRSTEA Montpellier / UMR ITAP ELSA

Freshwater resource has been recognized as being a safeguard subject within the Area of Protection (AoP) natural resources (WULCA resource group framework). Besides depletion also long-term pollution threatens the sustainability of freshwater resources, but currently no LCIA model links emissions to potential damage on freshwater as a natural resource. This study proposes a characterisation model to assess the potential impacts on freshwater resources generated by persistent changes in water quality caused by chemical emissions. The relevance of this new approach regarding the methodological issues of long-term (toxic) impacts is also discussed. As recommended in the WULCA freshwater resources framework the concept of recovery period is used: when the recovery period lasts longer than a given period of time, potential impacts to freshwater resources (i.e. affecting freshwater availability for future generations) need to be considered. Based on literature review, we set the time period at 100 years, which requires a dynamic fate model. The dynamic fate factor is calculated with a (currently non-public) dynamic version of the USEtox® model. This provides the time-integrated pollutant mass remaining in the freshwater compartment (at continental scale) after 100 years m>100 (in kg.day). Then, the quantification of the effect factor is based on the concept of adsorption processes with activated carbon. This allows for an indicator based on physical properties of the pollutant which is (i) substance specific, (ii) dependent on the level of pollution (i.e. chemical mass in freshwater), and (ii) proportional (although not covering energy requirements) to the effort necessary to fully remediate the persistent pollution. Therefore, the substance-specific characterization factor has a unit of mass equivalent of activated carbon per kg emitted [$kg_{equ.AC}/kg_{em}$] and represents a midpoint. It does not describe the effects of a specific behaviour of future generations when facing water pollution, but rather indicate the potential effort required to recover the polluted freshwater resources depending on the persistence of the pollution and its difficulty in being remedied. Thus, this approach provides a new perspective allowing more transparent results for the differentiation of safe, long-term water supply issues (AoP natural resources) and potential (short-term) toxicity effects (AoP human health).

133

Towards global regionalized characterisation factors for water consumption impacts on instream freshwater ecosystems

M. Damiani, IRSTEA Montpellier / UMR ITAP, ELSA; P. Roux, Irstea / ITAP ELSA-PACT; E. Loiseau, Irstea; M. Núñez, TU Berlin / Sustainable Engineering; N. Lamouroux, H. Pella, Irstea / MALY; A. Hélias, Montpellier SupAgro / LBE ELSA; R.K. Rosenbaum, National Research Institute of Science and Technology for Environment and Agriculture - Irstea / UMR ITAP

Several life cycle impact assessment (LCIA) models have been proposed to quantify potential water consumption impacts on freshwater ecosystems. In existing mechanistic models, mostly based on species-discharge and species-area relationships, ecological requirements are not taken into account. This implies considering equal response to stress for different taxa, overlooking the relationships between species and their habitat, along with other aspects of biodiversity (e.g. abundance). In this study we want to show the importance of habitat modelling to describe the impact pathways of water consumption on ecosystem quality. We propose a new mechanistic LCIA model based on freshwater physical habitat and discuss the applicability at the regional and global scale. Water consumption may alter stream discharge and other related physical variables. Habitat suitability equations can be used to quantify physical habitat availability for freshwater species in Weighted Usable Area (WUA). Starting from WUA equations, a Habitat Change Potential (HCP) midpoint indicator for river fish species, guilds and invertebrates is proposed and applied in France at Q50 (wet season) and Q90 (dry season) flows. HCP represents the change of available habitat area deriving from river discharge alteration. At the river reach scale, HCPs from different taxa have been aggregated under different perspectives in order to test the results' sensitivity to negative and positive effects of hydrological alteration. A spatial aggregation has been also performed at watershed and sub-watershed. Subsequently, the global HCP model's applicability has been discussed. HCP is highly correlated with river size. The aggregation at reach scale is driven by specific taxa and by positive HCP scores (habitat loss). The result of the aggregation at watershed is consistent with existing evaluations of hydromorphological pressures in Europe. The main challenge in applying HCP globally is due to hydrological and hydraulic data availability. It is however possible to find convergence between European and extra-European species habitat preferences. The proposed model is a promising effect factor for mechanistic impact characterisation which should be integrated with fate factor models describing hydrological alteration at a compatible spatial resolution. Since habitat models are based on species abundance, HCP represents the first step towards developing biodiversity damage indicators complementary to species loss.

134
The use of dynamic stock model to the definition of characterisation factors for biotic resources depletion
A. Hélias, Montpellier SupAgro / LBE ELSA; J. Langlois, Université Paul-Valéry Montpellier 3 / CEFE UMR CNRS Université de Montpellier Université PaulValéry Montpellier EPHE Université PaulValéry Montpellier Montpellier cedex France; P. Fréon, IRD, emritus scientist
Biotic natural resources have received little attention by the LCA community and this tempers the use of LCA for fish based food and feed products. Current LCA methods do not assess the impacts of biotic removal on biotic stocks very well. The dynamic of biotic stocks (i.e. population) is a well-studied topic in theoretical ecology and it is commonly used for fish stocks management. The main of the present work is the use of this knowledge to assess biotic resources depletion in LCA. This is illustrated by the definition of characterisation factors (CF) for fish stocks depletion. The relationship between inventory and impact is often determined by the marginal effect, where the CF is used for quantifying a marginal change in impact according to a marginal change in inventory. Here, the Depleted Stock Fraction (DSF) is considered to be the assessed impact while the elementary flow of the inventory is a mass of fish removed from the biomass stock. The marginal approach is applied to the Schaefer model, a commonly used model in fisheries representing the dynamics of the biotic stocks. It combines catches, current biomass and maximum intrinsic growth rates of the stock (population). To determine these parameters for most of the world fisheries, catch time series were used for species of FAO Global Fishery and Aquaculture Statistics. This allows assessing all fisheries of the world in the LCA framework with a midpoint biotic resource depletion impact. To our knowledge, this work is one of the first assessments of biotic depletion in LCA, based on a model of population dynamics. We are confident this will bring improvements to the LCA of fish-based products, allowing for the comparison of different fishery alternatives with respect to the use of marine biotic resources. One of the extensions of this present work could be the definition of CFs for terrestrial biotic resources where similar dynamic stock models are used. This advocates the use of a similar approach based on population model dynamics for both terrestrial and marine biotic resources.

135
Accounting for soil quality effects of agricultural land management in LCA
B. Lieselot, R.A. Alvarenga, Ghent University / Department of Sustainable Organic Chemistry and Technology; V. Van linden, Flanders research institute for agriculture, fisheries and food / Technology and Food Science Unit; I. Roldán-Ruiz, Flanders research institute for agriculture, fisheries and food; J. Dewulf, Ghent University / Department of Sustainable Organic Chemistry and Technology
The growing demand for food and feed has put a pressure on the agricultural sector, which resulted in agricultural intensification. This entails many adverse environmental impacts and takes a huge toll on land resources; which can result in land degradation. Especially farm management can affect soil quality (e.g. soil organic carbon, SOC) by for instance the choice of crop rotations. The agricultural sector is therefore forced into a more sustainable approach. To assess the environmental sustainability, life cycle analysis (LCA) is a powerful tool. As soil quality is an inherent aspect of agricultural sustainability, life cycle impact assessment (LCIA) methods should account for the effect of agricultural land management on soil quality. Despite a lot of efforts that have already been made, there are still several research needs on the integration of soil quality effects in LCIA methods. Ideally, inventory data are translated into the effect on the environment by indicators moving along the cause-effect chain from mid- to endpoint towards an area of protection (AoP). Although soil quality changes are often related to the AoP ecosystem health, we focus on the AoP natural resources as we refer to the impact of land management on soil by the long-term ability to produce biomass. To improve the usefulness in the agricultural sector, different land use intensities should be distinguished. We therefore introduce three interdependent LCIA indicators. At early midpoint, SOC changes (SOC[2], indicator 1) are used to indicate the long-term effect of agricultural land use on soil. This can result in biomass productivity losses (BPL, indicator 2). At endpoint, we propose additional land requirements (ALR, indicator 3) as indicator, which corresponds to the area needed to produce the yield that has been lost. To calculate characterization factors (CFs), we chose as reference situation the highest achievable SOC stock and yield, which are calculated for each initial soil quality stock. The models RothC and EU-Rotate_N are used to quantify SOC stocks and yields, respectively. CFs are developed for several rotation systems in Flanders. Though, the elaborated framework is generally applicable and allows the calculation of CFs for other regions. Thanks to the use of an achievable reference situation and a distinction between land management strategies (good, bad and standard practices), the indicators can be a useful tool to strive for a more sustainable agriculture.

136
Poster spotlight: MO093, MO094, MO106

Environmental fate of emerging contaminants in the water cycle: analytical challenges and engineered solutions (III)

137
Full scale WWTP balancing with passive samplers offers new insights in xenobiotic elimination processes
T. Galle, Luxembourg Institute of Science and Technology; C. Koehler, M. Bayerle, D. Pittois, Luxembourg Institute of Science and Technology LIST; A. Christen, J. Hansen, University of Luxembourg / Faculty of Science, Technology and Communication
Calculating elimination rates for full scale wastewater treatment plants is very demanding because it requires the knowledge of mixing regimes to match inflow to out flow volumes which translates very often in sampling campaigns that span over several days. While a certain progress in understanding elimination processes of xenobiotics has been gained from laboratory reactor tests, reliable and large datasets from real-life full scale plants are still missing due to the cumbersome sampling effort. Here we implemented the use of passive samplers to establish WWTP balancing with the simple concept of using recalcitrant compounds like carbamazepine to normalize sampling rates from in- and outlets of treatment plants and hence be able to directly calculate elimination rates. The method was validated in a pre-study with parallel autosampling and then applied to 18 WWTPs representing a large range of design properties such as hydraulic and sludge retention times. Normalizing with carbamazepine and lidocaine proved to be robust since both inlet-outlet ratios were well correlated and elimination rates of the investigated compounds fell into the ranges documented in literature. Furthermore it was possible to identify patterns of elimination by applying a cluster analysis and several compound elimination rates were found to negatively correlate with sludge retention time and hence to be more related to active biomass of the sludge. Inlet loads of the compound could be calculated by calibrating the more invariant outlet concentrations to passive sampler masses and then back calculating to inlet loads via the elimination rates. Population equivalent loads proved to be within expected ranges from the literature and non-domestic sources could be identified. Passive sampling might hence close the gap of investigation in xenobiotic behaviour on full-scale treatment plants and serve as well as a routine performance surveillance tool.

138
Screening of wastewater-borne pharmaceuticals and their phototransformation products in rivers
S. Perez, N. Montemurro, IDAEA-CSIC / Environmental Chemistry; D. Barcelo, IIQAB-CSIC / Department of Environmental Chemistry
Pharmaceuticals are continuously discharged into the rivers from wastewater treatment plants. Hundreds of wastewater-borne pharmaceuticals have been detected in river samples but their concentrations along the river change constantly due to additional inputs and natural attenuation processes. Apart from biodegradation, drugs can undergo phototransformation reactions by either direct or indirect photolysis including reaction with singlet oxygen ($^1O_2$), hydroxyl radical (?OH), peroxy radicals (?OOR), photo-excited organic matter, and other reactive species.To evaluate these processes in a river, usually laboratory studies are

performed in a first stage and then in the next step studies are conducted directly in the natural environment. In our group, we proposed a workflow using the combination of HRMS and processing software for evaluating the phototransformation of pharmaceuticals on a single compound basis under simulated and real environmental conditions. In contrast to this compound-by-compound approach, in the new approach presented here, degradation was not assessed for a single compound but instead a cocktail of human drugs was subject to the transformation process. Following the identification of photo-TPs, a list of suspect TPs was created and used to screen them in SPE-concentrated river water samples. For the generation of photo-TPs, reconstructed surface water was spiked with a cocktail of 34 pharmaceuticals at concentrations of 10 µg/L and exposed to artificial light in a sunlight simulator. UPLC-HR-MS (QExactive) was used to identify the photo-TPs. Subsequently surface water samples originating from rivers were screened for their presence. For the enrichment of the potential photo-TPs, water samples were preconcentrated on four SPE cartridges connected in series and then analyzed using the same system mentioned above. With this methodology more than 30 photo-TPs were detected in the irradiated reactor samples. As of the time of submission of this abstract, the identification of some of the photo-TPs was still underway. Several photo-TPs of our database were detected in the extracts of the surface water samples. Thus this approach highlights that UPLC–HRMS is a powerful tool for qualitative analysis, allowing the search for photo-TPs. With the detection of some photo-TPs we have provided evidence for photolysis and thus underpinning the importance of natural attenuation processes in rivers.

139
Degradation of a polymer probe exposed to different wastewater environments: Linking chemical transformations and potential microbial consumers
A. Ginebreda, CSIC - Spanish National Research Council / Environmental Chemistry; M. Vila-Costa, B. Zonja, IDAEA-CSIC / Environmental Chemistry; N. Montemurro, IDAEA CSIC Barcelona / Dipartimento di scienze agro-ambientali e territoriali; S. Pérez, A. Martínez-Varela, IDAEA CSIC / Environmental Chemistry; D. Rivas, IDAEA CSIC / Eviromental Chemistry; D. Barcelo, IIQAB-CSIC / Department of Environmental Chemistry
The objective of the present contribution was to link the chemical transformations observed in a polymer probe exposed to the different aquatic environments found in a WWTP with the microbiological communities present in situ. Phylogenetic composition of free living bacteria was compared to polymer-attached microbial communities. Specifically, chemical degradation and microbial communuity characterization was carried out in the influent flow (IN), in the secondary aeration tank (AER) and in the denitrifying anaerobic bioreactor (ANA). Polycaprolactonediol (PCLD; average MW=1250) was selected as suitable candidate polymer probe. The method was tested in four WWTPs by exposing the polymer probes *in situ* at the secondary and nitrifying/ denitrifying conditions. The progress of biodegradation after several days of exposure was reflected on changes in the MALDI-TOF MSI mass spectra obtained across the sample's surface. Peaks were identified using liquid chromatography coupled to high-resolution mass spectrometry (QExactive, Thermo Scientific), which evidenced the occurrence of three different degradation pathways (Fig.1). Results were further examined using different image processing tools enabling to observe differences between degraded and non-degraded areas. Microbial biomass was collected from water surrounding the polymers (free-living communities) while communities attached to the polymers were collected scratching the surface of the polymer with a sterile spatula. The nucleic acids were extracted by phenolization process. One microlitre of the extracted DNA was used to amplify the bacterial 16S rDNA by PCR.In general, composition of free-living bacteria was significantly different than polymer-attached microbial. Bacteroidia, Spirochaetes and unclassified bacteria co-dominated the free-living communities in AER and ANA, although metabolisms under oxic and anoxic/nitrifying conditions differ quite dramatically. Polymer-attached in the same waters were dominated by deltaproteobacteria, a class of proteobacteria characterized by sulfate-reducing bacteria although also harbors aerobic phylotypes. Free-living microbial communities in IN were dominated by Bacteroidia, belonging to the phylum Bacteoidetes whereas their polymer-attached counter part was dominated by Sphingobacteriia (a class of Bacteroidetes). Results suggest that polymers select specific microbial groups that benefit from consumption of PCLD that can be used as carbon and energy source.

140
Optimization of Laccase Catalyzed Iodine Synthesis as Enzyme Based Disinfectant
g. yüksek, Universite de Sherbrooke / Civil Engineering
In addition to traditional and new generation water pollutants, microbial contamination is still one of the major problem which has affected the potential recycle and reuse of wastewater for domestic or industrial usage. Selection appropriate treatment approach has played an important role not just to reach discharge limits, it is also important to decrease human footprints in the environment. Some hydrolytic and oxidative enzymes (i.e., Laccase) is capable of oxidizing unreactive iodide to reactive iodine, when they can play roles to degrade recalcitrant pollutants in wastewater. The resulting iodine represents a powerful antimicrobial compound. The aim of this study is investigating the potential of acetophenone and phenolic organic contaminant acetaminophen as mediator in a laccase mediator system to generate disinfectant iodine. The stability of reaction can be changed depending on the pH, temperature and multiple compound existence and system optimization is required to stabilize iodine synthesis. In this study, two different free laccases and insolubilized as cross-linked enzyme aggregates have been tested. Also, Iodine synthesis is investigated with different KI (0-100 mM) concentrations and different enzyme activities (5, 10, 30 and 40 Unit /L) for 5 hours. Compounds were injected in distilled water as well as in the influent and effluent samples of wastewater treatment plants to see synthesis of iodine while the micropollutants have been removed in Laccase Mediator System. In the experimental sets, removal of persistent compounds were determined by LC-MS/MS. I_2 generation determined spectrophotometrically and disinfection effect of iodine measured by fecal coliform tests. 0.35 maximum mM/L iodine concentration could be synthesized during experiments. During iodine production, while phenolic compounds' concentrations were decreased (50% acetaminophen removal in real effluent wastewater treatment plant), removal of non-phenolic compounds such as naproxen were also observed (50%). The results have shown that the biocatalytic generation of I_2 was possible using laccase-mediator system. Iodine production was affected by the initial laccase activity and mediator concentration. Laccase catalyzed bactericidal activity in municipal wastewater was also assayed without the addition of any mediator assuming that wastewater already contains mediators such as acetaminophen. Using that system, non-fecal coliforms present in the tested wastewater were removed.

141
Halogenated methanesulfonic acids in drinking water - Identification, standard synthesis, and analysis
D. Zahn, Hochschule Fresenius / Chemistry and Biology; A. Harloff, Hochschule Fresenius, University of Applied Sciences; R. Meusinger, TU Darmstadt / FB Chemie; T. Frömel, Hochschule Fresenius, University of Applied Sciences; T.P. Knepper, Hochschule Fresenius, University of Applied Sciences / Chemistry and Biology
Persistent, bioaccumulative, and toxic (PBT) substances have been monitored since the 1960s. PBT substances are usually not well water soluble and tend to adsorb to sludge, soil and other particular matter, and thus spread slowly in the environment. However, as a consequence of their high environmental stability, they can eventually reach even remote areas like the arctic, and pose a threat to apex predators due to their ability to accumulate in the food chain. Persistent, mobile, and toxic (PMT) environmental contaminants, however, do not accumulate in the food chain, but are much more mobile in aquatic environments, and thus, they spread faster throughout the environment. Drinking water consumption might be a major human exposure pathway for PMT substances. While environmental contaminants of low and intermediate polarity have been thoroughly investigated, so far only little is known about the most polar, and thus potentially most mobile, water contaminants. This gap in knowledge might be caused by difficulties in the analysis of very polar organic chemicals, especially in their enrichment from aqueous matrices. PMT substances might be, among others, pharmaceuticals, personal care products, or industrial chemicals, however, a significant fraction may also be (dead-end) transformation products (TPs), and thus a substantial share of them might still be unknown. In a non-target screening approach dedicated to the identification of mobile, potentially drinking water relevant organic contaminants, we identified chlorinated and brominated methanesulfonic acids (MSAs) as novel water contaminants and estimated the concentrations of the most prevalent congeners to be in the 100 ng/L range for some drinking water samples. Accurate quantification, however, was hindered by the lack of commercially available reference materials. Thus, we synthesised chloromethanesulfonic acid, dichloromethanesulfonic acid, bromomethanesulfonic acid and bromochloromethanesulfonic acid as well as ^{18}O-trifluoromethanesulfonic acid (as internal standard) and included these analytes in a sample pre-treatment and hydrophilic interaction liquid chromatography – tandem mass spectrometry (HILIC-MS/MS) method dedicated to the analysis of very polar water contaminants. With this method, we monitored chlorinated and brominated MSAs throughout four drinking water treatment plants and in several tap water samples taken from high population areas in different countries.

142
Poster spotlight: MO272, MO273, MO274

Building of large-scale inventories of emissions and resources and applications for environmental footprints of territories, nations and sectors

143
Framework for building national inventories of toxic emissions to air, water and soil, in Europe
A.S. Leclerc, DTU / Management Engineering; S. Sala, M. Secchi, A. Cerutti,

European Commission Joint Research Centre / Bioeconomy unit; A. Laurent, DTU / Division for Quantitative Sustainability Assessment DTU Management Engineering
The European INventory of Existing Commercial chemical Substances (EINECS) lists over 100 000 chemical substances used on the market. Over 16 000 chemical substances have been registered in REACH since 2008. In comparison, only ca. 3 000 substances are associated with characterization factors in life cycle impact assessment (LCIA) to express their potential toxic impact on human health (cancer and non-cancer effects) and freshwater ecosystems. Because of human activities, those pollutants may enter the environment in several different ways: they are emitted to air from the combustion of materials, released through wastewater from industries and households, applied to soils together with manure and pesticides, etc. Combined with the limited availability of release data, the sheer number of substances and the large variety of emission sources are challenges that one needs to overcome to quantify the overall toxic impacts of a country. Here, we therefore propose an updated methodology to build national inventories of toxic emissions in EU Member States in 2000-2014. The framework builds on earlier works and differentiates environmental compartments (air, water and soil) as well as anthropogenic sources (industries, households, manure and pesticides application on agricultural soils). It relies on existing and publicly available data, and extrapolation techniques are developed and used to fill in the gaps across countries in the entire period 2000-2014. The resulting harmonized inventories cover more than 500 substances, including both organics and inorganics such as persistent organic pollutants and heavy metals. Albeit still limited in substance coverage, it is thus possible to analyze the contribution of each substance and anthropogenic source to the toxic impacts on human health (human toxicity) and freshwater ecosystems (freshwater ecotoxicity), using LCIA methods such as the consensus model USEtox.

144

Combining economic modelling and LCA to assess regional policies: key learning points from a case study on the French forestry sector

T.B. Beaussier, INRA; E. Loiseau, Irstea; S. Caurla, INRA

Economic modelling is increasingly used in Life Cycle Assessment (LCA) to perform consequential LCA for the environmental assessment of product and services. Economic models can also provide significant enhancements for assessing the effects of regional policies, such as in territorial LCA approaches. Among them, equilibrium models appear as a good compromise to assess both socio-economic and environmental impacts of regional policies in an exhaustive and representative way. However, there are still some bottlenecks when trying to combine both approaches in practice. For instance, the levels of system aggregation and disaggregation can differ between the two types of modelling and methodological developments are required to ensure a consistent combination while limiting the time spent to collect data. This talk aims at providing insights on the combination of a partial equilibrium model, the French Forest Sector Model (FFSM), and LCA to assess the eco-efficiency of two regional policies supporting local wood industries in the French East Region. Two approaches will be used to combine economic modelling outputs with Life Cycle Inventories (LCI), i.e. i) Extended Environmental Input Output modelling and ii) a method based of MFA (Material Flow Analysis) and process-based LCA. Eco-efficiency ratios based on economic and environmental impacts allow identifying scenarios with best environmental performances. In addition, this combination allows considering supply and demand dynamics, and thus the socio-economic effects of a decision. Using two different approaches, we are able to compare strengths and weaknesses of both types of combinations and discuss them considering policy assessment results, system representation and system boundaries. Thus, our work provides both insights on down-to-ground policy analysis and methodological developments on combining economic modelling with LCA. Here, economic modelling outputs are used as LCA inputs but more integrated modelling could be performed for completeness and optimization purposes. Perspectives on a stronger coupling will also be discussed.

145

A regional life cycle approach for assessing the climate change mitigation potential of biobased systems

S. OKeeffe, Helmholtz centre for environmental research - UFZ / BEN; S. Majer, DBFZ, Deutsches Biomasseforschungszentrum gemeinnützige GmbH / Bioenergy Systems; U. Franko, Helmholtz Centre for Environmental Research, UFZ / Department of Soil Physics; D. Thraen, Helmholtz Centre for Environmental Research UFZ/ Deutsches Biomasseforschungszentrum gemeinnützige GmbH, DBFZ / BEN

\n *Keywords: Regional, spatial, biobased economy, GHG* While traditional life cycle assessment is a powerful tool, for spatial applications, it is limited. With the ever increasing drive towards regional biobased circular economies, as a means of ensuring future climate change mitigation, there is a need to produce more regional and spatially representative life cycle assessments of biobased systems and bioeconomy regions. “RELCA”, a REgional Life cycle inventory approach, was developed to assess the regional and spatial variation in the environmental performance of bioenergy production within a focus region. Through the use of catchment delineation, conventional geographical modelling is combined with life cycle software to assess the potential environmental burdens of regional bioenergy configurations (i.e. bioenergy plants and their biomass catchments). RELCA was used to assess the climate mitigation potential of biodiesel, for the region of Central Germany. With this approach we showed, for the focus region, that the mitigation potential changed between the different biodiesel configurations, due to their location within the region. When compared to a fossil diesel comparator (83.8 CO_2eq. /MJ), the climate change mitigation potential of the regional biodiesel ranged between 53%-62%. When the results were compared to the typical RED (Renewable Energy Directive) values, a 13-31% greater mitigation potential than the RED was observed. The latter, illustrating that regional variability cannot be captured with a simple regional average value or default value. Additionally, scenarios were used to test the mitigation potential of reduced nitrogen fertiliser application during the biomass production phase. The results of the scenarios indicated that while reduced nitrogen fertiliser led to emission reduction per hectare, the greater land area input required to meet the demand of the biodiesel plants, led to an increase in emissions ranging from 1.14-5.71 g CO_2eq./MJ biodiesel. Thus, highlighting that assessments of biobased systems should consider configurations of biomass and conversion plants in order to determine appropriate mitigation strategies. Therefore, the ability to account for the heterogeneous geographical characteristics found within a region, using life cycle approaches, is important to support more sustainable regional resource management.

146

LCA_WIND_DK: temporally, geographically and technologically-sensitive life cycle inventories for the Danish wind turbine fleet

R. Besseau, Mines ParisTech / Centre Observation, Impacts, Energie (OIE); R. Sacchi, Aalborg University / Planning Department; P. Pérez-Lopéz, Mines ParisTech, PSL Research University / Centre Observation Impacts Energy O.I.E.; I. Blanc, Mines ParisTech / Centre Observation Impacts Energy O.I.E.

The environmental performance of a wind turbine is usually calculated as the ratio of the life cycle impacts occurring during the manufacture, installation, maintenance and dismantling of the plant, to the electricity it produces during the use phase. The modelling of the life cycle inventory in each phase should ideally cover the temporal, geographical and technological dimensions of the product system under study. Assumptions are commonly used to simplify and handle variable aspects of the inventory. While this approach provides generic one-size-fit-all inventories, it may disregard important characteristics of the wind turbine leading to biased end-results. As these assumptions are prone to differ from one study to another, the results become hardly comparable. With more than 1,500 wind turbine models on the market and a high variability of sites and manufacture periods of the different installations, it makes the environmental assessment of wind turbine fleets a daunting task. LCA_WIND_DK is an on-line tool that provides the environmental footprint of Danish wind turbines based on systematic individual cradle-to-grave life cycle inventories using manufacturer’s data. The temporal context is considered through the evolution of the electricity mix used for manufacturing wind turbines as well as the evolution of recycled content in materials over time. The spatial dimension is also accounted with geographical parameters determining the amount of material required, such as the distance from shore and sea depth for offshore installations. Additionally, the supply chain is adapted to select the relevant origin of the material and energy suppliers. Finally, the approach considers the registered electricity production for past and present wind turbines and assesses the future production from site-specific weather re-analysis data and power curves. Denmark, where wind power contributed to 45% of the gross annual electricity production in 2016, is a prominent choice to demonstrate the benefits of such comprehensive modelling based on spatial, technological and site specific LCAs. The approach generates a life cycle assessment for each of the 11,000 wind turbines that compose the Danish national fleet over the 1980-2030 period. The results, through the on-line tool, are showcased as a map, where the individual performance of each of the past, present and future wind turbines can be consulted, as well as the performance of the whole fleet at a given year.

147

Assessing environmental impacts of individual households: A large-scale bottom-up LCA-model for Switzerland

A. Froemelt, ETH Zurich; R. Buffat, ETH Zurich / Institute of Cartography and Geoinformation; N. Heeren, S. Hellweg, ETH Zurich / Institute of Environmental Engineering

Besides governmental consumption, household consumption is the main driver of economy, and is thus ultimately responsible for the environmental impacts that occur over the whole life cycle of products and services that households consume. Therefore, assessing environmental footprints of households is an important basis to identify environmental policies. This study aimed to develop a comprehensive regionalized bottom-up model for Switzerland that is able to assess the environmental impacts induced by individual households. The purpose of this model is to provide a virtual platform for detailed scenario analysis which shall support effective political decision making on different scales. Three existing bottom-up models were merged: a building stock energy model, an agent-based transport simulation and a household consumption model. All of them were tested and evaluated beforehand. The physically-based building energy model establishes simplified energy balances for each residential building based on spatially and temporally resolved climate data, building characteristics and 3D-geometries. It provides estimates of space heating, hot water and electricity demand for each Swiss household. The mobility sub-model builds upon the results of an agent-based traffic simulation framework which was applied to Switzerland and reproduces mobility patterns of Swiss inhabitants in space and time. The third sub-model pursues a data-driven approach and enables the quantification of consumption of food, consumables, and other goods and services for each Swiss household by means of data mining techniques. Linking these sub-models with environmental background data allowed for computing an environmental profile for each household in Switzerland. The application of this model to the current situation of Switzerland reveals interesting differences between individual households, different regions and different consumption areas. By covering the variability of household behavior and quantifying the demands and environmental footprints of households within a certain area, the model delivers important insights for local policymakers to derive targeted environmental strategies tailored to the specific problems and household types in a region. Furthermore, the high resolution of all three sub-models permits testing of policies and in-depth analyses of scenarios, ranging from detailed building refurbishment programs to future mobility solutions such as autonomous vehicle systems.

148
Poster spotlight: MO109, MO110, MO113

Mechanistic effect modelling for risk assessment: applications, use in a regulatory context and future directions

149
Modelling ecological scenarios for the assessment of chemical effects on stream communities
A. Gergs, Bayer AG - Crop Science Division / Department of Environmental, Social and Spatial Change; S. Classen, K. Ladermann, Research Institute gaiac; T. Strauss, M. Hammers-Wirtz, Research Institute gaiac / gaiac - Research Institute for Ecosystem Analysis and Assessment
The ecological risk assessment of chemicals (ERA) aims at quantifying the likelihood of adverse ecological effects posed on populations and the communities they comprise. Effects caused by the exposure of organisms to chemicals can however to a great extent depend on environmental scenarios as well on the states, behaviours and interactions of organisms with consequences for individual life history, population responses and community dynamics. In this regard, our major objective is to suggest how to model stream ecological scenarios for ERA. We suggest to employ ecological classifications as defined within the Water Framework Directive. Here, the ecological scenario is a virtual representation of an ecosystem, which involves both abiotic components (habitat scenario) and biotic components (the functional and life history scenario). Technically, we integrate spatial explicit habitat information in form of raster maps, temporal information on abiotic factors like temperature and chemical exposure, functional trait data bases, dynamic energy budget models and process based effect models to simulate macroinvertebrate and fish assemblage dynamics. In model applications, we explore to what extent the ecological scenario will affect the adverse outcome of chemical exposure.

150
Robust implementation of TKTD models with Bayesian inference
V. Baudrot, Université Lyon 1; S. Charles, University Lyon 1 / Laboratory of Biometry and Evolutionnary Biology
The application of toxicokinetic-toxicodynamic (TKTD) modeling proved to be of particular interest in strengthening the Environmental Risk Assessment (ERA) of chemicals compounds (e.g., REACH dossier accounting for toxicity of industrial discharge, evaluation of impacts of Plant Protection Products (PPPs), ...). TKTD models describe the time-course of processes leading to toxicity at the level of organisms. These models include all mechanisms from the toxicokinetics part describing the compound fate from external concentration to internal kinetics (e.g., exposure, uptake, elimination, biotransformation, internal distribution), and translate the internal concentration into toxicodynamics covering alteration of cells and organs functioning that can eventually lead to a toxic effect at the organism level (e.g., mortality, reduced reproduction, abnormal behavior) then affecting the population dynamic.\nFor survival analysis of organisms in response to a chemical stressor, the General Unified Threshold model of Survival (GUTS) is today recognized as a suitable and powerful TKTD framework incorporating two complimentary death mechanisms: Stochastic Death (GUTS-SD) and Individual Tolerance (GUTS-IT), from which a large range of existing models can be derived.\nIntergovernmental institutions as the OECD have acknowledge the necessity of TKTD models for ERA improvement, but while an integrative mathematical framework as GUTS offers an efficient theoretical approach, its practical use is challenging (from model implementation to parameter estimation), especially with time-variable exposure. The Bayesian approach has multiple advantages as (i) using all data provided by the experiments, (ii) taking into account the knowledge from experts and/or previous studies, (iii) being relevant for complex model with small dataset since there is no degree of freedom, and (iv) a clear handling of uncertainties by providing distributions of parameter posteriors.\nTo ease the access of Bayesian fitting of GUTS models based on ordinary differential equations, we compared several implementation of GUTS models using two software dedicated to Bayesian statistics in connections with the widespread statistical language R (JAGS and Stan). Then, we embedded those algorithms within two R packages with the core idea to be user friendly (e.g., using experiment design to define priors). A side result is to propose a step-by-step approach to perform Bayesian statistics in ecotoxicology.

151
Can TKTD-models describe and predict synergistic interactions in Chironomus riparius?
K. Dalhoff, University of Copenhagen / Department of Plant and Environmental Sciences; G. Bellisai, European Food Safety Authority EFSA; E. Neira, N. Cedergreen, University of Copenhagen / Department of Plant and Environmental Sciences
The azole fungicides propiconazole and prochloraz are known to enhance the toxicity of pyrethroid insecticides like α-cypermethrin during co-exposure. The development of these synergistic actions in the waterfly *Daphnia magna* have recently been modelled using toxicokinetic (TK) and toxicodynamic (TD) models in order to describe the underlying mechanisms for the enhanced toxicity. The purpose of the current study was to test if the same conceptual TKTD-framework for synergistic interactions in *D. magna* can be applied to the midge larvae *Chironomus riparius* to describe development in survival rates and the underlying mechanisms over time when co-exposed to azole fungicides and pyrethroid insecticides. Toxicity of the individual compounds was tested using a pulsed concentration response design with an initial 24 hour exposure period followed by six days of recovery in clean medium. To assess the combined toxicity of the azoles and α-cypermethrin were a range of tests conducted with co-exposure to 1, 3, 10, 30, or 100 µg L^{-1} of propiconazole or prochloraz and 2.5, 5.0, or 10.0 µg L^{-1} of α-cypermethrin. For the TK-modelling will uptake and elimination rates of the individual compounds in *C. riparius* be measured to parameterize the TK-model before relating the internal concentrations to the observed survival rates We hypothesise that the synergistic interactions can be described and modelled by adding a synergy parameter "*s*" to the biotransformation rate constant for α-cypermethrin and that the value of this "*s*" parameter will depend on the azole exposure concentration. The preliminary results indicated time-dependent synergistic interactions in *C. riparius* as previously observed in *D. magna*, but also a higher sensitivity of *C. riparius* towards the fungicides with 168 h EC_{50}-values for the 24 h pulse exposure of 1.06 ± 0.27 and 0.28 ± 0.10 µmol L^{-1} for propiconazole and prochloraz, respectively. This is surprising as previous non-published data indicated that *C. riparius* has an approximately 10 fold faster initial elimination rate of the azoles compared to *D. magna*. We expect that our TKTD models will be able to explain these kinetic differences and how they relate to the observed toxicity. We furthermore hope that models can predict survival of *C. riparius* over time when exposed to azole and pyrethroid pulses with varying time intervals between the pulses.

152
Integration of temperature-dependent TKTD kinetics in individual-based population modelling - A case study with Chaoborus crystallinus
T. Strauss, Research Institute gaiac / gaiac - Research Institute for Ecosystem Analysis and Assessment
The toxicokinetic-toxicodynamic (TKTD) model framework GUTS is increasingly used and becoming the standard effect model in regulatory risk assessment. However, this model is mostly used without the temperature dependency of TKTD kinetics, and effect measurements are usually performed in the laboratory at 20°C. This approach is rather unrealistic, especially in outdoor scenarios with significantly different water temperatures over the year: On the one hand, in cases with low water temperatures during autumn and winter, the toxic effects can be reduced or delayed, while on the other hand, the degradation of the substance is often slowed down, which increases the exposure time. But also at higher temperatures than 20°C, increased toxic effects on organisms are to be expected. In this presentation, the influence of seasonal temperatures on toxicity at the population level is exemplarily examined. For this purpose, an individual-based population model for the phantom midge *Chaoborus crystallinus* is used. This has been intensively tested using outdoor aquatic mesocosm studies, and has been coupled to the TKTD framework GUTS, which has now been extended by a temperature dependency based on laboratory data. Temperature dependencies of lethal effects on *C. crystallinus* larvae exposed to the pesticide chlorpyrifos and the fate of chlorpyrifos were measured in the laboratory in the range of 4-20 °C. These data are used to parameterize the GUTS, and subsequently to simulate the population dynamics of *Chaoborus* under variable outdoor temperature conditions. The Chaoborus model was used with and without the implemented temperature dependency of the effect model to assess whether toxic effects on a *Chaoborus* population are increased or decreased depending on the temperature conditions. Therefore representative summer and winter exposure scenarios were selected.

Using temperature dependencies for the relevant biological and toxicological processes, this modelling approach allows a more realistic risk assessment of pesticides for populations in the field.

153
Assessing lethal and sublethal effects from time variable exposure for different life-stages with the DEB model: an example for a Pyrethroid in rainbow trout
E. Zimmer, IBACON GmbH; T. Preuss, Bayer Ag / Environmental Safety; S. Norman, RidgewayEco; B. Minten, ADAMA Deutschland GmbH; V. Ducrot, Bayer Ag / Environmental Safety Ecotoxicology
The study investgates effects of beta-cyfluthrin on juvenile rainbow trout (*Oncorhynchus mykiss*) using TK-TD modelling. As part of the risk assessment modelling is used as a supporting tool to back up the experimental results and as an investigation tool to better undertand the mechanisms of effects of beta-cyfluthin. Beta-cyfluthrin is acting as neurotoxicant in fish for which the severity of effect depends on the magnitude and duration of the exposure peak. To address these characteristics, the effects of beta-cyfluthrin on rainbow trout were evaluated with two independent early life stage tests (ELS): a standard Tier 1 study with constant exposure and a Tier 2c study under time variable exposure. Observed effects differed in these two studies. Under constant exposure, severe mortality and significant growth effects were observed while under peak exposure, no effects on survival were observed, and only negligible effects on growth were found. The model was successfully calibrated using the constant exposure experiment, and then accurately predicted the effects observed in the peak-exposure assay. The model reveals that the internal concentration in the fish does not pass the threshold for an effect on survival. This helps to explain why no mortality is observed in the peak exposure experiment. The no effect threshold for sublethal effects is passed in the modelling under constant exposure, which is consistent with the observations. In the peak experiment, the duration of the effect on the feeding behaviour is insufficient to induce large effects on growth in weight or lengths, because beta-cyfluthrin is rapidly removed from the body and the fry have enough reserves to cope with reduced feeding over a short period. The modelling supports the experimental finding that under realistic exposure conditions, short term effects on the feeding behaviour do not lead to growth or survival effects, and gives a mechanistic explanation for this observation. We were able to derive a mechanistic explanation for the results from laboratory experiments conducted with three different early life-stages of the trout, and for different exposure profiles to beta-cyfluthrin. The model shows that results from both laboratory studies are consistent. This validated model has the potential to be used to make accurate *in silico* predictions of effects on fish early life stages from time-variable exposure profiles.

154
Prediction of effects on chemicals on three-spined stickleback populations in mesocosms
V. David, INERIS; B. Goussen, University of York / Environment; J. Porcher, INERIS / INERIS UMRI SEBIO ECOT; R. Beaudouin, INERIS / Models for Ecotoxicology and Toxicology METO
To improve environmental risk assessment, mechanistic models predicting the impacts of toxicants on populations such as individual-based models (IBMs) was suggested as relevant tools. Furthermore, IBMs can be coupled with DEB (Dynamic Energy Budget) models which describe physiological processes of an organism. However, the development of DEB-IBMs requires a large number of data on the organism and the population dynamics which make them difficult to build. To this aim, data from mesocosm experiments can be of great interest for developing and calibrating DEB-IBMs. One of the species that can be used in mesocosm experiments is the three-spined stickleback (*Gasterosteus aculeatus*). Furthermore, the ecology and biology of this teleost fish is relatively well-known and a DEB model for this organism has already been developed. In this study, we used data from several mesocosm experiments to describe stickleback populations under control conditions, and exposed to three concentrations of an endocrine disruptor, the Bisphenol A (BPA, 1, 10 and 100 µg/L). First, using two set of experiments in control conditions, different ways of integrated temperature and food data was tested in order to assess the relevance of the DEB model calibrated with laboratory data for sticklebacks in mesocosms. Then, the DEB-IBM was developed and calibrated and simulated endpoints of the population dynamics in control conditions were compared to the observed enpoints of the population dynamics in control conditions or exposed to BPA. We showed that the DEB model successfully predicted the growth of mature male and female sticklebacks for two set of experiments in control conditions. Furthermore, the calibrated DEB-IBM successfully predicted dynamics of stickleback populations during mesocosm experiments in control conditions. Indeed, the different descriptive variables of the populations (population size, male, female and juvenile frequencies, lengths and coefficient of variations) were well described and were used to compare with the endpoints of mesocosms exposed to BPA. In conclusion, simulated enpoints of stickleback populations can thus be used as a baseline to compare exposed populations to BPA in order to improve environmental risk assessment. In a second step, the DEB-IBM could be adapted in order to introduce the effects of toxicants such as BPA on the individuals and thus extrapolate the effects at the population level.

New Horizons in Particulate Polymer Analysis: Micro- and Nanoplastics and Tire Rubber Detection, Characterisation and Impacts in the Environment

155
Atmospheric Microplastic's: A novel method for the identification of microplastic's in the inhalable size range.
J.M. Levermore, MRC-PHE Centre for Environment and Health / Department of Epidemiology and Biostatistics,; T. Smith, Kings College London / Department of Geography; S. Wright, F. Kelly, Kings College London / MRC-PHE Centre for Environment and Health
Microplastics (microP) are a class of persistent omnipresent contaminants found in aquatic, atmospheric and terrestrial environments. Current investigations focusing on atmospheric microP have identified microP fibres >50µm in size. For atmospheric microP to have the potential to directly impact human health, research must now focus on the presence of microP in the inhalable size range (< 10µm). We present a novel analytical method compatible with the Multi-vial cyclone sampler (MVCS), for assessing whether microP down to an inhalable size range are airborne. An automated Raman Spectral Imaging (RSI) protocol has been developed for chemical analysis of sampled atmospheric particulate matter (PM). This approach removes operator bias while allowing for the chemical identification of all microP >3µm in size in a sample. To validate RSI for the identification of microplastics (RSI analysis was conducted using an in-house program developed by Dr. Frederic Festy (KCL). Pre-identified features unique to polystyrene (1000.9 cm^{-1}, 1030.7 cm^{-1} and 1602.1 cm^{-}1) were fit to the dataset using a Pearson-based cluster analysis to indicate spatial feature intensity (ImageJ). The identification of 4 and 10µm beads was successful and an operator based particle count detected 163 particles per 100 µL; this resulted in an 85% recovery rate. However, 2µm polystyrene beads were not identified as they were found to be below the limit of detection. This automated RSI protocol facilitates the identification of microP >3µm in size amongst environmental PM. An improved identification rate could be achieved by eliminating the dilution step. This RSI protocol will be utilised for the analysis of samples collected from a month-long monitoring campaign at an urban background site in London, UK.

156
Analysis of polystyrene based microplastics in the environment
G.F. Schirinzi, IDAEA-CSIC / IDAEA; M. Farre, IDAEA-CSIC / Environmental Chemistry; m. farré-urgell, IDAEA-CSIC; D. Barcelo, IIQAB-CSIC / Department of Environmental Chemistry
Marine anthropogenic litter is a severe environmental problem. Wastes discarded or deposited in aquatic environments (including rivers, estuaries and coastal waters) usually consist of metal, glass, wood and plastic, being the 80% of these plastic wastes. One of the main issues is the extreme stability of plastic wastes. Under environmental conditions, the erosion of these materials generates smaller fragments some of them in the nano- and microscopic scale, which are known as nanoplastics (NPLs) and microplastics (MPLs), respectively. The quantitative analysis of these plastic particles is particularly difficult because of their physicochemical properties (low solubility, a wide range of molecular weights, etc.) and potential contaminations sources in the laboratory. For these reasons, different approaches should be considered to find a standardised protocol for the determination of MPLs and NPLs in the environment. In this context, this study was focused on the investigation and practical comparison and combination of different analytical tools for the quantitative and qualitative analysis of MPLs/NPLs using: (1) techniques to assess the physicochemical properties such as Thermogravimetric Analysis or *TGA*, Differential Scanning Calorimetry or *DSC*, and Fourier-Transformed Infrared Spectroscopy or *FT-IR*); (2) quantitative and qualitative information by techniques based on direct mass spectrometric as high resolution mass spectrometry with Electrospray Ionization (*ESI*), Atmospheric Pressure Chemical Ionization (*APCI*), Atmospheric Pressure Photoionization (*APPI*), Matrix-assisted Laser Desorption Ionization (*MALDI*), Desorption Electrospray Ionization (*DESI*) and Direct Analysis Real-Time (*DART*). These studies have been carried out using as a representative polymer the polystyrene (PS), which is one of the most frequently used for plastics production. Finally, LC-APPI-HRMS complemented by other techniques such as TGA, DSC and FT-IR allow obtaining qualitative and semi-quantitative information about of the whole spectrum of polymers, which may be present in the environment.

157
Uptake, egestion and accumulation of microplastic in mussel after an experimental exposure
B. Fernández, Instituto Español de Oceanografia / Marine Pollution and Biological Effects Department; M. Albentosa, Instituto Español de Oceanografia / Marine Environment and Environmental Protection Area. Fisiology and Ecotoxicology of Bivalve Molluscs Department
Filter feeding invertebrates such as mussels are especially susceptible target species to microplastic (MP) ingestion. Field and laboratory studies have reported that MP are ingested by mussel. Once ingested, MP may be egested through defecation,

retained in the digestive system, and/or transferred through the haemolymph to other body tissues (translocation). However, the knowledge on the ingestion and egestion of MP and on the accumulation of MP within different organs of mussel is limited. In this context, a laboratory experiment was conducted to investigate the kinetic of uptake and egestion of MP and its accumulation in digestive gland of mussel. To this end, individual mussels, *Mytilus galloprovincialis*, were exposed in a single dose to two nominal concentrations (2 and 4 $mm^3\ l^{-1}$, Low and High MP dose, respectively) of microalgae (MA) (*Isochrysis galbana*, clone t-ISO) and MP (high-density polyethylene, HDPE) of similar size (Results showed no differences between the uptake kinetic of MP and MA, indicating a similar capture efficiency and acceptability for both types of particles by mussel. After 120 hours of the exposure, mussels had egested around a 80% of the MP ingested. The highest volume of MP (around 30% of the MP ingested) was quantified in faeces collected after 24 hours of the exposure. Then, lower volume of MP was recorded in faeces collected after 48 hours (around 20%) and 120 hours (8%) of the exposure. The diameter of the MP particles egested decreased with time. The highest particle diameter (about 9 µm) was observed in the MP egested after 4 hours of the exposure. This may be related to a size-dependent rejection of larger MP particles in pseudofaeces. Results showed that after 120 hours of the exposure the 6 and 2% of the MP ingested was accumulated in the digestve gland of mussels exposed to the Low and High MP dose, respectively. The diameter of this MP (around 3 µm) was significantly lower than that of the MP ofered (8 µm) and the MP egested (6-9 µm). This suggested a specific removal through faeces of larger MP particles and the retention of smaller ones in the digestive system.

158
Analysis of tire wear particles in environmental samples using TED-GC-MS
P. Eisentraut, Bundesanstalt für Materialforschung und -prüfung; E. Dümichen, Bundesanstalt für Materialforschung und -prüfung / 5.3 Mechanics of Polymers; A.S. Ruhl, TU Berlin / Department of Water Quality Control; M. Jekel, TU Berlin; M. Albrecht, M. Gehde, TU Chemnitz; U. Braun, BAM- Federal Institute Materials Research and Testing / 5.3 Mechanics of Polymers
Tire and road wear particles (TRWP) as environmental contaminants have received interest since the 1960s[1]. TRWP have adverse effects on human health[2]. Multiple cities in the EU are violating legal threshold values for atmospheric pollution to which TRWP contribute. Therefore, financial penalties as well as consequences like vehicle bans in metropolitan regions are discussed. TRWP can be regarded as microplastics, because the rubber component of TRWP is mainly polymer (natural and synthetic). With regional differences, the contribution of TRWP to the microplastics emissions to the environment can reach up to 60%[3]. Analysis of TRWP is challenging because of the high variance in compositions of the tires. Published analytical methods suffer from unspecific marker compounds, small sample size or low sensitivity[4-6]. The topic of this presentation is the analysis of TRWP using the recently developed method TED-GC-MS (thermal extraction desorption gas chromatography mass spectrometry)[7]: Sample materials are heated in a thermogravimetric analyzer. The decomposition products are purged with nitrogen through a heated coupling device to a solid phase adsorber bar. After the adsorber is loaded with an excerpt of the decomposition products, an auto-sampling robot transports the adsorber to a thermal desorption-GC-MS for further analysis. Rubber materials were provided by TUC, Sigma Aldrich and Avokal. The tire samples included used and unused materials provided by TUC, Umweltforschungszentrum Leipzig (UFZ) and former BAM projects. A bitumen and an asphalt material, provided by BAM were analyzed. As tire-free matrix materials, BAM reference materials and suspended matter from rivers provided by Umweltbundesamt (UBA) were chosen. Environmental samples with expected TRWP pollution were provided by TUB and consisted of lake sediment from Berlin / Germany and various stages of a filter system receiving street runoff. Various potential marker compounds for tires were identified. They include characteristic decomposition products of elastomers, antioxidants and vulcanization agents. Advantages and draw-backs of these marker substances will be evaluated. Emphasis is given to the presence/absence of these in tire-free environmental matrices and in bitumen and asphalt samples. In the next step, we analyzed environmental samples and detected signals of decomposition products from tire materials. Method parameters and options for quantitative analysis will be discussed.

159
Determination of tire wear particles based on elemental composition
P.E. Kloeckner, Helmholtz Centre for Environmental Research GmbH - UFZ / Analytical Chemistry; S. Wagner, Helmholtz Centre for Environmental Research GmbH - UFZ / Department of Environmental Geoscience; A.S. Ruhl, TU Berlin / Department of Water Quality Control; P. Eisentraut, Bundesanstalt für Materialforschung und -prüfung; U. Braun, BAM- Federal Institute Materials Research and Testing / 5.3 Mechanics of Polymers; M. Albrecht, TU Chemnitz; T. Reemtsma, Helmholtz Centre for Environmental Research / Department Analytical Chemistry
In this contribution we present the analytical method development based on Zn and S content and apply the analytical method to determine tire wear particle concentrations along the treatment path of road runoff. Tire wear particles have been recognized as an important environmental pollutant. Analytical methods for the quantification of tire wear particles in environmental samples are still under development and struggle with multiple sources or insufficient stability of markers. We developed an analytical method which allows quantifying tire wear particles in road runoff, sediments and surface waters. Tire wear particle quantification is based on elemental composition and distinct elemental ratios. The analytical method aims at i) tire wear particle enrichment using density separation followed by ii) microwave assisted acid digestion iii) and elemental detection of zinc, sulphur and carbon. A stepwise method development including an analytical method verification by determination of the rubber content is presented. In particular, S and Zn are present in characteristic concentrations in tires. Zn and S contents were determined in 30 tire samples as an internal reference. The average S content in the analysed tires was 15400 mg/kg (± 6000 SD), while the average Zn concentration was 8500 mg/kg (± 1700 SD). The average S:Zn ratio in the tires was 1.9 (± 0.9 SD). Furthermore, the developed method was applied to field samples. Samples were taken from the intake water of a treatment facility (raw water), from the sedimentation basin, the inlet of the soil retention filter as well as from the soil itself. Isolation of tire wear particles by density separation was achieved by use of a heavy liquid, sodium polytungstate mixed with MilliQ water. Separated fractions were acid digested with microwave assistance and elemental analysis was conducted by ICP-MS and ICP-OES. Elemental content of the particulate fraction in the water samples was analysed after filtration only, since the amount of solids was too low for the density separation procedure. *Acknowledgement* - The authors thank the BMBF for funding the MiWa project (reference number 02WRS1378H) and BWB for provision of samples.

160
Are we speaking the same language? Towards a definition and categorization framework for environmental plastic debris
M. Wagner, Norwegian University of Science and Technology / Department of Biology; N.B. Hartmann, Technical University of Denmark (DTU) / DTU Environment; A. Verschoor, RIVM / Centre for Safety of Substances and Products; T. Hüffer, University of Vienna / Department of Environmental Geosciences; M. Hassellöv, University of Gothenburg / Department of Marine Sciences; R.C. Thompson, Plymouth University / School of Marine Science and Engineering
The occurrence and accumulation of plastic debris is a global environmental issue, with potential consequences affecting the economy, wildlife and human health. However, there is currently a lack of consensus on the definition and categorisation of environmental plastic debris, including macro-, micro- and nanoplastics. The lack of clarity in terminology regarding plastic debris, in particular microplastics, results in confusion and misunderstandings. This is problematic both for legislative measures as well as for general coherence and data comparability between studies. While finding a common language appears beneficial, any definition should be well-justified as it will ultimately shape the direction of future research and legislation. To help decide whether a consensus definition and categorization framework for plastic debris is valuable and if so how this might look, the scientific community needs to engage in a critical discussion. The aim of our presentation is to foster such discourse in the SETAC community by providing impulses and sharing our thoughts rather than providing definitive answers. In our presentation, we will use a new format with two presenters jointly discussing the advantages and disadvantages of a definition. Further we will discuss our ideas on relevant components of a definition/categorization framework. To get immediate feedback by the community, we will use online polling asking specific questions to the audience throughout the presentation. This will cover opinions on the need of a definition, acceptance of certain defining and categorizing criteria and questions on special cases with high uncertainty. The aim of this is to get an ad hoc idea on where consensus may be easy to achieve and areas which are controversial. Finally, we will present an online platform (www.microplastics.eu, currently under development) that we will use to perform a large-scale survey on a consensus definition of environmental plastic debris. In addition, the platform will host a module for discussing the questions mentioned above and a module for networking. This platform can be used by the audience and the wider community to further discuss the impulses we give and share their opinions and input.

Advances in environmental risk assessment of oil spills and offshore oil & gas operations (III)

161
Behavioral and physiological responses of bicolor damselfish and mahi-mahi to olfactory cues following crude oil exposure
L. Schlenker, RSMAS, University of Miami / Marine Biology and Ecology; M.J. Welch, James Cook University; E.M. Mager, RSMAS University of Miami; J.D. Stieglitz, RSMAS University of Miami / Marine Biology and Ecology; D.D. Benetti, RSMAS University of Miami / Department of Marine Ecosystems and Society; P.L. Munday, James Cook University; M. Grosell, RSMAS University of Miami / Marine Biology and Ecology
In fishes, olfactory cues provide information about predators, prey, and conspecifics that is crucial to survival; however, olfactory sensory neurons are directly exposed to the environment and are susceptible to damage from aquatic contaminants. The 2010 Gulf of Mexico oil spill overlapped with the habitat of

pelagic and reef fishes, including mahi-mahi (*Coryphaena hippurus*) and bicolor damselfish (*Stegastes partitus*). To date, within the marine teleost group, nothing is known about how crude oil exposure affects the detection of olfactory cues or if crude oil can be detected and avoided. To address these questions, the time that control and oil-exposed bicolor damselfish spent in a chemical alarm cue and the time that control and oil-exposed mahi-mahi spent in diluted crude oil was examined using a two-channel flume choice system. Control bicolor damselfish avoided a conspecific chemical alarm cue, while oil exposed conspecifics did not avoid the cue ($p < 0.001$). Control mahi-mahi did not distinguish between seawater and crude oil, however oil exposed mahi-mahi spent a greater proportion of time in crude oil than the control fish ($p < 0.01$). Moving forward, an electro-olfactogram technique will be used to measure the generator potential from the olfactory epithelium of bicolor damselfish and mahi-mahi to detect the response to olfactory cues following oil exposure. This research was made possible by a grant from The Gulf of Mexico Research Initiative. Grant No: SA-1520; Name: Relationship of Effects of Cardiac Outcomes in fish for Validation of Ecological Risk (RECOVER).

162

A wide range of endpoints are impacted by oil exposure in early and later life stages of marine fish

M. Grosell, J.D. Stieglitz, RSMAS University of Miami / Marine Biology and Ecology; R.M. Heuer, University of Miami / Marine Biology and Ecology; C. Pasparakis, Rosenstiel School of Marine Sciences / Marine Biology and Fisheries; L. Schlenker, RSMAS, University of Miami / Marine Biology and Ecology; J.T. Magnuson, University of North Texas / Biology; D. Nelson, University of North Texas; E. Xu, University of California Riverside; D. Schlenk, University of California Riverside / Department of Enviromental Sciences; D.A. Crossley, University of North Texas / Department of Biological Sciences; A.P. Roberts, University of North Texas / Department of Biology Institute of Applied Science; D.D. Benetti, RSMAS University of Miami / Department of Marine Ecosystems and Society

Oil toxicity to fish has traditionally been attributed mainly to impacts on cardiac function and development in early life stage fish. Such impacts include pericardial edema and reduced stroke volume and cardiac output. However, additional sensitive endpoints include accelerated metabolic rate, accelerated yolk sac depletion, and alterations of buoyancy in developing embryos. Furthermore, brief exposures during embryonic development lead to reduced swim performance, reduced maximal oxygen uptake, and reduced visual acuity in later stage juveniles. Juvenile fish exposed to oil show altered olfactory responses, reduced prey capture ability, and higher susceptibility to predation, likely due to altered central nervous system function. Even adult marine fish are sensitive to brief, low-level oil exposures, showing reduced aerobics scope and swim performance. The lower swim performance in adult oil-exposed fish with a normally developed heart, is due to reduced cardiac output driven by reduced stroke volume. Work on isolated cardio-myocytes show that sarcomere shortening upon electrical stimulation is reduced by acute oil exposure which likely explains the reduced stroke volume observed in intact, oil exposed fish. Such reductions in cardiomyocyte contractility are likely related to impaired cellular calcium cycling also suggested by RNAseq data. This research was made possible by a grant from The Gulf of Mexico Research Initiative. Grant No: SA-1520; Name: Relationship of Effects of Cardiac Outcomes in fish for Validation of Ecological Risk (RECOVER).

163

Investigating the endocrine disruptive and genotoxic potential of crude oil samples using adapted in vitro toxicity tests

S. Johann, Institute for Environmental Research RWTH Aachen University; L. Nuesser, RWTH Aachen University, Institute for Environmental Research / Department of Ecosystem Analysis; L. Spencer, Institute for Environmental Research RWTH Aachen University / Department of Ecosystem Analysis; D. Altin, BioTrix; H. Hollert, RWTH Aachen University / Institute for Environmental Research; T. Seiler, RWTH Aachen University / Ecosystem Analysis

In case of an acute oil spill the decision on optimal response techniques to achieve the best environmental benefit is based on important knowledge about regional ecology, the key oil compounds as well as the ecotoxicological risk of the spilled oil. It is known that for petroleum hydrocarbons not only narcosis is the mode of action for toxic effects but also dioxin-like or estrogenic activity can act as the toxicity mechanisms. Hence, it is important to include these mechanism-specific endpoints in the toxicity profiling of crude oils. This study focuses on the endocrine disruptive and genotoxic potential of a naphthenic North Atlantic crude oil. Furthermore, a third generation dispersant used as a chemical response action to combat oil spills at sea is integrated to evaluate the dispersant's toxicity and the influence of the dispersant on the toxicity of the spilled oil. To investigate the toxicity of this complex and variable sample type, low energy water accommodated fractions (LEWAF) of crude oil as well as chemically enhanced water accommodated fractions (CEWAF) of crude oil and dispersants were prepared according to Singer et al [3]. The protocols of standardized methods investigating endocrine disruption and genotoxicity were modified in respect to the sample type containing hydrophobic and volatile compounds. In order to avoid cytotoxic false negative results due to cytotoxicity masking sublethal effects, the cell viability was investigated. It was found that the LEWAF sample was less cytotoxic than the CEWAF sample. For an initial screening of the endocrine disruptive potential of crude oil WAFs, the ERα-CALUX® was performed. First results indicate that sample compounds of the LEWAF and CEWAF do interact with the estrogen receptor (ER) in the highest test concentrations. As part of genotoxicity investigations the micronucleus assay with ZF-L cells was performed. It was observed that both LEWAF and CEWAF exposure resulted in higher micronucleus formation compared to the unexposed control. The CEWAF exposure led to the highest micronuclei induction occurring already at much lower exposure concentrations than used for LEWAF exposure. In general, it can be concluded that the mechanism-specific mode of actions estrogenic acticity and genotoxicty can contribute to crude oil toxicity. Further investigations will include additional endpoints to generate a more comprehensive toxicity profile of the selected crude oil.

164

Impacts of Oil Exposure on Mahi Embryos

C. Pasparakis, Rosenstiel School of Marine Sciences / Marine Biology and Fisheries; L.E. Sweet, Environmental Protection Agency USA; E.M. Mager, University of North Texas / Department of Biological Sciences; J.D. Stieglitz, RSMAS University of Miami / Marine Biology and Ecology; D.D. Benetti, RSMAS University of Miami / Department of Marine Ecosystems and Society; A.P. Roberts, University of North Texas / Department of Biology Institute of Applied Science; M. Grosell, RSMAS University of Miami / Marine Biology and Ecology

The *Deepwater Horizon* spill coincided with the spawning window of many ecologically and economically important fish species, such as mahi-mahi (*Coryphaena hippurus*). Aside from the acute mortality elicited by this event, additional sublethal effects may have imparted more subtle yet ecologically significant consequences on populations of pelagic fishes as a whole. The maintenance of embryo buoyancy is critical to survival; and aids in promoting dispersal by facilitating drift through ocean currents and positioning newly hatched larvae in the upper water columns where planktonic food is plentiful. We found that co-exposure to oil and additional environmentally relevant stressors, such as high temperature and UV-radiation, affect the timing and duration of negative buoyancy in mahi-mahi embryos. Further, premature negative buoyancy was coupled with significantly faster sinking rates and increased energy depletion, likely resulting in detrimental consequences for these developing fish. The mechanisms behind untimely buoyancy change are unknown, but our findings suggest a behavioral avoidance response as well as an inability to maintain buoyancy due to diminished energy reserves. This research was made possible by a grant from The Gulf of Mexico Research Initiative. Grant No: SA-1520; Name: Relationship of Effects of Cardiac Outcomes in fish for Validation of Ecological Risk (RECOVER).

165

Crude oil impairs heart cell function in the pelagic mahi-mahi (Coryphaena hippurus)

R.M. Heuer, University of Miami / Marine Biology and Ecology; H.A. Shiels, G.L. Galli, University of Manchester / Faculty of Biology, Medicine and Health Sciences; G.K. Cox, J.D. Stieglitz, RSMAS University of Miami / Marine Biology and Ecology; D.D. Benetti, RSMAS University of Miami / Department of Marine Ecosystems and Society; M. Grosell, RSMAS University of Miami / Marine Biology and Ecology; D.A. Crossley, University of North Texas / Department of Biological Sciences

Crude oil from the *Deepwater Horizon* spill of 2010 has been shown to have a number of cardiotoxic effects across life stages, species, and levels of organization in marine fish. Over the last decade, the use of the mahi-mahi (*Coryphaena hippurus*) to study these cardiovascular impairments has been particularly important, since this pelagic species is both ecologically and economically important in the Gulf of Mexico. Mahi exposed to environmentally-relevant crude oil concentrations have shown compromised intact animal performance, including reductions to maximal swimming speed and maximal metabolic rate. In addition, *in-situ* studies have revealed a ~40% reduction in cardiac output following oil exposure in mahi. Although cardiotoxic effects have been widely reported, the mechanisms underlying these impairments remain understudied. In the present study, we examined the impacts of crude oil on isolated mahi heart cells to better understand these mechanisms. Contractility of mahi ventricular heart cells was measured via sarcomere shortening using an IonOptix cell recording system. The first objective was to examine cardiomyocyte contractility over range of crude oil exposures. The second objective was to examine the impacts of crude oil contractility over a range of stimulation frequencies representative of heart rates observed in mahi (~100-180 beats per minute). Exposure to crude oil was found to significantly reduce heart cell contractile function, but was not found to be dose-dependent in the tested range of concentrations (3.0, 6.4, and 12.9 ug l^{-1} ∑50 PAH). Crude oil was also found to impair contractility over a range of stimulation frequencies (1.5, 2.0, 2.5, 2.5, 3.0 Hz; 3.6 ug l^{-1} ∑50 PAH). In addition to contractility, other mechanical aspects of cell contractile function were also examined. Efforts to assess the role of circulating catecholamines (adrenaline) as a potential protective mechanism against these impairments is currently ongoing and will also be presented. This research was made possible by a grant from The Gulf of

Mexico Research Initiative. Grant No: SA-1520.

166

microRNA and messenger RNA networks in early life stages of pelagic and nearshore fish species exposed to Deepwater Horizon oil

D. Schlenk, University of California-Riverside / Department of Environmental Sciences; G. Xu, UC Riverside / Department of Environmental Sciences

Developmental cardiotoxicity is a common phenotype observed in a number of fish species following exposure Polyaromatic hydrocarbon (PAH) or oil. While many PAHs elicit cardiotoxicity through activation of the aryl hydrocarbon receptor (AhR). Additional pathways of toxicity have been observed including downregulation of genes that regulate potassium and calcium channels in embryonic and larval stages of development. While functional inhibition the channels has been observed following exposure to oil and non-AhR activitng PAHs, mechanisms associated with downregulation has not been observed. MicroRNAs (miRNA) play key roles in a number of diverse biological processes including heart development in vertebrates. To test the hypothesis whether miRNA changes may regulate ion channel genes, embryos and larvae of mahi-mahi (*Coryphaena hippurus*) were treated with High Energy Water Accommodated Fractions (HEWAF) made from source and weathered *DHW* oil. miRNAs and mRNA were sequenced from the same pooled animals and expression compared using advanced bioinformatics with subsequent target organ predictions based on their interactions. Gene ontology (GO) analysis on the target mRNAs was consistent with pathway analysis of miRNAs, predicting disruption of cardiovascular system development after oil exposure and showed that specific miRNA–mRNA interactions may contribute to these effects (Figure 1). Oil caused an overexpression of miR-133a, miR-34, and miR-15b (Figure 2). Enhanced expression of miR-133a correlated to the decrease in the expression of KCNH2 mRNA, which controls the potassium ion transporter that has been observed to be reduced in the cardiac phenotype in multiple fish species following oil treatment. In addition miR-34 and 15b, were also upregulated and informatic analyses with mRNAs were consistent impairment of eye development (Figure 2). Ongoing analyses of dose response treatments at early hatch larval stages (48 hpf) will provide additional data that will enhance our knowledge of the impacts of oil on fish development. This research was made possible by a grant from The Gulf of Mexico Research Initiative. Grant No: SA-1520; Name: Relationship of Effects of Cardiac Outcomes in fish for Validation of Ecological Risk (RECOVER).

Alternative Approaches to Animal Testing for Ecotoxicity Assessments

167

Early life stages of a vertebrate species as an alternative model for the study of stressors in marine environment

M.J. Araujo, CESAM & DeBio / APPLEE; R.J. Rocha, Department of Biology & CESAM - University of Aveiro / Department of Biology and CESAM; C. Quintaneiro, Department of Biology & CESAM - University of Aveiro; A.M. Soares, University of Aveiro / department of Biology & CESAM; M. Monteiro, Aveiro University / Biology

Early life stages of marine vertebrates have been scarcely used in ecotoxicity testing. The Senegalese sole (*Solea senegalensis* Kaup, 1858) is a common vertebrate occurring in Eastern Atlantic coastal areas. Eggs of this marine vertebrate can be obtained from aquaculture rearing facilities and used in laboratory as testing organisms. At the end of the first month of life this species completes a metamorphosis, changing from bilateral to flatten shape morphology. Early life stages of aquatic vertebrates are windows of development considered highly sensitive to anthropogenic contamination, including in marine environment. Organic compounds, such as pesticides and personal care products have been increasingly used and directly released to the aquatic ecosystems or indirectly released from wastewater treatment plants. Besides, human activities have been indirectly changing abiotic conditions, such as ultraviolet (UV) radiation. Therefore, in this work we aim to understand the effects of different stressors to early life stages of *S. senegalensis*, namely of UV radiation and of the organic compounds 4MBC, Carbendazim, Linuron and Triclosan, which have potential endocrine disruptor activity. Mortality, malformations, development, growth, behaviour and biochemical markers were evaluated as endpoints in two periods of exposure to stressors: a first initial period between egg stage and 96 hpf and a second period during the nearly 15-day full metamorphosis progression of *S. senegalensis*. Exposure to UV radiation and to the four organic compounds (compounds 4MBC, Carbendazim, Linuron and Triclosan) was performed. Our results suggest that the use of *S. senegalensis* for ecotoxicity testing requires the evaluation of effects at different development stages. Initial egg stages globally display a higher sensitivity to stressors, presenting lower LC_{50} and EC_{50} values. Besides, biochemical markers (cholinesterases and oxidative stress) were differently affected, depending on *S. senegalensis* life stage. Significant alterations of normal behavioural pattern were observed in response to stressors exposure, confirming behaviour as a sensitive and relevant tool in ecotoxicology studies. The increasing environmental levels of the contaminants tested may lead to adverse effects on highly sensitive life stages of marine vertebrate species.

168

Predicting in vivo toxicity from in vitro transcriptional responses following chemical exposure

D. Basili, University of Liverpool / Institute of Integrative Biology; M. Knöbel, Eawag / UTOX; A. Sawle, Cancer Research UK Cambridge Institute / Department of System Biology; J. Herbert, P. Antczak, University of Liverpool / Institute of Integrative Biology; K. Schirmer, Eawag / Environmental Toxicology; A. Cossins, F. Falciani, University of Liverpool / Institute of Integrative Biology

Alternatives to in vivo animal testing in ecotoxicology aim to increase the throughput of chemical safety assessment whilst reducing the number of animals used. The use of in vitro systems is more cost-effective, practical and expedient. However, it is still unclear whether such alternative methods provide the level of information gained from the use of a whole-living system. In vitro to in vivo extrapolation relies on measuring the effects of chemicals on cultured cells or biological molecules to predict how exposure to those compounds might cause adverse effect in animals or people. In this study, we investigated whether the transcriptional state of a trout gill cell line (Oncorhynchus mykiss, RTgill-w1) exposed to a given chemical can be used as a biosensor to predict toxicity in a zebrafish embryo (Danio rerio). More specifically, we developed a regression model linking gene signatures that are independent of compound lipophilicity and predictive of toxicity. We show the ability of residual analysis to identify excess toxicity and to accurately predict in vivo toxicity for most of the chemical MoA in the panel. Our results support the view that gill cell line has the potential to replace zebrafish embryo in toxicity testing

169

Combining computational modelling with in-vitro cellular responses in order to predict chemical impact on fish growth

K. Schirmer, Eawag / Environmental Toxicology; H. Mottaz, R. Schoenenberger, Eawag Swiss Federal Institute of Aquatic Science and Technology / Environmental Toxicology; J. Stadnicka-Michalak, EPFL Swiss Federal Institute of Technology / Environmental Toxicology

A paradigm shift in chemical risk assessment is needed due to the time-consumption and ethical controversies of current chemical testing on animals. Within this project, we propose that the chemical effects on cell population growth, measured over few days, can be used as proxy for chemical impact on fish growth, which needs weeks to occur. In particular, we linked information on the proliferation and survival of a fish gill cell line (in vitro) to the effect of chemicals on fish growth (in vivo). Research was divided into two phases. In the first phase, we have tested in vitro several chemical concentrations that correspond to those used in in vivo experiments. In the second phase we have upgraded our approach so that no prior knowledge about chemical concentrations tested in vivo was required: in vivo data were needed only to validate the model but not to decide which chemical concentrations should be tested in vitro. More than ten organic chemicals (including fungicides, herbicides, insecticides, industrial compounds and pharmaceuticals) were tested for different fish species (rainbow trout, fathead minnow and zebrafish). The results indicate a very good agreement between measurements and predictions determined for different species of fish, being exposed in vivo from 7 to 62 days, depending on the species and test design. Results moreover confirm that it is possible to predict chemical impact on fish growth without prior knowledge on concentrations that are used in in vivo studies for chemicals that do cause an effect on fish weight as well as for those that do not. Therefore, in spite of several assumptions and simplifications, combining in vitro experiments with computational modelling can result in a powerful strategy for screening chemicals to determine their effects on fish. In addition, considering the simplicity, rapidity and low costs of this approach, we believe that it can be an encouraging step toward alternatives to long-term whole organism toxicity testing.

170

Ecological Threshold for Toxicological Concern (eco-TTC) - Applications for Environmental Risk Assessment in Various Contexts

M.R. Embry, ILSI Health and Environmental Sciences Institute (HESI); M.G. Barron, U.S. EPA / Gulf Ecology Division; A. Beasley, The Dow Chemical Company / TERC; S.E. Belanger, The Procter & Gamble Company / Environmental Stewardship and Sustainability Organization; M.A. Bonnell, Environment and Climate Change Canada / Ecological Assessment Division; D.T. Chang, United States Environmental Protection Agency / National Exposure Research Laboratory; K.A. Connors, The Procter & Gamble Company / Environmental Stewardship and Sustainability Organization; D. De Zwart, DdZ Ecotox / Centre for Sustainability Environment and Health; B. Farr, ILSI Health and Environmental Sciences Institute (HESI) / Health and Environmental Sciences Institute HESI; S.A. Hughes, Shell Health - Americas / Shell Health Risk Science Team; A. Kienzler, European Commission - Joint Research Centre / DG Joint Research Centre IHCP EURL ECVAM; T.J. Norberg-King, U.S. EPA / NHEERL/Mid-Continent Ecology Division; R.R. Otter, Middle Tennessee State University / Biology; H. Sanderson, Aarhus University; P. Wilson, SANOFI

The Threshold for Toxicological Concern (TTC) is well-established for assessing human safety but has only recently been explored in the ecological context. Ecological Thresholds for Toxicological Concern TTC (eco-TTC) summarize the

wealth of ecotoxicological information as Predicted No-Observed Effect Concentrations (PNECs) on diverse chemical substances in the form of probability distributions. These enable the prediction of untested chemicals based on a structural attribute, mode of action, or functional use. The approach may be useful for assessing chemicals at early tiers of the risk assessment process, providing hazard perspective on chemicals that lack QSARs, guiding product development discussions, and assisting read across or category justifications.An ecotoxicological database of approximately 120,000 records was developed based on recent assessments of published data and international chemical management programs. This ecotoxicity data is associated with physical chemistry data and curated taxonomic information for the organisms tested, including a process to conclude acute and chronic effects as well as identify the PNEC for exposed ecosystems based on depth and breadth of data. Several mode of action schemes are also included to facilitate development of a best approach for grouping compounds.To make these data accessible and useful to stakeholders, the dataset was transitioned from Microsoft Excel and Access into a modern MySQL format, allowing for a for format that is relational and scalable, facilitating easy access, sharing, and integration with other datasets and tools. The dataset is accessed via a web-based query system that is integrated with PNEC calculator and probability distribution tools. The novel interface allows users to explore the data, upload additional datasets, derive threshold values based on specific criteria, and explore the potential use and application of the ecoTTC concept. An international workshop was held to discuss and evaluate the feasibility of the eco-TTC approach, which included evaluation of several case-studies based on particular decision-contexts (e.g., prioritization and screening, chemical risk assessment, site specific risk assessment, mixtures, product development, criteria development). This presentation will highlight the discussions and conclusions from a recent multi-stakeholder workshop, including exploration of how this approach could be applied and integrated into evaluation strategies.

171
Mode of action diagnosis by normalized multiple endpoint assessment in zebrafish embryos
E. Teixido, Helmholtz Center for Environmental Research - UFZ GmbH / Department Bioanalytical Ecotoxicology; N. Klüver, Helmholtz centre for environmental research - UFZ / Department of Cell Toxicology; O. Kerkhof, Helmholtz centre for environmental research - UFZ / Department Bioanalytical Ecotoxicology; M. Leonard, lOREAL SA; T. Kießling, Scientific Software Solutions; R. Altenburger, UFC Centre for Environmental Research / Department Bioanalytical Ecotoxicology; S. Scholz, Helmholtz Centre for Environmental Research / Department Bioanalytical Ecotoxicology
The diagnosis of a specific or reactive mode of action (MoA) is crucial for the prediction of adverse effects using alternative test systems. While baseline toxicity can be easily predicted using regression-based QSARs, the identification of non-baseline toxicants requires assessment of endpoints that can be related to the MoA of a chemical and ultimately assign an adverse outcome pathway. Such a MoA analysis could be possible using the zebrafish embryo test (ZFET) extended by various endpoints that capture different MoAs. In the ZFET, as alternative system, the detection of MoA-related endpoints has been discussed to improve its predictive capacity for acute and chronic fish toxicity, and for human developmental toxicity. We hypothesized that using a battery of endpoints in the zebrafish embryo test would allow to differentiate between baseline toxicity, formation of methemoglobin, neurotoxicity, heart rate inhibition, and developmental toxicity. Therefore, we compared the toxic ratios and endpoint-specific effect concentrations (EC50) of 12 compounds representing 5 broad MoA groups with 2, respectively 4 (neurotoxicity) compounds per MoA. In order to compensate for differences in the toxicokinetics and mortality, the effect concentrations were normalized by the LC50 of each compound. It was shown that the toxic ratio and effect concentrations for behavior, heart rate inhibition and chorda malformations were able to differentiate the selected compounds according to their anticipated MoA. Using a threshold for the normalized effect concentration a decision tree was developed that allowed to assign a MoA to a compound. A major bias of the selected approach could be the variability associated with visual phenotype assessment – which may depend on the experience and accuracy of the observer. Therefore, we developed a software named FishInspector that enables a more unbiased assessment of malformation using images of zebrafish embryo. Using the software and a system for automated positioning of zebrafish embryos we compared the phenotypes of 25 compounds with known developmental toxicity outcome in rats and/or rabbits. In contrast to the previous analysis, we applied a different normalization approach based on the most sensitive endpoints. The analysis indicated that the developmental toxicity observed for inhibitors of cyclooxygenase may not be related to the pharmacological MoA, given the diverse phenotype patterns observed for this class of compounds.

172
Poster spotlight: MO158, MO159, MO190

Migratory bird species at risk - the role of pesticides and other chemicals

173
CMS talk setting the scene for the CMS working group on poisoning and outlining CMS needs in terms of scientific input from SETAC
B. Heredia, UNEP/CMS / Avian Unit

174
Main scientific gaps in knowledge of risk from pesticides to [migratory] wildlife globally, and potential contribution of WTIG to CMS questions - POSTER SPOTLIGHT MO456
C.A. Bishop, Environment and Climate Change Canada / Wildlife Research Division

175
Main scientific gaps in knowledge of risk from rodenticides to [migratory] wildlife globally, and potential contribution of WTIG to CMS questions - POSTER SPOTLIGHT MO457
P. Berny, VETAGRO-SUP / Toxicology

176
Main scientific gaps in knowledge of risk from Pb ammunition and shot to [migratory] wildlife globally, and potential contribution of WTIG to CMS questions - POSTER SPOTLIGHT MO458
R. Cromie, Wildfowl & Wetlands Trust

177
Main scientific gaps on knowledge of NSAIDs [migratory] wildlife globally, and potential contribution of WTIG to CMS questions - POSTER SPOTLIGHT MO459
M. Taggart, University of the Highlands and Islands / Environmental Research Institute

178
Main scientific gaps on knowledge of deliberate poisoning to [migratory] wildlife globally - POSTER SPOTLIGHT MO460
M. Odino, Independent Environmental Services Professional

179
Questions and discussion

180
Regulatory view describing the extent to which [if any] regulation takes into account neighbouring country/regional use of compounds, accounts for how local use might affect migratory species, how field data on migratory species might feed into regulatory
R. Sharp, EFSA - European Food Safety Authority / Pesticides Unit

181
Panel discussion with audience and presenters focusing on how SETAC can interact with CMS usefully to provide scientific evidence and expertise

Challenges in setting, meeting and measuring specific protection goals for plant protection products

182
Towards a more holistic environmental risk assessment approach of crop protection products as tools in agriculture
P. Dohmen, BASF SE / Environmental and Consumer Safety, Ecotoxicology
Currently, very complex risk assessment approaches are conducted for crop protection products. This is largely justified as pesticides are intentionally introduced into ecosystems and since these substances are intended to have effects on “crop pests” and side effects on non-target species cannot be fully avoided. However, doing so, we often ignore the context, the reasons why crop protection products are utilized. This ignorance can lead to failing the overall targets or lead to sub-optimal or even wrong decisions. This will be explained in more detail in the following presentation advocating for a more holistic approach in environmental risk assessment. Agriculture per se does have an impact on the environment and

does reduce biodiversity at the sites used for food production. This, in principle, is true for any kind of agriculture. As a consequence, it is irrational to demand that agriculture, respectively the different agricultural practices, shall have '*no impact'* on the environment. Accordingly, regulations require that '*no unacceptable'* impact may occur. To define what constitutes an acceptable impact and what not, the 'Ecosystem Services' concept is considered a suitable approach. This is not a call for ignoring the impact of crop protection products. However, for achieving our targets of sustainable efficient and sufficient local food production with minimal environmental impact we need to have a broader approach and evaluate the cost/benefits of all agricultural tools equally against the background of food produced per area. Scientific tools to go for such more complex holistic approaches are partly available, but more efforts are needed to develop practical and manageable concepts allowing an overall assessment on a landscape level. In addition, the regulatory options have to be broadened from mere risk assessment of chemicals towards a landscape level assessment of food production, and risk management has to include landscape management options. Ecotoxicological research is undoubtedly important; however, in order to achieve the overall target of feeding human populations in a sustainable way and maintaining ecosystem health, we need to cover all aspects of 'stress ecology' (impacts of humans on ecosystems) beyond the impact of chemicals. Therefore this call for a more holistic environmental risk assessment approach.

183
Identifying ecosystem services-based protection goals.
L. Maltby, Y. Pan, The University of Sheffield / Dpt of Animal   Plant Sciences
There is an increasing interest in the use of ecosystem service-based approaches for assessing the risk of environmental contaminants to ecological systems. Ecosystem functions become ecosystem services when they are utilized and valued by people. Therefore, the first step in implementing an ecosystem services approach to ecological risk assessment it to identify what portfolio of services are required, by whom and where they should be protected. But whose preferences should contribute to identifying the services to be protected and how should preferences be assessed? These questions are particularly pertinent when considering the role of the general public in protection goal prioritization. Here we use information from case studies in the UK and China to investigate the importance of: who you ask (do different sections of society have different preferences?); what they know (how does prior knowledge influence preferences?); how preferences are assessed (e.g. stated v revealed preferences). We demonstrate that all three factors can have an impact on which freshwater ecosystem services are preferred by the general public and therefore should be prioritize for protection.

184
ECPA over-arching Specific Protection Goals proposal for EFSA Non-Target Terrestrial Plants, Non-Target Arthropods and Soil Organisms Guidance Documents based on EFSA Ecosystem Services approach
K. Romijn, Bayer CropScience AG
Currently there are 3 Scientific Opinions from EFSA that are waiting to be developed into Guidance Documents ie Effects of pesticides on Non Target Terrestrial Plants, Non Target Arthropods and Soil Organisms. Whilst each of these Scientific Opinions makes proposals for SPGs, the European Commission and Member States should agree on the on SPGs before they can be taken forward to be used in the Guidance Document development phase. The purpose of this paper is to provide industry input for consideration and discussion during this process. In an earlier EFSA Scientific Opinion, it was recommended by EFSA that Specific Protection Goals (SPGs) should be based on the principle of Ecosystem Services utilising 6 dimensions: ie biological entity, attribute, magnitude, temporal and geographical scale of the effect, and the degree of certainty that the specified level of effect will not be exceeded. Whilst this EFSA Scientific Opinion is a good basis for setting SPGs going forward, the experience with the EFSA Bee Guidance Document shows there is a need to reconsider how the principles described in this EFSA SPG Opinion are applied to SPG setting in individual Guidance Documents. In the case of the EFSA Bee Guidance SPG it was not the definition of "negligible effects" on colony strength that was the controversial issue but the translation of this into a numerical value (< 7%) without robust scientific justification. The use of 7% suggested there was data to support it but in fact it was still a judgement, i.e. it is a hidden "judgement". The suggestion that there is an arithmetic relationship between large (>35%), medium (15-35%) small (7-15%) and negligible (3.5-7.5%) is in itself surprising for a biological system particularly one with complex feedback loops such as in honeybee colonies. To avoid the difficulty of selecting a specific hard numerical SPG value upon which consensus between different scientist and stakeholders is very hard to reach, it is recommended to use an expert judgement qualitative approach adapting the EFSA Ecosystem Services approach. The predicted impact of any effect of a PPP on an invertebrate/plant population should be described using expert judgement, which combines the predictions of all 4 EFSA dimensions above (attribute/nature of effect, magnitude, temporal and spatial scales of effects) as well as the number and importance (eg keystone species) of species potentially affected, and the frequency of occurence.

185
Protection goals for non-target terrestrial plants: Is in-field protection of beneficial weeds achievable?
J. Davies, Syngenta / Environmental Safety; L. May, A. Russell, A. Seville, D. Stock, Syngenta
EFSA's Scientific Opinion addressing the state of the science on the risk assessment of plant protection products for non-target terrestrial plants (NTTPs) was published in 2014. The Opinion defines non-target plants as all plants growing outside fields and those growing within fields that are not the intended pesticide target. It also states that non-crop plants growing in in-field areas provide ecosystem services including food web support, aesthetic value, genetic resources and endangered species, which require protection from the adverse effects of plant protection products. As such, The Opinion advocates the protection of plant species growing in-field that under current agricultural practice would be considered target weeds growing in the crop. Since 2014, options for protecting ecosystem services provided by non-crop plants growing in-field, have been discussed in various stakeholder workshops and publications. Proposed options for protecting in-field ecosystem services include (a) compensation mechanisms whereby designated areas are set aside specifically for NTTPs, and (b) beneficial weed protection, which involves the control of those weed species that compromise crop yield while not affecting other species that are considered to have ecosystem service value. This second option relies upon the availability of highly selective herbicides and/or other methods of selective weed control. In order to inform this debate, the feasibility of beneficial weed protection will be considered from three perspectives: (a) issues relating to the definition of beneficial weed species (b) potential agronomic consequences of protecting beneficial weeds and (c) challenges facing the discovery and registration of selective herbicides. The issues outlined in this presentation will illustrate that the feasibility of protecting weeds in-field requires investigation of complex species definitions and agronomic consequences while the discovery and availability of new herbicides with the required selectivity is considered unlikely, particularly under the current regulatory framework in the EU.

186
Specific Protection Goals and the Assessment of Key Drivers in the Aquatic Environment: Are we doing the right thing?
R. Benstead, Fera Science Ltd. / Centre for Chemical Safety and Stewardship; D. Phillips, Fera Science Ltd / Environmental Sciences; P. Gilbertson, Fera Science Ltd; J. Chinn, Centre for Crop Health and Protection (CHAP)
When designing High Tier Assessments, the underlying concept is a progression from simple and conservative laboratory exposures, towards those that more closely resemble the 'Final Reference' (the actual ecosystem), so that the risk assessment can be refined by reducing the Assessment Factor that accounts for uncertainty. In the aquatic environment, the focus of High Tier Assessments should be the Specific Protection Goals as defined for surface waters; Biodiversity and Ecosystem Services. Biodiversity is a common and important General Protection Goal, and Ecosystem Services, as they pertain to human health and welfare, are comprised of 'provisioning services' (e.g. drinking water), 'regulating services' (e.g. water purifying microbial communities), 'supporting services' (e.g. organisms contributing to nutrient cycling) and 'cultural services' (aspects of the water landscape that conserve species and promote wellbeing). To this aim, EFSA Guidance requires that a surrogate system for the Final Reference (termed the Surrogate Reference Tier) scientifically underpins the assumption that the endpoints assessed at High Tier are closer to the actual objectives of the adopted protection goal. It is accepted that Low Tier assessments do not measure the SPG directly, and therefore at High Tier, endpoint assessment should rely on ecotoxicological and ecological data that are more directly relatable to the 'target image' of the aquatic community. This surrogate system should be relevant for the surface water at risk, and endpoints should account for the ecological traits of the important aquatic species at risk. The EFSA PPR Panel's solution was to identify for each Key Driver (taxonomic group or other ecological entity), a Surrogate Reference Tier that is based on the most sophisticated experimental or modelling risk assessment currently available to address the SPG. Consequently, scientists should ensure that Aquatic High Tier experimental approaches are (a) the most sophisticated approach available and (b) represent the 'target image' of the biodiverse aquatic communities that support the relevant ecosystem services. Perhaps in this context, the most important assessment endpoints are those that reflect functional and trait based effects, and those that preserve the biodiversity of those communities that ultimately present the 'target image' and therefore closer relevance to the Final Reference.

187
Is "biodiversity" a measurable study endpoint?
F.M. Bakker, Eurofins-Mitox
The general protection goal addressed by current and future risk assessment schemes for Plant Protection Products is Biodiversity. In addition to this there are specific protection goals that may conflict with the general protection goal. Specific protection goals are generally phrased in terms of Ecosystem Services and expressed in human currency there is a risk that economic motives may prevail in their prioritization. After all, who is to judge e.g. whether preservation of pest control capacity outweighs the preservation of aesthetic value? Biodiversity is a concept with many facets and the quantification of biodiversity is no straightforward exercise. A vast array of indices exists (see e.g. Magurran 2004)

and not a single one would capture the essence on its own. The two basic parameters involved in biodiversity measures are the number of species and their abundance. Their correct estimation is core to biodiversity assessments and their balance describes community structure in terms of e.g. dominance, evenness and diversity. Similarity indices have been developed that enable the comparison of (sub)habitats, which seems a promising lead to assessing effects on biodiversity in experimental designs. With this contribution we explore different approaches to quantifying effects on community structure and functioning caused by plant protection products in an experimental setting. We apply and compare multivariate statistical approaches, similarity indices and a combination of univariate statistics and species richness assessments and discuss how these findings may address the general issue of effects on biodiversity.

Innovative techniques for enhancing and monitoring microbial activities for in situ remediation of contaminated sites

188

Evaluation of plant-driven biostimulation of soil microbiota for the setup of a site-tailored rhizoremediation process in a historical PCB-polluted soil

L. Vergani, University of Milan / DeFENS; F. Mapelli, University of Milan-DeFENS / Department of Food, Environmental and Nutritional Sciences; E. Terzaghi, University of Insubria (Como) / Department of Science and High Technology, Como; G. Raspa, Sapienza University of Rome / Department of Chemical Engineering Materials and Environment; O. Uhlik, University of Chemistry and Technology, Prague; E. Zanardini, C. Morosini, University of Insubria / DSAT; A. Di Guardo, University of Insubria / Department of Science and High Technology; S. Borin, University of Milan / DeFENS

The Site of National Priority (SIN) Brescia-Caffaro is a highly polluted area in Northern Italy presenting mixed and uneven soil contamination by metals and organic pollutants, in particular polychlorinated biphenyls (PCBs). In order to evaluate the biostimulation performance of different plant species and soil treatments for the development of a suitable rhizoremediation strategy, an experimental trial including ten vegetated treatments and their non-planted controls was set up for 18 months in greenhouse conditions. Molecular fingerprinting was applied to unveil the ability of different plants/soil treatments to shape the structure of soil microbial communities. The results showed a succession over time in both bacterial and fungal assemblages. Only the diversity of the bacterial community was, nevertheless, significantly and differentially influenced according to the applied treatment. The stimulation effect on the organic matter hydrolytic activity of the soil microbiota was evaluated using fluorescein diacetate as a proxy. All the vegetated treatments showed a significant increase in activity after 18 months from planting, demonstrating effective biostimulation of the soil bacterial communities, putatively enhancing their degradation capacity and, consequently, sustaining rhizoremediation. Aiming to select bacterial strains to be exploited for autochthonous bioaugmentation coupled to rhizoremediation, we established a collection of isolates from the soil biostimulated by *Phalaris arundinacea.* This species cultivated in conditions of redox cycle showed to stimulate the highest increase in soil hydrolytic activity after 3 months from planting. Moreover, when the 18-month biostimulated soil was incubated with ^{13}C-labelled 4-chlorobiphenyl, the production of $^{13}CO_2$ indicated metabolic activity of biphenyl and possibly the presence of PCB-degrading populations. All the isolates were identified as *Actinobacteria* and were characterized for PCB-degradation and plant growth promotion. In particular, three *Rhodococcus* sp. strains significantly promoted lateral root development in the model plant *Arabidopsis thaliana* and depleted PCBs from the cultivation medium according to the results of a resting-cell assay, thus representing ideal candidates to sustain PCB-rhizoremediation through a site-tailored bioaugmentation approach.

189

Enhancement of Biological Reductive Dechlorination by in situ Adsorption onto Colloidal Activated Carbon: from the Lab to the Full Scale Application

M.P. Papini, F. Arjmand, Università La Sapienza / Chemistry; P. Ciampi, C. Esposito, Università La Sapienza / Department of Earth Sciences; M. Carboni, P. Goria, J. Birstingl, Regenesis Ltd; S. Rossetti, B. Matturro, Water Research Institute Italian National Research Council IRSACNR; M. Bacchi, P. Foglietto, Rete Ferroviaria Italiana Spa - RFI

The University of Rome “La Sapienza” has been commissioned to evaluate strategies for the management of the contaminated areas of the new High Speed Railway Station of Bologna (Italy), where a historical Chlorinated Aliphatic Hydrocarbons (CAHs) contamination has been found in two aquifers and characterized by a long-term monitoring activity (PCE, TCE and *cis*-DCE - concentrations ranged between 10-100 µg/L). The Italian environmental legislation is among the most restrictive in Europe with some of the most stringent target levels especially concerning the CAHs. A thorough investigation of the site has been carried out (geological, chemical and biological) and integrated with a microcosm study. Based on the results, biological reductive dechlorination was recognized as a potential approach for the site remediation but the extremely low CAHs concentration and the consequent kinetic limitation made it unfeasible for the site. Thus, the possibility to use a new dispersed colloidal activated carbon technology (Plumestop™, Regenesis) together with an electron donor to create an in-situ adsorption/biologically-reactive zone was deeply investigated and finally adopted as the site-specific remediation approach. The full-scale remediation plan was approved by the local authorities and completed by the end of 2016. This was the first example of a completed full-scale application of this approach in Europe and the monitoring results after more than one year appear particularly encouraging. A very good reduction rates within only a few weeks from the application was observed in all the treated zones. Together with classical chemical analyses, microbiological tools, such as qPCR and CARD-FISH, were used to verify the enhancement of the biological reductive activities induced by the simultaneous injection of activated carbon and electron donor.

190

An innovative bioelectrochemical reactor for in-situ treatment of groundwater contaminated by monoaromatic petroleum hydrocarbons

E. PALMA, CNR-IRSA; M. Daghio, A. Franzetti, University of Milano - Bicocca / Department of Earth and Environmental Sciences; M.P. Papini, Università La Sapienza / Chemistry; F. Aulenta, National Research Council / Water Research Institute (IRSA)

A major share of world energy production, derives from fossil fuels, such as oil. According to OPEC (Organization of the Petroleum Exporting Countries), world oil demand growth is expected to rise by 1.53 mb/d in 2017. The large-scale use and countless applications of petroleum compounds, frequently lead to environmental contamination, as a consequence of petroleum transport, storage and refining, or accidents. Groundwater contamination by petroleum hydrocarbon is a serious problem, with nearly 50% of groundwater contamination being due to petroleum-deriving products such as mineral oil, chlorinated hydrocarbons, monoaromatics (e.g., BTEX) and polycyclic aromatic hydrocarbons (i.e., PAH). Accidental petroleum spills may result in severe environmental problems, hence requiring the development and implementation of suitable remediation strategies. In recent years, microbial electrochemical technologies (MET) have attracted considerable attention as an effective and sustainable remediation technique. In MET the microorganisms catalyze oxidation or reduction reactions by using solid-state electrodes as terminal electron acceptors or donors. The discovery that carbon-based electrodes can be used as terminal electron acceptors during the anaerobic oxidation of a variety of organic substrates has raised the possibility that they could be employed *in-situ* to accelerate the anaerobic oxidation of environmental contaminants, such as petroleum hydrocarbons in soils and groundwater. Here we describe a novel bioelectrochemical reactor configuration, named the “bioelectrochemical well”, that is suitable for *in-situ* treatment of petroleum-contaminated groundwater. A lab-scale prototype of the “bioelectrochemical well [1]” has been realized and operated in a continuous-flow regime using first toluene and then a mixture of BTEX as model contaminants. The performance of the bioelectrochemical reactor was characterized in terms of degradation rate and yield. GC-MS analysis was also carried out in order to shed light on the “electrogenic” pathway of contaminants biodegradation. This study was financially supported by Fondazione Cariplo in the framework of the project BEvERAGE - BioElEctrochemical RemediAtion of Groundwater plumes (2015-0195). [1] Palma E., Daghio M., Franzetti A., Petrangeli Papini M., Aulenta F. The bioelectric well: a novel approach for in situ treatment of hydrocarbon-contaminated groundwater. Microb Biotechnol., 2017. doi: 10.1111/1751-7915.12760.

191

Identification of major HMW-PAH degrading communities during active bioremediation of a PAH-contaminated soil

J. Vila, Instituto de Recursos Naturales y Agrobiologia; M. Grifoll, Universitat de Barcelona / Dept. Genètica, Microbiologia i Estadística; M. Aitken, University of North Carolina / Environmental Sciences and Engineering; J. Ortega-Calvo, Instituto de Recursos Naturales y Agrobiologia / Agroquimica y Conservacion del Suelo

Biodegradation of polycyclic aromatic hydrocarbons (PAHs) in soils is generally constrained by their low availability to microbial communities. As a result, a residue with unpredictable concentration of PAHs remains after bioremediation, with a major composition in high molecular weight (HMW) compounds (four or more rings). We analyzed the active microbial processes associated with the dissipation of PAHs during a lab-scale bioremediation of a creosote-contaminated soil. Treatment under stimulated conditions resulted in an extensive reduction (93%) of the total PAH concentration. Low molecular weight (LMW) compounds (2- and 3-rings) were mainly depleted during the first month of incubation. In contrast, degradation of HMW-PAHs started thereafter following biphasic kinetics. Despite the 16S rRNA gene copy numbers (bacterial abundance) remained constant throughout the 5-month treatment period, the number of 16S rRNA gene transcripts (bacterial activity) dramatically increased (from 10^6 to 10^{10} copies/g dry soil) during the initial 45 days, associated with major PAH removal. 16S rRNA gene pyrosequencing revealed distinctive profiles for total and active communities that evolved with time. Gene expression analysis of ring hydroxylating dioxygenases, together with changes in pyrosequencing libraries, identified members of *Pseudomonas* as the main LMW-PAH degraders. In contrast, dioxygenases of

Gram-positive bacteria, associated to *Mycobacterium*, were mainly active during the last two months of incubation, when only residual fractions of HMW compounds were degraded. Community analysis during the period of major HMW-PAH removal identified members of the recently described order *Immundisolibacterales* and members of *Sphingobium* as the main active populations. Their role on HMW-PAH removal was confirmed by DNA-SIP. Members of *Spingobium* were the major phylotypes associated to fluoranthene assimilation, while members of *Immundisolibacteriales*clearly predominated in incubations with ^{13}C-pyrene and ^{13}C-benzo[*a*]anthracene. Interestingly, members of *Mycobacterium,* traditionally associated to HMW-PAH degradation were not detected in either of the incubations. Our results indicate the minor contribution of mycobacteria to the degradation of the more labile fraction of HMW-PAHs. Their increase in activity during the late incubation phase, when degradation kinetics were drastically attenuated, suggested their potential action on the residual fraction of contaminant.

192

Stable Isotope Raman Microspectroscopy and Surface-Enhanced Raman Scattering: Analysis of Microorganisms at Single Cell Level

N.P. Ivleva, Technical University of Munich / Chemistry Department, Chair of Analytical and Water Chemistry; R. Weiss, M. Seidel, Technical University of Munich; R. Niessner, Technische Universität München / Chemistry Department, Chair of Analytical and Water Chemistry; M. Elsner, Technical University of Munich / Chemistry Department, Chair of Analytical and Water Chemistry

Microorganisms play a vital role in most ecosystems. They are essential for global biogeochemical cycles and for biodegradation of soil and water pollutants. Therefore, it is crucial to develop reliable and sensitive methods for the detection, discrimination and identification of microorganisms as well as for analysis of their activity. Raman microspectroscopy (RM) in combination with a stable isotope approach (SIRM) is an emerging tool for the nondestructive characterization of the molecular and isotopic composition (due to a red-shift in Raman spectra for heavier stable isotopes) of microorganisms at the single cell level.[1-3] SIRM allows for *in situ* investigations of ecophysiology and metabolic functions of microbial communities. Furthermore, the sensitivity of RM and SIRM analysis can be significantly improved (in the range of $10^3 – 10^6$, max. up to 10^{11}) due to surface-enhanced Raman scattering (SERS), e.g. by using of Ag nanoparticles synthesized *in situ*. In contrast to RM and SIRM (where whole-organism fingerprints for bacteria are obtained), SERS is more selective and provodes information on cell surface substances. We applied SIRM and SERS for analysis of unlabeled, ^{13}C- and ^{15}N-labeled single bacterial cells.[4-6] Single cell SIRM analysis was carried out for the *Deltaproteobacterium spez.* strain N47, a strictly anaerobic sulfate-reducer, degrading the recalcitrant environmental pollutant naphthalene. For the ^{13}C-labeled N47 cells peak pattern from isotopologues of phenylalanine with 0, 2, 4 and 6 ^{13}C atoms was found even though this strain is a strict anaerobe growing on ^{13}C-naphthalene. Additionally, our results suggest an incorporation of hydrogen carbonate from the medium into biomass during growth of strain N47 on naphthalene. Furthermore, SERS analysis of *E. coli* revealed that the SERS signal intensity and the number of hits depend on different factors (storage time, presence of D_2O) and can reflect the metabolic activity of cells. Our findings can open new possibilities for the application of SERS (in combination with a stable isotope approach) to probe for the activity of polutant degradating microorganisms at the single cell level. [1] Berry B, et al. 2015. Proc Natl Acad Sci USA 112: E194-E203. [2] Wang Y, et al. 2016. Curr Opin Biotechnol 41: 34-42. [3] Ivleva NP, et al. 2017. Anal Bioanal Chem 409: 4353-4375. [4] Kubryk P, et al. 2015. Anal Chem 87: 6622-6630. [5] Kubryk P et al. 2016. Analyst 141: 2874-2878. [6] Weiss R, et al. in prep.

193

Flux chambers data for the estimation of the biodegradation rate in the subsurface at hydrocarbon contaminated sites

I. Verginelli, University of Rome Tor Vergata / Department of Civil Engineering and Computer Science Engineering; R. Pecoraro, Versalis; R. Baciocchi, University of Rome Tor Vergata

The occurrence of aerobic biodegradation in the subsurface by ubiquitous soil microbes has been shown to reduce and in some in cases eliminate the impacts of petroleum hydrocarbon vapours from contaminated soil or groundwater on indoor and outdoor air quality. The occurrence of natural attenuation in the subsurface is generally evaluated by employing multi-level soil gas sampling through the installation of nesty probes, by applying the so-called “gradient method”. In this way, it is possible to assess the vertical concentration profiles of vapors and oxygen above the source of contamination and hence to evaluate the attenuation factors in terms of soil gas fluxes through the subsurface In this work, we propose a novel approach based on the combination of the data obtained from standard source characterization with dynamic flux chambers measurements. The natural attenuation rates are calculated as difference between the flux of contaminants estimated with a non-reactive diffusive model starting from the concentrations of the contaminants detected in the source (soil and/or groundwater) and the effective emission rate of the contaminants measured using dynamic flux chambers installed at ground level. The reliability of this approach was tested in a versalis site characterized by the presence of BTEX in soil and groundwater, using dynamic flux chambers. The site is characterised by the presence in the subsurface (mainly in groundwater) of BTEX and light petroleum hydrocarbons. The flux of volatile organic compounds (VOCs) from the subsurface was estimated using 14 “dynamic” chambers, by measuring with a canister the concentration of vapours collected over a period of approximately 6 hours. Before starting the measurement, the achievement of steady-state conditions inside the chamber was assured by purging at least 3 to 4 chamber volumes of an inert gas. The measurements in the 14 sampling points were repeated in 4 seasonal campaigns. The obtained results highlighted that the traditional methods based on the application of a non-reactive diffusive model with the concentrations measured in the soil and/or groundwater can lead to an overestimation of the emission rates of BTEX from the subsurface in some cases up to 4 orders of magnitude. Furthermore, the BTEX loss rates for the investigated site were found to be up to almost 0.5 kg/year/m^2. These rates are in line with the values reported in the recent literature for natural source zone depletion.

New frontiers in Life Cycle Inventory data collection and modelling

194

The end of an era: is data and model exchange across LCA software tools finally possible?

M. Vieira, PRe Sustainability; K. Cenian, PRe Consultants; A. de Schryver, European Commission; A. Genest, ifu Hamburg; L. Zampori, European Commission / Joint Research Centre; C. Wolf, Tier3 Solutions GmbH; M. Dupriez, RDC Environment; S. Horlacher, thinkstep; E. Mieras, PRe Sustainability

In the context of the Environmental Footprint (EF) pilot phase, the European Commission requested the development of 70 representative products/organisations (RPs/ROs) so they are consistent with the requirements of the final product environmental footprint category rules (PEFCRs) and organization environmental footprint sector rules (OEFSRs), they use the prescribed EF-compliant secondary datasets, and can be made available for free to any user applying a PEFCR/OEFSR and are easily usable in the major existing LCA software tools. An implicit prerequisite is that software tools give equal results when calculating the impact of the same RPs/ROs. At the start, a document was established that defined the minimum requirements as well as the vetoes for modeling. This way, potential problems for implementability of the models in the different software tools were identified and their use was banned. Five software tools were included, GaBi, openLCA, RangeLCA, SimaPro, and Umberto. The next step was to test the EF impact assessment (IA) results of all EF-compliant secondary datasets. This way, differences coming from their implementation or of the EF IA method could also be identified before testing the results of the models. Furthermore, the development of an extension to the International Reference Life Cycle Data System (ILCD) format (eILCD) enabled importing and exporting models between software tools. Finally, the models developed for RPs and ROs will be tested in the five software tools. The problems faced in importing, calculating and comparing the results of the developed models in the different software tools together with suggestions on how to address the issues will be presented. Some claim different results in different software tools used, often generally pointing to different “software”, without being more specific. The reason may be rather bound to methodology, age, version, flow list and import-export-interface aspects, or even a combination. This work is the basis to enable the reduction of software-system related issues and makes it easier to detect and prevent mistakes. Most importantly, now there is the commitment of the 5 tool developers to make available to all users in the course of 2018 a compliant import and export interface for the eILCD format. We believe this is a crucial step towards the exchange of data and models across software tools and the comparability of results.

195

LCA using real time information: the case of DEA-enabled monitoring of WWTP lifecycle environmental performances

A. Marvuglia, Luxembourg Institute of Science and Technology (LIST) / Environmental Research and Innovation (ERIN); D. Torregrossa, Luxembourg Institute of Science and Technology LIST; E. Benetto, Luxembourg Institute of Science and Technology (LIST) / Environmental Research and Innovation

Life cycle assessment (LCA) is undergoing the effects of a data abundance era, which poses old (data storage) and new (data mining, computational speed) challenges. The deep integration of Internet of Things (IoT) in product- and service-oriented manufacturing systems has enabled a Big Data support for lifecycle modelling along the entire value chain, and the emergence of open-access LCA repositories now allows harnessing the power of crowd-sourced information. However, how to use this huge amount of data in a consistent way to obtain more precise, spatially and temporally differentiated life cycle inventories (LCIs) and life cycle impact assessment (LCIA) results is still not an easy task. In the case of wastewater treatment, the larger and larger availability of on-line measurements coming from sensors installed in many wastewater treatment plants (WWTPs) should theoretically enable daily benchmarking, ultimately allowing faster correction actions, when needed. However, WWTPs data still finds very limited use and is often simply stored. The aim of this work is to showcase an application of temporally differentiated LCA performed with real-time high frequency data and present a methodology for the on-line assessment of the shift in the performance of WWTPs. The implemented methodology performs a DEA-based benchmark coupled with LCA to evaluate the environmental impacts linked to the operation of a WWTP. A web application of the system is currently being developed using the Shiny R package. It will enable plant managers to calculate and visualize LCIA results in real time by modifying customizable parameters.

196

Enhancing Land Use Change modelling with IO data

J. Schmidt, Aalborg University / Department of Planning; M. De Rosa, BONSAI / Agroecology

Land Use Changes (LUC) are responsible for around 11% of global GHG emissions, nearly the same as the transport sector. This is about half of the GHG emissions from coal-based electricity production worldwide. Nevertheless, LUC are often excluded from LCA studies because ascribing the LUC to their drivers distinguishing between production sectors is challenging and requires a complex global inventory data modelling. In order to address this, 2.-0 LCA consultants has been developing a model for indirect LUC (iLUC) modelling in LCA since 2011 as part of a crowdfunded project. Recently, the model has been integrating into the multiregional hybrid Input-Output model EXIOBASE, thus providing an unprecedented level of detail in iLUC modelling. Differentiation between use of land among regions of the world is based upon information on potential land productivity in different locations. The IO data allow identifying the land supplied by each country linking the production trends with the land use trends. The agricultural and land use module in EXIOBASE make use of FAOSTATdata, which provide time series on area and production per crop. The data allow modelling the global supply of land to the global market for land, distinguishing between land expansion (land transformation) and land intensifications (increased production per unit of land). The land transformation and intensification LCA activities are populated with data on carbon stocks of different land use types in all countries, and time series of fertiliser use in all countries. The current version of the model (version 4.3) includes the following elementary flows: emissions of CO_2, N_2O, NO_x, NO_3^-, NH_3 and resource inputs of accelerated denaturalisation caused by transformation of land. The iLUC model can be combined with any life cycle impact assessment (LCIA) model. Overall, the results show that for agricultural crops, iLUC increases the GHG emissions with 100-200%, for beef cattle 20-60%, for pigs 40-80%, for dairy products 40-60%, for wood products 50-300%, and for primary plastics 2-15%. The model is location agnostic and can be applied to any decision-making context concerning long-term effects of small-scale changes. The iLUC framework is now integrated in the global EXIOBASE model, thus accounting for all crops in all countries in the world. It has already been applied to more than 50 LCA studies and on several product categories.

197

WSmix: a globally regionalised Water Supply mix framework with current and prospective databases for use in LCA

S.O. Leão, IRSTEA Montpellier / UMR-ITAP ELSA; P. Roux, Irstea / ITAP ELSA-PACT; M. Núñez, TU Berlin / Sustainable Engineering; E. Loiseau, Irstea; G. Junqua, Ecole des Mines dAlès / LGEI; A. Sferratore, Société du Canal de Provence; Y. Penru, SUEZ groupe / CIRSEE; R.K. Rosenbaum, National Research Institute of Science and Technology for Environment and Agriculture - Irstea / UMR ITAP

Freshwater comes from different sources that are unevenly distributed in the world and different water users (e.g. domestic, agriculture, industry) need different water qualities provided by local (surface, groundwater, rain), external (inter-basin transfers) and alternative sources (e.g. sea water). Water from these sources are withdrawn and processed via water technologies using the local electricity mix. The combination of water sources and technologies results in a regional water supply mix (WSmix) for each specific use. Current LCI databases do not include these mixes when modelling processes leading to a poor representation of water supply systems and related environmental impacts in LCA. Furthermore, changes in water sources, caused by changes of climate and socio-economic factors, will occur in the future. These changes should be considering in LCA of products or infrastructure with long lifespans. This work aims to develop a WSmix framework for modelling current and prospective WSmix (WSmix and P-WSmix) and an inventory database for direct use in LCA. To demonstrate the relevance of including WSmix and P-WSmix in LCI databases, case studies have been conducted. To develop the WSmix framework, system boundaries have been defined and variabilities in classification and terminology of water sources and users have been harmonized. Also, a water source (or origin) mix (WOmix) database for different users has been developed and a technological matrix has been established to link water sources to water production technologies and energy use. To develop the P-WSmix, a methodology based on algorithms enabling to obtain prospective WOmix (P-WOmix) is proposed. Data on water demand and water availability projections for different scenarios and time horizons have been used. The WSmix includes a framework, a WOmix database and technological matrix. The P-WSmix includes also a framework, a P-WOmix database and electricity mix and technology evolutions. The WOmix database covers 93 countries at different spatial scales for various users. The P-WOmix covers 73 countries at national scale for two users under different scenarios and time horizons. It has been shown that the environmental impacts of supply public water are highly dependent on the country and change over time. The inclusion of WSmix and P-WSmix in LCI databases is relevant for a more consistent water-use related impact assessment and for the LCA of infrastructures or products with a long life span.

198

The evolution of database- and tool development for Agri-footprint

B. Durlinger, L. Kuling, Blonk Consultants

From performing individual Life Cycle Assessment (LCA) studies for specific products, the field is moving towards automated LCAs for full product portfolios and tool and database development. This ongoing evolution is a result of the increased quality and availability of background databases as well as an increased acceptance of LCA as the measurement and monitoring tool for environmental impact. However, a point has been reached where existing LCA software and data structures have become a limiting factor for further development. Therefore, we would like to present our recent developments regarding database and tool development for LCA purposes. Existing LCA software frameworks have become limiting in our database development, because they only have a limited set of calculation features and interfacing capabilities. Also, the data structure of existing LCA software has proven to be limiting. For example, there is no explicit distinction between a process, products/substances, and exchanges. This can result in loss of valuable information. Therefore, we have decided to develop our own database infrastructure and accompanying calculation and import/export modules, that provide enhanced flexibility. This allows for more freedom, we can now make our own choices on how data is stored, what types of analyses can be performed and how this information is presented to a user. In addition, we see a trend where LCA analysts are becoming more and more interested in advanced tools that utilise Life Cycle principles. For Agri-footprint 2018 we are therefore developing a completely new framework in a Python/Django environment that aims leverage the past developments of Blonk Consultants and Agri-footprint and utilise them to develop a cloud based Life Cycle Inventory datastore and calculation engine to support and improve both our internal data developments and to serve as a backbone for custom tools for users. With this presentation we hope to contribute to the advancement of LCA databases and tools by providing insight in recent Agri-footprint developments.

199

Poster spotlight: TU097, TU098

Behavioural Ecotoxicology: Unravelling behavioural responses to chemical contaminants in the environment

200

Do laboratory assays predict behaviour in the wild? A study with pharmaceutical pollutants

E. McCallum, Umea University; A. Sundelin, J. Fick, Umea University / Department of Chemistry; A. Alanärä, Swedish University of Agricultural Sciences SLU / Department of Wildlife, Fish & Environmental Studies; G. Hellström, Swedish University of Agricultural Sciences SLU; T. Brodin, Umea University / Department of Ecology and Environmental Science

Concern over the impact that pharmaceuticals have on wild aquatic organisms has increased over the past decade. Laboratory studies have shown that pharmaceuticals can cause sub-lethal changes to animal behaviour and physiology; however, few studies have addressed whether effects documented in the laboratory extend to the natural environment. We exposed fish to one of two pharmaceuticals (temazepam and irbesartan) commonly detected in the environment at two doses (80 ng/L, 1500 ng/L; 200 ng/L, 20000 ng/L, for temazepam and irbesartan, respectively). We then assessed how exposure affected fish behaviour in the laboratory (scototaxis to measure anxiety and activity responses) and in the field (downstream dispersal using PIT tags). We found no evidence that either pharmaceutical treatment affected behaviour in the laboratory scototaxis assay. In contrast, fish exposed to both the high and low doses of temazepam dispersed faster downstream when compared to control fish. Irbesartan exposure did not affect fish behaviour in the field. Across all treatments, we also found that activity in the laboratory correlated with migration speed, indicating that fish that were more active in the laboratory also moved faster downstream in the wild. We discuss our findings in relation to differences in tissue bioconcentration for both pharmaceutical compounds in the muscle tissue of fish in our study. Our results emphasize the importance of measuring how pollutants affect ecologically relevant behaviours in the field alongside standard and efficient laboratory assays.

201

Exposure to the widespread androgenic steroid 17β-trenbolone alters behaviour in fish

M.G. Bertram, Monash University / Biological Sciences; M. Saaristo, J.M. Martin, T.E. Ecker, C.P. Johnstone, B.B. Wong, Monash University / School of Biological

Sciences
The capacity of pharmaceutical pollution to alter behaviour in wildlife is of increasing concern to the scientific community. A major pathway of these contaminants into the environment is the treatment of livestock with hormonal growth promotants (HGPs), highly potent veterinary pharmaceuticals that can enter aquatic ecosystems via effluent runoff. Hormonal growth promotants are designed to have biological effects at low doses, often act on physiological pathways that are evolutionarily conserved across taxa, and have repeatedly been detected in ecosystems worldwide. However, despite being shown to cause altered development, reproduction and morphology in various non-target species, relatively little is known about the potential of HGPs to alter ecologically important behaviours, especially across multiple contexts. Here, we investigated the effects of short-term (21-day) exposure to field-detected levels (average measured concentration: 16 ng/L) of 17β-trenbolone-—a potent growth-promoting veterinary pharmaceutical repeatedly detected in freshwater systems—on a suite of ecologically important behaviours in female eastern mosquitofish (*Gambusia holbrooki*). We found that fish exposed to 17β-trenbolone- were more active and exploratory in a novel environment (i.e. maze arena), while boldness was not significantly affected. Further, when tested for sociability, exposed fish were again more active and exploratory, and spent less time associating with a shoal of stimulus (i.e. unexposed) conspecific females. Lastly, when assayed for foraging behaviour, exposed fish spent a greater total amount of time within a foraging zone containing of an array of prey items (chironomid larvae) than did unexposed fish, entered this zone more frequently, and were more likely to feed. Further, a significant effect of exposure was detected on the total number of prey items consumed, although treatment-induced increases in foraging behaviour were dependent on female size. Taken together, these findings highlight the potential for sub-lethal levels of veterinary pharmaceuticals detected in the environment to alter sensitive behavioural processes in wildlife across multiple contexts, with possible ecological and evolutionary implications for exposed populations.

202
Selective grazing behaviour of chironomids between three microalgal species under pesticide pressure
J.N. Neury-Ormanni, Irstea / EABX-CARMA; C.N. Doose, INRS - Centre Eau Terre Environnement; B. CHAUMET, Irstea; N. Mazzella, Irstea Bordeaux / UR EABX; N. Majdi, Ecolab / UMR 5245 CNRS; J. Vedrenne, S. Morin, Irstea Bordeaux / UR EABX; W. Traunspurger, Bielefeld University / Animal Ecology
The herbicide diuron and the insecticide imidacloprid are amongst the most frequently detected pesticides in French rivers, and each is known to affect many aquatic organisms. However, it is less examined whether and how both pesticides together might induce multistress conditions, which could induce indirect effects such as modification of biological interactions within freshwater microbial communities. This study was undertaken to determine the effect of diuron and imidacloprid alone and in combination on the feeding behavior of chironomid larvae. A first experiment measured the impact of the different contamination conditions at environmental concentrations (5µg L^{-1} for each pesticide) on the grazing rate of chironomids on three microalgae species, independently. Therefore, two diatom species, *Gomphonema gracile* (two different morphotype: normal (GG) and teratogen (GT)) and *Planothidium lanceolatum* (PL), and one green algae *Pseudokirchneriella supcapitata* (PS) were offered as food, during 24h. Protein and lipid contents in microalgae were analysed subsequently. Each pesticide condition elicited a different grazing rate in chironomids with regards to algal species and their nutritional quality, with a general preference for *Gomphonema gracile* with teratogen shape and *Pseudokirchneriella supcapitata.* In a second experiment (cafeteria), food selectivity of chironomids was determined under similar contamination conditions during 4h: Under diuron, larvae switched equally among microalgae, then were as mobile as in the control without pesticide. However, imidacloprid and the pesticide mixture condition altered chironomids' movements and grazing behaviour. In these experiments, we highlighted that chironomids feeding behaviour and food preferences are impacted by pesticides. Herbicide and insecticide exposure, alone or in combination, had contrasting effects on grazing, both directly on the larvae or indirectly (food selectivity according to its quality). Our study illustrates the value of considering the impacts of toxicants on target and non-target organisms across trophic levels to improve ecotoxicological risk assessment in an ecosystemic perspective.

203
Environmental levels of anxiolytic pharmaceuticals alter migration of Atlantic salmon in both lab and field
T. Brodin, Umea University / Department of Ecology and Environmental Science; G. Hellström, Swedish University of Agricultural Sciences SLU; J. Klaminder, Umea University / Ecology and Environmental Science; F. Finn, Umea University; A. Lagesson, Umea University / Department of Ecology and Environmental Science; M. Jonsson, Umea University; J. Fick, Umea University / Department of Chemistry
Humans consume more pharmaceuticals than ever and consumption is set to rise. As a consequence, increasing amounts of pharmaceuticals are released into waterways worldwide with virtually no knowledge of how they might affect aquatic ecosystems. Some conspicuous effects of these *emerging contaminants* are already evident including the feminization of fish by contraceptive residue. However, recent work suggests that important effects of pharmaceuticals in aquatic environments are much more widespread than currently believed, and that these effects may result in major changes in species interactions, population survival and ecosystem functioning. In several earlier laboratory studies, we have shown that concentrations of pharmaceuticals presently found in waterways alter important behavioural traits in both aquatic macroinvertebrates and fish, and that this in turn affects both feeding efficiency and predation risk. These results suggest that pharmaceutical contamination of aquatic environments may change species interactions, in particular predator-prey interactions, with severe ecosystem-effects as potential consequence. Recently our research focus has turned towards realistic large-scale studies in lakes and rivers using acoustic telemetry to test if findings from the lab also hold in natural settings. Here I present results from one such study comparing effects of environmental levels of the anxiolytic pharmaceutical Oxazepam on migration pattern of Atlantic salmon (Salmo salar) in the lab and the field. In the lab, salmon exposed to the drug migrated approximately twice as fast as unexposed salmon and the subsequent field-study generated similar results, validating the results found in the lab. This pharmaceutically induced change in migration-intensity has the potential to be a key determinant between survival and mortality of salmon individuals and as such important for population persistence as migration intensity is believed to be adapted to the environmental conditions of the river in question. The overall finding of recent studies suggests that effects of pharmaceutical contamination of natural systems might be much more widespread than we predict based on conventional ecotoxicological tests.

204
Can personality influence the response of fish to environmental contaminants?
M. Oliveira, University of Aveiro; M. Sampaio, T. Santos, University of Aveiro / Biology Department & CESAM; A.L. Machado, University of Aveiro / CESAM Department of Biology; M. Barbosa, Department of Biology & CESAM - University of Aveiro / Biology; I. Domingues, University of Aveiro / CESAM Department of Biology
Stress is a determinant factor reducing animal welfare. Currently, it is recognized that animals react differently as a function of their personalities, or stress coping styles (i.e. consistency in behavioural and physiological responses across time or contexts). However, the role of personality in modulating individual response to environmental contaminants have received limited attention, despite the recognition that personality traits associated with a shy-bold continuum play an important role in animal fitness. Knowing that pharmaceuticals can interfere with personality, one question arises: what is the role of personality on animals' response to contaminants? In this research, zebrafish was sorted according to their exploration of a novel environment into shy and bold individuals, and subsequently exposed during 96h to carbamazepine, a human pharmaceutical, suggested as a marker of anthropogenic pollution. Assessed responses included behaviour (distance swan, position in the tank and time spent swimming) and biochemical markers associated with oxidative stress, neurotransmission and energy metabolism. Overall, our results showed significant differences between control shy and bold organism with behaviour endpoints demonstrating to be very sensitive to stressor conditions. Although carbamazepine alone did not show considerable effects in the assessed endpoints, strong interactions were found between personality and pharmaceuticals, supporting further studies.

205
Effects of fluoxetine on anxiety-related behaviours and physiology in a songbird
S.E. Whitlock, Environment Department, University of York / Environment; R. Shore, Centre for Ecology & Hydrology (NERC); J. Lane, Animal and Plant Health Agency; K. Herborn, Newcastle University / Centre for Behaviour and Evolution; M.G. Pereira, Centre for Ecology & Hydrology / Lancaster; K. Arnold, University of York / Environment
Several species of bird are known to forage directly on invertebrates at wastewater treatment plants (WWTPs), representing an exposure route to a range of contaminants including pharmaceuticals. The selective serotonin reuptake inhibitor (SSRI) fluoxetine is heavily prescribed and has been widely detected at WWTPs. Since fluoxetine is commonly prescribed for anxiety, we hypothesised that the antidepressant could modulate anxiety behaviour and physiology in exposed birds. Anxiety is an important state which arises in response to a real or perceived threat, enabling the individual to respond appropriately. Contaminants with the potential to alter anxiety-related behaviours are thus of concern to wildlife. We conducted a study to investigate the effects of chronic exposure to an environmentally relevant concentration of fluoxetine in a model songbird, the Eurasian starling (*Sturnus vulgaris*). We used a combination of behavioural and physiological endpoints to assess the effects of fluoxetine, specifically: 1) object neophobia, 2) exploration in a novel environment, 3) activity levels, 4) faecal corticosterone (CORT) metabolite concentration; and 5) leg skin temperature. Compared with pre-treatment data, fluoxetine-treated birds became less neophobic on average after six weeks of dosing, indicating a decrease in anxiety behaviour. There was no such reduction in neophobia in the control group. After six weeks of dosing, control birds became more active on average but fluoxetine-treated birds showed no increase in activity,

indicating increased lethargy in the fluoxetine birds relative to controls. There was no clear effect of treatment on exploratory tendency. Finally, infrared thermography showed that fluoxetine-treated birds had significantly colder legs compared with controls. This indicated that, as observed in humans, fluoxetine causes vasoconstriction, which in birds will affect the ability to thermoregulate. This study provides further evidence that low, environmentally relevant concentrations of pharmaceuticals can cause sublethal changes to behaviour and physiology that are predicted to impair the capacity of wildlife to respond appropriately to environmental changes.

Can trends in wildlife populations revolutionise our understanding of the impacts of chemicals on the environment?

206

Does single compound risk assessment protect from mixture effects and multiple stress?

P. Von der Ohe, UBA - Federal Environment Agency / IV 2.2 Pharmaceuticals

There is clear evidence that stress from anthropogenic activities can have profound local and regional effects on aquatic communities. To what extent chemicals are responsible remains largely unknown. The question whether a single compound risk assessment can protect from further deterioration of our water resources is discussed in the light of current mixture toxicity frameworks and multiple stress considerations. Here we present a European wide risk assessment of organic chemicals, based on regulatory monitoring data at about 6,000 monitoring sites available from the European Environment Agency (EEA). For the more than 600 mostly industrial substances, including many detergent ingredients such as benzotriazol, the available quality standards were collated or predicted from reliable QSAR models. Results showed that organic chemicals are likely to exert long-term effects on sensitive species in more than ¾ of the sampling sites with multi-year samplings. In this study, we analyzed the potential cumulative effects of multiple exceedances of the PNEC in consecutive years as well as from various substances. The monitoring programs considered in this study often include only a subset of the chemicals expected. Hence, our assessment is likely to underestimate the actual risk. Nevertheless, the results show that multiple exposures at each site is rather the rule than the exception. Finally, we discuss whether chemicals from WWTP have significant effects on aquatic invertebrate communities as compared to effects from local habitat. For that purpose, we analysed two data sets on macro invertebrate communities where we have \n a) similar chemical stress, but differing habitat quality and b) similar habitat quality but differing chemical stress. The results indicate that in direct comparison, chemical stress induced larger effects as compared to habitat degradation. Our results therefore clearly indicate that chemical pollution is still a large-scale environmental problem that requires far-reaching, holistic mitigation measures to preserve and restore ecosystem health. It also suggests that current risk assessment approaches on a substance by substance bases are likely not protective for the environment.

207

Threshold trends in wildlife taxa: challenging and evaluating our chemical- and environmental risk assessments of chemicals and their mixtures

L. Posthuma, RIVM / Centre for Sustainability, Environment and Health; D. De Zwart, DdZ Ecotox / Centre for Sustainability Environment and Health; J. Postma, Ecofide; M.C. Zijp, RIVM / Centre for Sustainability, Environment and Health

“Big data” are a potential goldmine for studying and contextualizing chemical and environmental risk assessments, as they enable relating predicted risks to observed impacts. This can confirm or challenge our chemical risk assessments, by checking whether Environmental Quality Standards provide sufficient safety, and whether or not mixture exposures cause problems not captured in the widely used ‘single-chemical plus safety factor’ approaches. Digging the goldmine asks for – first – data collation, and then: a powerful design which can be borrowed from epidemiology. This presentation is a recent gold-digging trial, with surprising results. We collated biomonitoring and mixture exposure data from the Dutch Waterboards, as collected due to obligations of regulatory frameworks (such as the Water Framework Directive). We quantified for each sample, based on measured chemical concentrations, the mixture toxic pressure at EC50-level. Outcomes were expressed as multi-substance Potentially Affected Fraction (msPAF-EC50) of species via Species Sensitivity Distribution modelling (SSD). Earlier research suggested that this proxy – higher values of which are interpreted to imply a higher potential for species loss – has a gross absolute relationship with species loss in various regional studies. In the current study, we overlaid the SSD-model basis (all species are unequal in their sensitivity to exposures) with this finding, by analyzing taxon-specific threshold values. That is, we determined the taxon-specific msPAF-EC50 beyond which species abundance starts changing when toxic pressure rises, in a downwards (sensitive) or upwards (indirect opportunistic response) direction. The results show a series of species-specific mixture exposure thresholds, and an overall assemblage-level (aggregated) exposure threshold. These outcomes are compared to simpler approaches, such as quantile regression on the species assemblage level, as well as multi-stressor statistics. We conclude that the set of species-specific and assemblage-level thresholds bear important contextual information to judge our risk assessment and management foundations, be it in the realm of prospective Chemical risk assessment or of the retrospective Environmental risk assessment and management.

208

How much do improvements in wastewater treatment benefit downstream macroinvertebrate populations?

A. Johnson, CEH Wallingford / Wallingford; F. Edwards, Centre for Ecology Hydrology Maclean Building; M.D. Juergens, Centre for Ecology and Hydrology / Wallingford; H. Vincent, Centre for Ecology Hydrology Maclean Building

The River Ray, which is a small tributary of the Thames (UK), offers an unrivalled opportunity to examine the impact of changes in wastewater treatment on the resident aquatic wildlife. This opportunity exists because the waste from the 170,000 plus people of Swindon discharges into this small 12 km long waterbody, such that the downstream mean annual flow is composed of 65-80% treated effluent. The downstream monitoring sites showed a sustained improvement in macroinvertebrate diversity starting from 1991 onwards. This sustained improvement for macroinvertebrates coincided with a 10-fold drop in ammonia, halving of biodegradable organics, (BOD) and improvement in dissolved oxygen associated with the conversion of the Swindon plant from trickling filter to nitrifying activated sludge. There were no dramatic changes in metal concentrations over the key early 1990s’ period unlike the main sanitary determinands. Whilst there was no change in overall flows, winter water temperatures downstream of Swindon rose over the course of the 30 year monitoring period. We could not identify a notable benefit to the macroinvertebrate community from the application of tertiary granular activated charcoal treatment (GAC) lasting from 2008 to 2014. No new macroinvertebrate families appeared during this period. The steady improvement in macroinvertebrate diversity in an effluent dominated river implies that current chemicals in domestic wastewater are not noticeably harmful to these organisms. This implied that provided we can achieve a 90th percentile BOD below 5 mg/L, NH_3 below 0.6 mg/L and DO above 60% saturation, aquatic wildlife macroinvertebrate diversity will steadily improve (within the limits of habitat suitability). The small river has a population of stickleback and other fish but their changes in abundance are not known. The recovery of macroinvertebrate diversity is slow and has not yet returned to the expected range for such a river, however, it was noted that the habitat is not ideal. This project has demonstrated the immense power and value of consistent long-term parallel chemical and wildlife monitoring. Andrew C. Johnson, François Edwards, Monika D. Jürgens, Helen Vincent Centre for Ecology and Hydrology, Wallingford, Oxfordshire, OX10 8EB, UK ajo@ceh.ac.uk Keywords: macroinvertebrates, biodiversity, wastewater, GAC Track 7, Session 7.2 Preference Platform

209

Biometric parameters of the bream (Abramis brama) as indicators for long-term changes in environmental quality - results from the German ESB

D. Teubner, Trier University / Biogeography; M. Paulus, M. Veith, Trier University; R. Klein, Trier University / Biogeography

Fishl health depends upon the state and quality of the aquatic environment. Changes in physical condition of fish may therefore be attributed to changes in environmental quality. Based on time series of 20 years of biometric data of bream from multiple sampling sites of the German environmental specimen bank (ESB), we assessed which biometric parameters and indices of bream are suitable indicators for long-term changes in fish health and environmental quality. The length and weight of individuals of a defined age, hepatosomatic index and with restriction the condition factor and lipid content of bream are reliable indicators for long-term changes of fish health and hence hint at long-term changes of environmental quality. We show examples for current trends of these indicators in German river systems. Our results confirme the high value of biometric parameters for monitoring of long-term changes in state and quality of aquatic ecosystems.

210

The burden of being a slow-life cycle species: freshwater fish population dynamics in France, correlations to species life traits and implications in ecotoxicology

R. Santos, Hepia, University of Applied Sciences Western Switzerland / Ecology and Engineering of Aquatic systems research group; N. Poulet, Agence Française pour la Biodiversité / Pôle Ecohydraulique, AFB-IMFT; A. BESNARD, Centre d Ecologie Fonctionnelle et Evolutive / Biogéographie et Ecologie des Vertébrés

Population dynamics of aquatic species and ultimately their population growth rate (λ), must be known to properly define species conservation status and plan appropriate conservation strategies. It is also essential to understand how inter-specific variations in demographic and life history traits influence population dynamics. For this purpose, we implemented an integrative approach focused on 18 common freshwater fish species representing 94 % of fish abundance and 88 % of fish biomass sampled since 1990 in 546 monitored sites in France. Abundance and biomass growth rates were estimated with space-state models and fish length trends with quantile regressions. To further study correlations between fish abundance, biomass, fish-length trends and fish life traits (life history strategies, species trophic position, habitat preferences and thermal tolerance) we performed multivariate analyzes. The present work demonstrates that during the last decades, 10 species exhibited significant decline in abundance, 2 species were in expansion and fish abundance remained stable for 6 species. The correlation between biomass and abundance growth rates was also very high (R^2=0.93). The intra-specific trends in fish length over the studied period also showed a severe decrease among the largest individuals (quantile 0.75 and 0.90) and was correlated to severe biomass decline in several species. This result reflect progressive alterations in the population size / age structures suggesting that a decrease in growth and survival might be responsible of the pattern here observed. Among the demographic and ecological traits we investigated, generation time and fish maximum length were the most correlated to species population growth rates indicating the decline of slow generation time species. These results are discussed with regards to global pressures which could explain large scale decline of periodic species with a focus on chemical pressures which could explain body growth decrease, juvenile and adult survival alterations due to micropollutant exposures. Further attention will be paid to discuss conservation measures and life stages which should be protected in priority to favor periodic species recovery.

211
The use of natural historical collections to reconstruct temporal trends of the exposure to major contaminants in different white-tailed eagle (Haliaeetus albicilla) populations
J. Sun, Antwerp university / Department of Biology; J.O. Bustnes, Norwegian Institute for Nature Research / Fram Centre; A. Covaci, University of Antwerp, Toxicological Center / Toxicological Centre Dep of Pharmaceutical Sciences; B. Helander, Swedish Museum of Natural History / Environmental Research & Monitoring; G. Malarvannan, University of Antwerp / Toxicological Center; J. Søndergaard, Aarhus University / Arctic Research Centre; K. Thorup, A. Tøttrup, Natural History Museum of Denmark; M. Eens, University of Antwerp / Department of Biology; I. Eulaers, Aarhus University / Department of Bioscience
Understanding temporal trends of persistent contaminants in wildlife is critical for the assessment of potential associated past, present and future health risks. We have established a retrospective examination of Mercury (Hg) and several organohalogen contaminants (OHCs) in Swedish, Norwegian and Greenlandic white-tailed eagle (*Haliaeetus albicilla*) using body feathers obtained from natural history and ornithologist collections. We analyzed feathers for Hg, polychlorinated biphenyls (PCBs) and polybrominated diphenyl ethers (PBDEs), as well as stable carbon and nitrogen isotopes. The aims of the present study were: 1) to retrospectively reconstruct temporal trends of major contaminants in relation to anthropogenic emissions and regulations, 2) to interpret how dietary habits (using stable isotope proxies) may have affected the temporal trends, and 3) to document a potential spatial gradient from high (Sweden) to low (Greenland) anthropogenic contaminant input. Strong evidence for spatiotemporal patterns was found based on preliminary results for Hg and OHCs in the Swedish and Norwegian populations. Swedish white-tailed eagle feathers showed concentrations associated with adverse physiological effects, thus supporting the earlier observed dramatic decline in productivity. Hg concentrations were much lower in the Norwegian feathers, while the peak PCB exposure is of the same order of magnitude as the average PCB exposure in Swedish white-tailed eagle during 1970s (median = 3,293 ng g^{-1}). Clear declining trends were observed for both the Swedish and Norwegian time series after 1970s. We did not detect PBDEs before 1970 in the Swedish or Norwegian time series, although concentrations increased steeply during the 1980s, decreasing trends were observed afterwards. Hg concentrations in the Greenlandic time series were stable before 1940, but showed an increasing trend from 1980 to 2013. These results suggest that changes in historical and recent anthropogenic emissions strongly shape temporal changes of persistent contaminants in avian top predators. In addition, the relative interplay between local versus long range transport have likely affected the spatial pattern of contamination levels. The here presented preliminary results will be complemented with data for the Greenlandic populations as well as emission and stable isotope data to disentangle the relative effects of dietary habits and anthropogenic contaminant sources.

Environmental effects of metals: Improvements to risk assessment by considering speciation and bioavailability (I)

212
Findings of a SETAC Technical Workshop on Bioavailability-based Water Quality Criteria
C.E. Schlekat, NiPERA; K. Gallagher, U.S. EPA / Office of Water; B.A. Stubblefield, Oregon State University / Environmental and Molecular Toxicology
A workshop entitled “Technical Workshop on Bioavailability-based Water Quality Criteria” was held under the auspices of the Society of Environmental Toxicology and Chemistry on 3-8 December 2017 in Pensacola, Florida, USA. The goal of the workshop was to evaluate the state-of-the science regarding metals bioavailability models and their use from a regulatory perspective. A second, but equally important, goal was to provide a scientific forum for discussions that could inform the further incorporation of bioavailability concepts into regulatory approaches for deriving water quality criteria/standards for metals. The state-of-the-science regarding the ability to accurately predict the toxicity of metals in the environment to aquatic organisms has increased rapidly in recent years. Workshop participants reviewed the current state-of-the-science on aquatic bioavailability information for metals and on methods to model bioavailability under a range of environmental conditions. Participants also discussed the technical challenges associated with applying bioavailability-based approaches, especially in a regulatory context. The outcome of the workshop will support expanded incorporation of metal bioavailability information into global regulatory frameworks. Approximately 40 invited scientists representing industry, government, and academia participated in the workshop. Participants were divided into five groups with each group addressing one of the following meeting objectives: Review of the state-of-the-science regarding the issue of metal bioavailability and toxicity to aquatic organisms, Determine the extent to which available biotic ligand models (BLM)/multi-linear regression (MLR)-based models/or other alternative approaches offer a means to address metals bioavailability and toxicity and to which they are protective of aquatic life, Develop technical information regarding the type and quantity of data necessary to develop and apply bioavailability-based aquatic toxicity models, Develop recommendations for approaches to validate bioavailability-based models, and Provide recommendations for suggested measures of acceptability for bioavailability-based aquatic effects models. Workshop findings will be presented and will later be published in the form of a SETAC “Summary document” and a series of manuscripts to be submitted to a SETAC journal.

213
Modifying factors for nickel speciation and toxicity in seawater
W. Chen, Wilfrid Laurier University; S. Sherman, Wilfrid Laurier University / Biology; J. McGeer, Wilfrid Laurier University / Department of Biology; R.C. Santore, Windward Environmental, LLC; T. Blewett, University of Alberta; G. Merrington, WCA Environment Limited; D. Smith, Wilfrid Laurier University / Department of Chemistry
Nickel (Ni) toxicity in seawater is of increasing concern because of coastal Ni mining and processing activities. Determining Ni speciation is vital to understanding and predicting Ni toxicity and ultimately for bioavailability based nickel risk assessment. Application of existing freshwater bioavailability based approaches for nickel in salt water predicts negligible binding of Ni to dissolved organic matter, but there are several examples of toxicity tests in natural seawater that are protective compared to artificial seawater control samples. Determining (1) the source of this protective effect (2) how geographically wide-spread protective saltwater sources are, and (3) to be able to predict Ni speciation and toxicity, are the objectives of this project. As a test of Ni toxicity protection by ligand complexation in salt water media, defined solutions of artificial seawater (ASW) containing different model compounds (i.e. citric acid, EDTA, L-tryptophan, glutamic acid, and histidine) were titrated with Ni to determine speciation. In addition, Ni speciation was determined in real saltwater samples of diverse geographic origin from the east coast of the United States and Canada. The divalent Ni free ion in these synthetic and real seawater samples was quantified using Ion Exchange Technique (IET) with Ni measured by Graphite Furnace Atomic Absorption (GFAA). The measured Ni^{2+} values were compared with model predictions (i.e. Visual Minteq) for evaluating the feasibility and applicability of the IET method for Ni in seawater. For the most part IET-measured [Ni^{2+}] agreed very closely with model predictions. In the same defined solutions, 96-hour Ni embryo toxicity tests were performed for a sea urchin (*S. purpuratus*). The dose response curves were expressed both as total dissolved Ni concentration ([Ni_D]) and free Ni concentrations from IET ([Ni^{2+}]). If the Ni toxicity is explained by [Ni^{2+}], all the toxicity response curves of different model ligands will overlap and this was in fact observed for the majority of samples. The results of this research contribute to the development of bioavailability-based prediction models for estimating the impacts of Ni in marine water. Funding was provided by Natural Sciences and Engineering Research Council of Canada (NSERC), Vale Canada and NiPERA Inc.

214
Acute bioavailability models for nickel: Development and regulatory application
K. De Schamphelaere, Ghent University (UGent) / Applied Ecology and Environmental Biology; P. Van Sprang, I. Vercaigne, ARCHE; A. Peters, wca; C.E. Schlekat, NiPERA; E.R. Garman, NiPERA / Ecotoxicologist
Chronic bioavailability models for nickel are well-established. For example, the Annual Average Ni Environmental Quality Standard (AA-EQS) under the European Union’s (EU) Water Framework Directive (WFD) is based on normalization of chronic Ni ecotoxicity data using chronic Ni bioavailability models. Acute Ni models have been developed for invertebrates, fish, and algae, but they have received less attention than chronic bioavailability models. However, given that acute Ni toxicity can vary by greater than 17-fold for aquatic invertebrate species tested in variable water chemistries, normalization of acute Ni ecotoxicity data is an important consideration for the determination of Maximum Allowable Concentrations (MAC) under the WFD. The goal of this study was to test if the existing acute Ni bioavailability models can be used to predict acute Ni toxicity to both model and non-model species and to demonstrate how they can be applied to derived MAC values. Acute Ni ecotoxicity data (eight species from 13 different studies) were identified from the literature. Data were accepted for analysis if acute Ni toxicity for a species was tested in >2 test waters differing in physico-chemistry.

Acute Ni bioavailability models (3 invertebrate models, 2 algae models, and 1 fish model) were used to evaluate the ecotoxicity data. To simplify the normalization process, an "average animal" bioavailability model was developed using a weighted average of parameters for existing models. Because crustaceans are typically among the most sensitive organisms to Ni exposure, and because the fish model did not capture pH effects on acute Ni toxicity to crustaceans very well, an "average crustacean" model was also developed. The arithmetic mean of parameters for the 3 crustacean models was used to construct the "average crustacean" model. Both the "average animal" and "average crustacean" models reduced intraspecies variability considerably among the available Ni ecotoxicity data. For example, the "average animal" model predicted 98% of the 193 individual acute ecotoxicity data points within a 3-fold error, and 90% within a 2-fold error. The "average crustacean" model performed similarly, although this model clearly showed a better ability to predict the effect of pH on Ni toxicity to cladocerans. The models were applied to an acute Ni ecotoxicity dataset to derive bioavailability-based MAC for European water bodies with typical ranges of water chemistry.

215

Bioavailability and bioaccumulation of uranium: From lab experiment to modelling

A. Husson, Mines ParisTech / Hydrodynamics and reactions team (HR); M. Leermakers, Vrije universiteit Brussel / Department Analytical, Environmental and Geochemistry; M. Descostes, AREVA Mines / R&D; V. Lagneau, Mines ParisTech PSL Research University / Geosciences Hydrodynamics and reactions team (HR)

Environmental regulations are becoming more and more ecosystems-oriented. In this respect, the habitat's faunal biodiversity is targeted, meaning the contaminant's bioavailability is taken into account. The latter depends on the geochemical conditions of the aquatic system, which define the chemical speciation. For several elements, Environmental Quality Standards (EQS) and Sediment Quality Standards (SQS) have been proposed. For non-priority elements such as Uranium, EQS and SQS should be supplemented with national standards. As the bioavailable fraction in sediments is dependent on geochemical factors and sediment mineralogy, it is important to investigate the role of sediment composition on the bioaccumulation of uranium in benthic organisms. In the present study, laboratory experiments were performed on the bioaccumulation of uranium in the larvae of the nonbiting midge *Chironomus riparius* exposed to specific mineralogical phases (pure Quartz (Q), 10% Kaolin/ 90% Q, 10% Smectite/ 90% Q, 10% Ferrihydrite (FOH)/ 90% Q and a mixture of the 4 mineral phases (3.3% Kaolin/ 3.3% Smectite/3.3% (FOH) and 90% Q) spiked with uranium at two different concentration levels. During a ten days' exposure experiment, the uptake of uranium in the chironomid was investigated and the concentrations of uranium in sediment, overlying water, pore water were measured as well as the composition of major ions and physicochemical parameters. Diffusive Gradients in Thin Films (DGT) devices were deployed simultaneously to investigate the relationship with the uptake of uranium in the chironomid larvae Sediment to porewater partition coefficients (K_D) range from 10 L/Kg for quartz to 60000 L/Kg for FOH. After ten days exposure, the highest Biota Sediment Accumulation Factor (BSAF) is found for quartz (11), followed by Ferrihydrite (9) and Smectite (8) and is the lowest for the mixed composite sediment (1). DGT labile uranium porewater concentrations account for 70-100% of the uranium in porewater for all mineral phases except the quartz, where C_{DGT} only accounts for 10% of the uranium in porewater. Uranium accumulation on the DGT units is correlated with the accumulation in the chironomids. Results obtained by lab experiments are used in a geochemical speciation code (CHESS) to model the uranium sorption behaviour and chemical speciation in the aqueous phase. These results are compared with the proposed regulations by IRSN on uranium bioavailable chemical species.

216

Empirical Investigations into the Toxicity and Bioavailability of Aluminium to Aquatic Species

B.A. Stubblefield, Oregon State University / Environmental and Molecular Toxicology; A.S. Cardwell, Oregon State University / Faculty Research Assistant; W.J. Adams, Red Cap Consulting; R. Gensemer, GEI Consultants / Ecological Division; R.C. Santore, Windward Environmental, LLC; E. Nordheim, Aluminium REACH Consortium

Implementation of the Registration, Evaluation, Authorisation, and Restriction of Chemicals (REACH) regulation over the past years has resulted in: 1) the generation of large amounts of empirical toxicity data and 2) increased our understanding of the relationships between water physicochemical parameters and aluminium (Al) toxicity. REACH requirements necessitate the production of data describing the chronic toxicity of regulated chemicals to a variety of aquatic organisms. To address possible data gaps in the Al database, a series of chronic toxicity tests were conducted with freshwater organisms. Aluminium toxicity is a function of its speciation and this is a function of water pH. Previous chronic toxicity tests with Al were typically conducted under acidic test conditions and few studies have been conducted at pHs more typical of natural surface waters. The studies reported here investigated the chronic toxicity of Al at pH 6.0 to 8 freshwater species. The species tested were the great pond snail (*Lymnaea stagnalis*), a rotifer (*Brachionus calyciflorus*), an aquatic oligochaete (*Aeolosoma* sp.), a midge (*Chironomus riparius*), an amphipod (*Hyalella azteca*), an aquatic plant (*Lemna minor*), and two fish, the fathead minnow (*Pimephales promelas*) and zebrafish (*Danio rerio*). Chronic test durations ranged from 48 hours to 35 days. The most sensitive species was the zebrafish (10% effect concentration (EC10) of 98 µg/L total Al) based on growth effects. The least sensitive species was *Lemna minor*, with an EC10 of 2175 µg/L total Al as total dry weight. A series of chronic and short-term chronic tests conducted with *Ceriodaphnia dubia*, fathead minnows, and the algae (*Pseudokirchneriella subcapitata*) suggest that modifying factors such as pH, dissolved organic carbon (DOC), hardness, and temperature have a large impact on the bioavailability and toxicity of Al to aquatic organisms.

217

Main factors responsible for the environmental degradation of rivers in a basin dedicated to gold mining using ecological predictive models. Case study Ponce Enríquez.

C. Sanga, Escuela Superior Politecnica del Litoral (ESPOL) / Departamento de Ciencias Químicas y Ambientales; L. Dominguez, P. Almeida, Escuela Superior Politécnica del Litoral ESPOL / Departamento de Ciencias Químicas y Ambientales; J.C. Pindo, M. Arias, G. Loayza, Escuela Superior Politécnica del Litoral ESPOL / Facultad de Ciencias de la Tierra

The irreversible effects that the environment has suffered due to anthropogenic activity has reduced the availability of water for living beings, both in terms of quantity and quality. Industrial, agricultural, mining and urban development activities have in many cases led to the generation of pollutant discharges that threaten the health of our ecosystems. The impacts of mining activity on aquatic ecosystems have been widely documented, reporting the deterioration of water quality, the impact of biodiversity, as well as the release of heavy metals of potential accumulation in organisms and subsequent biomagnification through the food chain. Although it is known that non-technical mining activity affects the environment, it is necessary to identify and prioritize those factors related to mining activity that have a greater impact on the ecosystem (eg extraction, cyanidation, amalgamation). The identification of these factors would allow environmental control authorities to prioritize management actions focused on those parameters with the greatest impact, thus mitigating the impact of this activity on aquatic ecosystems. To illustrate this, the present study conducted in the Ponce Enríquez area seeks, through the construction of predictive models based on decision trees, to discriminate those environmental factors responsible for the environmental degradation observed in rivers and streams in the study area. In order to determine the environmental quality of the sites visited, the application of the BMWP / Col index was used, which is based on the structure of the macroinvertebrate community present. Additionally, a set of environmental variables of water and sediment quality were used as indicators of environmental pressure of the activities carried out in the basin, which is predominantly dedicated to the extraction of gold. Artisanal gold mining in the Ponce Enríquez area is a social technical problem that is affecting aquatic ecosystems and the ecosystem services they provide. The main stressors of the deterioration of the ecological quality of the rivers studied are calcium, copper, the total suspended solids and the modification of the characteristics of the substrate, all related to the mining development and climate change. Through the ecological predictive model it was possible to determine the permissible levels of calcium in the rivers to improve the environmental condition of 30% of the stations sampled. (Ca

Microplastics in freshwater and terrestrial systems - fate, monitoring and biological interactions (I)

218

Closing the gap between small and smaller: Towards a framework to analyse nano- and microplastics in aqueous environmental samples

P. Bauerlein, KWR / Analytical and Environmental Chemistry; A. van Wezel, KWR Watercycle Research Institute / Chemical Water Quality and Health; S. Mintenig, Utrecht University / Copernicus Institute of Sustainable Development; A. Koelmans, Wageningen University / Aquatic Ecology and Water Quality Management; H.M. Ohlendorf, CH2M

Detecting nanoplastics and determining actual concentrations and sizes of plastic present in the environment is essential to assess the risks that are posed by plastic particles. Microplastics have been detected globally in various aqueous ecosystems. The determination of nanoplastics is hampered due to the high methodological challenges. But yet, its formation was proven experimentally and, for the first time, its presence in marine surface water samples confirmed. Building on those results, we further propose a framework that is able to consistently determine a broad size spectrum of plastic particles in an aqueous environmental sample. The results from initial tests confirm the general applicability of individual techniques to, firstly, sample and, secondly, detect plastic sizes and polymer types. To obtain representative results, a sampling strategy is needed to concentrate plastic particles. Crossflow ultrafiltration is applied to concentrate nanoplastics from 100 into 0.5 L and yields in a reproducible particle recovery of 54 ± 2 %. Microplastics are detected using FTIR-microscopy which is limited to a minimum particle size of 28 µm. For nanoplastics field- flow- fractionation, that reveals information on the particle sizes, and pyrolysis GC-MS, that is used to identify the polymer types, are applied. Under the given settings the latter requires a mass of approximately 100 ng to identify polystyrene in an environmental sample by which this technique seems promising for the detection of nanoplastics. The pre-concentration by crossflow ultrafiltration reduces the determined detection limits, and enables the identification of polystyrene for an original concentration of 20 µg L^{-1} in an aqueous samples. Finally, we propose an approach to estimate polymer masses based on the two-dimensional microplastic shapes that are recorded during the analysis with FTIR-microscopy. By this, the results of spectroscopic and thermal degradation analyses (e.g. pyrolysis GC-MS) can be combined and compared.

219

Trace particulate plastic analysis in environmental systems: synthesis and utility of metal doped nano- and microplastic particles and fibers

F. Schmidt, M. Schmiedgruber, Eawag Swiss federal Institute of Aquatic Science and Technology; D.M. Mitrano, Eawag - Swiss federal Institute of Aquatic Science and Technology / Environmental Risk and Management

Research on particulate plastic (nano- and microplastic particles and fibers) and their distribution in the environment has intensified in recent years; but truly quantitative analysis, even at the bench scale, has remained elusive in part due to the analytical difficulties in detection. Synthesizing plastic particles with a metallic, chemically entrapped tracer can provide a robust way to more easily, accurately and quantitatively detect particulate plastic in complex environmental and biological media. In this study, a suite of methods to synthesize a variety of particulate plastics of various sizes (100 nm to 1 mm), surface morphologies/charges and polymers (polystyrene, polyester, polypropylene and low density polyethylene). Each variant has an embedded metallic fingerprint (Pd, In, Au; approximately 0.5% metal/wt) which can be used to detect plastic by common analytical techniques, such as ICP-MS. To highlight the utility of this approach, dilute concentrations of particulate plastic were measured in various media including river water, municipal wastewater treatment plant effluent and wastewater sludge. Nanoplastics, for example, were detected three orders of magnitude lower in number concentration than similar particles with a fluorescence label. After ensuring the metal tracer was stable over time, digestion methods were developed which took into consideration the specific polymer and metal in question to achieve a minimum of 85% recovery in every matrix. Furthermore, the particulate plastics synthesized were spiked into mixed liquor in batch experiments representing different stages of a municipal wastewater treatment plant to determine how various particulate plastics move through the system. In this way, some conclusions could be drawn concerning particle behaviour, aggregation and the likelihood of microplastics and the potential for microplastics to be discharged to freshwater within wastewater treatment effluent. Beyond the case study specifically highlighted here, these metal laden particulate plastics are suitable to study fate, transport, eco-toxicity and interactions with organisms at trace concentrations. By using these materials, bench scale and pilot scale studies can be used as a bridge to understand the environmental processes that dominate (particulate) plastic fate, transport and interactions with biota until analytical techniques to measure native particulate plastics of small sizes and in trace concentrations have matured.

220

Detection of polymers in treated waste water using TED-GC-MS

C. Goedecke, K. Altmann, Bundesanstalt für Materialforschung und prüfung; C. Bannick, Umweltbundesamt; E. Kober, Technische Universität Berlin; M. Ricking, UBA Umweltbundesamt; T. Schmitt, Berliner Wasserbetriebe; U. Braun, BAM-Federal Institute Materials Research and Testing / 5.3 Mechanics of Polymers

The presence of large quantities of plastic waste and its fragmentation in various environmental compartments are an important subject of current research. In the environment, (photo-) oxidation processes and mechanical abrasion lead to the formation of microplastics. However, until now, there are no established quality assurance concepts for the analysis of microplastic (< 5 mm) in environmental compartments, including sampling, processing and analysis [1-4]. The aim of the present work is the development of suitable examination methods and protocols (sampling, sample preparation and detection) to qualify and quantify microplastic in urbane water management systems. At first a fractional filtration system for sampling and the analytical tool, the so-called TED-GC-MS (thermal desorption gas chromatography mass spectrometry) were developed. The TED-GC-MS method is a two-step analytical procedure which consists of a thermal extraction where the sample is annealed and characteristic decomposition products of the polymers are collected on a solid phase. Afterwards these products are analysed using GC-MS [5]. The developed fractional filtration for sampling and the TED-GC-MS for detection were used for quantitative analysis to screen the waste water influent and effluent of a Berlin waste water treatment plant for the most relevant polymers, polyethylene (PE), polypropylene (PP), polystyrene (PS), polyethylene terephthalate (PET) and polyamide (PA). The results of the study revealed that the polymeres PE, PS and PP were detected in the effluent, and PE and PS were find in the raw waste water of the sewage treatment plant in Ruhleben, Berlin. Differences in polymer types and amounts were detected at different sampling dates and within different sieve fractions. Much higher amounts of polymers were observed in the raw waste water. The peak areas of the decomposition products, used for quantification of the polymers, were adjusted using so-called response factors since the TED-GC-MS method is more sensitive for PP and PS than for PE. It has been shown that PE is the most dominant polymer in the samples. Comparing the masses of polymers in the effluent and in the raw sewage, a removal of 97 % of the polymers in the water treatment plant can be assumed. These results are consistent with the literature where removal rates between 98-99 % were described [6-7].

221

Soil and sludge: A time and cost-effective method for extracting microplastics from complex, organic-rich environmental matrices

R. Hurley, NIVA - Norwegian Institute for Water Research; A. Lusher, NIVA Norwegian Institute of Water Research / Marine and Freshwater Research Centre; M. Olsen, Telemark University College; L. Nizzetto, NIVA

There is very little existing work on the analysis of microplastics in organic-rich substrates such as soil or sewage sludge. The organic components, as well as the complexity of the solid matrix, complicates the extraction process. No standardised methodology has thus far emerged. This study aims to establish an effective extraction technique appropriate for the monitoring of microplastic contamination in soil and sludge samples. Four main protocols (including two temperature and concentration variants) were tested for the removal of organic material followed by a density separation process. This approach was selected to afford comparability with existing sediment microplastic analyses. The selected reagents were: peroxide oxidation (60°C, 70°C), Fenton's reagent, NaOH (1 M, 10 M) and KOH. The methods testing procedure was split into three phases: 1. Effect of reagents on test polymers; 2. Efficacy of reagents in reducing organic matter content in soil and sludge; and 3. Extraction efficiency using spiked soil and sludge samples including density separation. 7 reference polymers were used: HDPE, LDPE, PP, PS, PET, PC and PA-66, which account for >70% of global plastic demand. Degradation was observed for several of the reagents and selected polymers. NaOH treatments caused the most significant changes, including degradation of PET and PC. Small degradative changes occurred as a result of peroxide oxidation and KOH treatments. Fenton's reagent caused no alterations to the tested polymers. NaOH and KOH treatments were not effective in sufficiently reducing organic material whilst peroxide oxidation and Fenton's reagent removed the majority of organic material. Based on these results, Fenton's reagent was identified as the optimum treatment and was further optimised. Phase 3 tested revealed high extraction efficiencies for this protocol. This technique represents a time- and cost-effective approach suitable for application in routine monitoring of complex environmental substrates whilst facilitating the collection of important particle information.

222

Mapping microplastics in sludge during a country-wide investigation of wastewater treatment plants

A. Lusher, NIVA Norwegian Institute of Water Research / Marine and Freshwater Research Centre; R. Hurley, Nowegian Institute for water research; M. Olsen, C. Vogelsang, NIVA Norwegian Institute for Water Research

Incorporation of anthropogenic particles into sludge has been highlighted as a major route for the transport of pollutants into the environment. Here we present the results of a nationwide survey investigating the incorporation of microplastics into sludge from domestic wastewater treatment plants which operate different waste water and sludge treatment technologies. The main objective of this project was to characterize, map and compare results between different types of wastewater treatment plants. Particles were extracted using organic matter removal followed by a density separation procedure following. Plastics were identified in sludge samples from all treatment plants investigated. The overall average microplastic concentration was 6 077 particles kg^{-1} (d.w.) (1701 – 19 837) or 1 176 889 particles m^{-3} (470 270 – 3 394 274). Particles from sludge consisted of beads (37.6%), fragments (31.8%) fibres (28.9 %) and glitter (1.7 %). Most of the particles were clear in colour (41%). Ten percent of the overall particles extracted were tested using FT-IR. All particles (n= 60) were confirmed to be plastics. Polyethylene particles were the most common (30.5%) followed by polyethylene terephthalate (26.7%) and polypropylene (20.3%). 62% of plastics were extracted during the low density (1 g cm^{-3}) separation steps and 38% were extracted at high density (1.8 g cm-3). Results between WWTPs were highly variable and there was no clear difference between average microplastic concentrations across the different treatment technologies. Based on this study and details on the application of sewage sludge in Norway, it can be estimated that approximately 446 billion microplastic particles are spread on agricultural soils, 27 billion microplastic particles are added to green areas and 112 billion microplastic particles are sent to soil producers per year. This equates to over 584 billion microplastics that are released into the environment via sewage sludge application each year in Norway.

223

The Influence of Weathering on the Sinking Behavior of Microplastic in Freshwater and all Surface Waters

H. Arp, NGI / Environmental Technology; L.B. Olsen, D. Issler, NGI; N. Berrojalbiz, NGI / Environmental Chemistry; S. Wongsoredjo, X. Shen, E. Toorman, KULeuven

Key to understanding the fate of microplastic particles in fresh water are the changes they undergo as a result of various weathering processes, like biofilm growth, aggregation, UV exposure and physical stress. Some of these weathering

processes may cause changes in the density and shape of individual plastic particles, or aggregates. This can be a driving factor for the ultimate fate of microplastic, as it could cause floating microplastic to sink or be suspended below the water surface. However, the factors controlling the buoyancy or sinking velocity of different microplastic are not as well-known as they are for other particles/particulates, like phytoplankton and sedimentary material. Herein we present the results of sinking experiments for a suite of microplastic, covering different shapes (spheres, fibres, irregular), microplastic densities and surface water properties, considering a range of ambient conditions (temperature, salinity and turbulence). The microplastic were compared before and after exposure to weathering processes in the lab and outdoors. The results obtained in this sinking experiment were compared to theoretical expectations, based on literature equations that describe the relationship between the drag coefficient and particle Reynolds number. A key advantage of this relationship is that it is independent on the type of plastic and properties of water, and would therefore apply to both freshwater and saline waters alike, as it would to all types of microplastic.

Air Pollution, Biomonitoring and Human Health (I)

224

Particulate matter in indoor academic environments: chemical composition, sources, infiltration from outdoor

L. Tofful, CNR / Institute of Atmospheric Pollution Research; S. Canepari, Sapienza University of Rome / Chemistry; M. Catrambone, F. Marcovecchio, M. Giusto, CNR / Institute of Atmospheric Pollution Research; S. Pareti, CNR / institute of atmospheric pollution reasearch; T. Sargolini, CNR / Institute of Atmospheric Pollution Research; c. perrino, CNR Institute of Atmospheric Pollution Research / Institute of Atmospheric Pollution Research

We present the results of the first part of an experimental study carried out in an number of academic environments, ranging from small laboratories to very wide classrooms. The study was aimed to evaluate the mass concentration and the chemical composition of indoor atmospheric particulate matter (PM_{10} and $PM_{2.5}$) and its relationship with a number of parameters. These include: concentration and chemical composition of outdoor particles, mixing properties of the lower outdoor atmosphere, volume and floor of the classroom, distance from the street, presence/absence of the students, season. Two type of sampling schedules were applied. The first one differentiates among working days, nights and week-ends during a 6-week winter period and a 4-week summer period (Special Observation Periods – SOPs). This schedule was planned to highlight the differences due to the presence of the students and teachers. The second one (Long-Term Sampling) consists in twelve 1-month PM_{10} samplings carried out by using very-low flow-rate samplers. It was envisaged to obtain a general picture of the effect of the above parameters during a whole calendar year. Both sampling schedules were applied to six indoor and four outdoor sites, all inside or around the same building. At each site and for both schedules the sampling were simultaneously carried out on Teflon, quartz and polycarbonate filters. Teflon filters were used for the determination of the collected mass (by gravimetry), of the elemental content (total content by energy dispersion X-ray fluorescence, bioavailable and residual fractions by inductively coupled plasma mass spectrometry), of anions and cations (by ion chromatography), of elemental and organic carbon (by thermo-optical analysis) and of the bioaerosol content (by propidium iodide staining and epifluorescence microscopy observation). We report here the results obtained during the winter SOP and during the winter part of the Long-Term Sampling. The composition of PM in the indoor environments was dominated by the organic fraction, with a relevant contribution of the bioaerosol, mainly in the coarse fraction. The infiltration of particles from outside constituted a significant source of inorganic species. A vertical gradient was observed for soil components. A relationship of the concentration and composition of indoor PM with the volume of the classroom, height from the ground, presence/absence of the students and distance from the street has been highlighted.

225

Source apportionment of major species and metals in PM2.5 in urban sites under industrial influences in northern France

F. Ledoux, University of Littoral Côte dOpale / Unité de Chimie Environnementale et Interactions sur le Vivant UCEIV EA4492; A. Kfoury, University of Balamand / Department of Environmental Sciences; G. Delmaire, University of Littoral Côte dOpale / Laboratoire Informatique Signal de la Côte dOpale LISIC EA4491; G. Roussel, University of Littoral Côte dOpale / Laboratoire Informatique Signal Image de la Côte dOpale LISIC EA4491; D. Courcot, Université du Littoral Côte dOpale / Unité de Chimie Environnementale et Interactions sur le Vivant UCEIV EA4492

$PM_{2.5}$ have been related to various adverse health effects, mainly due to their ability to penetrate deeply and to convey harmful chemical components inside the body. The North of France is one of the most densely populated area in Europe and is known as an industrialized region especially in the field of metallurgy, organic chemistry, and glassmaking. Furthermore, its strategic position in the heart of Europe means that this area is subject to major transportation activities by road and also by sea. In this context, the objective of this work was to acquire a better knowledge on the exposure level to major species and metals in $PM_{2.5}$ and on the identification of their sources in urban area influenced by particulate emissions from anthropogenic sources. Sampling was performed using Digitel® DA80 high volume samplers between november 2010 and april 2011 in three medium cities located in northern France, Dunkerque (Dk, coastal urban and industrial site), Boulogne-sur-Mer (BL, coastal and urban site) and Saint-Omer (StO, inland urban and industrial site). $PM_{2.5}$ composition was analyzed for major elements, trace elements, water soluble ions and total carbon, respectively.Species concentrations were examined according to different ways including temporal evolution, concentration and pollution roses.. The impact of such sources on major species and metal concentrations in $PM_{2.5}$ was then quantified using a weighted non-negative matrix factorization based receptor model that considers constraints on chemical profiles (CW-NMF). NO_3-, SO_4^{2-}, NH_4^+ and TC were found as the major components of $PM_{2.5}$ (between 93% and 95%) and significant seasonal differences were evidenced. Trace elements (Cr, Zn, Ni, As, Ag, Cd, Cu, Mn, Pb, V, Sn, Rb, Sr, Bi, Ba, Co, Sb and Ti) only correspond to 0.30% to 0.45% of the $PM_{2.5}$ mass according to the sites. The CW-NMF model identified 8 common source profiles at the 3 sites: secondary nitrates, residential and road traffic, secondary sulfates, fresh and aged sea-salts, heavy fuel oil combusion, non-exhaust traffic and crustal. In addition, 4 industrial source profiles were identified in Dk and/or StO : diffuse emissions and sintering stack of an integrated steelworks, electric steelmaking and glassmaking activities. Despite their low contribution to $PM_{2.5}$, such industrial sources were the main contributors of metals at the two sites.

226

Estimating the contribution of deposition in the total exposure to PAH's in order to derive save deposition reference values

J. Bierkens, VITO / Sustainable Health; L. Geerts, M. Van Holderbeke, VITO NV; K. De Brouwere, VITO NV / Health; A. Standaert, VITO; C. Cornelis, VITO / Environmental Risk and Health; T. Fierens, VITO

Partitioning of PAHs between the particulate and the gaseous phases strongly influences their fate and transport in the atmosphere and human exposure. Dry and wet deposition processes are major sources for PAHs in soil and crops. Our aim was to determine the proportion of the overall burden of environmental and dietary exposure to PAHs that is attributable to deposition in order to derive save deposition reference values. To this end, the fate and human exposure was modelled using the MERLIN-Expo, a software tool that allows to model lifetime exposure, integrating exposure through multiple pathways. Model simulations were based on recent yearly average concentrations in air and particulate matter (PM_{10}) in Belgium. The conceptual model implemented, included inhalation, soil and dust ingestion as well as dietary exposure via the consumption of vegetables, meat and dairy products. Toddlers were chosen as the receptor as they are considered a vulnerable group. Dietary exposure to PAHs via crops was simulated using 3 plant uptake model representing leafy vegetables, fruits and grain, respectively. A cattle model taking its inputs from a grass and maize model was used to calculate concentrations in meat and dairy products. Concentrations in fish were modelled as an external fixed food source. The calculated concentrations for all these food categories were matched to Belgian consumption data in order to calculate dietary exposure. Estimation of the contribution of deposition in the overal exposure was done by comparing a baseline exposure scenario based on recent atmospheric measurement data with a scenario where dry and wet deposition fluxes were arbitrarily set to zero. Save deposition reference values were derived by comparing the calculated overall exposure for the baseline scenario with the oral Tolerable Daily Intake (TDI). In case of exceedance, the deposition rates were iteratively adjusted until the calculated exposure equaled the TDI. Simulations were ran for 16 priority EPA - PAHs + benzo(j)fluoranthene. For only 4 PAHs sufficient measurement data are available in the EFSA database to perform a reality check. Of these 4 carcinogenic PAHs, only the calculated exposure to benzo(a)pyrene exceeded the TDI with a factor 4, implying that current deposition rates might be too high. More deposition measurement data for B(a)P are required to confirm these results. *(The authors thank the Flemish Agency for Health and Care for their support)*

227

A bioassay-directed analysis as a biomonitoring tool to assess the endocrine-disrupting potency of indoor air multi-contamination

S. Laborie, EPHE; E. Moreau-Guigon, EPHE, PSL / UMR METIS; F. Alliot, EPHE / UMR Metis; M. Bimbot, Univ. Paris-Sud / UMR ESE; A. Desportes, EPHE / UMR METIS; V. Huteau, Univ. Paris-Sud / UMR ESE; M. Chevreuil, EPHE / UMR METIS 7619; L. Oziol, University of Paris-Sud / UMR CNRS 8079

Air quality is currently assessed by monitoring a few pollutants involved in the development of several human diseases. However, it remains difficult to identify with any certainty the molecule responsible for a given biological effect, owing to human co-exposition to many bioactive micropollutants, which can also interact with each other. In this way, in vitro bioassays might be relevant biomonitoring tools to assess the air quality, as they integrate these “cocktail” effects. Furthermore, the pulmonary exposure to semi-volatiles endocrine-disrupting compounds (EDCs) may cause hormonal disruptions observed in humans, especially indoors where they spend 80 % of their time. By using cellular bioassays, we have previously shown that bioactive EDCs tend to concentrate indoors, especially in the gaseous phase. The concomitant chemical analysis of a wide range

of EDCs led to the same conclusions, except in cold season during which the indoor gaseous phase concentrated fewer target EDCs than in summer along with presenting a higher endocrine-disrupting effect, especially estrogenic. In order to identify the bioactive compound families responsible for this endocrine-disrupting potential, a bioassay-directed analysis was developed and may represent an advanced biomonitoring tool for air quality. The gaseous phase was collected in a Parisian day nursery during cold season (winter 2014) and in sufficient quantities (6 consecutives samplings of 15 days) to implement the entire fractionation process by semi-preparative liquid chromatography: split of the initial organic extract into three fractions, each fractionated into three subfractions. All these samples and the multiple controls were submitted to the chemical analysis (68 target EDCs) and the biological analysis (estrogenicity measured by transactivation cellular assay). After applying various selection criteria for the quantified target EDCs in bioactive fractions (concentration level, intrinsic estrogenicity, lipophilicity), major families found indoors (phthalates, synthetic musks, alkylphenols, parabens) stood out as playing a role in the estrogenicity of the gaseous phase, especially the lead compounds butylbenzylphthalate, galaxolide, butyl-paraben and nonylphenol. The data suggest however the involvement of non-studied EDCs in this biological effect. Our data confirm that bioassays represent suitable biomonitoring tools to assess air quality, whether they are associated with the chemical analysis or not.

228

The Modifying Effects of Ambient Air Pollution on Indoor Air Quality, Impacts on Human Health

H. Rickenbacker, University of Pittsburgh / Civil & Environmental Engineering; W. Collinge, M. Bilec, University of Pittsburgh / Civil and Environmental Engineering

As energy efficient buildings and communities continue to grow, energy retrofits and new designs need to be made in concert with improvements in indoor air quality (IAQ). Although thermal renovations may decrease heat loss and reduce energy expenditures, inversely, envelope improvements (airtightness) can impact infiltration and air exchange rates, and magnify the effects and intensity of internal air pollutants. In this context, a pilot study was conducted to evaluate the human health risk associated with exposure to indoor air pollution in eight buildings ranging from green certified (i.e., LEED Platinum, Living Building) to historic archetypes. The buildings are located in an energy conservation district, which is situated downwind of major industrial point sources. Indoor and ambient measurements of particulate matter ($PM_{2.5}$, PM_{10}), black carbon, ozone (O_3), total volatile organic compounds (TVOC), carbon monoxide, carbon dioxide (CO_2), relative humidity, and formaldehyde (HCHO) were collected on a seasonal basis. The heterogeneity in ventilation type along with the negative effects of deteriorating and aged mechanical systems had on indoor air quality was distinct; however, the counterintuitive findings implied that green and naturally ventilated buildings underperform when compared to some the conventional buildings within our study. The CO_2 sensors used in most green buildings respond to the number of occupants within a space but does not consider ambient concentrations of criteria air pollutants (i.e., $PM_{2.5}$, O_3, NO_2) before increasing outdoor air volume. Natural ventilation systems then supply outdoor air to indoor spaces in the absence of mechanical filtration, and in turn compromise the health and well-being of building occupants. Additionally, green and high-performance buildings are equipped with state-of-the-art HVAC systems that work in tandem with occupancy sensors to optimize energy use when buildings are occupied and minimize energy use when buildings are vacant (overnight). To this end, with our limited sample size, our results show a constant increase and elevated TVOC and HCHO levels overnight. In this given case, the use of occupancy sensors did not allow proper flush-out of indoor environments and interior finishing prior to the building being opened for operations, and as a result, increases exposure over the lifetime of the building.

229

INNOVATIVE AND LOW-COST MONITORING TECHNIQUES FOR EVALUATING THE SPATIAL VARIABILITY OF PM COMPONENTS: VALIDATION AND FIELD APPLICATION

L. Massimi, Sapienza University of Rome / Environmental Biology; c. perrino, CNR Institute of Atmospheric Pollution Research / Institute of Atmospheric Pollution Research; M. Conti, Sapienza University of Rome; S. Canepari, Sapienza University of Rome / Chemistry

A new very-low volume sampler has been developed with the purpose of allowing spatially-resolved determination of atmospheric particulate matter (PM) and of its chemical components. The low-cost, automatic and self-powered device assures long-term (1-2 months) collection of PM on membrane filters, suitable for subsequent chemical analyses. The device has been validated during a 1-year study focused on the concentration of PM_{10} mass, ions, levoglucosane, polycyclic aromatic hydrocarbons (PAH) and elements. It showed very good performance in terms of repeatability of the samplings, which is the essential characteristic to build a reliable network. The samplers have been employed, for the first time, to evaluate the spatial variability of PM_{10} mass concentration and its main chemical components in the area of Terni, a urban/industrial hot-spot sited in an intramountain depression of Central Italy. Lichen transplants have been exposed at the same sites of the samplers in order to evaluate the potential of lichens as biomonitors for PM spatially resolved analyses. The meteorological conditions of Terni basin, which limit the dispersion and enhance the accumulation of atmospheric pollutants, are ideal to test and validate new experimental methods for the acquisition of spatially-resolved data.

The added value of using invertebrate species in ecotoxicology: new insights for environmental risk assessment (I)

230

Transgenerational effects of a parental exposure in the sentinel species Gammarus fossarum

P. Cribiu, ENTPE, IRSTEA LYON; A. Devaux, INRA-CNRS / UMR LEHNA USC INRA IGH ENTPE; K. Abbaci, H. QUEAU, N. Delorme, L. Garnero, Irstea Lyon / UR MALY Laboratoire Ecotoxicologie; S. BONY, INRA - CNRS / UMR LEHNA USC INRA IGH ENTPE; a. chaumot, Irstea / UR MALY Laboratoire Ecotoxicologie

Since the 80s, the development of molecular biomarkers is an important component of ecotoxicology. Unfortunately, field studies that univocally link biomarker responses to fitness impacts and finally to population level are scarce (1). This is mainly due to the discordance in time scales between toxicological and ecological responses. In previous laboratory studies exploring the effects of high contamination levels of single molecules, a relationship has been established between genotoxic impacts in gametes of the sentinel species *Gammarus fossarum*, and impairment in embryo production. However, such a link was not observed after exposure to complex mixtures in the field at more environmentally realistic concentrations (2). Taking advantage of the availability of biomarkers measured in multiple scale in this species, from the molecular level (primary DNA damage, global DNA methylation) to physiological one (feeding rate, molting success, vitellogenesis) and life history traits (growth, fertility, embryonic survival), along with the possibility to conduct rearing culture in the lab (time to puberty about 4 months), the objective of this study was to assess whether biomarker responses recorded in adult gammarids exposed to a chemical stress could be predictive of the fitness of their progeny (*i.e.* transgenerational effects). For this, the consequences of an exposure in the lab of genitors to environmentally relevant concentrations of cadmium were evaluated in F1 and F2 individuals reared in uncontaminated conditions. In complement, a field exposure experiment through *in situ* caging of the adult F0 followed by the assessment of the subsequent effects in F1 and F2 progeny is currently in progress. References (1) Forbes VE, Calow P, Sibly RM. 2008. The extrapolation problem and how population modeling can help. Environmental Toxicology & Chemistry 27:1987-1994. (2) Lacaze E, Geffard O, Goyet D, Bony S, Devaux A. 2011. Linking genotoxic responses in Gammarus fossarum germ cells with reproduction impairment, using the Comet assay. Environmental Research111:626-634

231

Species differences of bioaccumulation, biotransformation and synergistic effects of two fungicides in two aquatic invertebrates

Q. Fu, Eawag, Swiss Federal Institute of Aquatic Science and Technology / Department of Environmental Chemistry; A. Rösch, Eawag / Environmental Chemistry; C. Vignet, Eawag, Swiss Federal Institute of Aquatic Science and Technology / Environmental Toxicology; D. Fedrizzi, Eawag Swiss federal Institute of Aquatic Science and Technology; N. Cedergreen, University of Copenhagen / Department of Plant and Environmental Sciences; J. Hollender, Eawag / Environmental Chemistry

Numerous micropollutants have been detected concurrently in aquatic systems, but little is known about the mixture effects of micropollutants in aquatic organisms and the underlying mechanisms. Azole fungicides are known to act synergistically with other chemicals by inhibiting cytochrome P450 (CYP) catalyzed detoxification and thereby enhance the effect of already toxic substances in different organisms. Two widely used fungicide classes co-occur in surface water are strobilurin and azole fungicides. This study aimed to investigate the species' sensitivity to both fungicide classes and to gain mechanistic insights on potential synergistic effects of azoles on strobilurin fungicides in non-target organisms. Therefore, we determined the toxicokinetics of a strobilurin fungicide azoxystrobin and an azole fungicide prochloraz in two aquatic invertebrate species *Gammarus pulex* and *Hyalella azteca*. Furthermore, we explored median lethal concentrations (LC_{50s}) of azoxystrobin in the presence and absence of prochloraz, the inhibition strength (IC_{50s}) of prochloraz, and its effect on the locomotory behavior of the two species. Bioaccumulation of azoxystrobin were similar in both species with bioaccumulation factors (BAFs) approximately 5 L kg_{ww}^{-1}, while bioaccumulation of prochloraz was different in two species, with BAFs 57 and 110 L kg_{ww}^{-1} in *G. pulex* and *H. azteca*, respectively. Many biotransformation products were found for both fungicides in both species, of which taurine and malonyl conjugates were only identified in *H. azteca*. Most BTPs result from oxidation and conjugation reactions, which occurred at the (*E*)-methyl *β*-methoxyacrylate group of azoxystrobin and imidazole ring of prochloraz. Prochloraz inhibited the CYP-catalyzed biotransformation of azoxystrobin in both species, leading to higher internal azoxystrobin concentrations and suggesting synergistic effects. The half maximal inhibition concentration of prochloraz $IC_{50,\ PRZ,\ AZ}$ was 0.1 and 0.02 µM in *G. pulex* and *H. azteca*, respectively. The LC$_{50s}$ of azoxystrobin alone were 157 and 200 µg L^{-1} in *G. pulex* and *H. Azteca*, respectively. Prochloraz significantly decreased the LC$_{50}$s of azoxystrobin in both species. Video-tracking of the locomotory behavior suggested that prochloraz induced hyperactivity in *G. pulex*, but not in *H. azteca*. Overall, results suggests *H. azteca* comprise more diverse biotransformation reactions and *G. pulex* tended to be more sensitive than *H. azteca* toward prochloraz effects.

232

Use of Gammarus sp. for toxicity testing. A case study with the growth regulator insecticide fenoxycarb.

H. Arambourou, Irstea Lyon / Freshwater system, Ecology and Pollution Research Unit; a. chaumot, Irstea / UR MALY   Laboratoire Ecotoxicologie; N. Delorme, K. Abbaci, Irstea Lyon / UR MALY   Laboratoire Ecotoxicologie; P. NOURY, Irstea Lyon / Ecotoxicologie; R. Tutundjian, Irstea Lyon; E. Vulliet, Institute of Analytical Sciences; G. Daniele, ISA / Biology; C. Barata, CSIC / Environmental Chemistry; I. Fuertes, Institute of Environmental Assessment and Water Research IDAEA CSIC; V. Debat, MNHN / Institute of Systematics, Evolution and Biodiversity

Gammarus sp. (Amphipoda) are widely distributed across European freshwater systems. In the present study, we evaluated the effect of a fenoxycarb exposure on *Gammarus* sp.. More specifically, i) we assessed the sensitivity of the embryo stage, ii) we identified embryogenesis' sensitive period, iii) we evaluated the sensitivity of the reproductive period and iv) we compared the response to fenoxycarb exposure among three European gammarid species. Fenoxycarb is a growth regulator insecticide, analog of the insect juvenile hormone, used for pest management and for veterinary purpose. This study demonstrated that 5 and 50 µg L^{-1} fenoxycarb can alter embryonic development of *G. fossarum*. The gastrulation phase was particularly sensitive. Moreover, exposure to 5 and 50 µg L^{-1} fenoxycarb strongly altered the pre-copulatory behavior in *G. fossarum* and a 50 µg L^{-1} exposure prevented the production of viable embryos. These results highlighted the deleterious effects of the insect growth regulator fenoxycarb on gammarid embryogenesis and reproduction, which could have severe repercussions on population dynamics. The response to the toxic exposure was dependent on the study gammarid species which underlined the importance to consider species with broader phylogenetic representation to better assess insect growth regulator effects.

233

Adaptation of Gammarus pulex to agricultural insecticide contamination in streams

N. Shahid, Helmholtz Centre for Environmental Research UFZ; J.M. Becker, Helmholtz Centre for Environmental Research UFZ / System-Ecotoxicology; M. Krauss, W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; M. Liess, UFZ Center for Environmental Research / System-Ecotoxicology

Exposure to pesticides affects non-target aquatic communities, with substantial consequences on ecosystem services. Adaptation of exposed populations may reduce the effects of pesticides. However, it is not known under which conditions adaptation occurs when only a low toxic pressure from pesticides is present. Here, we show that *Gammarus pulex*, a dominant macroinvertebrate species in many agricultural streams, acquires increased tolerance to pesticides when recolonization from non-contaminated recovery area is low. Populations in the field that were exposed to pesticides at concentrations several orders of magnitude below considerable acute effects showed almost 3-fold higher tolerance to the neonicotinoid insecticide clothianidin (mean EC$_{50}$ 218 µg L^{-1}) compared with non-exposed populations (mean EC$_{50}$ 81 µg L^{-1}). This tolerance of exposed populations increased from 2- to 4-fold with increasing distance to the next recovery site (0 to 10 km). We conclude that the development of tolerance for non-target species may occur at very low concentrations, much below those affecting sensitive test organisms and also lower than those predicted to be safe by governmental risk assessment frameworks.

234

The use of antifouling biocides in a changing world: combined impact of nanoengineering biocides and thermal stress in a coral species

V.L. Ferreira, University of Aveiro / Biology Department   and CESAM; M.D. Pavlaki, University of Aveiro / Department of Biology; M. Monteiro, Aveiro University / Biology; F. Maia, Smallmatek - Small Materials and Technologies, Lda.; R. Martins, Department of Biology, University of Aveiro / Department of Biology and CESAM; J. Tedim, University of Aveiro / Department of Materials and Ceramic Engineering CICECO; A.M. Soares, University of Aveiro / department of Biology & CESAM; R. Calado, University of Aveiro / CESAM Department of Biology; S. Loureiro, Universidade de Aveiro / Biology

The use of antifouling agents to prevent organism's adhesion onto surfaces continues to be used worldwide. While the European Union already authorized the use of DCOIT (4,5-Dichloro-2-octyl-2H-isothiazol-3-one) biocide, data on its toxicity to non-target organisms is still scarce. Given the advances in the encapsulation of biocides in smart-releasing systems, this work aims to access the impacts of a new antifouling approach, DCOIT encapsulated in silica nanocontainers (SiNC@DCOIT), toward the non-target species *Sarcophyton cf. glaucum*, a coral that is also a model of the cnidarian-algae symbiosis found in some marine invertebrates. Elevated seawater temperatures, as predicted by global climate change scenarios, are described as a major cause of corals reef decline. Due to DCOIT photosynthesis inhibition properties, a joint effect of these two stressors (warmer seawater and DCOIT) may occur in the ocean. Toxicity assays were performed by exposing monoclonal coral fragments (n=5) for seven days, at two different temperatures (present day conditions—26ºC—and forecasted scenario for 2100—30.5 °C), to 50 µg DCOIT L^{-1} for free-DCOIT or SiNC@DCOIT and 196 µg SiNC L^{-1} (nanocontainer control). A negative control was added for each temperature. Photosynthetic parameter (F_v/F_m) was measured using a Pulse Amplitude Modulated fluorometer (PAM), with the behavioural endpoint (% polyps open) being scored and the biochemical parameters (both in animal and microalgae fractions) being determined by measuring the activity of catalase (CAT), glutathione-S-transferase (GST) and lipid peroxidation (LPO). Results revealed a decrease on *Fv/Fm* values at 30.5 ºC, when compared to 26 ºC (from 0.65 to 0.60), but only in corals exposed to free-DCOIT the temperature effect was significant throughout the days. By the end of the assay the polyps were mainly open at 26 ºC, whereas at 30.5 ºC they were closed. Regarding enzymatic activities, significant increase on the GST of both animal and microalgae fractions was found in 30.5 ºC groups. On the controls, the raise of 4.5 ºC in water temperature was responsible for a two or three-fold increase in algae and animal GST activity, respectively. The raise on temperature also induced an increment in CAT activity. Regarding LPO, high variability among samples was found. Overall, under thermal stress, the toxicity of DCOIT is enhanced and the negative impacts associated with the use of this biocide will likely be magnified in the warmer oceans.

235

Assessing interspecific variation in Imidacloprid toxicity in earthworms

A. Robinson, Centre for Ecology & Hydrology; E. Lahive, Centre for Ecology and Hydrology; S. Short, P. Kille, Cardiff University; D. Spurgeon, Centre for Ecology & Hydrology

A. robinson[1], (alerob@ceh.ac.uk), S. Short[2], E. Lahive[1] , P. Kille[2],D. SpUrgeon[1] [1] Centre for Ecology and Hydrology, Maclean Building, Benson Lane, Wallingford OX10 8BB, UK [2] School of Biosciences, University of Cardiff, Main Building, Museum Avenue, Cardiff CF10 3AT, UK Widespread interspecific variation is seen in the sensitivity of soil invertebrates to chemical pollutants (e.g. pesticides). Since chemical risk assessments are performed on the basis of tests in a small number of reference species there is a danger of significantly underestimating the effect a toxicant may have in the environment. In order to understand these differences a robust and scientifically based ecotoxicological framework for interspecies ecotoxicological extrapolation is needed. In this study a 30 fold difference in the EC50 reproduction values of imidacloprid was observed across 5 species of earthworm (*Eisenia fetida, Lumbricus rubellus, Dendrobaena octahedra, Apporectodea caliginosa and Amynthas gracilis*) with *A. gracilis* being the most sensitive and *L. rubellus* the least. The role of toxicokinetics in determining interspecific variations in sensitivity is interpreted by assessing the Accumulation, Distribution, Metabolism, and Excretion (ADME) of the chemical into the body and to the neurological tissues that are the common target using radiolabelled compounds and cold chemistry. The contribution of toxicodynamic traits to variations in sensitivity was assessed through genome analysis to identify 1) the number, nature and activity of key receptor genes present, and 2) molecular docking affinities as affected by the amino acid substitutions present in different species receptor homologues. Finally, to assess how these interactions affect the key biochemical and physiological parameters lead to overt toxicity, we use gene expression, biochemistry and life-cycle measurements. By combining these different approaches and identifying key traits, we seek to improve interspecies extrapolation, better predict species vulnerability, and thereby improved the basis for soil species protection during chemical registration.

Solutions for emerging pollutants - Towards a holistic chemical quality status assessment in European freshwater resources (I)

236

Multiple exposure to pesticides and other emerging pollutants – problems and solutions for healthy ecosystems and humans

M. Santen, G. Ungherese, Greenpeace

Industrial pollution is a severe threat to water resources around the world, particularly in the Global South factories release hazardous chemicals that impact our precious water resources - causing long term devastation to human health and the environment. Rivers supply vital resources, including drinking water, crop irrigation, and food. They also serve as a critical support system for industrial activity. In the past decades Greenpeace did several investigations on persistent chemicals like pesticides and industrial chemicals polluting waterbodies. Producing our food within an agricultural system highly dependent on synthetic-chemical pesticides doesn't come without consequences. The impacts of industrial agriculture like Apple and fruit production are widespread, ranging from contaminated soil and water, to impacts on bees and other beneficial insects, as well as on farmers, their families and consumers. Starting in 2011 investigations in the

context of Greenpeace's detox campaign have found a wide range of hazardous substances in the waste waters of textile production or in the effluent of communal wastewater treatment plants (WWTPs) from industrial zones in China, as well as in nearby rivers. Case Studies on per- and polyfluorinated chemicals show that PFAS (perfluorinated alkyl substances) are widespread compounds of environmental concern. Because of their well-recognized hazardous properties, long chain PFASs have been subject to increasing regulation. In 2015 Greenpeace conducted 8 expeditions in remote areas, snow and lake water samples were taken at 10 remote high altitude sites showing that these persistent chemicals are present everywhere on the planet. In 2017 Greenpeace Italy carried out PFASs analysis in wastewaters, analysis revealed PFASs presence in all tested samples of rivers and drinking water collected in schools and public fountain . It is not too late to act – but new rules and responsibilities are required. The use of pollution control or wastewater treatment does not deal effectively with all hazardous substances, and only postpones the need for more effective measures. The problem has to be tackled at its source. The Detox campaign challenges top textile brands to work with their suppliers and eliminate PFAS and all other hazardous chemicals across their entire supply chain, and the entire life-cycle of their products. The growing concern about Europe's massive pesticide use goes hand in hand with an increasing need to search for ecological solutions. To be effective, action needs to be based on knowledge, which requires transparency as a first step, the quantities of hazardous substances used and discharged to be reported and monitored, with full availability of data to the public. [1]
http://gain.fas.usda.gov/Recent%20GAIN%20Publications/Fresh%20Deciduous%20Fruit%20Annual_Vienna_EU-27_10-28-2011.pdf [1]
http://www.greenpeace.org/international/Global/international/publications/toxics/Water%202011/dirty-laundry-12pages.pdf [1]
https://www.greenpeace.de/sites/www.greenpeace.de/files/20121203-Toxic-Threats-China-engl..pdf ^[1] Greenpeace (2015), Footprints in the Snow: http://detox-outdoor.org/assets/uploads/Report%20RAE/RAE_report_08_2015_english_final.pdf ^[1] Greenpeace Italy (2017) Pfas in Veneto: inquinamento sotto controllo? (in italian)
http://www.greenpeace.org/italy/Global/italy/report/2017/Inquinamento/PFAS-in-Veneto.pdf ^[1] Greenpeace Italy (2017) Non ce la beviamo. Presenza di PFAS nell'acqua delle scuole venete (in italian)
http://www.greenpeace.org/italy/Global/italy/report/2017/Inquinamento/Report_Non_ce_la_beviamo.pdf

237

Benefits of international Science & Policy cooperation to promote a paradigm shift in water quality and safety assessment framework

A. Hebert, VEOLIA Environnement Recherche et Innovatio / Environment and Health; S. Rinck-Pfeiffer, Global Water Research Coalition; B. Escher, Helmholtz Centre for Environmental Research GmbH - UFZ / Cell Toxicology; F.D. Leusch, Griffith University / Australian Rivers Institute; P.A. Neale, Griffith University / School of Environment; A. van Wezel, KWR Watercycle Research Institute / Chemical Water Quality and Health; M. Dingemans, KWR Watercycle Research Institute; M. Meeker, Water Environment & Reuse Foundation (WE&RF)

Bioanalytical tools hold great promise as an additional tool of our current water monitoring strategies. In vitro bioassays, which are increasingly being applied in water quality assessment, provide relevant and robust predictive biosystems able to specifically and quantitatively measure early adverse effects of contaminants in water, including providing a measure of mixture effect, even in low doses, where individual components of the mixture alone would not show an effect. They provide comprehensive and high-throughput monitoring systems for a wide range of water contaminants, without the use of experimental animals. Smart combinations of chemical & biological analytics can lead to reduced uncertainty in safety assessments, especially with regards to endocrine disruption, oxidative stress as other relevant primary adverse outcome pathways triggered by environmental mixtures of water micropollutants. Gathering the experts worldwide, recent large scale projects delivered several methodological advances leading to a comprehensive framework including the most promising panel of assays and expanded effect-based trigger values (EBT) for both drinking water and environmental waters (GWRC Endocrine Toolbox II, FP7 DEMEAU, FP7 Solutions, BRAVE initiative). These innovations could contribute to strengthen the safety of conventional water treatment plants and be integrated in future regulations. They also could provide robust monitoring frameworks to promote alternative water schemes as promoted by the Blue Print Initiative in Europe to better safeguard water resources and the WHO Potable Reuse Guidance document. While leading players in Australia, Europe and US recommend to incorporate predictive tools in the water cycle regulatory monitoring (Water Research Australia, US (CA), Canada, RIVM, EAWAG, KWR, UFZ, EU-JRC and EU DG-Env, WHO and GWRC), these bioanalytical tools need to be more comprehensively validated and benchmarked across the entire water cycle and against human and ecological health outcomes before they can be adopted in regulatory frameworks. A critical next step will be to derive further EBT for an expanded scope of bioassay endpoints. Several strategies for the derivation of EBT have been proposed but there remains a lack of acceptance and harmonization across the field to allow better acceptance of these innovative water quality and safety frameworks. Covering a wide range of issues including water quality and quantity management and the management of water-related risks, the OECD is endeavouring to capture science as policy recommendations that derive from its past and recent work on water in a single, consistent and action-oriented policy. By hosting a collaborative task-force or expert working group including GWRC experts and gathering international organizations such as WHO, UNESCO and the OECD we can get to benchmark these new effect-based trigger values, and contribute to the water challenge by targeting Water effect-based guidelines. Complementary tasks could also be taken up by such Science to Policy interface as a supportive action to better explain and disseminate the associated benefits for stakeholders as citizen towards their health protection, municipalities and local authorities, water professionals and institutional bodies.

238

Chemicals of emerging concern (CEC) in the water cycle – a regulatory perspective

M. Helmecke, Umweltbundesamt (UBA)

Environmental authorities increasingly need to address the challenge of contaminants of emerging concerns found in the water cycle. The German Environment Agency has assessed entry paths, critical characteristics of chemicals and the existing legislation to derive potential measures to minimize micro-pollutants in the aquatic environment. A holistic and precautionary approach is needed that combines measures at the source, during the usage of products and chemicals as well as end-of-pipe measures. The EU Water Framework Directive and the Marine Strategy Framework Directive pose a legal frame to achieve good status of all waterbodies and prohibit any further deterioration. Environmental Quality Standards (EQS) are defined and used for the assessment of chemical status. Further provisions are defined in regulations specific to pesticides, biocides, pharmaceuticals and chemicals under REACH. These existing legal provisions need to be continuously developed and supplemented in order to reflect new knowledge and best available technology regarding micro-pollutants. This also includes more holistic approaches for the assessment and monitoring of chemicals. The review of the Water Framework Directive can provide a suitable window of opportunity in this regard as agreed by the European Water Directors in 2016. However, there are challenges regarding the inclusion of new approaches to a regulatory context.

239

Non-target Screening for Holistic Chemical Monitoring and Compound Discovery: Open Science, Real-time and Retrospective Approaches

E. Schymanski, University of Luxembourg / Luxembourg Centre for Systems Biomedicine (LCSB); R. Aalizadeh, National and Kapodistrian University of Athens / Department of Chemistry; N. Alygizakis, Environmental Institute; J. Hollender, Eawag / Environmental Chemistry; M. Krauss, T. Schulze, Helmholtz centre for environmental research - UFZ / Effect-Directed Analysis; J. Slobodnik, Environmental Institute; N.S. Thomaidis, National and Kapodistrian University of Athens / Department of Chemistry; A.J. Williams, US EPA / ORDNCCT

Non-target screening (NTS) with high resolution mass spectrometry (HR-MS) provides opportunities to discover chemicals, their dynamics and effects on the environment far beyond the current 45 "priority pollutants" or even "known" chemicals. Open science and the exchange of information (between for example scientists and regulatory authorities) has a critical role to play in the continuing evolution of NTS. Using a variety of case studies from Europe, this talk will highlight how open science activities such as MassBank.EU (https://massbank.eu), the NORMAN Suspect Exchange (http://www.norman-network.com/?q=node/236) and NORMAN Digital Sample Freezing Platform (http://norman-data.eu) as well as the US EPA CompTox Chemistry Dashboard (https://comptox.epa.gov/dashboard/) can support NTS. Further, it will show how initiatives such as near "real time" monitoring of the River Rhine and retrospective screening via so-called "digital freezing" platforms have opened up new potential for exploring the dynamics and distribution even of as-yet-unidentified chemicals. Collaborative European and international activities facilitate data exchange amongst analytical data scientists and enable quick, effective and reproducible provisional compound identification in digitally archived HR-MS data. This is leading to new ways of assessing and prioritizing the next generation of "emerging pollutants" in the environment, enabling a pro-active approach to environmental assessment unthinkable only a few years ago. *Note: This abstract does not reflect US EPA policy.*

240

Toxicological profiling of water samples with in vitro bioassays and assessment using effect-based trigger values

B. Escher, Helmholtz Centre for Environmental Research GmbH - UFZ / Cell Toxicology; R. Altenburger, UFC Centre for Environmental Research / Department Bioanalytical Ecotoxicology; S. Aït-Aïssa, Institut National de lEnvironnement Industriel et des Risques (INERIS); P.A. Behnisch, Biodetection Systems BV; W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; F. Brion, INERIS / Ecotoxicology Unit; S. Buchinger, Federal Institute of Hydrology / Department G Biochemistry Ecotoxicology; W. Busch, Helmholtz Centre for Environmental Research - UFZ GmbH / Bioanalytical Ecotoxicology; S.E. Crawford, RWTH Aachen University / Institute for Environmental Research,

Dept. of Environmental Analysis; T. Hamers, VU University Amsterdam, Institute for Environmental Studies (IVM) / Department of Environment and Health; K. Hettwer, new diagnostics GmbH; K. Hilscherova, Masaryk University, Faculty of Science, RECETOX / Research Centre for Toxic Compounds in the Environment RECETOX; H. Hollert, RWTH Aachen University / Institute for Environmental Research; R. Kase, Swiss Centre for Applied Ecotoxicology Eawag-EPFL; C. Kienle, Ecotox Centre Eawag-EPFL; J. Legradi, Vrije Universiteit Amsterdam; J. Tuerk, IUTA, Institute of Energy and Environmental Technology; R. van der Oost, Waternet / Onderzoek en Advies; E. Vermeirssen, Ecotox Centre Eawag-EPFL / Aquatic Ecotoxicology; P.A. Neale, Griffith University / School of Environment
In vitro bioassays including cell-based bioassays and low-complexity whole-organism assays have been applied for decades in water quality monitoring. However, there is no common understanding what level or response is acceptable. As of now, bioassay results were only benchmarked against each other but not against an absolute measure of chemical water quality. The EU environmental quality standards (EQS) differentiate between poor and acceptable surface water concentrations for individual chemicals of concern but cannot capture the thousands of chemicals that are in water and their biological action as mixtures. We developed a method that reads across from existing EQS and makes additional mixture considerations to assure that the derived EBT are protective for complex mixtures as they occur in surface water. The EBT derivation method was applied to 48 *in vitro* bioassays with 37 of them having sufficient information to yield preliminary EBTs. 30 of those were considered robust enough to pursue further and for the remainder it is necessary to obtain more experimental data for single chemicals but also to derive more EQS values. To assess the practicability and robustness of the proposed approach, we tested the EBTs numerous case studies from the literature where wastewater treatment plants and surface water were evaluated with bioanalytical tools. In this presentation, we highlight specifically case studies from the EU project SOLUTIONS, where water quality was assessed in large streams (e.g., Danube), hot spots of contamination (e.g., disposal of untreated wastewater into the Danube in Novi Sad) and influence of wastewater treatment plant effluent into small creeks (case study of small Rhine tributaries in Switzerland). In many cases the proposed EBTs were able to differentiate wastewater from surface water and EBTs for different bioassays gave very consistent results indicating the benefit of a common derivation method. Despite the limitations due to limited effect data availability and limitations of the existing lists of EQS, the proposed generic methods to derive EBTs is a first step to harmonise existing approaches and explore various different options of a large diversity of *in vitro* bioassays commonly applied for water quality assessment.
Acknowledgement – This study was a joint effort of the EU project SOLUTIONS (grant 603437) and the workgroup bioassays of the NORMAN network.

241
Chemical gene interactions for associating contaminants with biological effects
A. Schroeder, University of Minnesota-Crookston / Math, Science and Technology; D. Martinovic-Weigelt, University of St. Thomas / Biology; G.T. Ankley, D.L. Villeneuve, U.S. EPA / National Health and Environmental Effects Research Laboratory
Evaluating the potential human health and ecological risks associated with exposures to complex chemical mixtures in the environment is one of the main challenges of chemical safety assessment and environmental protection. There is a need for approaches capable of integrating chemical monitoring and biological effects data to evaluate risks associated with chemicals present in the environment. We will present an approach that uses prior knowledge regarding the biological effects of individual contaminants to predict toxicity of mixtures and prioritize contaminants. More specifically, we use chemical-gene interactions networks to develop knowledge assembly models (KAMs; which is specific to the aquatic system of interest) based on chemical monitoring data and publically available chemical-gene interaction data. When only chemical data are available, KAMs allow for the development of site-specific hypotheses for follow-up biological effects testing. When transcriptomics data are available, KAMs can be used with statistical approaches, such as reverse causal reasoning approaches to prioritize risk and contaminants. Two brief examples using chemical-gene interactions and KAMs will be presented. The first example used chemical monitoring data from the effluent of a local wastewater treatment plant (WWTP) to develop chemical-gene interaction networks. The networks were used to develop hypotheses about the biological effects of the effluent. To test the network predictions, targeted gene expression, using quantitative polymerase chain reaction, was measured from adult male and female fathead minnows that were exposed to the effluent. The second example used prior knowledge about chemical-gene interactions to develop a KAM for detected chemicals at five locations near two WWTPs. Hepatic transcriptome data from fathead minnows exposed to site-water at each location were mapped to the assembly models to evaluate the likelihood of a chemical contributing to the observed biological responses using richness and concordance statistics. The use of chemical-gene interaction networks and KAMs have strong potential for associating chemical occurrence data to biological effects that, when integrated with adverse outcome pathway knowledge, can guide research and/or monitoring efforts related to the effects of contaminants in the environment. The contents of this abstract neither constitute nor necessarily represent official US EPA views and policies.

Anthropogenic and natural sources of environmental contaminants highlight the impacts of opposing and conflicting regulations

242
The triazole story: Clarification of sources, fate and footprint in the environment of the molecule 1,2,4-triazole
M. Blank, Bayer AG Crop Science Division / Environmental Safety; B. Harvey, Syngenta; B. Miles, BASF SE / Crop Protection, Environmental Fate Modelling; D. Liss, SGS Institut Fresenius GmbH / Agro
1H-1,2,4-Triazole (124T) is a key structural component of azole-fungicides, one of the world’s most widely used fungicide classes in agriculture. A crop protection industry taskforce (Triazole Derivative Metabolite Group, TDMG) is jointly addressing scientific and regulatory topics, covering the environmental fate, metabolism, ecotoxicology, toxicology, and risk assessment of 124T. So far in the regulation of crop protection its origin was interpreted as a result of the breakdown of the parent azole-fungicides only. 124T is toxicologically classified as R63. According to the EU Regulation, Directive 1107/2009, it is a «relevant» metabolite in groundwater and subject to a legal maximum concentration of 0.1 µg/l with respect to crop protection uses. Distinct restrictions on agricultural uses of azole-fungicides have been necessary to meet this hazard based limit value. In response to the large number and widespread use in the EU of registered azole products, regulatory authorities are asking for information about 124T’s potential leaching and actual concentrations in groundwater. The TDMG scientists discovered that 124T occurs ubiquitously in the environment, originating also from other anthropogenic sources than azole-fungicides, such as fertilizer additives (to avoid nitrate leaching) and commodity chemicals, and is also naturally formed. To investigate the different sources of 124T and elucidate the pathways of entry and distribution in the environment innovative approaches were needed. The TDMG scientists have therefore expanded their scientific scope into non-agricultural environments and residues in different matrices. The work is supported with a recent terrestrial field dissipation study using a ^{13}C stable isotope labelled azole-fungicide to enable differentiation between the different sources, a forest soil study to assess the background abundance of 124T in non-agricultural soils, and several groundwater monitoring studies. This work has confirmed the wide range of sources of 124T and shown that it is currently not possible to distinguish between their contributions to measured environmental concentrations. Consequently, relying on azole-fungicides as the only source of 124T-concentrations could result in a systematic over-estimation of the environmental exposure risk from pesticide usage. An improved understanding of the sources of the molecule in the environment is a pre-requisite for reliable and justified regulatory conclusions.

243
The triazole story: Assessment of the background abundance of 1H-1,2,4-triazole in selected German forest soils
M. Blank, Bayer AG Crop Science Division / Environmental Safety; H. Borchers, A. Chapple, Bayer Crop Science AG; M. Telscher, Bayer AG Division CropScience/Environmental Fate / Development Environmental Safety; O. Heinemann, Bayer AG Crop Science Division
1H-1,2,4-triazole (124T) is an ubiquitous occurring small molecule which originates from different anthropogenic sources in the environment or from biotic or abiotic degradation of triazole-fungicides. In addition, 124T potentially originates from natural sources like soil microorganisms (Fungi, Actinomycetes). Information about the natural background abundance of 124T in forest top soils of German origin is of importance for the assessment of the entry paths and occurrence levels into the environment by crop protection measures. In a GLP terrestrial field study, duplicate forest top soil samples from ten different locations and different forest types (beech, spruce, pine, oak) in Germany were sampled in 2012/2013 for analytical investigation of the 124T background. For this reason, remote areas without close contacts to agricultural areas were selected. In addition, at two of these ten sites the development of the 124T residue background level was investigated over the period of one year. The background abundance of 124T in the samples ranged from < 1.0 to 1.9 µg/kg in oak forest top soils, from 1.0 to 2.1 µg/kg in pine forest top soils, and from < 1.0 to 1.2 µg/kg in spruce forest top soils. In the selected beech forest top soils the background abundance of 124T was below 1.0 µg/kg. The background abundance of 124T in beech and spruce top soil samples taken from April 2012 to February 2013 showed fluctuations over time. These variations could not be associated to seasonal changes. Single values ranged from < 1.0 to 1.8 µg/kg in the beech top soil samples and from < 1.0 to 2.1 µg/kg in the spruce top soil samples. Overall, a background abundance of 124T could be detected in all forest top soil samples of German origin in areas where anthropogenic 124T sources (e.g. fertilizer additives, metabolites of triazole-fungicides, breakdown product of commodity chemicals) could be excluded. This indicates that the measured 124T residues originate from natural sources in the environment.

244

Challenges of a groundwater monitoring study design for a substance with multiple sources: determining risk for groundwater from 1,2,4-Triazole formed from fungicides used in arable crops in Germany

B. Harvey, Syngenta; M. Blank, Bayer AG Crop Science Division / Environmental Safety; B. Brumhard, Syngenta Agro GmbH / Registration; P. Edwards, Syngenta Ltd.; A. Kaune, Bayer AG Crop Science Division; D. Liss, SGS Institut Fresenius GmbH / Agro; B. Miles, BASF SE / Crop Protection, Environmental Fate Modelling; H. Resseler, Syngenta Agro GmbH; M. Schneider, SGS Institut Fresenius GmbH; H. Staudenmaier, BASF SE / Crop Protection, Environmental Fate

1H-1,2,4-Triazole (124T) is a widely occurring molecule with a number of anthropogenic sources, but also biogenic sources, all of which may result in the presence of the molecule in groundwater. The regulatory framework under which potential risks to environment and health are assessed, including applicable trigger concentrations, depends on the source. As a metabolite of triazole fungicides used in agriculture, 124T is regulated under EU Regulation 1107/2009 and subject to a regulatory trigger concentration of 0.1 µg/L in groundwater. At the same time, 124T is also used in agriculture as a nitrification inhibitor added to mineral fertiliser or slurry, making this a potential additional source contributing to overall soil load. The risk to groundwater from this use is however assessed under other regulations. To address regulatory concerns about the leaching risk for 124T from combinations of triazole fungicides used in agricultural practise, the industry group TDMG has conducted groundwater monitoring studies in Germany. As the aim was to evaluate the risk from triazole fungicides only, the challenge was to design studies to sample groundwater originating in areas with intensive triazole fungicide usage, but where applications of 124T-containing fertiliser and other potential sources could be reasonably ruled out. Existing wells from authorities' or water producer's monitoring networks were sampled in the studies, thus capturing a range of scenarios for leaching risk in real-world agricultural practice. To rule out other anthropogenic sources of 124T, for each potential monitoring well a stepwise screening approach was applied to ensure suitability, with in-depth farmer interviews to document relevant product applications and rule out use of 124T-containing fertilizer in the upstream infiltration area. In total 211 groundwater samples from 31 wells in different regions of Germany were analysed. All samples were < 0.1 µg/L, with 14 samples between 0.05 (=LOQ) and 0.08 µg/L. The results show that even with intensive use of triazole fungicides, the concentrations of 124T in shallow groundwater downstream from treated fields did not exceed the regulatory trigger of 0.1 µg/L. The presented approach is considered to be effective to obtain a realistic picture of groundwater exposure to 124T from triazole fungicides in agricultural practice. Ruling out other sources is challenging, but possible with site screening and engagement with the farmers.

245

Leaching of 1,2,4-triazole through agricultural fields in Denmark

A.E. Rosenbom, N. Badawi, Geological Survey of Denmark and Greenland / Geochemical; P. Olsen, Aarhus University AU / Department of Agroecology - Soil Physics and Hydropedology; S. Marcher, The Danish Environmental Protection Agency; A. Gimsing, The Danish Environmental Protection Agency / Pesticides and Gentechnology

The compound 1,2,4-triazole is a degradation product of many azole-fungicides and growth regulators used in agriculture. Leaching of 1,2,4-triazole from agricultural fields has been evaluated in Denmark in the Danish Pesticide Leaching Assessment Programme (PLAP; www.pesticidvarsling.dk), which comprise five agricultural fields (two sandy and three clayey till fields). The monitoring of 1,2,4-triazole began in 2014 and is still ongoing. 1,2,4-triazole is monitored in groundwater and in 1 m depth in water collected from tile drains and suction-cups. The known applied sources of 1,2,4-triazole in PLAP from 2014 to 2015 are the fungicides tebuconazole, epoxiconazole and prothioconazole, where the latter according to the EFSA conclusion only forms minor amounts of 1,2,4-triazole by degradation in soil. These pesticides together with other triazole-fungicides have been applied to the PLAP fields several times since 1999. The applications of tebuconazole and epoxiconazole have not resulted in unacceptable leaching of the active substances to the groundwater. Monitoring of 1,2,4-triazole in PLAP showed detections in groundwater, and some of the detections exceeded 0.1 µg L-1 (max. 0.26 µg L^{-1}). Due to the high background levels of 1,2,4-triazole before application of these triazole-fungicides, it was not possible to fully relate the detections to the specific application of fungicides, as there may be unknown sources like other triazole-fungicides used before 2014. A general decrease in the concentration of 1,2,4-triazole with depth, however, indicates a surface applied source.

246

The triazole story: Differentiation between different 1,2,4-Triazole sources using a 13C stable isotope labelled azole-fungicide

M. Blank, Bayer AG Crop Science Division / Environmental Safety; H. Borchers, A. Chapple, Bayer Crop Science AG; O. Naeb, SGS Institut Fresenius GmbH; O. Heinemann, Bayer AG Crop Science Division; S. Roulier, ADAMA

1H-1,2,4-triazole (124T) is a small and ubiquitously occurring molecule which originates from different anthropogenic sources (e.g. azole-fungicides, fertilizer additives, pharmaceuticals) but is also naturally formed. The environmental degradation pathway and footprint of 124T is complex, partly still unknown, and very difficult to correlate to individual sources. 124T is toxicologically classified as R63, so according to the EU Regulation, Directive 1107/2009, it is a «relevant» metabolite in groundwater. To investigate the different sources of 124T and elucidate the pathways of entry and distribution in the environment innovative approaches are needed. A GLP terrestrial field dissipation study investigated the dynamic of 124T produced from Tebuconazole (TBZ) in the field and aimed to gain DegT50 data for 124T and TBZ while differentiating between different 124T sources. In the study 13C labelled TBZ (13C-TBZ) was applied to bare soils in six different locations across Europe (Spain, Italy, UK, Germany, Belgium, and Denmark). The use of non-labelled triazole fungicides or N-stabilized fertilizers could be excluded for all sites since 2013. ^{13}C labelling allowed for the differentiation between 124T from TBZ and from other sources. Soil samples were collected at 15 sampling times from 0 to 360 days after application, in triplicates and in additional control plots. The soil specimens were analysed for residues of 13C-TBZ, unlabelled TBZ (12C-TBZ), ^{13}C labelled 124T (13C-124T), and unlabelled 124T (12C-124T). Residues of 13C-TBZ remained in the top-soil segments. There were no detects of 12C-TBZ in any of the investigated samples. 13C-124T as the degradation product of 13C-TBZ could be detected in all six trial sites in varying concentrations. Of special note, 12C-124T was detected in four of the six trials, even though the use of triazole fungicides on the trial plots could be excluded for a minimum of three years before the application. At one trial site, residues of 12C-124T were detected down to a depth of 100 cm in all plots with a maximum concentration of 117.6 g/ha. The data collected in this study confirm that in many cases the origin of 124T findings is not the use of azole fungicides, but other sources. Consequently, relying on azole-fungicides as the only source of 124T-concentrations could result in a systematic over-estimation of the environmental exposure risk from pesticide usage.

247

Overlooked sources of trifluoroacetate in the water cycle - consequences for drinking water supply and regulatory measures

K. Nödler, TZW DVGW-Technologiezentrum Wasser / Analysis and Water Quality department; M. Scheurer, DVGW Water Technology Center / Analysis and Water Quality; F. Freeling, DVGW Water Technology Center; J. Janda, O. Happel, F. Lange, H. Brauch, DVGW Water Technology Center / Analysis and Water Quality

Relevant amounts of trifluoroacetic acid (TFAA) are formed in the atmosphere by photochemical transformation of several refrigerants and subsequently introduced into the aquatic environment by wet deposition. TFAA occurs as trifluoroacetate (TFA) in the aquatic environment and is considered to be persistent and mobile. Both the acid and its potassium salt are manufactured and/or imported in large amounts in the European Union. Furthermore, TFA was identified as a dead-end biodegradation product of several pesticides. During a screening of surface waters in south-west Germany, high concentrations of TFA (up to 140 µg/L in a tributary of the River Rhine) were detected. As a consequence, concentrations of TFA at adjacent bank filtration sites and tap waters were also substantially elevated. The here presented study aims on source identification as well as on the assessment of treatment options (ozonation, chlorination, activated carbon filtration) for contaminated raw waters. Ozonation of model substances and wastewater samples was applied to reveal the TFA-forming potential of individual compounds and the presence of not yet identified TFA-precursors in waters bodies in general. Discharge of industrial wastewater was identified as the source of elevated concentrations of TFA in the Rhine tributary. Extended monitoring demonstrated that this contribution impairs the drinking water supply along the lower River Rhine. Ozonation, activated carbon filtration, and chlorination do not allow for considerable removal of TFA. Monitoring of wastewater treatment plants (WWTP) demonstrated that WWTP also emit TFA. One WWTP showed substantial formation of TFA and ozonation of WWTP-outflows led to inceasing concentrations. Tests on the formation of TFA by ozonation of the selected potential precursors demonstrated highly compound-specific TFA-yields between insignificant and ~40%. Sources and pathways of TFA may still lay hidden. As contaminated water resources cannot be efficiently treated by technical options typically applied in water utilities, this topic needs to be addressed for sustainable raw water management. Small molecules such as TFA may originate from different sources and their individual contributions may be subject of temporal and spatial variability. This must be taken into account with regard to risk assessment as well as for the regulation and authorization of chemicals.

Persistence & Biodegradation Assessment

248

Why biodegradable chemicals persist in the environment? A look at bioavailability

J. Ortega-Calvo, Instituto de Recursos Naturales y Agrobiologia / Agroquimica y Conservacion del Suelo

The biodegradability of a given chemical in the in the environment cannot be assessed properly without considering the chemical´s bioavailability to the degrading microbial populations. This is especially applicable to hydrophobic organic chemicals (HOCs), like polycyclic aromatic hydrocarbons (PAHs). With the aim of providing pathways for implementation into regulatory contexts, this overview contribution will examine the range of techniques and experimental models suitable for the assessment of HOCs biodegradability taking into account state-of-the-art bioavailability science (Environ. Sci. Technol. 49:10255-10264, 2015). During recent years, we have applied these techniques to study the microbial interconnections with bioavailability processes, involving pollutant phase exchange, microbial mobilization and cell attachment to interfaces. We can consider two groups of techniques; 1) Broadly applied methods to estimate the bioavailable contaminants using Tenax or passive sampling, methods also subject of standardization and 2) specific methods suitable to deeply characterize phase exchange with liquid mixtures and ^{14}C-labelled chemicals. Examples and applications of these approaches will be summarized. They include desorption extraction (Environ. Sci. Technol. 45:3019-3026, 2011; Environ. Sci. Technol. 48:10869-10877, 2014), passive sampling and dosing methods (Environ. Toxicol. Chem. 27:1526-1532, 2008; Environ. Pollut. 184:435-442, 2014; Environ. Pollut. 205:378-384, 2015), constant NAPL/water interfacial area method (Environ. Sci. Technol. 45:1074-1081, 2011; Environ. Sci. Technol. 51:11935–11942, 2017), and radiorespirometry and dual ^{14}C/residue analysis (Environ. Pollut. 159:3692-3699, 2011). In spite of these advancements, significant gaps of knowledge exist between bioavailability and biodegradation sciences. Still today, it is difficult to predict bioavailability of HOCs, for example, solely on the basis of basic parameters such as organic matter, black carbon or clay contents of a given soil or sediment, and the physicochemical constants of the chemicals (such as solubility in water, octanol-water or organic-carbon based distribution coefficients). This limitation even remains with improved assessments through determinations of chemical activity and bioaccessibility. This uncertainty not only applies to biodegradability in natural environments, but also to engineered remediation systems.

249

Strategy for ready biodegradability evaluation of poorly water-soluble organic compounds in aqueous media

C. Sweetlove, lOREAL SA / Research and Innovation; J. Chenèble, LOreal Research / Research and Innovation; Y. Barthel, Eurofins Expertises Environnementales / Eurofins Expertises Environnementales; J. Lharidon, LOréal Research & Innovation / Life Sciences Direction; T. Gerald, University of Nantes / Microbiology

The assessment of the environmental impacts of an environmental substance is based on ready biodegradability tests, demonstrating a rapid biodegradation in most environmental media. However, when these tests are applied to poorly water-soluble substances, difficulties are encountered, often related to their limited bioavailability towards the microorganisms inducing increased variability that we have studied. An innovative strategy has therefore been established in order to improve the assessment of the biodegradability in the natural environment of these substances. It has compared 24 methods of improving bioavailability methods (BIM) and initiated the revision of the international standard ISO 10634.

250

Impact of temperature on micropollutants removal in an activated sludge system

P. Meynet, Newcastle University / CEGS; R.J. Davenport, Newcastle University / School of Engineering; K. Fenner, ETH Zürich/Eawag

The investigation of the environmental fate of pollutants is essential for evaluating their ecological impact and human exposure, and is a priority for the European water framework. In particular, the high variability of micropollutants removal efficiency in biological treatment systems calls for a better understanding of how plant performances are affected by operational and environmental parameters, such as temperature fluctuations (e.g. daily and seasonal). Currently, environmental exposure assessment uses Arrhenius-based models to estimate biotransformation rates at different temperatures, despite they neglect potential compositional and functional variation of the microbial community. This work aims to evaluate the validity of such models, by exploring the effect of short-term temperature variation on micropollutant biotransformation in an aerobic sludge community. Laboratory batch reactors were seeded with activated sludge from a Swiss full-scale treatement plant and the biotransformation of 93 target micropollutants (6µg/L) was monitored over time at five different temperatures (4-40°C range). The experimental kinetic parameters were compared to model predictions. The microbial population was also characterised by high-throughtput sequencing to reveal community composition and activity during the biotransformations. Positive correlation of biotransformation rate constants with temperature was found in the 4-20°C range. At higher temperatures, the biotransformation potential decreased or reached a plateau for the majority of the compounds, and just a limited group showed a steady increase till 40°C. Consequently, model estimations could not accurately predict rate constants above 20°C, despite major risk assessment guidelines recommend Arrhenius model predictions in the 0-30°C range. The microbal community also showed significant shift in both composition and activity at higher temperatures, in agreement with the observed decrease in biotransformation potential. Contrarily, for compounds showing an Arrhenius-behaviour over the 4-40°C range, the biotransformation processes may be linked to basic living cell functions, less sensitive to temperature fluctuations. Our study highlights limitations in the applicability of Arrhenius-based models for the estimation of chemicals fate in biological systems, and the need to re-examine model parameters to assure more accurate predictions for potential chemical exposure in events of temperature fluctuations.

251

Findings from an international ring test for an improved marine biodegradation screening test

A. Ott, T. Martin, Newcastle University / School of Engineering; G. Whale, Shell Health / Risk Science Team; J. Snape, AstraZeneca UK Ltd. / AstraZeneca Global Environment; B. Rowles, Cefas Lowestoft Laboratory; R. Davenport, Newcastle University / School of Engineering

A series of standardised biodegradation screening tests (BSTs; e.g. OECD 301, 306) have been developed to measure the relative biodegradability of chemicals. Recently, regulatory emphasis has shifted from measuring biodegradation towards prioritisations on chemical persistence. In their current guise, BSTs are ineffective as screens for persistence. They are prone to high levels of variation and produce a large number of fails, many of which can be considered false negatives, wherby a chemical fails a test not because of its recalcitrance, but rather because the test itself has failed. An ECETOC funded workshop to discuss improvements to marine biodegradation testing was delivered in 2015. During this workshop, methodological improvements to BSTs were discussed, in addition to clarifying guidance on testing and interpretation of results obtained from marine BSTs. Methodologically: (i) increasing bacterial cell concentrations to better represent the bacterial diversity inherent in the sampled environments; and (ii) increasing test durations to investigate extended lag phases observed in marine assessments, were recommended to be validated in a multi-institutional ring test. This presentation will report the findings from an international ring test of an improved marine BST, whereby an improved marine BST comprising inocula concentrated by tangential flow filtration, a modified marine BST comprising seawater and a standard OECD 306 closed bottle test were compared across 13 laboratories in the UK, Norway, Germany, Italy, Canada, USA and Japan. Five test chemicals including a positive reference compound (sodium benzoate), a negative reference compound (pentachlorophenol) and three compounds with variable reported degradation (4-nitrophenol, triethanolamine and hydrolysed polyacrylamide), were used to provide a range of biodegradation potentials by which to validate the new method. Biodegradation data for the five chemicals, in the three test systems used, across the 13 participating laboratories will be presented. The need for clearer guidance on biodegradation testing and interpretation will be discussed, with particular reference to test variability and extended lag phases frequently encountered in marine biodegradation assessments. The role which microbial communities play in chemical biodegradation and the extent to which microbial community analysis can explain inter- and intra-laboratory variation in biodegradation test outcome will also be discussed.

252

Relevance of photolysis for the fate of pendimethalin in deeper water layers - results of a scale-up approach according to OECD TG 309

D. Hennecke, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Ecological chemistry; M. Kruse, Fraunhofer IME, Institute for Molecular Biology and Applied Ecology; J. Hassink, BASF SE / Environmental Fate

OECD TG 309 "Aerobic Mineralisation in Surface Water" is currently used under different regulatory frameworks for the persistence assessment of chemicals in surface water. The test is performed in batch to measure biodegradation at defined conditions. Other processes which might be relevant for the fate of a chemical in water like direct and indirect photolysis are not addressed. Since biodegradation is limited in the OECD 309 study, the consequences are critical for substances which are hydrolytically stable but sensitive to light. Within pesticide regulation direct photolysis studies are mandatory, indirect photolysis studies optional. In natural waters, which have to be used for OECD 309, both processes are relevant for photolytically instable compounds. Hence, beside direct photolysis in the upper layer of a water column, it is interesting to know until which water depth indirect photolysis might contribute to degradation since the light intensity decreases with increasing water depth. A simulation approach has been performed considering the major conditions required in OECD 309 but at a much larger scale. Stainless steel containers of 900 liter volume are filled with surface water taken from a natural lake and maintained at 20°C. The geometry of the container result in a water level of 140 cm and a surface area of 0.70 m^2. In contrast to OECD 309 the system is exposed to simulated sunlight and the water is not mixed by stirring or shaking. Sampling is performed in 5 different water depths using permanently installed steel tubes of different length in order to avoid mechanical mixing of the water body by the sampling procedure. A second container with same test setup but equipped with a lid of stainless steel served as dark control. The test is conducted as both pelagic and suspended-sediment setup. Test substance was ^{14}C-pendimethalin, which is known to degrade rapidly in aqueous systems under the influence of light. The purpose of the test was to determine if photolysis is a relevant process in natural water bodies under OECD 309 test conditions and up to which water depth this can be applied.

253

Poster spotlight: TU267, TU268, TU269

Integrating life cycle approaches towards a sustainable circular economy (I)

254

How can we measure a sustainable circular economy? Unveiling current indicators for the life cycle of products

H. Helander, A. Petit-Boix, S. Leipold, University of Freiburg / Chair of Societal Transition and Circular Economy

Currently, EU policy on circular economy (CE) aims to decrease environmental damage as well as secure the future supply of resources to support economic growth. Even though the implementation of new strategies might cause burden shifting, it is mostly assumed that an increased circularity of resources results in environmental benefits. At the same time, indicators suggested to assess CE progress often fail to provide an assessment of both CE goals and strategies from a sustainability perspective. A life cycle perspective provides a point of departure to address CE strategies, as the stages involved in the circulation of materials are clearly illustrated. Nevertheless, which indicators to assess is still to be defined to support the implementation of CE at any stage of the supply chain. This contribution aims to identify the type of indicators suggested to measure the progress towards a CE at a product level and to evaluate these in relation to the overarching goals and the implementation strategies of CE. To this end, we first define the main CE goals and implementation strategies identified in recent literature and translate these into measurable flows by creating a system model that accounts for each step in the product life cycle. Finally, we review the literature on CE indicators and classify them into CE goals and strategies, life cycle stages and flows addressed, and measurement units (i.e. economic, mass, energy or environmental impact). This contribution provides a consistent framework to compare and assess CE performance indicators at a product level. It also aims to detect shortcomings and underlying assumptions of existing indicators. Through a structural assessment of currently suggested CE performance indicators, we can define gaps and needs in the monitoring process of CE to ensure that progress contributes to the overarching goals of CE and in turn capture trade-offs between implementation strategies.

255

Making sense of circularity indicators with Multi Criteria Decision Analysis

M. Niero, Aalborg University / Department of Chemical and Biochemical Engineering & Department of Management Engineering; P.P. Kalbar, Indian Institute of Technology Bombay / Centre for Urban Science and Engineering (CUSE)

The focus of this study is on packaging, i.e. a sector with high priority for circular economy (CE) implementation, by exploring a situation where a company intends to compare the circularity performances of different products in order to identify which is the best option from a CE perspective. We considered six different packaging alternatives for beer in different contexts and calculated the following indicators to assess product-level circularity: i) the Material Reutilization Score (MRS), included in the Cradle to Cradle® certification program; ii) the Material Circularity Indicator (MCI) developed by the Ellen Mac Arthur Foundation and Granta and iii) the most relevant impact categories according to the Product Environmental Footprint Category Rules for beer product category, i.e. climate change (CC), acidification (Ac), and marine eutrophication (ME). The Multi-Criteria Decision Analysis (MCDA) methodology provides an integration approach to aggregate indicators representing performance of the product system with respect to various aspects such as material recyclability, recycled content and eco-efficiency. We argue that ranking the alternatives based on such approch will ease the identification of the best packaging alternative from a CE perspective. The MCDA is applied to process the indicator scores and subsequently obtain the ranking. Specifically, the compensatory approach based on the MCDA method TOPSIS (Technique for Order of Preference by Similarity to Ideal Solution) is used for ranking the six alternatives The results of the stand-alone application of the two types of indicator sets, i.e. those focusing only on circularity (MRS and MCI) and on life cycle assessment (CC, Ac, ME) is different. The ranking of the packaging is the same within the life cycle assessment indicators, but differs when MCI and MRS are considered. The implementation of the MCDA with different weighting sets shows that two alternatives are dominating i.e. have higher scores for all the indicators. These two alternatives are ranked in the first two positions in all the weighting schemes and hence the ranking is considered as stable. The use of MCDA in combination with several product-circularity indicators is thus recommended to support companies in the identification of the best alternative from a CE perspective.

256

Consistent allocation using archetypes of LCA Goal and Scope definitions

D. Schrijvers, ISM; G. Sonnemann, University of Bordeaux / ISM CyVi

Identifying a suitable allocation procedure is always a challenge in the modelling of the LCI. This is especially the case for metals, that are often mined as co-products and that could be used in multiple life cycles due to efficient recycling. PE International recommend how to apply allocation for the production and recycling of metals [1]. However, by recommending both partitioning and substitution for establishing the data for an attributional LCA, several inconsistencies are introduced. It becomes unclear to what research question LCA results respond if different modelling approaches are combined in a single LCA study. We developed archetypes of LCA Goal and Scope definitions – in the form of research questions – that aid in identifying a suitable and consistent allocation procedure. In order to identify an allocation procedure for an LCA study, several elements of the LCA goal and scope need to be clearly defined: the topic of the LCA, the perspective, the reason to conduct the study, and potential additional functions of the product system that are taken into consideration. We present a framework that shows how the allocation procedure is dependent on the different LCA approaches that are defined in the goal and scope of the LCA. Based on this framework, building blocks are derived that are used to formulate research questions. These research questions represent archetypes of goal and scope definitions. The presented framework shows that there is a relevant difference between system expansion and substitution, and that we must differentiate between process-oriented and product-oriented LCAs, which is not common practice. Furthermore, we show that all types of LCA approaches can be used to support decision-making, which is often only ascribed to consequential LCAs. It is concluded that it is not the *topic* but the *research question* of the LCA study that determines the most suitable allocation procedure. One LCA topic (e.g. 1 kg of recycled aluminium) can already be used for at least 13 different research questions. "What is the impact of 1 kg of recycled aluminium?" is not detailed enough to identify an LCA modelling approach. This paper shows the level of detail that is required to enable this. The importance of a research question is not always acknowledged in LCA practice, but is crucial for unambiguous interpretation and communication of LCA results. [1] PE International, Harmonization of LCA Methodologies for Metals, Ottawa, Canada, 2014.

257

Sustainability assessment of product lifetime extension through increased repair and reuse

E. Bracquene, J. Peeters, J. Duflou, KU Leuven / Department of Mechanical Engineering; W. Dewulf, KU Leuven Association/Group T / Department of Mechanical Engineering

The concept of circular economy is characterized by an economy that aims to keep products, components and materials at their highest utility and value at all times. Based on data collected by a reuse organisation, computers are often considered for self-repair by consumers. In order to increase the number of successful repairs and reduce the required time, current European policy aims to improve the repairability of products. The potential environmental benefits of reuse after repair is investigated by considering a baseline, recycling and reuse scenario. The baseline scenario is considered to be the worst case because it does not consider any recycling or reuse and it assumes all waste is incinerated. A professional use of 3 years is assumed with an annual electricity usage of 76 kWh from the average European grid. For disposal, a specific incineration dataset was calculated based on the assumed laptop composition using the available ecoinvent waste tools. In the recycling scenario the laptops are collected and sent to a recycling plant for material recovery. The production phase of the laptop is modified to reflect a closed loop for recovered materials. For this reason, only a limited number of materials are taken into account. The recycling scenario assumes the optimistic case that all laptops are collected. The pre-processing of the laptop is modelled assuming a manual depollution step followed by mechanical treatment. The end-processing is modelled assuming remelting in an average European electro furnace, aluminium production site, copper smelter and precious metal refining. In the third scenario, a second life of 3 years is assumed for the repaired laptop. The main environmental impact of the repair activity is the replacement of components. It is assumed the laptop requires a new hard disk drive and a new battery. No additional transport is assumed for self-repair. At end of life the laptop is assumed to be recycled, as described above. The case study presented in this paper indicates that repair of laptop's should be considered before discarding for material recycling. The potential benefits of material recycling for high-end or closed loop applications remain limited. If the extended life is at least 2 years, the number of components to be replaced considered in this case study does not impact the descision making. The results also show that increased energy efficiency of new products has a limited impact on the overall results.

258

Building - Rooftop Symbiosis at the next level. Improving urban agriculture through circular economy strategies

M.R. Salís, ICTA-UAB / Institute of Environmental Science and Technology & Department of Chemical, Biological and Environmental Engineering; A. Petit-Boix, University of Freiburg / Chair of Societal Transition and Circular Economy; G. Villalba, X. Gabarrell, Universitat Autònoma de Barcelona / Institute of Environmental Science and Technology & Department of Chemical, Biological and Environmental Engineering

Cities food supply has become an emerging problem with consequences like greenhouse gas emissions or land occupation. In this direction, new ways of producing food within the limits of cities have arisen as potential solutions. Integrated Rooftop Greenhouses (i-RTG) have the potential of a conventional greenhouse for producing vegetables, but they are located at the top of a building, where they can benefit from different residual flows that were previously being wasted. In ICTA-ICP building (4 floors), in the UAB campus (Bellaterra, Barcelona), a rainwater harvesting system stores rainwater on an underground tank, from which water is pumped to the rooftop to irrigate the crops with a hydroponic automatized system. In the third floor, high CO_2 concentrations (up to 820ppm) and more stabilized temperatures (between 15 and 22ºC) are reached. The transport of this air to the rooftop could benefit crop production by performing a CO_2 enrichment and providing more suitable temperatures to the plants. In this aspect, an open chamber made of steel and LDPE will be used to compare its environmental performance and production with a control crop, using life cycle tools. In this direction, previous LCA studies have stated that the fertilizers are one of the items that exerts the greatest impacts in i-RTG systems. Hence, different nutrient cycles could be optimized. In this sense, different literature express that half of the currently economic phosphate resources will have been used up in 50 to 100 years. To optimize P cycle, struvite has been defined as a potential source of this nutrient. With a circular economy perspective, wastewater treatment plant precipitated struvite will be used as the P source for crops in the i-RTG in two different ways: mixing struvite in the nutrient solution and by adding struvite in perlite sacks (hydroponic substrate). The goal of this contribution is thus to evaluate the CO2 and P cycles in an i-RTG through experimental and environmental studies by considering circular economy strategies. With these enhancements, urban agriculture will cut its environmental impacts, making it a more sustainable source of food for cities.

259
Chemical recycling of plastic packaging waste - A life cycle perspective on PET recycling
R. Meys, RWTH Aachen University / Chair of Technical Thermodynamics; S. Westhues, RWTH Aachen Univeristy; J. Klankermayer, RWTH Aachen University / Institute for Technical and Macromolecular Chemistry; A. Bardow, RWTH Aachen University
Plastic packaging waste is one of the priority areas inside the European action plan for the transition to a circular economy. To establish a circular economy, a potential large-scale avenue is chemical recycling of plastic packaging waste. However, preliminary assessments of chemical recycling technologies rate them inferior compared to mechanical recycling not only from an economic but also from an environmental point of view.\nBased on these results, we develop a method based on life cycle assessment to evaluate the environmental performance of chemical recycling to produce chemicals. The method calculates the maximal environmental impact reduction for chemical recycling technologies for future industry setups. To calculate the maximal environmental reduction, a general model of the life cycle of plastic packaging waste is developed from cradle-to-grave. The results are benchmarked against both mechanical recycling and energy recovery for the environmental impact categories climate change and fossil resource depletion. The basis of the calculations are four key system parameters: (i) the efficiency of the waste treatment technology and the environmental impacts of (ii) production of secondary materials, (iii) the waste treatment technology and (iv) the target chemical produced by chemical recycling.\nThe presented method is used to evaluate chemical recycling technologies for polyethylene terephthalate (PET). In this case study, we show the need to produce high value-added chemicals from chemical recycling technologies to possibly achieve an environmental benefit compared to mechanical recycling. To improve on mechanical recycling, chemical recycling needs to yield a chemical that is currently produced with an impact on climate change or fossil resource depletion exceeding 2.54 kg CO2 eq. or 1.58 oil-eq. per kg, respectively. To the best of the authors' knowledge, no chemical in patents or literature meets both of these threshold values. In contrast, chemical recycling is beneficial compared to energy recovery in all studied cases.\nThe presented method enables the easy and early-stage assessment of the maximal environmental reduction of chemical recycling. The case study shows that chemical recycling should target PET waste that is currently used for energy recovery or needs to transform waste from mechanical recycling to high value-added chemicals.

Informed substitution of hazardous chemicals for circular economy: science and practice

260
Substitution of PFOS under the Stockholm Convention
M. Janssen, Nat. Inst. Publ. Health Environ. / Centre for Safety of Substances and Products; S. Posner, Swerea IVF, P.O. Box 104, SE-431 22 Mölndal; K. Ohno-Woodall, Secretariat of the Basel, Rotterdam and Stockholm conventions UNEP, International Environment House, 11-13 Chemin des Anémones, CH 1219 Châtelaine, Geneva
In 2009 PFOS, its salts and PFOSF were added to the UNEP Stockholm Convention aiming at protecting human health and the environment from POPs. However, the addition contained twenty exempted uses, for which PFOS could still be produced and used. These exemptions were accepted, accompanied by a decision to evaluate these uses at regular intervals in order to facilitate a total phase out. Besides the evaluation, the Convention produces Guidance of alternatives to PFOS, which is regularly updated and meant to facilitate the Parties to the Convention in phasing out PFOS. The evaluation was carried out in 2014 under the POPs Review Committee. In 2016 the previous Guidance on alternatives was updated, followed by endorsement in 2017. The update focused on all known applications of PFOS, including the twenty exemptions. However, priority was given to the open applications in two pesticide uses, namely insect baits for control of leaf-cutting ants from *Atta* spp. and *Acromyrmex* spp. and insecticides for control of red imported fire ants and termites. Data on the evaluation and guidance update will be presented. The presentation will focus on the further need of PFOS and the specific uses in various Parties under the convention and possible alternatives among which chemical and non-chemical ones. The recommendations of the POPs Review Committee led to the decision to request the Parties using PFOS for ant baits to deliver data on production and use and monitoring data on emissions at the points of use. It was concluded that in phasing out a substance, it is necessary to understand the functional characteristics of the substance in that specific application and to follow a case by case approach. This enables to find alternatives and to define the proper measures. A multidisciplinaire approach is indispensable in this phase-out process.

261
Experiences of "Substitution in Practice"
C. Jönsson, Swerea IVF AB / Energy and Environment; S. Schellenberger, Stockholm University / Department of Applied Environmental Science (ITM); I. Cousins, Stockholm University / Department of Environmental Science and Analytical Chemistry (ACES); I.v. Veen, Institute for Environmental Studies (IVM) VU University Amsterdam / Chemistry and Biology; S. Roos, Swerea IVF AB / Energy and Environment; H. Holmquist, Chalmers University of Technology; P. Leonards, VU; G.M. Peters, Chalmers University of Technology / Department of Chemistry and Chemical Engineering; S. Posner, Swerea IVF, P.O. Box 104, SE-431 22 Mölndal; A. Hanning, Swerea IVF AB
Within the research project SUPFES (*Substitution of per Fluorinated compounds to Eliminate diffuse Sources*), research on the substitution of hazardous chemicals in consumer products is focused on identifying feasible solutions with better sustainability performance. Such substitution model, suggesting evaluation for both technical as well as environmental and health performance, requires an interdisciplinary approach to create and/or identify feasible alternative solutions. SUPFES SiP model include 1) characterisation of PFAS in use for different selected consumer products 2) initial alternative selection based on matching function criteria and toxicity and exposure assessment and 3) final selection of alternatives undergoing full environmental impact and technical performance assessment for specific scenarios. The SUPFES project has demonstrated an iterative model for practical substitution where in addition to evaluating the environmental and health performance of alternatives, the technical and economical performance are also included. It is clear that distinct substitution strategies will be required for PFASs in different textile products because of the range of performance requirements. Evaluating functionality of the differnt products revealed that it is critical to have a chemical (alternative) product that has the required functionality, but also to measure function in new ways matching the actual requirements. Hazard assessment vs risk assessment reveals challenges in situations were very toxic compounds may be present in an alternative chemical formulation, but the levels are either really low or absent in high quality products. Furthermore, there is clear lack of key information on what is in chemical products and what is released from these products (e.g. do we have polymer degradation leading to toxic degradation products or not). From the environmental and health assessments, the specific products representing scenarios of production and use, it was obvious that there is a need to make a trade of between protecting the user of the garment in certain working environments and high environmental impact. In addition, different environmental impact categories migth give contradictionary decsion support.

262
Implementing a life cycle perspective in chemical alternatives assessment - the case of per- an polyfluoroalkyl substances in textile applications
H. Holmquist, Chalmers University of Technology; S. Roos, Swerea IVF AB / Energy and Environment; S. Schellenberger, Stockholm University / Department of Applied Environmental Science (ITM); C. Jönsson, Swerea IVF AB / Energy and Environment; G.M. Peters, Chalmers University of Technology / Department of Chemistry and Chemical Engineering
Informed chemical substitution is about eliminating chemicals that give rise to unacceptable (eco)toxicological risks, while avoiding problem shifting within a product's or chemical's life cycle, or between types of impacts. For this reason, the life cycle perspective becomes crucial. Chemical alternatives assessment (CAA) has been increasingly in focus in the last years, and life cycle assessment (LCA) and life cycle thinking are part of the more comprehensive CAA methods available. However, more detailed guidance is lacking and few practical examples have been published. A substitution case of current relevance is the phase-out of hazardous per- and polyfluoroalkyl substances (PFAS) from durable water repellent (DWR) textile applications. Alternatives are sought which offer sustained technical performance but an improved environmental and human health profile compared to the hazardous PFAS. To support an informed substitution of hazardous PFAS, and complement our previous hazard assessment, we have conducted an LCA to compare environmental and human health impacts across DWR alternatives on a

functional basis. Based on this case we were also able to further elaborate on the inclusion of the life cycle perspective in a CAA framework by identifying both possibilities and challenges. We conclude that the inclusion of a life cycle perspective in CAA is crucial for an informed and sustainable substitution, as lack of life cycle thinking can lead to problem shifting. We show that LCA, with its focus on function, is a tool that can identify such problem shifting as well as the key chemical properties to be considered. Consideration of (eco)toxicological effects in such an assessment can however turn out to be difficult, especially for substances such as the PFAS if they are outside the domain of the LCIA model. In the case under study here we conclude that the DWR should be selected with three main considerations: (i) the intrinsic hazard properties of the chemistry, selecting the DWR associated with the lowest hazard but, (ii) providing the functionality as needed and, (iii) giving the garment the longest life length.

263

How much function do we need in textiles? Strategies for replacing PFASs based on end-user requirements

S. Schellenberger, Stockholm University / Department of Applied Environmental Science (ITM); P. Hill, University of Leeds / School of Design; O. Levenstam, University of Borås; P. Gillgard, Swerea IVF AB; R. Blackburn, P. Goswami, M. Taylor, University of Leeds; I. Cousins, Stockholm University / Department of Environmental Science and Analytical Chemistry (ACES)

Current approaches to substitute harmful chemicals could benefit from a broader perspective when it comes to the functionality they provide in consumer products[1]. Following the concept of "functional substitution" this study presents an evaluation of material properties of new durable water repellents (DWR) for textiles focusing on end-user requirements. Since the phase out of side-chain fluorinated polymers (SFPs) based on long perfluoroalkyl moieties that were associated with the release of persistent, biaccumulative and toxic perfluoroalkyl acids (PFAAs), a variety of new DWRs have been developed[2] including biodegradable materials that are based on renewable resources.[3,4,5]Due to their unusual properties to provide hydro and oleophobic fibre modifications SFPs based on long perfluoroalkyl chains were historically used on all kinds of different textiles applications. It is so far unclear if alternative DWRs can follow this "one solution will solve all" approach. By segmenting the textile sectors in terms of liquid repellency, this study sets out to outline the different requirements in case studies for functional outdoor clothing and occupational medical apparel. For functional outdoor clothing, a "bottom-up" strategy was chosen by using a survey to assess the consumers' needs and expectations. For occupational medical apparel, exposure scenarios to liquids were defined based on protection needs described by the Center for Disease Control and Prevention (CDC)[6]. Based on these demands, relevant liquids were chosen to evaluate repellent functionality provided by DWRs using established industrial test methods and by developing a new method to determine the roll-off angle for textiles. It has been found that some non-fluorinated DWRs based on green chemistry concepts showed excellent water repellence and also a resistance towards the penetration of liquids with intermediate polarity (e.g. orange juice and synthetic blood). When it comes to liquids with very low surface tension like gastric fluid even fluorinated SFPs had a clearly reduced surface repellency, but were the only materials that protected against liquid penetration. This study of chemical substitution based on chemical and textile functionality as well as end-user requirements pointed out the opportunities and limitations for functional substitution.

264

Analysis of the technical and economic feasibility of alternatives to lead gunshot

A. Mazzolini, D. Mottet, P. Simpson, C. Logtmeijer, C. Rheinberger, M. Blainey, ECHA / Risk Management Implementation Unit

An analysis of the technical and economic feasibility of alternatives to lead gunshot, has been prepared by ECHA as part of a REACH Annex XV Restriction Report on lead and lead compounds used in shot in wetlands. Lead has historically been used in cartridges because of its softness, low melting point, high density, relatively low price and high abundance. Because of these properties, lead is often considered to be the ideal material for use in ammunition. Steel gunshot (soft iron) is by far the most common alternative; others include bismuth and tungsten. The alternatives have a somewhat different ballistic behaviour, but they are still technically and economically suitable alternatives to lead gunshot. However, some adaptation is required by the shooter to use alternatives successfully, including the following: Adaptation of the shot size used as this would typically need to be increased to counter for the lower density of steel Awareness that shotguns are a short-range weapon: the shot material will have a limited influence on the ammunition performance, if fired at targets within a range of 35m Training should be done using shot of the same material as is intended for use in hunting/shooting This suggests that, in assessing hunting/shooting success, the individual skills of a shooter are more decisive than the type of ammunition used. The fact that several countries in the EU have implemented a full ban on the use of lead shot (for example Denmark and Netherlands) is evidence that alternative gunshot is suitable for both hunting activities and sports shooting. Steel shot is the most common alternative to lead gunshot due to its similar price per cartridge, making it the cheapest of the currently available alternatives. Some hunters may need to modify an existing shotgun to enable the use of steel gunshot. However, major gun manufacturers have confirmed that the vast majority of modern shotguns can fire alternative shot materials without any problem. In rare cases, a very old shotgun may need to be replaced or the hunter needs to use the more expensive bismuth or tungsten shot. The analysis of alternatives indicates that the use of alternatives to lead gunshot for hunting and shooting in wetlands is technically and economically feasible. While the availability of such alternative gunshot may currently vary across the EU Member States, it can be expected that a rise in demand triggered by an EU-wide regulatory action will be met on the supply side.

265

The road to successful substitution - case studies

N. Vallotton, N. Ball, Dow Europe GmbH / Toxicology Environmental Research and Consulting; H.M. Hollnagel, Dow Europe GmbH / Toxicology and Environmental Research and Consulting

Delivering innovative products and solutions to the market is a driver for research and development. Drivers of innovation include changes in the market demand or the availability of new technology. New substances or new products are continuously being evaluated for their performance, functionality and safety in a given application. Adaptation to the market demand thus often leads to substitution in the use of one substance for another providing improved functionally. Beyond the evaluation of the product's safety throughout its life-cycle, increased regulatory pressure such as possible changes in hazard classification plays a role in the selection of alternative candidate substances. In a limited number of cases, the evaluation of alternatives is required by regulatory frameworks, such as the authorisation process under the REACH regulation. However, substances of very high concern (SVHC) are identified solely based on their hazard profile (e.g. CMR or PBT properties), and do not consider findings from the overall safety assessment which includes an assessment of risk. Substitution driven by the hazard characterisation alone raises a number of questions in the search for alternative substances. Relevant candidates need to demonstrate equivalent or enhanced performance and functionality in the same range of applications and demonstrate a superior environmental and heath profile based on their hazard properties. However, there is a concern that substitutions based on hazard criteria may lead to regrettable substitutions, for example when it is unclear if a substitution presents a benefit in terms of overall risk to human health and the environment. Comparative risk assessment may prove to be complex as the substances typically do not have the same level of testing information to characterise the hazard. In addition, exposure may occur at greater levels when the uses require higher use rates or if processing and handling lead to higher exposures. Finally, identifying a candidate substance providing a broad range of functionalities allowing substitution in all application may prove to challenging. Case studies will be used to help illustrate the challenges faced by R&D scientists and the need to work closely with experts in disciplines as varied as chemistry, chemical engineering, EH&S specialist and application specialists during the long search for candidate substances having to meet value chain requirements in terms of performance and EH&S profile.

Big data analysis in ecotoxicology: how to get new information out of existing data?

266

EDAPHOBASE - soil biodiversity data warehouse and its applications in ecotoxicology

M. Ross-Nickoll, RWTH Aachen University, Institute for Environmental Research / Institute for Environmental Research; U. Burkhardt, Senckenberg Museum of Natural History Görlitz; J. Hausen, RWTH Aachen University Institute for Environmental Research, Aachen; H. Höfer, Staatliches Museum für Naturkunde Karlsruhe; S. Jänsch, ECT Oekotoxikologie GmbH; S. Lesch, Senckenberg Museum of Natural History (SMNG), Görlitz; J. Oellers, gaiac Research Institute for Ecosystem Analysis and Assessment, Aachen; R. Ottermanns, RWTH Aachen University / Institute for Environmental Research; F. Raub, State Museum of Natural History (SMNK), Karlsruhe; S. Rick, Senckenberg Museum of Natural History (SMNG), Görlitz; J. Römbke, ECT Oekotoxikologie GmbH; B. Scholz-Starke, RWTH Aachen University, Institute for Environmental Research / Institute for Environmental Research; A. Toschki, Research Institute gaiac; D.J. Russell, Senckenberg Museum of Natural History, Görlitz

In this talk we present the soil-zoological information System EDAPHOBASE, a taxonomic-ecological database system, developed within a joined research project funded by the German Federal Ministry of Education and Research (BMBF). It combines existing taxonomical primary data on soil organisms from collections, scientific literature and reports in a data warehouse. Up to now EDAPHOBASE contains more than 500000 observations, about 300000 sites, an 140000 taxa. Data can easily be imported, quality checked, published, queried and analyzed via a web application interface. Detailed analyses can be performed with the interactive web application EDAPHOSTAT which allows species-level analysis as well as definition of reference communities. Future development of EDAPHOBASE towards a pan-European soil-biology data warehouse is presented with the aims of (1) the development of a harmonized tool for the evaluation of ecological soil quality, (2) the collection and usage of existing data from different parties all over

Europe, (3) the provision of a reference base of the ecological quality of soils and (4) the coverage of relevant needs of as many as possible European policies. Finally, necessities for practical use in common agricultural policy, circular economy and for EU transboundary issues are discussed.

267

Diving into REACH database with Rstudio to produce input data for the USEtox model for thousands of chemicals

E. Saouter, EU Commission JRC / Sustainable Assessment UNit; F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Resources Bioeconomy Unit; A. Gissi, European Chemicals Agency; S. Proenca, EU Commission Joint Research; P. Karamertzanis, European Chemicals Agency; J. Provoost, European Chemical Agency ECHA; D. Versteeg, EcoStewardship LLC

In the context of the EU commission product environmental footprint activities (PEF) [1,2], the potential impact of chemicals emitted during the life cycle of a product is assessed via the USEtox multimedia fate model [3]. For each single chemical, this model requires dozens of physico-chemical parameters as well as data on ecotoxicity to freshwater aquatic life and toxicity to human for cancer and non-cancer endpoints. For PEF those data are required for thousands of chemicals using the most up-to-date information [4,5]. The EU commission Joint Research Centre has obtained from the European Chemicals Agency (ECHA) data on more than eight thousand chemicals. These data includes all the physico-chemical properties (166'926 test results),ecotoxicity (305068 test results) and human toxicity data (41'381 test results) available in the IUCLID 5.5 database (as of May 2016). Data were aggregated to calculate unique values for chemical properties and toxicity indicators for thousands of chemicals. The present paper focuses on the use of REACH data to calculate chemical Effect Factors. All the REACH registration data on physico-chemical, aquatic ecotoxicity and human toxicity were exported from the IUCLId 5.5 database into individual Excel files. Each Excel file was imported into the **R-studio** program [6] where data treatments / manipulations / calculations were performed. R allowed us to build code in step wise manner until we obtained the desired selection without impacting the structure of the original file (Excel). The final code can be released to the scientific community to be reapplied on the original files obtained from the REACH database. The ecotoxicity data extracted from the IUCLID database contained about 7500 substances covering both mono and multi-constituents as well as UVCB (Unknown or Variable composition, Complex reaction products or Biological materials). The database covers acute and chronic toxicity tests for various organisms with about 305'068 End-point study reports (ESR)

268

The effect of modelling decisions on macroinvertebrate sensitivity modelling

S.v. Berg, Wageningen University & Research / Aquatic Ecology and Water Quality Management; A. Focks, Alterra Wageningen University and Research Centre / Environmental Risk Assessment Team; C. Rendal, Unilever / Safety and Environmental Assurance Centre SEAC; E. Butler, Unilever; F. De Laender, Université de Namur ASBL / Research Unit in Environmental and Evolutionary Ecology; P. van den Brink, Alterra/Wageningen UR / Aquatic Ecology and Water Quality Management Group b Alterra

Main challenges in modern ecological risk assessment (ERA) lie in the simultaneous occurrence of species diversity and compound multiplicity. The recent development of trait-based sensitivity models has proven to be successful in tackling this problem. However, this methodology is one of the first of its kind, and has therefore to be regarded with caution. In this study, we explore multiple facets of this method in a critical manner. We start by implementing the original method, which will be used as the null method when comparing the different modelling alternatives. As input data, a toxicity, chemical classification, chemical characteristics, and a traits database are used. First, the relative sensitivity of each species to each compound is calculated. Next this relative sensitivity is averaged over all compounds belonging to the same Mode Of Action (MOA) class, resulting in a Mode Specific Sensitivity (MSS) value. Subsequently, exhaustive multiple linear regressions are made between MSS values and species traits, looking for traits which are best in explaining species sensitivity. The next step is to see the effect of different modelling decisions. As a first aspect, the effect of the used model selection criterion is studied. This is done by comparing the cross-validation error resulting from using the adjusted R^2 or the Akaike Information Criterion (AIC). As a second aspect, the effect of the used taxonomic level during species-traits matching is studied by comparing the adjusted R^2 of models resulting from species-traits matching done at family or at genus level. Third, two methods for exploiting species traits data are explored, trying to find out whether using the individual traits or summarising the traits with Principal Component Analysis results in models with a higher R^2. And finally, we take a look at the relationship between the collinearity threshold and the cross-validation error, since we expect that accepting higher collinearity results in worse models. For our dataset we find that i) using AIC as model selection criterion results in models which are better in predicting sensitivity outside the training data, ii) species-traits matching should ideally be done at the genus level, iii) the method chosen for trait predictors depends on the envisioned modelling purpose (mechanistic understanding or improved predictions), and finally, iv) that the collinearity threshold greatly determines the reliability of the resulting models.

269

New approach facing new challenges in Ecotoxicology: D counter

S. Abreu, University of Aveiro / Dep. Biology & CESAM; A.M. Soares, University of Aveiro / department of Biology & CESAM; R.E. Martins, M. Oliveira e Silva, University of Aveiro / DETI IEETA

Routine tests in Ecotoxicology are simple, relatively inexpensive and rapid methods. They can be used to compare the sensitivity of various bioassays to chemical pollutants but data are globally missing for marine organisms. In addition, studies have mainly been focus on only one species under the same exposure, but toxicity exposure involving several species are scarce. D counter is an innovative device that can be used in ecotoxicology assays involving not only one, but also two or more different species, and proving separated data from each of the species coexisting under simultaneous exposure, whenever chromatic differentiation can be achieved among the species. D counter has been mainly pointed to organisms with body sizes from 0.2mm to 3mm, but it also has been tested with larger organisms, and it can be applied either to freshwater, estuarine or marine species, being suitable in bioassays using for example, the brine shrimp *Artemia salina* nauplii (used within48 h of hatching) or neonates (less than 24 h old) from *Daphnia magna* or *D. longispina*, or even *Acartia tonsa* naupli. How it works? - First it is necessary to extract the characteristic signal from sets of organisms belonging to each of the species involved in the study; this is done with a software based pattern identification and recognition procedure using training sets of organisms of each specie, which will provide the tools for pattern recognition in the subsequent bioassays using (together) organisms from different species in the same exposure. Then, simply present the battery of exposures (mixed species or single) to the to the device by just pouring the (tens of) flasks to the serialization component in the D counter device. The dedicated software will procced with pattern recognition and differentiation, providing counting and body length for every single organism under test, either being from different species, presenting sub-totals – by species – and/or total counting when required. The application of these device in bioassays do not alter the requirements of the standardized methods. The device is easier to apply, saves time and it adds accuracy and objectiveness, comparing the sensitivity of various bioassays involving a single or several species, being tested independently or in mixed sets of organisms from more than one species.

270

Ceriodaphnia is equisensitive to Daphnia and should fulfil invertebrate regulatory toxicity requirements

K.A. Connors, S.E. Belanger, The Procter & Gamble Company / Environmental Stewardship and Sustainability Organization

The OECD 202 Acute *Daphnia* Immobilization Toxicity Test requires the use of *Daphnia magna* or another "suitable *Daphnia* species.. (e.g., *Daphnia pulex*)". The zooplankton *Ceriodaphnia dubia* is not considered a standard test species for chemical registration in Europe despite the availability of ISO and USEPA standard acute and chronic test methods and its wide use and acceptance in other countries. Standard acute assays conducted with *C. dubia* submitted to fulfill REACH dossiers can only be used as supporting or weight of evidence studies and not as key studies for the purposes of fulfilling registration requirements. Previous comparative work by Versteeg at al (1997) and data from the USEPA web-ICE tool suggests that *D. magna, D. pulex, and C. dubia* are acutely equisensitive. Here, we employ a big data approach to critically evaluate the comparative species sensitivity between *D. magna* and *D. pulex* and contrast that to the comparitive species sensitivity of *D. magna* and *C. dubia*. Toxicity data were collected from the Ecological Threshold of Toxicological Concern (ecoTTC) project database and Procter & Gamble's internal data. Geometric means were used to summarize information when multiple studies existed for a given species and chemical. Species comparisons were constructed using orthoganol regression. A total of 207 chemicals that had both *D. magna* and *D. pulex* acute toxicity data were identified (5443 studies). This orthoganol regression has a slope of 0.881 and an intercept of 0.484. As both *D. magna* and *D. pulex* are accepted OECD 202 standard test species, this inherent biological difference in sensitivity is accepted under the regulatory guidelines. A total of 193 chemicals that had both *D. magna* and *C. dubia* toxicity data were identified (5465 studies). The orthoganol regression for the acute toxicity studies has a slope of 0.919 and an intercept of 0.599. The relative species sensitivity differences between *D. magna* and *D. pulex* and *D. magna* and *C. dubia* are of the same magnitude and have nearly identical slopes. Given the shared life history, geographic range, and acute/chronic sensitivity, we argue that both *D. magna* and *C. dubia* should be fully accepted for regulatory toxicity requirements regardless of geographic jurisdiction. To further the acceptability of this proposal, it would be useful to develop an OECD test guideline for *Ceriodaphnia* acute and chronic toxicity to supplement existing ISO, ASTM and USEPA standard guidelines.

271

Poster spotlight: TU001, TU002, TU003

Environmental effects of metals: Improvements to risk assessment by considering speciation and bioavailability (II)

environmental realism of laboratory toxicity studies.

272
Assessment and management of stormwater on sediment recontamination due to metal contaminants
I. Drygiannaki, Texas Tech University / Department of Civil Environmental and Construction Engineering; B. Rao, Texas Tech University / Deparment of Civil Environmental and Construction Engineering; M. Rakowska, Texas Tech University / Civil and Environmental Engineering; M. Bejar, Texas Tech University; D. Athanasiou, Texas Tech University / Civil, Environmental, and Construction Engineering; D.D. Reible, Texas Tech University / Civil and Environmental Engineering; G. Burton, University of Michigan / School for Environment and Sustainability; B. Chadwick, Us Navy Spawar Systems Center; G. Rosen, M. Colvin, SPAWAR Systems Center Pacific; R. Pitt, The University of Alabama; E. Strecker, B. Steets, M. Otto, Geosyntec Consultants
There is a lack of understanding on the association of diffuse sources, such as episodic storm events, in the metal recontamination of sediments. The study objective is to define the effect of metals associated with storm events, with regards to the sediment recontamination in the Paleta Creek near Naval Base San Diego (NBSD), in California. Two storm-events were captured for particle size characterization and metal analysis. Receiving and outfall waters were monitored for sediment deposition, contaminant water, and porewater, using auto-samplers which were triggered at each location during two different seasons. Sediments collected in outfalls, deposition traps and sediment deposits were also subjected to chemical analysis. The fractionated water and sediment samples were processed for metal extraction using the modified EPA method 3005A and 3050B, respectively, and were analyzed using ICP-MS and MERX-T. The samples were analyzed for a variety of metals, Cd, Pb, Cu, Ni, and THg, as a function of particle size, to present the percentages of clay, fine silt, coarse silt and sand, which represent particles that could be most directly related to recontamination potential. The results show that over time the contaminant loadings decreased due to reduction in particulate contaminants while the concentrations in finer, and dissolved fractions remained relatively constant. Cd and THg are associated with the largest particles in stormwater, but only Cd is strongly associated with sediment recontamination. Cu, Pb, and Ni are associated with the dissolved phase, fine silts and clay in stormwater and present moderate impact on sediment recontamination. In addition to showing a greater dissolved fraction it appears that the depositing loads are more influenced by resuspension and redistribution of sediment than stormwater. The THg load is relatively small and stormwater recontamination does not add appreciably to sediment THg loads. The particle associations in stormwater along with spatial distribution in sediment traps can identify sources, contributing locations and effective remedial approaches. The implications of the study, can be the development of identification tools that give information about the potential mobility-transport of the metals during storm events, identification of contributing locations, effective remedial approaches, and thus, help to propose best practices for stormwater and sediment management.

273
The effect of percolation and form on lead bioavailability and toxicity to Enchytraeus crypticus
L. Zhang, VU University Amsterdam / Animal Ecology; C. van Gestel, Vrije Universiteit Amsterdam / Ecological Science
In the standard toxicity tests, metals are spiked freshly into test soils as easily soluble metal salts. This may lead to an inaccurate estimation of metal toxity in soil, as it may not mimic the fate of metals in contaminated fields while the counterion could also have a toxic effect on soil organisms. The present study was set up to investigate the bioavailability and toxicity of lead nitrate ($Pb(NO_3)_2$) and lead oxide (PbO) to the potworm *Enchytraeus crypticus* freshly spiked and 18-months aged Lufa 2.2 soil, with and without leaching. Survival and reproduction after 21 d exposure were related to total, 0.01 M $CaCl_2$ extractable and porewater Pb concentrations in the soil and internal Pb concentrations in the surviving animals. For all treatments, 0.01 M $CaCl_2$ extractable and porewater Pb concentrations showed a slight decrease after percolation. $Pb(NO_3)_2$ was more toxic to the enchytraeids than PbO, both for survival and reproduction and in both freshly spiked and aged soils. LC50 for the effect on enchytraeid survival, based on total Pb concentrations in the soils, did not differ for PbO after percolation in freshly spiked soils and aged soils, but increased from 1380 and 500 mg Pb/kg dry soil to 1521and 608 mg Pb/kg dry soil in freshly spiked soils and aged soils, respectively for $Pb(NO_3)_2$. LC50 based on 0.01 M $CaCl_2$ extractable Pb concentrations presented an increase from 2.07 and 1.72 to 2.78 and 2.42 mg Pb/kg dry soil after percolation in freshly spiked soils and aged soils for $Pb(NO_3)_2$ and a slight decreased from 2.79 and 2.45 to 2.16 and 2.18 mg Pb/kg dry soil after percolation in freshly contaminated soils and aged soils for PbO. LC50 values related to internal Pb concentrations did not differ for both $Pb(NO_3)_2$ and PbO, and ranged from 75.6 to 81.1 mg Pb/kg dry body wt in all treatments, indicating that survival of *E. crypticus* was better expained from internal Pb concentrations in the worms than from total or available Pb concentrations in the soil. In general, percolation did not affect total or Pb availability in the soil for $Pb(NO_3)_2$, suggesting that the counterion might have influenced Pb toxicity when Pb salts were used in the standard toxicity tests. Thus, leaching the contaminated soils before testing or using the oxide form of metals might be good ways to get rid of the influence of counterions and increase

274
To leach or not to leach: Soil enzymatic responses to metal mixture species
F. Awuah, University of Saskatchewan / Toxicology Centre; S. Siciliano, University of Saskatchewan / Department of Soil Science; B.A. Hale, University of Guelph / School of Environmental Sciences
Presentation Type: Presentation preferred **Abstract Title:** To leach or not to leach: Soil enzymatic responses to metal mixture species **Authors:** F. K. Awuah[1], B. Hale[2] & S. Siciliano[1]. [1]University of Saskatchewan, Toxicology Center. [2]University of Guelph, School of Environmental Sciences. **Abstract:** In soil laboratory experiments, metal mixture studies are usually carried out with metals dosed as salts, followed by leaching with artificial rainwater to remove excess salts. In the leaching process, metals are lost unevenly, which affects the ratio of the mixtures in the soil. An efficient way of carrying out metal mixture experiments is by using the fixed ratio ray design. This design reduces the amount of experimental effort and allows the estimation of both additivity and interactions. In using this design, metal concentrations should be fixed in specific ratios, but this is compromised when soils are leached. Hence, an alternative method of dosing that allowed fixed ratio testing had to be determined. Two proposed alternatives were metal oxides and spinel minerals which were both abundantly found in aged metal salt spiked soils and field metal contaminated soils. The toxicity of the oxides and minerals to soil enzymes were tested and compared to the salts. The experiment was conducted with three Canadian soils (pH: 3,5,7), three metal species, five fixed metal mixture rays, and five metals (Pb, Cu, Co, Ni, Zn) at one dose. The activity of the soil enzymes ammonia monooxygenases, beta-glucosidases, acid-phosphatases and arylsulphatases were determined colorimetrically. Results showed that leaching alone significantly inhibits the enzyme ammonia monooxygenases in all three soils. The response of acid phosphatases to the metal mixture rays followed known paradigms of bioaccessible concentrations defining toxicity. However, the response of ammonia monooxygenases followed a pH-dependent hormetic toxicity across the three soils. Here, ray toxicity was highest in the soil with a pH value of 5 and vice versa for pH 3 and 7. Generally, metal salts were the most toxic form, and the spinel minerals were the least toxic. Metal oxides were chosen as a replacement for carrying out metal mixture studies in soils because no leaching was required and it was more toxic than the minerals. **Keywords:** Fixed ratio ray, metal oxides, spinel minerals

275
Soil moisture influences the avoidance behaviour of Folsomia candida and Enchytraeus crypticus in metal(loid)-contaminated soils
M. González-Alcaraz, Department of Biology & CESAM - University of Aveiro / Biology & CESAM; C. Malheiro, Department of Biology, University of Aveiro / Biology; D. Nunes Cardoso, CESAM, University of Aveiro / department of Biology & CESAM; S. Loureiro, Universidade de Aveiro / Biology
This study aimed to assess the effects of soil moisture content on the avoidance behaviour of the soil invertebrate species *Folsomia candida* (arthropod) and *Enchytraeus crypticus* (soft-bodied oligochaete) in metal(loid)-contaminated soils. Two metal(loid)-contaminated soils from Central Portugal were selected as test soils (mining soil with pH~5.9; agricultural soil with pH~4.8). Avoidance behaviour was evaluated in two-section vessels for 48 h at 20 °C. Lufa 2.2 soil was used as control soil. Avoidance tests were performed at different soil moisture contents (expressed as soil water holding capacity, WHC): 50% (standard conditions), 75% (to simulate floods) and 25% (to simulate droughts). Different soil moisture content combinations were tested (test soil WHC vs. control soil WHC): 1) 50% vs. 50%, 2) 75% vs. 75%, 3) 25% vs. 25%, 4) 50% vs. 75%, 5) 50% vs. 25%, 6) 75% vs. 50%, and 7) 25% vs. 50%. Porewater metal(loid) concentrations were analysed by ICP-MS in soils incubated at 50%, 75% and 25% WHC for 48 h at 20 °C. Soils incubated at 75% WHC had higher porewater metal(loid) concentrations than those moistened at 50% and 25% WHC. This was more pronounced in the agricultural soil (e.g. 2-50 fold higher Mn, Ni and Pb concentrations at 75% soil WHC). *F. candida* did not avoid both metal(loid)-contaminated soils when tests were performed at the same moisture content in test and control soils while *E. crypticus* did, but only at 50% soil WHC (~66-68% avoidance). When tests were performed at different moisture content in test and control soils the behaviour of both invertebrate species was mainly controlled by soil moisture content. *F. candida* had a preference for soils moistened at 50% WHC, regardless the soils were contaminated or not. *E. crypticus* avoided both metal(loid)-contaminated soils in all the soil moisture combinations tested (~10-100% avoidance), except when the control soil was at drier conditions than the test soils. The present study showed that: 1) porewater metal(loid) availability increased with increasing soil moisture content, especially in soils with higher acidity; 2) *F. candida* and *E. crypticus* differed in their capacity to avoid metal(loid)-contaminated soils; 3) *F. candida* preferred soils moistened at 50% WHC, regardless soils were contaminated or not; 4) *E. crypticus* could avoid metal(loid)-contaminated soils but its capacity was highly dependent on soil moisture conditions.

276
Manganese bioavailability in legacy contaminated soils by medieval

metallurgical wastes
F. Gimbert, H. Gauthier-Manuel, R. Colpaert, D. Radola, F. Choulet, Q. Petitjean, University of Bourgogne FrancheComté / UMR ChronoEnvironnement; A. Walter-Simonnet, University of Bourgogne Franche-Comté, UMR UFC/CNRS 6249; H. Laurent, DRAC Bourgogne Franche-Comté; A. de Vaufleury, University of Franche-Comte / Department of Chrono-Environment
In human history, socio-economic development was strongly influenced by metal resources through mining and metallurgical activities, for instance, of lead (Pb), silver (Ag), or iron (Fe). However, they can lead to a significant environmental contamination through the emission of metal-rich particles and wastes. In the region Burgundy Franche-Comté (eastern France), iron mining and metallurgical activities were dominant over the Middle-Age period, especially in the ancient district of Berthelange. Preliminary analyses highlighted anomalous manganese (Mn) concentrations in soils surrounding medieval slag heaps. Therefore, this study aims at assessing the origin and fate of this Mn using combined physical, chemical and biological tools. For this purpose, we carried out three interdisciplinary and complementary approaches: i) mineralogical characterisation of slags (identification and mapping of their composition by XRD and SEM-EDS); ii) chemical extractions for the assessment of total and available Mn concentrations in soils and iii) environmental bioavailability of Mn using toxicokinetics (28 days) in *Cantareus aspersus* snails exposed to soils from 10 ancient sites of slag deposit (dated from the 5th to the 11th century) or fed with slag fragments incorporated in their diet. We identified olivine (fayalite) as the main Mn carrier in slags where its concentration reaches 4.5 wt.% MnO. With time, slag weathering, as testified by the presence of serpentine, a decay product of fayalite, leads to the release of Mn which accumulates in soils (up to more than 8000 mg.kg^{-1}). Extractible concentrations of Mn from soils (mainly bound to organic matter and under reducible forms) are elevated and may represent a potential toxic exposure to soil invertebrates, raising the question of Mn bioavailability in soils and slag fragments. The modeling of Mn accumulation kinetics in *C. aspersus* snail tissues allowed to show that i) the Mn from medieval smelting wastes is bioavailable to such soil invertebrates and ii) the snail ability to efficiently regulate this element. Nevertheless, when animals are directly exposed to slag fragments incorporated in their diet, the physiological mechanisms of Mn management are rapidly overloaded and internal concentrations increase up to 3000 $mg_{Mn}.kg^{-1}$. Hence, in slags, although often considered as inert materials, Mn turns out to be bioavailable to snails, particularly through their alteration *in situ* (field) and *in vivo* (digestive tract).

277
Chemical and ecotoxicological effects of the use of residues from the pulp and paper industry for the remediation of soils degraded by mining activities
P. Alvarenga, University of Lisboa - Instituto Superior de Agronomia / DCEB; D. Rodrigues, Polytechnic Institute of Beja - Escola Superior Agrária / Tecnologias e Ciências Aplicadas; C. Mourinha, Polytechnic Institute of Beja / Department of Technologies and Applied Sciences; P. Palma, Instituto Politécnico de Beja / Department of Tecnologies and Applied Sciences; N. Cruz, Universidade de Aveiro / CESAM & Departamento de Química; L.A. Tarelho, Universidade de Aveiro / CESAM & Departamento de Ambiente e Ordenamento; S. Rodrigues, Universidade de Aveiro / CESAM & Departamento de Química
Biomass ash and biological sludge, both residues from the pulp and paper industry, in different mixture formulations, with and without the application of mixed municipal solid waste compost (MSWC), were used to improve the quality of soils affected by mining activities (Aljustrel mine, Iberian Pyrite Belt). The experiments comprised an incubation experiment, to allow the selection of the application doses, and a pot experiment, with *Agrostis tenuis* Sibth, to evaluate the possibility of establishing a plant cover in the amended soils. The effects on soil quality were assessed evaluating: (i) soil chemical properties, (ii) plant growth, (iii) immobilization of metals in the soil; (iv) the effects of the amendments on soil eluates toxicity, using organisms from different trophic levels: luminescence inhibition of *Vibrio fischeri*; 24-h mortality test with *Thamnocephalus platyurus*; 72-hours population growth of the green microalgae *Pseudokirchneriella subcapitata*; and *Daphnia magna* acute immobilization test; and (v) soil dehydrogenase activity. Contrary to non-amended control pots, it was possible to establish a plant cover with *A. tenuis* in pots where correctives were applied, but with some variability between replicates. Phytotoxicity was observed in some of the pots, which compromised plant analysis. The improvement in soil quality, in comparison to non-amended soils (controls) was further evidenced by the increase in the activity of dehydrogenase.The amendments were also able to correct soil acidity, and to increase extractable P and K. However, a significant increase in the organic matter, and N content, was only possible by the simultaneous application of MSWC. Total Cd, Cr, Cu and Zn concentrations in soil decreased in the higher application rates of the correctives, due to a dilution effect. In general, amending the soil with biomass ash and biological sludge decreased the toxicity of soil eluates towards the organisms used. The formulation with 30% of biological sludge (applied in 2.5, 5.0 and 10%, w/w, dry matter), presented a better performance, although inducing a slight toxic effect in the microalgae. Concerning the amendment with MSWC, and despite its beneficial chemical effects, toxicological results did not reflect this improvement, since the presence of MSWC did not promote the decrease of toxicity towards the microalgae. Further research is needed with different plants species, since *Agrostis tenuis* showed some phytotoxicological response. \n

Microplastics in freshwater and terrestrial systems - fate, monitoring and biological interactions (II)

278
Profile of microplastics in water and sediments of Antuã river in Portugal
M.O. Rodrigues, Department of Biology & CESAM - University of Aveiro / Department of Biology; F. Gonçalves, University of Aveiro / Department of Biology and CESAM; H. Nogueira, Universidade de Aveiro / Department of Chemistry; J.C. Marques, University of Coimbra / MARE, Dep. of Life Sciences, Coimbra University; N. Abrantes, University of Aveiro / CESAM/DAO; A.M. Gonçalves, MARE, Dep. of Life Sciences, Coimbra University/Biologia Department & CESAM, Aveiro University
The accumulation of plastics in aquatic systems, especially, microplastics (particles with < 5 mm) is of particular apprehension since they can reach high densities, derive from a variety of sources and can interact with biotic and abiotic environment. These micro debris (MPs) differ in their physico-chemical properties (e.g. size, shape, colour, density and polymer type) as well as in their origin (primary or secondary).Notwithstanding, the occurrence of microplastics (MPs) in freshwater systems is less understood than in marine environment. Hence, the present study aims to fill this knowledge gap providing new insights into MPs contamination in Antuã river in to water and sediment samples collected in March and October of 2016 in several stretches of the river. The abundance of MPs reached 14.3 ± 18.3 mg m^{-3} or 306.4 ± 472.1 items m^{-3} in water samples and 35.8 ± 25.7 mg kg^{-1} or 318.9 ± 246 items kg^{-1} in sediments.It shows that this river is severely influenced by MPs, especially in water compartment. Spatial and temporal distributions show different pattern according to seasonal conditions, proximity to urban areas and flow velocity. The water and sediment samples with the greatest abundances were São João da Madeira and Aguincheira, respectively. In water compartment, the highest abundance of MPs was observed in October, while in sediments an opposite pattern was observed. Analysis of plastics by Fourier transform infrared spectroscopy (FTIR) underline polyethylene (PE) and polypropylene (PP) polymers as the most common types covering more than 50% of all polymer types identified. Furthermore, the low medium high oxidation ratio for these particles were 54:38:8% indicating that fewer particles are highly oxidized. Foams and fibers were the most abundant type in São João da Madeira, while fibers and fragments are the most abundant in Aguincheira and Estarreja in water and sediment samples, respectively. Since Portugal is the 12th country in Europe with the highest plastics demand (~1 mt) and 10%-50% of plastic go to landfills, it is urgent to monitor its freshwater systems. This study emphasizes also the importance of rivers as potential carriage systems of MPs within environment. Further studies should be performed to identify point sources in order to mitigate the MP contamination in aquatic systems.

279
Microplastics in German rivers - first monitoring results
M. Hess, LANUV NRW / Water management, water protection; C. Laforsch, University of Bayreuth; P. Diehl, State Environment Agency Rhineland-Palatinate; H. Imhof, University of Bayreuth / Animal Ecology I; M. Loeder, University of Bayreuth; J. Mayer, Hessian Agency for Nature Conservation, Environment and Geology; H. Rahm, North Rhine-Westphalia State Agency for Nature, Environment and Consumer Protection; W. Reifenhäuser, Bavarian Environment Agency, Wielenbach; I. Schrank, University of Bayreuth; J. Stark, State Institute for Environment, Measurements and Nature Conservation Baden-Württemberg; J. Schwaiger, Bavarian Environment Agency / Aquatic Toxicology and Pathology
Plastics are an indispensable component of our daily lives due to their diverse applications. In consequence of improper handling or disposal, plastics can enter surface waters and persist over a long period due to their low degradability. About 4.8 to 12.7 million tonnes of plastic waste are released into the oceans each year. Rivers and wastewater discharges may contribute significantly to the contamination of the marine environment. Despite an obvious causal link between the (micro)plastic load of inland waters and marine ecosystems, European rivers have been investigated for the presence of microplastics (MPs) only recently. However, the analytical results of different studies are usually not comparable among each other due to different methods of sampling, processing and analysis of microplastics. In Germany, five federal states initialised monitoring programmes to get a first overview on the microplastic load of inland water systems: Baden-Wurttemberg, Bavaria, North Rhine-Westphalia, Hesse and Rhineland-Palatinate in cooperation with the University of Bayreuth. Monitoring was synchronised in terms of sampling, processing and chemical analyses. The programme comprised microplastic monitoring in two large river basins (Rhine and Danube), including tributaries of various sizes, thereby covering a wide range of hydrological conditions and anthropogenic influences. A total of 52 measuring points distributed over 25 rivers and streams were examined for MPs near the water surface. MPs were sampled via MantaTrawl and analysed by FTIR spectroscopy. More than 4,000 out of 19,000 particles (> 20 µm) could be clearly identified as plastic particles and were characterized in terms of polymer type, size and shape. To our knowledge, the study of the five federal states in cooperation with the

University of Bayreuth represents one of the most comprehensive measuring programs in fluviatile systems regarding the number of sample sites and the analytical accuracy. Excerpts of the study, focussing on particle number, distribution of size classes, particle shapes and polymer types at the water surface of rivers in western and southern Germany will be presented.

280

Exploring the relation between plastic concentration and river discharge in an urban river

S. Wagner, Helmholtz Centre for Environmental Research GmbH - UFZ / Department of Environmental Geoscience; P.E. Kloeckner, Helmholtz Centre for Environmental Research GmbH - UFZ / Analytical Chemistry; C. Schmidt, Helmholtz Centre for Environmental Research GmbH - UFZ / Hydrogeology; T. Reemtsma, Helmholtz Centre for Environmental Research / Department Analytical Chemistry

Rivers play a major role in transport of plastic debris from inland sources into the marine environment Presently the relevance of various individual sources and emission pathways of plastic in rivers such as wastewater treatment plants, combined sewer overflows, surface runoff and littering can hardly be quantified. Therefore plastic emission from sub-catchments are determined by integral approaches. This study examines plastic particle concentration upstream (P1) and downstream (P2) of an urban subcatchment and establishes relationships between river discharge and plastic concentration (c-Q relationship). Suspended material > 500 µm was sampled at two sampling sites in the Parthe River, (Leipzig, Germany) upstream (P1) and downstream (P2) of an urbanized area under different discharge conditions during 17 campaigns each for 24 h. Plastic material was extracted and quantified in the suspended matter using particle size pre-fractionation, density separation and particle cleanup followed by Raman spectroscopy. Plastic particle mass and number concentration were determined and it was observed that plastic concentration and load increased in the urban subcatchment. To explain the observed concentration and load increase, plastic input in both subcatchments was related to the catchment attributes population, catchment size, urban area, and river length revealing that population determines plastic emissions. The log-log c-Q plots of total plastic mass and particle number concentrations show an enrichment pattern at P2, hence increasing concentration with increasing discharge (positive slope b of the regression). At P1 no significant c-Q relationship was observed. This indicates that in the rural sub-catchment increasing discharge does not drive an increased mobilization of plastic material. The c-Q relationship was applied to estimate the yearly plastic emission based on river discharge data.

281

Microplastic pollution in upstream river catchments

T. Stanton, M. Johnson, P. Nathanail, The University of Nottingham / School of Geography; R.L. Gomes, The University of Nottingham / Faculty of Engineering; W. Macnaughton, The University of Nottingham / School of Biosciences

Microplastic particles (< 5 mm) are known to pollute large lakes and river systems globally, where their presence has been associated with wastewater treatment plants and centres of population and industry. However, the extent to which they pervade upstream catchments is comparatively underrepresented in the study of freshwater microplastic pollution. Results presented here form part of a year-long project that aims to quantify the spatial and temporal variation of microplastic pollution in rural headwaters and urban rivers that do not receive wastewater treatment effluent across England's Midlands, as well as in atmospheric fallout. FTIR analysis of three months' samples identified microplastic particles in 30 litre water samples taken from the headwaters of the River Trent and its tributaries. It has also been used to identify non-fibrous microplastic fragments in rural and (sub)urban atmospheric fallout. Moreover, spherical particles that resemble those used in cosmetic / personal care products have been identified in rivers that do not receive wastewater treatment effluent, some of which have proven not to be polymer-based following FTIR analysis. This brings into question the source, and chemical composition, of spherical particles that have previously been visually identified as plastic spheres likely derived from cosmetic particles. The findings of this study have identified the need for the more extensive consideration of upstream catchments and reaches of rivers that do not receive wastewater treatment effluent in the study of freshwater microplastic pollution. The work conducted here suggests that, though wastewater treatment facilities and large centres of population and industry are suitable predictors of microplastic pollution, the cumulative contribution of headwaters and tributaries are likely to inflate a river's microplastic load.

282

Microplastics in stormwater ponds

F. Liu, Aalborg University / Civil Engineering Department; K.B. Olesen, K.B. Olesen, Aalborg University / Department of Civil Engineering; M. Simon, Aalborg University; N. van Alst, J. Vollertsen, Aalborg University / Civil Engineering Department

Stormwater runoff contains pollutants from land surfaces. As the majority of production and comsumption of synthetic polymers is happened on land, microplastic (MP) is one group of problematic pollutants in urban stormwater runoff. However, MPs in stormwater has barely been investigated. A large part of the urban stormwater runoff is treated in retention ponds prior to discharge. This study looked into the occurrence, composition and concentration of MP in stormwater ponds, aiming to evaluate the retention efficiency of MP by these systems. The results will contribute to the understanding of MP emission from urban areas, and potential impacts on adjacent environmental compartments. Seven stormwater ponds in Denmark were selected as study sites. Surface water was collected using a pumping system equipped with a 10 µm mesh stainless filter. Sediment samples were collected using an Ekman bottom grab sampler. The filters from the water samplings were pre-oxidized by H_2O_2, followed by enzyme treatment. secondary oxidation was applied afterwards. Inorganic particles were separated by density separation using $ZnCl_2$. Sediment samples were freeze dried and incubated in SDS solution. Enzymes were then added, followed by oxidation. Inorganic materials were reduced by density separation using $ZnCl_2$. After extraction, particles (from 10 to 2000 µm) were concentrated in 50% ethanol solution. A sub-sample was deposited onto a ZnSe window and dried. The window was scanned by micro-Fourier Transformed Infrared Spectroscopy imaging applying an Agilent Cary 620/670 system with a 128x128 pixel FPA. The software MPhunter was used to interpret spectrums. MPs were detected in water phase of all ponds. The most abundant polymers were PP, PE and PS. The highest concentration in terms of particle number was 10.8 particle/L, while the other 6 ponds ranged from 0.07 to 2.45 particle/L. For MP mass the highest concentration was 1.2 µg/L while the other 6 ponds ranged from 0.06 to 0.4 µg/L. High density polymers were also detected in some of the ponds, this is likely due to the resuspension of sediment by wind and traps of MPs by organisms. Sediment samples are still under processing and will be included in the final presentation. Nevertheless, the water samples have shown that stormwater pond do not detain all MPs, particularly for low-density polymers. With the high mobility, discharges from stormwater ponds will potentially affect adjacent environmental compartments. \n

283

Towards a more realistic assessment of microplastics as pollutant transporter: a combined experimental and modelling study

S. Seidensticker, C. Zarfl, O. Cirpka, P. Grathwohl, Eberhard Karls Universität Tübingen / Center for Applied Geoscience

Freshwater environments are contaminated with various compounds. In such systems, hydrophobic organic contaminants are often associated with particles such as microplastics (MP) which are ubiquitously detected and have raised concern. The release of pollutants from such particles is a combination of two different diffusive fluxes. External mass transfer governed by diffusion through an aqueous boundary layer on the one hand and internal mass transfer limited by the intraparticle diffusion coefficients on the other hand. Which of these mechanisms controls the kinetics depends on various factors, such as partition coefficients, particle properties, boundary conditions, and time. The aim of this study was to identify if and how observations of pollutant release from MP under laboratory conditions can be transferred to field conditions. We formulated a coupled mass-transfer model to consider both, external and internal mass transfer, and derived an analytical solution via Laplace transformation. For model evaluation, we performed batch experiments with different wastewater contaminants with varying hydrophobicity and at different amounts of dissolved organic matter, which changes the overall partitioning between the MP and the water phase. We measured equilibrium partition coefficients and release kinetics over 240 hours. Based on experimental data and the analytical solution of the model, characteristic times of mass transfer were calculated. These are proxies for the equilibration time and can be used to assess the relative importance of the two mass-transfer processes. Results show, that mass transfer for hydrophilic compounds usually is limited by intraparticle diffusion whereas for hydrophobic compounds it is externally limited. For intermediate compounds, a shift from internal to external dominance was observed. Calculated characteristic times show that under lab conditions the overall equilibration time decreased with increasing partition coefficient while under field conditions the opposite is the case. Thus, a simple first-order approximation of mass transfer would not be enough to transfer experimental results to field conditions adequately. Rather, it is necessary to consider true intraparticle diffusion. Application of our model to different particle sizes, shapes, materials and for varying particle concentrations underlines the fundamental differences between lab and field and allows the transferability between these different boundary conditions.

Air Pollution, Biomonitoring and Human Health (II)

284

Analysis of the contribution of a coal-fired power plant to PM10 concentrations in four sites in Southern Italy

D. Contini, Istituto di Scienze dellAtmosfera e del Clima, CNR / Division of Lecce; D. Cesari, E. Merico, Institute of Atmospheric Sciences and Climate, CNR; F.M. Grasso, A. Dinoi, Institute of Atmospheric Sciences and Climate, CNR / Division of Lecce; A. Genga, M. Siciliano, University of Salento; M. Berico, A. Malaguti, ENEA / SSPT-MET-INAT Via Martiri di Monte Sole 4, 40129 Bologna, Italy

This study is aimed to perform a source apportionment of PM_{10} collected simultaneously in four sites located in the Puglia region (South-Eastern Italy). The sites are located in the area surrounding the “Federico II” coal-fired power plant. The studied area included the territory of the Province of Brindisi, close to the coal-fired power plant “Federico II”, and in the territory of the Province of Lecce at about 26 km SSE of the power plant. The Lecce site was included to assess the impact of the power plant emissions at middle distances. The measuring sites are Lendinuso (LN), Cisternino (CI), Torchiarolo (TR) and Lecce (LE). The Lecce site is the Environmental-Climate Observatory managed by ISAC-CNR, regional station of the Global Atmosphere Watch (GAW-WMO) program. Daily PM_{10} samples were collected at the different sites during measurement campaigns in different seasons (summer, autumn and winter) between 2013 and 2016. Specifically, three measurement campaigns were performed simultanously at the four sites in 2016 and the dataset was enriched with previously available data collected in 2013 and 2015 at the sites in the province of Brindisi (LN, CI, and TR) for a total of 457 daily samples. Collected samples were chemically analysed to determine 19 species: the carbonaceous components (EC and OC); the water soluble ions Cl^-, NO_3^-, SO_4^{2-}, Na^+, NH_4^+, K^+, Mg^{2+}, Ca^{2+}; the elements Al, Si, Ti, V, Mn, Fe, Ni, Cu and Zn. Measured data was used for source apportionment of PM_{10} based on a receptor-oriented model approach that integrates the results obtained using two receptor models (Positive Matrix Factorization – PMF and Chemical Mass Balance - CMB), with those obtained using the CALPUFF dispersion model. This approach allows to estimate the primary contribution of the power plant to PM_{10} and to obtain an estimation of its contribution to secondary sulphate.

285

Air pollution toxicology: is it the right time to leave the bench for the field? A case study integrated approach

M. Gualtieri, ENEA / MET-INAT; F. Costabile, CNR / ISAC - Italian National Research Council, Institute of Atmospheric Science and Climate, Rome, Italy; M. Grollino, ENEA / SSPT-TECS-BIORISC Via Anguillarese, 301, 00123, Rome, Italy; P. Avino, INAIL / Department of Technological Innovations, Via IV Novembre 144, 00187 Rome, Italy; E. Cordelli, G. Raschellà, ENEA / SSPT-TECS-BIORISC Via Anguillarese, 301, 00123, Rome, Italy; A. Malaguti, E. Petralia, ENEA / SSPT-MET-INAT Via Martiri di Monte Sole 4, 40129 Bologna, Italy; T. La Torretta, M. Stracquadanio, ENEA; M. Manigrasso, INAIL / Department of Technological Innovations, Via IV Novembre 144, 00187 Rome, Italy; A. Wiedensohler, Leibniz Institute for Tropospheric Research / Permoserstrasse 15, 04318 Leipzig, Germany; G. Cremona, ENEA; K. Weinhold, Leibniz Institute for Tropospheric Research / Permoserstrasse 15, 04318 Leipzig, Germany; L. Di Liberto, CNR / ISAC - Italian National Research Council, Institute of Atmospheric Science and Climate, Rome, Italy; C. Consales, ENEA / SSPT-TECS-BIORISC Via Anguillarese, 301, 00123, Rome, Italy; M. Berico, ENEA / SSPT-MET-INAT Via Martiri di Monte Sole 4, 40129 Bologna, Italy; M. Aufderheide, CULTEX LABORATORIES GmbH / Feodor-Lynen-Straße 21, 30625 Hannover, Germany; G. Gobbi, CNR / ISAC - Italian National Research Council, Institute of Atmospheric Science and Climate, Rome, Italy; G. Zanini, ENEA / SSPT-MET Via Martiri di Monte Sole 4, 40129 Bologna, Italy

Air pollution (AP) is recognize as the most important environmental issue affecting human health. In Europe AP is responsible for 500.000 premature deaths mainly due to non-communicable diseases and disabilities. The epidemiological associations have already evidenced significant relationship between increases in risk factors for selected human diseases and air pollutants concentration. Finally IARC has classified outdoor air pollution as carcinogenic to humans (Group 1). During the last decades several toxicological studies have investigated the molecular biological mechanism of air pollutant effects specifically particulate on matter (PM). These studies worked mainly with *in vitro* or *in vivo* models exposed to PM samples previously collected on filters, then detached and resuspended in suitable media. This procedure, although extensively applied, has always posed the question about the representativeness of extracted PM in comparison to airborne PM. However, the lack of exposure systems directly working under environmental conditions made this experimental set-up widely accepted. In the last years, also thank to Nanotoxicology studies, innovative exposure modules have been proposed which are able to convey air-dispersed particles on cultured cells. The majority of the application so far reported, however deal with the exposure under laboratory condition of engineered nanoparticles or other molecules of interest. Here we report the results obtained by the exposure of bronchial epithelial cells cultured at the air-liquid-interface (ALI) under environmental condition to environmental pollution by means of an exposure module (CULTEX® RFS module). The data demonstrate that the maximal feasible exposure evaluated for the CULTEX® system is representative of the dosimetry calculated for human exposure. The toxicological evaluation evidenced the absence of cytotoxic effects and absence of significant release of inflammatory release. Ongoing analysis are focused on the differential expression of selected genes of interest. Altogether our results show that the time is arrived to leave the warmth of the laboratory bench and to start toxicological evaluation in field campaign. Although the proposed approach still require an extensive evaluation to assess all the pros and cons we reckon that the toxicological data obtainable under really representative environmental conditions may be more representative to understand the biological processes activated by air pollution.

286

Indoor and Outdoor air contamination by endocrine disruptor pollutants in the North part of France

E. Moreau-Guigon, EPHE, PSL / UMR METIS; P. Desmettres, ATMO Hauts-de-France; F. Alliot, EPHE / UMR Metis; E. Escat, N. Dufour, B. Rocq, ATMO Hauts-de-France; M. Chevreuil, EPHE / UMR METIS 7619

The atmosphere is the main environment with which humans have the most important exchanges. However, human activities have led to air pollution (ozone, particulate matter) but also an air contamination by a broad range of pollutants. To date, few data exist on air contamination by endocrine disruptor compounds (EDCs) in France. With the experience acquiring in Paris region in a previous research, the research team and ATMO Hauts-de-France realised two studies in the North part of France about indoor and outdoor air contamination by EDCs. According to the methodology previously validated, several types of indoor environments (office, house, scholar building and day nursery) and several areas (rural/ forest, urban, industrial) were investigated over 2 years (2015 and 2016-2017). During each season, 7 or 5 sites (indoor and outdoor) were sampling during three successive 2-week periods. The device is composed to a TSP filter system and a cartridge containing XAD resin, connected to a flowmeter and a pump. 70 EDCs were analysed by LC-MS/MS, GC-MS/MS or GC-MS in gaseous and particulate phases separately. Whatever the site, in outdoor air as well as in indoor air, all EDCs were detected and concentrations range from 33 553 to 0.001 ng/m^3. Phthalates, PAHs, musks and alkylphenols are the main compound families. Urban and industrial sites are more concentrated than rural and forest ones. Furthermore, for most pollutants, indoor air is more contaminated than outdoor. The EDCs are mainly in gaseous phase and their concentration depend directly on potential sources of emission, on activities inside the building and urban density. For example, phthalates concentrations are linked to consumer products, building materials, furnishing… PAHs are coming from residential and tertiary heating and from road transport; alkylphenols and musks from detergents. Excepted few specific sites, the EDCs concentrations in air in the North part of France are in the same order than those in Paris region. In conclusion, the air contamination by EDCs is becoming a sanitary concern because French people spend 80% of time in indoor environment and young children, a particularly sensitive population, are the most exposed.

287

Air pollution and health: early biological effects in children exposed to air pollutants and genotoxic effect of PM0.5 in different Italian towns

S. Bonetta, S. Bonetta, University of Torino / Department of Public Health and Pediatrics; M. Moretti, M. Villarini, University of Perugia / Department of Pharmaceutical Sciences; L. Covolo, University of Brescia / Department of Medical and Surgical Specialties, Radiological Sciences and Public Health; A. De Donno, University of Salento / Department of Biological and Environmental Science and Technology; M. Verani, University of Pisa / Department of Biology; T. Schilirò, C. Pignata, E. Carraro, University of Torino / Department of Public Health and Pediatrics; U. Gelatti, University of Brescia / Department of Medical and Surgical Specialties Radiological Sciences and Public Health; M. Study Group, University of Torino, Brescia, Pisa, Perugia and Salento / Dep of Medical and Surgical Specialties Radiological Sciences and Public Health

Children are a high-risk group in terms of the health effects of air pollution, and early exposure during childhood can increase the risk of developing chronic diseases in adulthood. The aim of MAPEC_LIFE (Monitoring Air Pollution Effects on Children for supporting public health policy) project was i) to evaluate the associations between air pollution and biomarkers of early biological effects in children and ii) to propose a model for estimating the global risk of early biological effects due to air pollutants and other factors in children. The study was performed on 6-8-year-old children (n=1,149) living in 5 Italian towns in different seasons. Micronucleus (MN) frequency and DNA damage were investigated in buccal cells of children. Socio-demographic and lifestyle features were collected using a questionnaire. Child exposure to air pollutants was assessed analyzing $PM_{0.5}$ (chemical composition and genotoxicity) and collecting data on air quality. In winter, the 52.7% of children showed at least one MN in cells (0.44 MN/1000 cells). A significant difference was observed among the towns. In spring, MN frequency was lower than in winter (0.22 MN/1000 cells) and the difference between towns disappears. MN frequency resulted associated with benzene, $PM_{2.5}$, ozone, SO_2 and polycyclic aromatic hydrocarbons (PAHs). Season and town influenced MN. Environmental tobacco smoke and high BMI were positively associated with MN frequency, while adherence to Mediterranean diet was negatively associated. The DNA damage in children was higher in spring (179.02 au) than in winter (159.00 au) and significant differences were found among the towns (only in winter). DNA damage was associated with benzene, $PM_{2.5}$, SO_2 and NO_2 (winter) and with ozone (complete data-set). Season and town influence the damage. No association was observed with socio-demographic and lifestyle features. The levels of main pollutants were higher in the Po valley in winter. No genotoxic effect of $PM_{0.5}$ extracts was observed using A549 cells while BEAS-2B cells highlighted a light DNA damage in Torino, Brescia and Pisa (winter). The Ames test confirmed the low level of biological effect and the highest mutagenicity in Torino and Brescia. In conclusion, the assessment of biomarkers of early effect in the population is useful for a complete overview of the impact of air pollution exposure and results obtained can be used to propose some guidance for implementing policies of public health protection.

288

Source apportionment of PM near steel plant by electron microscopy

A. Genga, University of Salento / Dep. of Biological and environmental Sciences and Technology; M. Siciliano, University of Salento; C. Malitesta, T. Siciliano, Università del Salento

Source apportionment based on bulk chemical analysis often uses advanced statistical tools for a detailed source categorization. In contrast, in this study the source apportionment is based on properties of individual particles determined by scanning electron microscopy with energy-dispersed spectrometry (SEM-EDS). The receptor site is located near a steel plant in the Apulia Region, South Italy. A total of 5000 particles were analyzed by SEM–EDS and based on the morphology and chemical composition they have been classified into the following main groups: Alumosilicate particles; Silicium reach particles; Ca-rich particles; Biological particles; Carbonaceous particles; Soot; Kish flakes; Salts of Sodium Chloride (sea salt); Calcium sulfate; Metal particles; Secondary particles; Fe reach particles (Fe mixture and Fe oxides). All particles, which could not be classified into one of these groups, were listed as other particles. The particle groups observed in the present investigation can be assigned to different emission sources. Beside the chemical composition, information on the morphology and mixing state of the particles is helpful for discrimination of industrial emissions originating from high temperature processes from a natural soil component, for both iron-rich particles as well as silicates. In the present study, the following source categories have been distinguished: soil, industrial, secondary, biological, soot, Ca-rich particles, Ca sulfates. In industrial, soil and secondary, particles of different groups are merged. The source apportionment analysis performed with the single particle analysis let to investigate the dimensional and mass distribution of the sources in PM10-2.5, PM2.5-1, PM1 fractions, showing that the antropic sources are mainly present in the fine and ultrafine particles, while the natural sources are characterized by coarse dimension.

289

Oxidized transformation products of polycyclic aromatic hydrocarbons in secondary organic aerosol particles

A. Kramer, Oregon State University / Environmental and Molecular Toxicology; S.L. Massey Simonich, Oregon State University / Department of Environmental and Molecular Toxicology; A. Zelenyuk, Pacific Northwest National Laboratory; K. Suski, Pacific Northwest Laboratories; D. Bell, Pacific Northwest National Laboratory

Long-range atmospheric transport of polycyclic aromatic hydrocarbons (PAHs) in fine particulate matter ($PM_{2.5}$) remains a global health concern as transport models continue to fall short of accurate predictions. To improve modeling accuracy the determination of chemical speciation of PAHs within $PM_{2.5}$ is necessary. Secondary organic aerosol (SOA) particles sorb PAHs during formation and transport them as a large fraction of global atmospheric $PM_{2.5,}$ and the presence of PAH vapor has been demonstrated to increase the mass loading of atmospheric SOA. The oxidation of four PAHs were studied in laboratory generated α-pinene SOA experiments. Dibenzothiophene (DBT), phenanthrene (PHE), pyrene (PYR), and benz(a)anthracene (BaA) were measured along with their oxidation products in freshly formed α-pinene ozonolysis SOA grown in the presence of vapor phase PAH (PSOA). Ratios of oxidized transformation products was measured and changes in those ratios was observed during the aging of the SOA, as well as after exposure to ozone. In freshly formed PSOA, the sum of measured oxidized products was found to be equal to the measured amount of parent compound in all four systems. Characterization of aged particles provides evidence of continuing chemical reactions in PHE and PYR PSOA. DBT and PHE PSOA showed evidence that ozone exposure, performed in a flow-tube reactor, results in further oxidation. Data suggests the environment inside SOA particles are complex and dynamic, and need to be further explored. Implications of the presence of oxidized PAHs in long-range transport modeling will be discussed.

The added value of using invertebrate species in ecotoxicology: new insights for environmental risk assessment (II)

290

The role of the p38-activated protein kinase signaling pathway-mediated autophagy in cadmium-exposed monogonont rotifer Brachious koreanus

H. Kang, C. Jeong, J. Lee, Sungkyunkwan University

Autophagy is a ‘self-eating’ system that regulates the degradation of cellular components and is involved in various biological processes including survival and development. However, despite its crucial role in organisms the regulatory mechanism of autophagy remains largely unclear, particularly in invertebrates. In this study, conserved autophagy in the rotifer *Brachionus koreanus* in response to cadmium (Cd) exposure was verified by measuring acidic vesicle organelles using acridine orange (AO) and neutral red (NR) staining, and by detecting LC3 I/II on Western blot and immunofluorescence. We also demonstrated activation of p38 mitogen-activated protein kinase (MAPK) in response to Cd-induced oxidative stress, leading to the induction of autophagy in *B. koreanus*. This was further verified by analysis of MAPK protein levels and immunofluorescence of LC3 I/II after treatment with reactive oxygen species scavengers and inhibitors specific to MAPKs. We propose a p38 MAPK-mediated regulatory mechanism of autophagy in *B. koreanus* in response to Cd-induced oxidative stress. This study will contribute to a better understanding of autophagic processes in invertebrates and its modulation by environmental stressors.

291

Effects of triclosan (TCS) on antioxidant system and oxidative stress-mediated gene expression in the copepod Tigriopus japonicus

J. Park, J. Lee, Sungkyunkwan University

Triclosan (TCS) is an antimicrobial agent that has been widely dispersed and detected in the marine environment. However, the effects of TCS in marine invertebrates are poorly understood. In this study, the effects of TCS on life cycle parameter (e.g. mortality and fecundity) along with cellular reactive oxygen species (ROS) levels, GSH content, antioxidant enzymatic activities, and mRNA expression levels of oxidative stress-induced defense genes, were analyzed using model marine copepod *Tigriopus japonicus*. The no observed effect concentration (NOEC) and median lethal concentration (LC50) of TCS in the adult stage were determined to be 300μg/L and 437.476μg/L, respectively, while in the nauplius stages the corresponding values were 20μg/L, and 51.76μg/L, respectively. Fecundity was significantly reduced ($P < 0.05$) in response to TCS at 100μg/L. Concentration and time-dependent analysis of ROS, GSH content (%), and antioxidant enzymatic activities (e.g. GST, GPx, and SOD) were significantly increased ($P < 0.05$) in response to TCS exposure. Furthermore, mRNA expression of detoxification (e.g., CYPs) and antioxidant (e.g., glutathione S-transferase-sigma isoforms, Cu/Zn superoxide dismutase, catalase) genes was modulated in response to TCS exposure at different concentrations over a 24 h period. Our results revealed that TCS can reduce fecundity and induce oxidative stress with transcriptional regulation of oxidative stress-induced defense genes along with the activation of the antioxidant system in the copepod *T. japonicus*. Based on our investigation, TCS affects survival through oxidative stress with antioxidant and detoxification defense system in *T. japonicus*. In addition, two *CYP* genes (*CYP3026A3* and *CYP3037A1*) are likely to have a potential role as biomarkers in response to TCS in *T. japonicus*. This study will be helpful for a better understanding of how TCS affects on antioxidant defense and detoxification mechanisms in copepod.

292

The protective role of multixenobiotic resistance (MXR)-mediated ATP-binding cassette (ABC) transporters in biocides-exposed rotifer Brachionus Koreanus

Y. Lee, H. Kang, C. Jeong, J. Lee, Sungkyunkwan University

In aquatic organisms, cellular membranes act as the final physical barrier to xenobiotics, since the membranes are in constant contact with the ambient water column that contains various anthropogenic pollutants. In this respect, the efflux activities of membrane transporters can be considered as the first line of defense to xenobiotic exposure in aquatic organisms. Among the transporters, P-glycoprotein (P-gp) and multidrug resistance-associated protein (MRP) are ATP-binding cassette (ABC) transporters that confer multixenobiotic resistance (MXR) via their efflux activity, which enables a variety of xenobiotics to be expelled from cells. MXR has been proposed as the first line of defense against xenobiotics. In this study, the protective roles of P-gp and MRP in the rotifer Brachionus koreanus were examined in response to four biocides (alachlor, chlorpyrifos, endosulfan, and molinate) using fluorescent substrates and inhibitors specific to P-gp and MRP. The efflux activities of P-gp and MRP in the rotifer B. koreanus were increased by biocide exposure, since the fluorescence intensities of the accumulated P-gp and MRP fluorescent substrates were lower in response to different biocides. Thus, exposure of rotifers to the four biocides resulted in increased P-gp and MRP activity. Moreover, the rotifers became more sensitive to the biocides, with reduced survival and slower population growth rates, when P-gp or MRP was inhibited. These findings suggest that P-gp and MRP are involved in the defense system in response to biocide exposure. Furthermore, the transcriptional levels of the genes encoding P-gp and MRP were examined to uncover the mechanism by which MXR is regulated. Taken together, our results demonstrate a crucial role of the MXR efflux system in the defense response to biocides, thereby providing a better understanding of rotifer defense mechanisms on the molecular level.

293

CRISPR/Cas9 genome editing generates Daphnia magna (loss of function) mutants for TRH and ABCB1 genes. Implications for (eco)toxicological testing.

C. Rivetti, IDAEA CSIC Barcelona / Environmental Chemistry; B. Campos, Unilever R&D / Environmental Chemistry; B. Pina, IDAEA-CSIC / Department of Environmental Chemistry; H. Watanabe, Osaka University / Biotechnology; Y. Kato, Osaka University / Department of Biotechnology; C. Barata, CSIC / Environmental Chemistry

Unravel the toxicological mode of action of action of pollutants to non-target keystone species may allow us to model and predict environmental risks of similar acting chemicals. OMICs technologies approaches developed in model ecotoxicological species have allowed us to determine the mechanisms of action of many chemical contaminants. There is, however, the need for validated physiological studies applying reverse genomic tools. Here we present results on six CRISPR-Cas9 mutated *Daphnia magna* clones: three of them bearing mutations on the tryptophan hydrolase gene (TRH), the rate limiting enzyme of serotonin synthesis, and other three having the transporter protein gene ABCB1 mutated. Bi-allelic *in-del* TRH mutants lack serotonin and have their growth rates impaired. Bi-allelic indel ABCB1mutants had lower transporter activity. Chronic toxicity tests with the selective serotonin reuptake inhibitor fluoxetine indicated that effects of this drug enhancing offspring production was independent of serotonin. Acute toxicity test indicated that the transporter ABCB1 is involved in the detoxification of ivermectin and its total or partial knockout dramatically increased its toxicity. These results provide the first evidence over for the use of reverse genetics in Daphnia to study the mechanisms of action of toxicants opening new avenues of research in a stress physiology perspective. This work was supported by the Spanish Government grant (CTM2014-51985-R.)

294
Assessment of Effects and Recovery of Chaoborus Populations in a Novel-Concept Microcosm Experiment
C. Gamblin, R. Cockroft, AgroChemex Environmental Ltd; W.R. Jenkins, W R Jenkins / Regulatory Affairs Ecotoxicology; S. Norman, RidgewayEco
Larvae of *Chaoborus* spp. (phantom midges) which inhabit water bodies in the agricultural landscape are very sensitive to synthetic pyrethroid insecticides and are known to often be the most sensitive taxon in microcosm studies. *Chaoborus* are holometabolous dipterans and from egg hatch, larvae develop through four aquatic instars before pupation and adult emergence. A concurrent study conducted on the same site (unpublished) elucidated that the species used in the study were multivoltine so *Chaoborus h*ave an almost all-year-round potential for re-colonisation. A new type of microcosm study was conducted to assess the extent and rate of recovery of *Chaoborus* populations in microcosms treated with a synthetic pyrethroid. Novel elements included spatial seperation of treated and control systems by a distance of 100 m and non-invasive monitoring of larvae and pupae. The test material was applied at a single rate on two occasions with a 14-day interval to ten microcosms containing predominantly egg rafts and post-overwintering fourth instar larvae of *Chaoborus obscuripes*. Ten untreated microcosms with similar populations of *Chaoborus* were established upwind of the treated units and these, together with indigenous *Chaoborus*, served as a potential source of adult insects for re-colonisation of the treated units. Three microcosms in each group were covered with insect-proof netting to prevent natural re-colonisation in order to assess the extent of population recovery from within microcosms. The numbers and developmental stages of larvae along with the numbers of pupae and presence and numbers of egg rafts were monitored throughout the study using non-invasive methods, from the week before the first application in May 2017 to the end of August 2017. These assessment methods were successful in enumerating the larvae, pupae and extent of adult emergence over time. The findings from the study show that the early instar larvae of *Chaoborus* in populations were impacted by treatment, but that later instars were able to survive and pupate, and that adults emerge. Recolonisation was relevant to the 'open-field' given that the control microcosms were a substantial distance (100 m) from the treated microcosms. The results show that populations impacted by synthetic pyrethroids are re-established in less than 8 weeks after the first application.

295
Poster spotlight: TU108, TU109, TU110

Solutions for emerging pollutants - Towards a holistic chemical quality status assessment in European freshwater resources (II)

296
Linking chemical pollution and effects – How to identify drivers of toxicity?
W. Brack, M.A. Hashmi, Helmholtz Centre for Environmental Research-UFZ / Effect-Directed Analysis; M. Koenig, Helmholtz Centre for Environmental Research GmbH - UFZ / Cell Toxicology; M. Krauss, Helmholtz centre for environmental research - UFZ / Effect-Directed Analysis; M. Muschket, UFZ-Helmholtz Centre for Environmental Research / Effect-Directed Analysis; m. muz, UFZ - Helmholtz Centre for Environmental Research / Cell Toxicology; T. Schulze, Helmholtz centre for environmental research - UFZ / Effect-Directed Analysis; B. Escher, Helmholtz Centre for Environmental Research GmbH - UFZ / Cell Toxicology; C. di Paola, RWTH Aachen University / Department of Ecosystem Analysis; H. Hollert, RWTH Aachen University / Institute for Environmental Research; A. Tindall, Watchfrog S.A.
European water resources are contaminated with complex mixtures of ten thousands of chemicals among them many non-regulated compounds of emerging concern but also unknown chemicals. Chemical monitoring, however, typically considers only a very small fraction of chemicals focusing on 45 priority substances according to Water Framework Directive (WFD) together with some additional River Basin Specific Pollutants. These chemicals often do not explain effects in toxicity tests and impacts on freshwater communities. Thus, we suggest a consistent tiered approach to identify drivers of toxicity in complex environmental mixtures employing mass balance and multivariate statistical approaches as well as effect-directed analysis. The approach is demonstrated using several case studies in the Rivers Danube, Rhine, Mulde and Holtemme as examples. A specific focus is given on endocrine disruption and mutagenicity. While natural and synthetic steroids were confirmed to play a key role for endocrine disruption, the fluorescent dye Coumarin 47 has been identified as a so far unknown environmental pollutant with great anti-androgenic potency *in vitro* and *in vivo*. In a water body with direct impact of industrial effluents individual aromatic amines probably from dye production could be identified as the drivers of mutagenicity. In contrast, mutagenic effects detectable in the River Rhine receiving multiple contaminations from many sources were driven by mixture effects of industrial and natural compounds with low individual potency but strong synergistic effects when occurring together.

297
Toxic mixtures in time-the sequence makes the poison
R. Ashauer, University of York / Environment
It is generally agreed that "the dose makes the poison" – that chemicals can be toxic or non-toxic depending on their dose. This principle assumes that once a chemical is cleared out of the organism (toxicokinetic recovery), it no longer has any effect. However, it overlooks the other more subtle process of re-establishing homeostasis, toxicodynamic recovery, which can be fast or slow. We tested four combinations of substances and found a clear difference in toxicity when the exposure order of two toxicants was reversed, while maintaining the same dose. When toxicodynamic recovery of the organism was slow relative to the interval between exposures it resulted in carry-over toxicity and so caused this sequence effect. We provide evidence of carry-over toxicity amongst chemicals acting on different targets and when exposure is several days apart. It is therefore not only the dose that makes the poison but also the exposure sequence.

298
How to deal with mixtures of pollutants in water resource management?
R. Altenburger, UFC Centre for Environmental Research / Department Bioanalytical Ecotoxicology; M. Faust, Faust & Backhaus Environmental Consulting; A. Kortenkamp, Brunel University London; W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis
Chemicals in the aquatic environment do not occur in isolation but as mixtures. Their compositions, concentrations and effects are highly dynamic with regard to their temporal and spatial occurrence. Current approaches of the EU water framework directive for assessing chemical and ecological quality do not reflect the emerging challenges. The SOLUTIONS project (Brack et al. 2015, STOTEN 503:22) deconstructed the general challenge into three questions (i) How to identify priority mixtures, (ii) How to identify drivers of mixture risk, (iii) How to measure the impact of mixtures (Altenburger et al. 2015, STOTEN 512:540). Now, there are options for utilising the advanced scientific knowledge for answers by either amending existing regulatory procedures of the EU water framework directive or by establishing novel assessment approaches. Priority mixtures can be conceived as a means to reduce the complexity of all real world situations into simplified archetypical scenarios. This might be achieved through modelling of typical emissions from different source types and by complementary information from chemical suspect pattern analysis. Identifying drivers of mixture risk can be tackled by various approaches all of which rely on a combination of chemical and biological information. Methods range from effect-directed analysis to compound class grouping by effect categories. To determine the impact of mixtures, multiple lines of evidence are emerging. They comprise of translating contamination information into expected adverse effect, effect-based monitoring using panel of bioassays and utilising trait-based parameters for analysing ecological monitoring data. In conjunction, they can be used to strengthen causal links between chemical and ecological status assessment. All these approaches were exemplarily tested within the SOLUTIONS project and should pave the way for improved water resource management.

299
A mixture risk assessment for pollutants that reach humans via the water – fish exposure route
A. Kortenkamp, Brunel University London; M. Faust, Faust & Backhaus Environmental Consulting
An important route for human exposures to substances present in freshwater is through the consumption of fish. To protect humans against this route of exposure, the WFD defines specific quality standards for priority substances. In general, combined exposures are not considered, with the exception of quality standards for mixtures of specific contaminant groups, such as poly-chlorinated dioxins (PCDD) or polybrominated diphenyl ethers (PBDE). However, the possibility of combination effects across these pollutant groups is not currently considered. We present an advanced tiered mixture risk assessment for these groups of pollutants, first by using Water Framework Directive Quality Standards defined for PCDD and

PBDE in fish. We then make an attempt to expand the assessment by integration across these pollutant groups. While the definition of Quality Standards for specific pollutant groups is a step in the right direction, our analysis shows that more efforts are needed to protect humans from possible combination effects across pollutant groups. Such efforts are currently hampered by data gaps concerning common toxicities likely to arise in humans.

300

An Advanced Methodological Framework for the Identification of Priority Pollutants and Priority Mixtures of Pollutants in European Freshwaters

M. Faust, Faust & Backhaus Environmental Consulting; R. Altenburger, UFC Centre for Environmental Research / Department Bioanalytical Ecotoxicology; T. Backhaus, University of Gothenburg / Department of Biological and Environmental Sciences; W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; V. Dulio, INERIS; J. van Gils, DELTARES; A. Ginebreda, CSIC - Spanish National Research Council / Environmental Chemistry; A. Kortenkamp, Brunel University London; J. Munthe, IVL Swedish Environmental Research Institute Ltd; J. Slobodnik, Environmental Institute; K. Tollefsen, NIVA / Ecotoxicology and Risk Assessment; A. van Wezel, KWR Watercycle Research Institute / Chemical Water Quality and Health

We present a proposal for an advanced methodological framework for identifying priority pollutants and priority mixtures of pollutants in European freshwaters. The proposal was developed by the EU project SOLUTIONS. It aims to tackle major shortcomings of current prioritisation procedures under the EU Water Framework Directive (WFD): (i) For most aquatic pollutants the high data demands for a conclusive risk assessment cannot be met. Significant risks from so-called emerging pollutants may remain undetected. The WFD does not include an effective mechanism to close such knowledge gaps. The introduction of a watch-list mechanism for up to 10 substances provided a minor improvement but no fundamental change to this situation. (ii) Individual pollutants are assessed as if they would occur in isolation, largely ignoring the fact that they are part of complex multi-constituent mixtures. Environmental quality standards that have been established for single priority pollutants may not be sufficiently protective against mixture effects. Regulatory approaches for effectively tackling the problem are missing. The advanced framework integrates all available lines of evidence (LOE) on significant risks. This includes evidence from (i) ecological monitoring (field observations on so-called biological quality elements), (ii) effect-based monitoring (*in vitro* or *in vivo* testing in the lab or onsite), (iii) chemical monitoring in combination with so-called component-based mixture risk assessment approaches, (iv) integrated modelling of co-exposure and resulting mixture risks. Where one or more lines of evidence identify groups of pollutants presenting a significant risk, these should be subject to prioritisation for risk reduction measures. Where appropriate, such groups may be reduced to few mixture components or even one single component which can be demonstrated to explain most of the overall risk, so-called drivers of mixture risks. Wherever conclusive evidence on significant risks and needs for risk reduction cannot be reached because all possible LOEs are somewhere blocked by significant data or knowledge gaps, mixture components of potential concern are not left unnoticed but they are prioritised for further research and testing. Some elements of the advanced methodological framework may be readily applicable under the existing WFD. Full implementation, however, requires changes in the legal text, as detailed in Brack et al. 2017 (Sci Total Environ 576:720-737).

301

A diagnostic toolbox for ecological effects of pollutant mixtures: a case study application using in situ experiments with microbial communities

T. Backhaus, University of Gothenburg / Department of Biological and Environmental Sciences; A. Arrhenius, University of Gothenburg / Biological and Environmental Sciences; R. Behra, Eawag / Department of Environmental Toxicology; T. Seiler, RWTH Aachen University / Ecosystem Analysis; P. van den Brink, Alterra and Wageningen University; B. Deutschmann, RWTH Aachen University / Department of Ecosystem Analysis ESA; N. Corcol, University of Gothenburg / Department of Biological and Environmental Sciences; A. Focks, Alterra Wageningen University and Research Centre / Environmental Risk Assessment Team; J. Hollender, Eawag / Environmental Chemistry; H. Hollert, RWTH Aachen University / Institute for Environmental Research; H. Segner, University of Bern / Centre for Fish and Wildlife Health; I. Teodorovic, University of Novi Sad / Department of Biology and Ecology, Laboratory of Ecotoxicology (LECOTOX); A. Tlili, Eawag / Department of Environmental Toxicology; B. Wagner, Swiss Federal Institute of Aquatic Science and Technology (EAWAG) / Department of Environmental Toxicology

A toolbox for the detection of ecological impacts of chemicals was developed using a statistically supported, transparent and formalized weight of evidence (WOE) approach. It integrates four lines of evidence (LOE's): (i) predictive mixture modelling, (ii) effect-directed analysis (EDA), (iii) *in situ* tests, and (iv) field-based monitoring studies. A systematic and quantitative method was developed for the aggregation of multiple *in situ* tests into LOE III, using an aggregated response index, which we termed the "average biomarker response" (ABR). The results of the four separate LOE's are finally integrated using a systematic decision matrix that provides the main overarching conclusions that can be drawn from a given set of data and that pinpoints to critical data gaps. Here we first present an overview of the toolbox. Afterwards, we present a case study that used *in situ* experiments with photoautotrophic biofilms (periphyton) in wastewater impacted streams. Chemical-analytical profiles initially showed clear differences of the micropollutant load in the water up- and downstream of the entry point of a sewage treatment plant effluent. These chemical-analytical data were evaluated for their potential ecotoxicological effects using predictive mixture toxicity modeling approaches. Based on outcome of this LOE, we hypothesized that clear ecological effects on the structure and function of the exposed microbial communities should be present. Indeed, these were then confirmed using the concept of pollution-induced community tolerance (PICT). In the end, the study allowed us to demonstrate that (i) the STP effluent actually caused ecological impacts on the exposed microbial community, (ii) a subsequent upgrade of the STP plant with actived carbon filtration led to a recovery of the community that was driven by a lowered overall toxic pressure, (iii) PSII inhibitors were the mixture toxicity drivers, and (iv) that ecologically relevant effects go beyond a mere toxic unit summation. The presented work was a joint effort of the EU funded project SOLUTIONS, the ERAfresh project that was funded by the Swiss Federal Office for the Environment, and the IMPROVE project, which is funded by the Swedish Research Council.

Derivation, Validation and Implementation of Environmental Quality Benchmarks

302

Questioning annual average concentrations for plant protection products - TKTD modelling of real exposure profiles

M. Junghans, Swiss Centre for Applied Ecotoxicology EAWAG - EPF / Ecotox Centre; R. Kuhl, E. Zimmer, IBACON GmbH; I. Werner, Ecotox Centre Eawag-EPFL / Department of Anatomy Physiology and Cell Biology; R. Ashauer, University of York / Environment

The EU Water Framework Directive defines two environmental quality standards for assessing the chemical status of surface waters. The MAC-EQS defines the Maximal Allowable Concentration that should never be exceeded and the AA-EQS defines a concentration that shoudn't be exceeded by the Annual Average concentration. While the MAC-EQS should account for the acute toxicity of a substance, the AA-EQS is based on chronic eco-toxicity studies. For substances with highly fluctuating environmental concentrations like plant protection products the use of the annual average is disputed. Hence, in Switzerland it was suggested to use 14-day time-weighted average (TWA) concentrations for assessing compliance with quality standards for chronic toxicity. This approach is based on the average duration of chronic eco-toxicity tests and Haber's rule. We assess the suitability of this approach for retrospective risk assessment by applying toxicokinetic-toxicodynamic (TKTD) modelling on high resolution exposure profiles of plant protection products measured in Swiss streams. The TKTD modeling is a proxy for the actual time-course of toxicity under time-variable exposure and is based on 7 species, 7 substances and 5 exposure profiles from 5 streams. The results confirm the suitability of the time integral of 14 days. The prediction of actual toxicity for the most toxic periods is very consistent with the toxicity modeled for the TWA. The deviations are on average less than factor 2 for each organism group tested. In addition to mortality for crustaceans and fish, only a small selection of sub-lethal effects was considered, namely reproduction and growth in water fleas and population growth in duckweed and algae. The results also show that comparing quality criteria for protection against acute effects (MAC-EQS) to time-proportional 3-day mixed samples is appropriate.

303

Revision of 62 Environmental Quality Standards - lessons learned

M. Korkaric, Swiss Centre for Applied Ecotoxicology / Department of Environmental Toxicology; M. Junghans, Swiss Centre for Applied Ecotoxicology EAWAG - EPF / Ecotox Centre; R. Kase, Swiss Centre for Applied Ecotoxicology Eawag-EPFL; I. Werner, Ecotox Centre Eawag-EPFL / Department of Anatomy Physiology and Cell Biology

Environmental Quality Standards (EQS) are ecotoxicologically based threshold values that aim to prevent harmful effects of pollutants on ecosystems. Similar values exist in Switzerland and the Federal Office for the Environment recently comissioned the revision of existing EQSs for 62 substances to ensure that they are based on the current state of science. This study aimed to analyse the underlying reasons for numerical changes of EQSs and to highlight knowledge gaps. As for the original EQS derivation, relevant data were retrieved from databases, the public literature, and registration dossiers, and were, in some cases, provided by manufacturers. The reliability and relevance of ecotoxicological data were assessed using the CRED method. EQS derivation then largely followed the EU-Technical Guidance for Deriving EQS. After the revision, 60 AA-EQSs and 58 MAC-EQSs were proposed. The EQS revision did not generally lead to either lower or higher EQSs. AA-EQSs increased in 13 cases (max./median fold change =9.6/3) and decreased in 18 cases (65/4.9). The MAC-EQSs increased in 21 cases (50.6/1.8) and decreased in 9 cases (22.7/2.4). Most EQS were derived deterministically, using the assessment factor (AF) method. Due to an increase in data for some substances, the number of AA-EQSs and MAC-EQSs derived using Species Sensitivity Distributions (SSDs) increased from 2 to 5 and from 7 to11, respectively. For AA-EQS derivation, AFs were reduced in 12 cases and increased in only 6 cases. For the MAC-EQS derivation, AF were reduced in 5 cases and increased only in one case. Our study demonstrates that EQSs based on small data sets are more prone to large numerical changes when revised. Hence, an update often reduces the uncertainty associated with the derived EQSs, as evident from application of lower AFs and more frequent EQS derivations based on SSDs. This is likely to make EQSs more robust against larger changes in future revisions. Nevertheless, for the majority of the substances considered in this study, data sets were insufficient to construct SSDs. This is mostly due to a lack of studies using non-standard test species and species from important taxonomic groups, such as amphibians or insects. In some cases, and depending on the mode of action of a substance, this factor alone prevented the use of lower AFs. Finally, recommendations regarding assessability and quality of ecotoxicity data from industry studies and from the scientific literature are presented.

304

Endocrine disrupting properties: how far and consistent they are considered deriving Water Framework Directive Environmental Quality Standards ? A case study tackling French and EU EQS values

A. JAMES-CASAS, INERIS / Toxicological Ecotoxicological Assessment of chemical Substances ETES; A. BOTHAMY, INERIS; S. ANDRES, INERIS / Toxicological Ecotoxicological Assessment of chemical Substances

The concern raised by endocrine disrupting properties of chemicals should be tackled in diverse regulatory frameworks, among which the Water Framework Directive Common Implementation Strategy (WFD CIS). In this context, endocrine disruption (ED) is quoted several times as an issue for deriving water quality thresholds in the European Commission Technical Guidance for Deriving Environmental Quality Standards (TGD EQS). However, even if this guidance introduces ED properties as a reason for growing concern, it does not properly recommend any specific methodological approach to consider these properties while deriving EQS values. In order to evaluate the usefulness of such a recommendation and the extent to which it should be implemented, a state of the art of how ED properties are currently been taken on board in the EQSs already derived at EU and national level was made. To begin with, the work consisted in carrying out an inventory of substances for which an EQS had been derived and a factsheet describing the reasoning behind value was available. These substances made up the universe of 178 substances on which further work was led. Then, an analysis was made of these substances EQSs derivation to categorise them according to how ED properties where reported and taken on board if necessary for protection of wildlife and human health. This work led to 4 groups of substances. Group 1 contains substances whose EQS values do not consider ED effects, and which need to be reassessed as a matter of priority. Substances for which EQS derivation has considered ED effects but whose rationale does not clearly explain this are grouped together in Group 2 and should be verified. Group 3 corresponds to substances whose ED characteristics have been considered by an additional safety factor and / or a study that covers ED effects. Group 4 contains all substances that have no ED effects demonstrated from now on. No action is required for these last two categories. This state of play and categorisation work made it possible to prioritise substances for which EQS should be updated first as regards their ED potential. Also, this work gives more insight in how to derive EQSs as regards ED potential in order to further propose recommendations for a harmonisation of the methodology in the future.

305

Bringing water quality benchmark derivation approaches into the 21st century

R.A. van Dam, Environmental Research Institute of the Supervising Scientist / Department of the Environment and Energy; G.E. Batley, CSIRO Land and Water / Centre for Environmental Contaminants Research; R. Fisher, Australian Institute of Marine Science; D. Fox, Environmetrics Australia; A.J. Harford, Environmental Research Institute of the Supervising Scientist / Dept. of the Environment and Energy; C.L. Humphrey, Environmental Research Institute of the Supervising Scientist / Department of the Environment Energy; A. Proctor, Australian Antarctic Division / Department of the Environment and Energy; J. Stauber, CSIRO / CSIRO Land and Water; M.S. Warne, Coventry University / Centre for Agroecology, Water and Resilience

The most common method for deriving water quality benchmarks (WQBs) for toxicants is the use of a species sensitivity distribution (SSD) to estimate a concentration that is protective of x% of species. Although variations exist in the specifics of the methods employed by jurisdictions around the world, the fundamental SSD approach is similar and, moreover, has not changed markedly over the past 20 years, despite a significant body of published research aimed at improving or developing new derivation methods. The recent revision of the Australian and New Zealand SSD-based derivation method has re-highlighted numerous limitations of the SSD approach for certain data situations and toxicant types; for example, small sample sizes, model choice and fit, and accommodating different routes of exposure (e.g. for persistent, bioaccumulative and toxic compounds) and specific mechanisms of toxicity (i.e. bimodality). However, areas for improvement of WQB derivation methods extend beyond just refining SSD-based approaches, to the use of non-SSD approaches and weight of evidence approaches that give consideration to both laboratory- and field-effects data. Other opportunities for improvement exist in the acquisition of data for WQBs (e.g. type and acceptability of toxicity data), as well as the application of WQBs in water quality management. Thus, it is important to identify and target the limitations that, if addressed, will yield the biggest benefits to environmental protection. Experience has shown that a ‘one size fits all’ approach to WQB derivation does not work, and an alternative is to provide flexibility and adopt approaches that do the best job in the face of the specific conditions and uncertainty posed by different situations. However, this may increase the complexity of the derivation process and, thus decrease understanding and adoption by users. This presentation will examine a variety of questions associated with WQB derivation methods, including: What research efforts have been undertaken to significantly evolve the way we derive WQBs? To what extent have the outcomes of this research been adopted in formal derivation methods? Why has the fundamental SSD approach not changed markedly over the past 20 years despite various proposed improvements? Do we need to move to a new generation of WQB derivation approaches and, if so, what are the aspects that will have the biggest impact, and the criteria that would make new approaches amenable for adoption?

306

The quest for consistent environmental protection: the challenge of variable water quality guidelines between regulatory jurisdictions

G. Merrington, A. Peters, wca; S. Kosmala, WCA Environment Limited

One of the tools used by regulatory jurisdictions to deliver environmental protection is Water Quality Guidelines (WQGs) or Environmental Quality Standards. These are thresholds expressed as a chemical concentration, with an associated summary statistic (e.g. maximum) and period (e.g. “annual average”). These thresholds are used around the world for a number of purposes including to assessing water quality status, classification and also as a starting point for setting effluent discharge permits. Historically, most regulatory jurisdictions across the globe have, at the very least, WQGs for some trace elements. The percieved challenge for many in the regulated community, especially multinational organisations, is the lack of transparency in derivation and implementation of WQG within a regulatory jurisdiction and the inconsistent environmental protection levels between regulatory jurisdictions, despite having the same protection goals. Here we will address the veracity of that perception and attempt to understand its source. There is limited consitency in environmental protection, as judged by WQG for the same trace element, across regulatory jurisdictions. The absence of robust protocols (or any protocols that can be reviewed) for the derivation is a major factor as is the inevitable lack of resources and time for many regulators to update WQG and account for new scientific developments. There are unfortunate repetitive cycles of derivation that each jurisdiction goes through for the same substances and perhaps there is benefit in sharing knowledge and understanding across jurisdictions that would deliver consistent and transparent levels of environmental protection.

307

A Call for Greater International Collaboration in Developing Environmental Quality Benchmarks: Many Hands Make Lighter Work!

M.S. Warne, Coventry University / Centre for Agroecology, Water and Resilience; K.M. Leung, The University of Hong Kong / The Swire Institute of Marine Science; G. Merrington, wca

Environmental quality benchmarks (EQBs, also variously called guidelines, standards, criteria) are an internationally accepted means of protecting ecosystems from the adverse effects of toxicants. As such, numerous countries, states/provinces, regions, academics and consultants have developed EQBs. As a result there are numerous EQBs for the same suite of chemicals (e.g. copper, lead, benzene), each slightly different. This is a huge waste of resources. These differences arise because of the differences in the methods used to derive the EQBs, which govern what is considered acute and chronic, what data can be used, and the magnitude of assessment factors etc. The current situation with the derivation of EQBs has been compared to that of toothbrushes – “everyone has one and no-one else wants to use anyone else’s” and disagreements arise about whose “toothbrush” is the best, whether particular features are “arbitrary” etc. We believe that such views are unhelpful and missing the point. There have been previous calls for increased collaboration between jurisdictions and even calls to harmonise the derivation methods with the ultimate goal of having a single global derivation process and a single set of global benchmarks. While having a single derivation method and set of benchmarks is a lofty goal it is also extremely unlikely to occur within the foreseeable future. The greatest gains are possible in the parts of the derivation process that takes the most time and effort. At least 90% of the time and effort spent on deriving an EQB is used to assess which ecotoxicity data are suitable and of appropriate quality to use. We therefore advocate that efforts should focus on these methods. Possible ways to reduce effort include: acceptance of other jurisdictions assessments; international acceptance of an existing method, developing a new assessment method and establishing an international archive for assessed data. Other components of the derivation process that could also be relatively easily be harmonised and would have a significant impact in reducing effort will also be discussed. A realistic plan for achieving these gains will be set out.

Microbial community ecotoxicology in environmental risk assessment and ecosystem monitoring

308

The impact of anthropogenic activities on bacterial and viral diversity in the Eastern Mediterranean Sea

A. Tsiola; P. Pitta, Hellenic Centre for Marine Research Crete / Institute of Oceanography and Environment; S. Fodelianakis, G. Michoud, King Abdullah University of Science and Technology; A. Pavlidou, E. Rousselaki, N. Simboura, Hellenic Centre for Marine Research; C. Zeri, Hellenic Centre for Marine Research / Institute of Oceanography; I. Karakassis, University of Crete / Department of Biology; G. Kotoulas, Hellenic Centre for Marine Research; D. Daffonchio, 4King Abdullah University of Science and Technology / Biological and Environmental Sciences and Engineering Division; M. Tsapakis, Hellenic Centre for Marine Science / Institute of Oceanography

The Eastern Mediterranean Sea is a low-nutrient low-chlorophyll marine ecosystem [1] but some variability within the basin does exist. Indeed, several coasts are influenced by anthropogenic processes, and specifically in the Greek coasts these include industrial, harbor, agriculture, mariculture activities, urbanization and tourism [2]. Our hypothesis was that prokaryotic and viral community diversity is differently affected in contrasting coastal systems by anthropogenic pressures. We used 16S rRNA gene amplicon and whole virome sequencing at stations characterized by different chemical features based on the “National monitoring project for the implementation the Water Framework Directive (2000/60/EE) in Greece” [2]. We focused on viral auxiliary metabolic genes and the influence of heavy metals (Cu, Cd, Co, Ni, Pb, Zn, Cr and Hg). Significant differences were found at the genus level between the sampling stations. Proteobacteria were dominant in all stations, while Bacteroidetes were more pronounced in some of the stations. Rare phyla were higher in Echinades and Patraikos Gulf. 16S rRNA patterns resembled abiotic variables, and especially the patterns of heavy metals Cd, Cd, Cu and Pb. The highest concentrations of NO_2^-, NO_3^-, NH_4^+, PO_4^{-3}, SiO_2 and chlorophyll a were found in stations influenced by extensive industial, agricultural and maricultural activities. The 3 stations of Amvrakikos Gulf were highly variable in terms of community structure. Significantly lower relative abundance of Verrucomicrobia and Bacteroidetes in the “control” than in the “impact” station in Kefalonia (inside and outside the influence of the fish farms, respectively) was seen. Bacterial 16S rRNA analysis revealed significant differences between stations along the Greek coast, suggesting that each station hosts a different community. Analysis of viral metagenomes will show if community composition reflects the anthropogenic activities in these areas, and if lysogeny (i.e. viral integrase and auxiliary metabolic genes’ abundance) is a prevalent life strategy. [1] Krom MD, Emeis K-C, Van Cappellen P. 2010. Why is the Eastern Mediterranean phosphorus limited?. Prog Oceanogr 85:236-244. [2] Pavlidou A, Simboura N, Rousselaki E, Tsapakis M, Pagou K, Drakopoulou P, Assimakopoulou G, Kontoyiannis H, Panayotidis P. 2015. Methods of eutrophication assessment in the context of the water framwork directive. Cont Shelf Res. 108: 156-168.

309

Impacts of stormwater on microbial community structure and function in estuarine sediments

K. Dafforn, Macquarie University / Evolution and Ecology Reserach Centre; P. Steinberg, University of New South Wales / Centre of Marine BioInnovation; S.L. Simpson, CSIRO Land and Water / Centre for Environmental Contaminants Research; J. Potts, P. Scanes, NSW Office of Environment and Heritage; S.C. Birrer, University of New South Wales; M. Sutherland, NSW Office of Environment and Heritage; V.X. Sim, University of New South Wales; T. Lachnit, University of Kiel; S. Swarup, National University of Singapore; S. Kjelleberg, Nanyang Technological University / The Singapore Centre on Environmental Life Sciences Engineering; M. Doblin, Department of Environmental Sciences / Department of Environmental Sciences; G. Birch, Sydney University / School of Geosciences; P. Gribben, University of New South Wales; E.L. Johnston, University of New South Wales / Evolution and Ecology Research Centre

Estuaries are diverse and productive ecosystems that are subject to high levels of disturbance. They are exposed to multiple stressors such as legacy contaminants in sediments and ongoing inputs of nutrients and metals *via* stormwater, but we still have little understanding of the consequences for ecosystem functioning. We surveyed sediment communities at four locations with large stormwater drains in Sydney Harbour, Australia. Locations were either poorly-flushed embayments or were well-flushed open channels. Sediment was collected monthly during base rainfall (< 5mm/day) for 4 months from 3 sites within each location at increasing distance from the stormwater drain (0, 200 and 1000 m). We also collected sediments after 2 large rain events (>150mm in 24h). Next-generation sequencing was used to characterize the microbial community and sediment was subsampled for metals, total organic carbon, total nitrogen and phosphate. Sediment cores were also collected to measure biogeochemical processes including primary productivity, community respiration and nutrient cycling. We observed major shifts in the microbial community related to exposure to legacy contaminants and new stormwater contaminant inputs. We also found trends of decreasing community respiration rates away from storm drains and lowest rates of primary production during base rainfall. The results have implications for future management of stormwater in estuaries and increase our understanding of how to conserve crucial sediment community diversity and function.

310

Drought as environmental driver on ciliates and micrometazoa communities in a multistressors scenario. An experimental approach

J. López-Doval, F. Romero, V. Acuña, S. Sabater, ICRA Catalan Institute for Water Research

Climate change will affect agriculture practices and productivity because increased intensity and frequency of pest events, increased sensitivity to stress and diseases and changes in phenology, that means a future increase in the use of pesticides and, in consequence higher risk of freshwater pollution. In addition climate change will lead to higher severity of drought events and higher temperatures. Ciliates and micrometazoans in freshwater ecosystems play an important role in the processing of organic matter and as basal resource for consumer organisms. The present work aims to study how these environmental and chemical stressors, and their interactions, in a future scenario of climate change can affect these communities in freshwater sediments. To study how drought (D), warming (T) and a realistic environmental mixture of pesticides (P) can affect the communities of ciliates and micrometazoa in river sediments, we developed an experiment with a factorial design in experimental indoor channels with natural sediment from a pristine river (24 channels, 3 replicates, 8 experimental conditions). The community was exposed to the stressors for 7 weeks. Diversity of Ciliophora and micrometazoa communities was studied twice during the experiment (one week before and after 7 weeks of chronic exposure). Significant changes in community composition between pre- and post- exposure were observed for all treatments. Community was dominated by micrometazoans in all treatments in terms of density, but a trend of increasing the percentage of ciliates in those treatments with stressors was observed, indicating a possible advantage of ciliates in stressed environments. At the end of the experiment total density was significantly higher respect to control in D, DP and TDP treatments ($p < 0.001$ Dunnet’s test) while diversity was significantly higher in D, TD and TP conditions ($p < 0.001$ Dunnet’s test). Taking into account the community structure, flow reduction was the most important factor causing significant differences in community composition (PERMANOVA $p< 0.001$). The mixture of pesticides at realistic environmental concentrations did not cause any effect on the studied communities. Our results suggest that flow reduction is the main driver for changes in micrometazoa and ciliate communities in our experimental system while pesticides and temperature produce significant effects only in combination with drought.

311

Linking pesticide pollution with periphyton quality in agricultural streams: a fatty-acids approach

N. Corcoll, University of Gothenburg, Sweden / Biological and Environmental Sciences; A. Håkansson, University of Gothenburg / Department of Biological and Environmental Sciences; A. Nilsson, University of Gothenburg / Section of Ophtalmology, Dept. Clinical Neuroscience, Institute of Neuroscience and Physiology, Sahlgrenska Academy; K. Johansson, University of Tartu / Institute of Technology; H. Spångfors, Halmstad University; M. Kahlert, SLU Swedish University of Agricultural Sciences / Department of Aquatic Sciences and Assessment; J. Kreuger, Swedish University of Agricultural Science / Centre for Chemical Pesticides; T. Backhaus, University of Gothenburg / Department of Biological and Environmental Sciences

Although the environmental risks associated with pesticide pollution in agricultural streams are quite well documented, little is know about its potential effects on periphyton quality. Periphyton provides many of the essential polyunsaturated fatty acids (PUFA) that are needed for organisms at higher trophic levels in river food webs. This study aims to assess the effects of pesticide mixtures on periphyton quality *in situ*. Three streams (Höje å, Skivarpsån and M42) located in the agriculturally dominated region of Skåne (SE Sweden) were sampled in September and October 2016. The effects of pesticide pollution were assessed by passive field sampling coupled with laboratory ecotoxicity tests, by mixture toxicity modelling to predict which chemical stressors were potentially driving the toxicity, and by examining the fatty acid profiles, pigment content and algal diversity of periphyton communities. Results from water chemical analyses clearly showed higher levels of nutrients and pesticide polution in Skivarpsån and M42 than in Höje å. Ecotoxicity tests using the passive sampler extracts demostrated that the pesticide mixture occurring at Skivarpsån and M42 were toxic for periphyton communities from Höje å, causing an inhibition of the photosynthetic activity up to 63% and 53%, respectively. Cluster and principal component analyses based on pigments content, algal diversity and fatty acid profiles, clearly separated the periphyton from the three river sites studied. Algal biomass from periphyton of pesticide polluted streams (Skivarpsån and M42) was hgher than in Höje å. The nutritive quality of the periphyton differed among streams, and fatty acids considered high-quality such as eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) were also more abundant in pesticide polluted streams (Skivarsån and M42). Overal, even though results from the lab show that the mixture of pesticide pollution in the studied

streams might be toxic for periphyton (i.e. inhibiting the photosynthetic activity), being herbicides the driving chemical stressors. Results from the field, indicate that when the levels of pesticide pollution are low and co-occur with hgh levels of nutrients pollution, nutrients migh mask pesticides effects on periphyton quantity and quality because compensatory effects from nutrients.

312

Estrone and triclosan mixture alters soil metagenomics during degradaton

D.L. Carr, Texas Tech University / Biological Sciences; E. Osuji, Texas Tech University / Biological Science

Wastewater derived from domestic use commonly contains mixture of pharmaceutical and personal product (PPCP), but its persistence and accumulation in the soil and the consequences for soil microbial community processes are poorly understood. Estrone and triclosan are two common PPCPs of domestic wastewater. Soil microbial communities degrade a variety of PPCPs however; most studies have only addressed single compound designs neglecting the reality of their co-occurrence in nature. In this study, we examined the interaction between estrone and triclosan mixture, their potential to persist and disrupt soil microbial community composition and function. Soil was spiked with estrone, triclosan, and a 1:1 mixture of estrone: triclosan, and incubated for 90 days in the dark at 27°C. We examined soil microbial function dynamics using commercial Biolog EcoPlates™. Microbial degradation rates were compared over the 90 days' incubation period using high performance liquid chromatography. Metagenomic analysis by 16S rRNA was used to determine changes in microbial community over time. There was significant increase in substrate activity and substrate richness in all treatments. Each microbial community utilized different carbon substrates by day 90 whereas they had exhibited similar substrate utilization at day 0. Estrone and triclosan as single compound treatments exhibited half-lives of 6.8 days (estrone) and 26.7 days (triclosan). The rate of degradation of the binary estrone:triclosan mixture was the same as the individual compounds. There was a decrease in species diversity between control at day 0 and all other treatments at day 90 with establishment of unique OTUs in each treatment group at day 90. Metagenomic analyses indicate distinct communities by treatment 90 days after exposure even though Bacillus sp. was dominant in all the day 90 treatments. Soil microbial communities are adept at degrading estrone and triclosan in this soil whether occurring singly or as a binary mixture thus preventing accumulation in soil and subsequent contamination of ground water.

313

Poster spotlight: TU014, TU015, TU016

Integrating life cycle approaches towards a sustainable circular economy (II)

314

Region-specific life cycle inventories for tailings disposal in ecoinvent v3

D.A. Turner, EMPA / Technology and Society Lab; G. Doka, Doka Life Cycle Assessments; A. Haarman, EMPA Technology & Society Lab / Technology & Society Lab; R. Hischier, EMPA / Technology and Society Lab

Tailings, a waste material produced during mineral concentration (beneficiation), often contain significant quantities of mobile toxic metals and are typically produced in large quantities. To manage these wastes, tailings are commonly stored behind dammed impoundments, known as "tailings ponds". These ponds pose a significant long-term pollution risk as metals may leach out into the surrounding environment, potentially over very long timeframes. The management of tailings therefore represents an important environmental burden for primary metal production. To help life cycle assessment (LCA) practitioners quantify this important environmental burden, the ecoinvent database contains – since 2009 and the release of version 2.1 – a global average life cycle inventory (LCI) dataset for sulphidic tailings disposal, developed using a dedicated tailings emissions model. However, the dataset was intended only to serve as a first generic estimate and is based on highly aggregated data that attributes an identical burden to each kilogram of waste, regardless of its composition. Given their relevance to the overall impacts of primary metals production, access to more detailed, region-specific LCI data on tailings disposal is crucial for a more comprehensive and adequate integration of primary metals in LCA studies. Here, we present an extended model for assessing the long-term emissions from tailings disposal. The model can heed a specific tailings composition and local climate data, allowing for the creation of site- or region-specific inventories. Based on an exhaustive literature survey of data on tailings compositions and leachate concentrations from different mine sites worldwide, the model was used to develop new country- and region-specific datasets for tailings disposal from a range of ore types, which will contribute to improving the quality and hence reducing uncertainties in LCA studies worldwide. Our presentation will give an overview of the extended model and related datasets, which will be integrated in a later version of ecoinvent. We will also highlight its improvements compared to the previous model by presenting the results of an LCA case study of region-specific primary metals production in order to demonstrate the important differences between global average and situation-specific calculations in such an important sector as primary metals production.

315

Closing the copper cycle in the EU-28: scenario analysis and potentials for GHG emissions reduction

L. Ciacci, Alma Mater Studiorum - University of Bologna; F. Passarini, Alma Mater Studiorum - University of Bologna / Dept. of Industrial Chemistry

Copper is widely used in modern society, finding application in traditional end-uses such as plumbing, infrastructure, and transportation, but it is also an essential material in emerging green energy technologies. Europe (i.e., EU-28) has modest natural deposits and strongly depends on imports to meet the domestic demand. In light of such an extended import reliance and possible supply shortages, end-of-life recycling can secure access to secondary copper forms and support the implementation of a circular economy. In addition, as copper recycling is generally less energy intensive than primary copper production, closing the elemental cycle through recycling would result in significant environmental benefits. However, despite a well-established industry network in the copper value chain, the EU-28 is still far from perfect recycling highlighting wide margins for improvements. Some of these potentials for copper circularity and environmental benefits were explored combining four well-regarded UNEP scenarios with material flow analysis. For each scenario, the copper demand and supply in the region was modeled to 2050. We commented the results in the case of stationary end-of-life recycling performance and under the hypothetical implementation of a near-perfect recycling condition. Life cycle assessment indicators were modeled to evaluate the resulting energy savings and greenhouse gas emissions reduction. The results show that copper recycling can contribute significantly to reduce the energy requirements and mitigate greenhouse gas emissions associated with the regional copper industry. However, for three out of the four scenarios the current recycling performance seems not to be enough to close the copper cycle. Fundamental constraints are likely to limit the implementation of a circular economy unless dramatic changes occur in the current pattern of copper production, consumption and recycling at end-of-life.

316

Multi-Objective Reverse Supply Chain Network Design of Fluorescent Lamps with Piecewise Linear Functions

C. Lee, L. Papageorgiou, University College London / Department of Chemical Engineering; N. Shah, Imperial College London / Department of Chemical Engineering

In this study, a reverse supply chain model has been developed to support strategic decision making problems associated with its network design and process operation. The examined networks comprise multi-echelons, including disposer markets, collection facilities, recycling plants and reuse markets connected by a transportation mode. The system is modelled as a multi-objective mixed-integer linear programming (MILP) optimisation problem allowing the inherent trade-offs between the conflicting economic and environmental objectives to be explored. The economic function is net economic value calculated by the total cost minus production savings. Total cost includes capital and operation costs required to operate the supply chain network. The production saving is the revenue obtained from selling secondary products. In contrast, the ecological objective function is based upon net environmental value. This is achieved by adopting the principles of LCA, expanding the network boundaries to incorporate a set of life cycle stages and using the Korean Eco-Indicator method to assess the environmental impact of the network and avoided burdens. In addition, the environmental and economic performances of reverse supply chain networks are assessed through the development of a spatially explicit model that combines logistics and Geographic Information Systems. The model is used to address strategic decisions involving the location, number and capacity of collection and recycling facilities; selection of recycling technologies; and assignment of transportation links required to satisfy returns and demand at the markets. At the operational level, optimal recycling profiles and flows of material between various components within the supply chain are determined. Furthermore, the model considers the economies of scale (large, medium and small) of collection & recycling facilities and transportation links, and explores whether centralised recycling is favoured over decentralised recycling. The applicability of the proposed models is explored within a South Korea context in order to determine the optimal reverse supply chain configuration of fluorescent lamps.

317

The use of Life Cycle Assessment to adjust consumption taxes: The concept of a Damage and Value Added Tax

B. Timmermans, Université Libre de Bruxelles; W. Achten, Université Libre de Bruxelles / Department Geosciences, Environment and Society

The purpose of this presentation is to provide a short insight about a study examining the principles and feasibility of a shift from Value Added Tax (VAT) or sales tax to a Damage and Value Added Tax (DaVAT) partially based on the life cycle assessment of products and services. With this shift, goods and services that seriously harm the environment and human health will be priced up, those that have less impact will be priced down. The DaVAT system relies on three essential points: i) Apply VAT (or consumption taxes in general) to all goods and services and reduce its multiple rates to one single low rate (e.g. 3%) called *Uniform VAT* (UVAT); ii) Add to UVAT a per-unit tax called *Global Damage Tax* (GDT) calculated on the basis of environmental impacts assessed by means of specific or generic LCAs. In the case of potentially high-polluting products or industries, a specific LCA will be automatically imposed; iii) In order to reflect environmental, social or ethical concerns specific to a country, another damage tax termed *Specific Damage Tax* (SDT) is proposed that extends beyond LCA. DaVAT is the sum of UVAT, GDT and SDT. DaVAT is conceived not as an additional burden but rather as a shift of taxation, as the rate of the old consumption taxes can decrease proportionally to the increase of GDT. DaVAT is also designed in such a way that the erosion of tax revenues, when pollutant releases would decline, is offset by the extension of the tax to all goods and services and by the possibility to gradually re-increase the UVAT rate when the number of highly-polluting products decreases. To reduce the variance of the LCA results used for this purpose, the DaVAT system should use common databases, apply the same inventory, characterization, normalization and weighting methods, as well as refer to the same impact categories and the same cut-off rules. LCA as a whole can stay as it is, but for use in DaVAT specific guidelines should be established. A rough assessment based on previous studies has been made of the costs of implementation, maintenance, administration and compliance of DaVAT, as well as of the risks of fraud, price changes and acceptability of the proposal. The presentation shall briefly outline the results of this assessment.

318

Towards global guidance on LCIA of mineral resource use - outcomes from the UN Environment Life Cycle Initiative task force

T. Sonderegger, ETH Zurich; M. Berger, Technische Universitaet Berlin / Chair of Sustaiable Engineering - Office Z1; R.A. Alvarenga, Ghent University / Department of Sustainable Organic Chemistry and Technology; V. Bach, Technische Universität Berlin / Chair of Sustainable Engineering; A. Cimprich, University of Waterloo; J. Dewulf, Ghent University / Department of Sustainable Organic Chemistry and Technology; R. Frischknecht, treeze Ltd.; J. Guinee, University of Leiden / Institute of Environmental Sciences; C. Helbig, University of Augsburg; O. Jolliet, University of Michigan; M. MOTOSHITA, National Institute of Advanced Industrial Sci. and; S. Northey, Monash University; D. Schrijvers, ISM; L. Tikana, DKI Copper Alliance / Life Cycle; A. Valero, Universidad de Zaragoza; M. Vieira, PRe Sustainability; S. Young, University of Waterloo

Primary mineral resources are of great relevance for industry and society. The environmental impacts caused by emissions to air, water, and soil resulting from mining and refining processes to produce usable materials are considered in relatively well established impact categories (e.g., climate change, acidification, and eco-toxicity). However, the assessment of impacts resulting from resource use on resource availability is still controversial – even though a wide range of methods and models are available. Therefore, this task force – comprising 48 experts from different fields and working under the umbrella of the UN Environment Life Cycle Initiative – focuses on methods assessing these impacts and works towards consensual guidance for method users and developers on what method type is best suited depending on the goal and scope of a particular LCA study. In a comprehensive literature review, more than 20 methods assessing impacts of resource use in LC(S)A have been identified. These methods have been clustered into four categories: methods assessing depletion of stocks, methods assessing "future efforts" resulting from ore grade decline as an (assumed) consequence of current extraction, methods using thermodynamic accounting (exergy and emergy approaches), and methods assessing supply risk of raw materials based on socio-economic aspects (like country concentration, political stability, etc.). Within the four clusters of methods, key axioms and methodological choices, like the resource stocks used in depletion methods, the reference environment used in exergy methods, or the socioeconomic aspects considered in methods addressing supply risk are discussed. Furthermore, methods are evaluated based on a comprehensive evaluation scheme developed within the Life Cycle Initiative that has been customized to the primary mineral resource context in an iterative process in the task force. The overall aim is to provide clear guidance on best practice for each method type. Furthermore, the review process includes a debate about what the scope of an (environmental) LCIA method should be and how problems related to resource use (provisioning capacity, availability for future generations, etc.) can best be assessed. Especially supply risk methods cover perspectives beyond "environmental" impacts and the task force keeps discussing how to map these methods in the framework of LCIA or how to use them complementary to "traditional" LCA.

319

Poster spotlight: TU214, TU215, TU237

Safe by Design: responsible and innovative research for safe and sustainable chemistry

320

Silica coating for the control of nano-reactivity

S. Ortelli, CNR ISTEC; M. Blosi, CNR; D. Gardini, CNR ISTEC; A. Costa, CNR

Nano-titanium dioxide (TiO_2) and nano-silver (Ag) and are among the materials most investigated for their technological importance and consequent interest in terms of their environment, health and safety (EHS) issues. In particular these particles cause alert for their capacity to generate, transport and release potentially toxicants such as metal ions and reactive oxygen species that can induce several negative effects, responsible for cytotoxicity. In this study we investigated silica coating as technique for control two recognised toxicity drivers for nano TiO_2 and Ag that are the exogenous production of ROS and the Ag^+/Ag total distribution. We evaluated the effect that silica coating had on physicochemical properties (size, shape and zeta potential), on toxicity drivers and in general on material performances. We first demonstrated that both at colloidal and dried state a matrix of SiO_2 surrounding TiO_2 and Ag nanoparticles was formed, even by simply playing with colloidal attraction between the two hetero-phases. The presence of silica coating seems to have two important effects for the control of ROS and Ag^+ toxicants, representing a safe by molecular design solution for the control of nano-reactivity: 1) Silica acts as dispersing/diluting matrix for TiO_2, decreasing the production of ROS, but improving the photocatalytic performances of pristine sample; 2) Silica act as reservoir for Ag^+ ions, decreasing the amount of immediately available fraction and so improving the range of concentration where the sample shows antibacterial properties despite to negligible cytotoxicity.

321

Framework for the optimal design of sustainable chemical processes

A. Gonzalez Garay, R. Calvo-Serrano, G. Guillén Gosálbez, Imperial College London / Chemical Engineering

Given its natural link between fundamental science, engineering and industrial practice, the chemical industry plays a key role in meeting the challenges of sustainable development. In particular, the use of computer aided tools for the generation of sustainable processes is essential to facilitate the transition towards a more sustainable chemical industry. In this work, we present a framework that incorporates sustainability principles in the design of chemical processes. The methodology proposed uses life cycle assessment to assess the sustainability of the processes; surrogate modeling and objective-reduction techniques to enhance the optimization os the processes; and data envelopment analysis (DEA) as multi-criteria analysis tool. The use of DEA facilitates the post-optimal analysis of the Pareto front by filtering and ranking the optimal designs that conform the Pareto frontier without the need to define explicit weights. In addition, DEA provides improvement targets for suboptimal alternatives that if attained would make them optimal. By comparing these suboptimal processes against the best technologies available, this targets can be used to guide retrofit efforts towards more effective actions. The capabilities of the framework are shown in a case study based on the production of methanol from CO_2 and hydrogen.

322

A decision framework for substances of very high concern at the interface of chemicals, products and waste

P. Wassenaar, National Institute for Public Health and the Environment (RIVM); P.G. Zweers, J.K. Verhoeven, National Institute for Public Health and the Environment RIVM; J. Spijker, RIVM; R.J. Luit, J.H. Ganzevles, National Institute for Public Health and the Environment RIVM; M. Janssen, Nat. Inst. Publ. Health Environ. / Centre for Safety of Substances and Products; L.C. van Leeuwen, C.W. Bodar, National Institute for Public Health and the Environment RIVM

Reuse and recycling of products are key elements towards a sustainable and circular economy. Besides the circular economy policy, a non-toxic environment is being pursued as well. Care should be given to the reuse and recycling of waste streams containing substances of very high concern (SVHC). Ideally, the presence of SVHCs in the design and production phase should be prevented by applying Safe-by-Design alternatives. However, we have to realize that we are still in an era in which we are faced with numerous (legacy) SVHCs in waste streams. For these waste streams, safe 'end-of-life' solutions have to be found in order to stimulate the circular economy and safeguard a non-toxic environment. Within this study, we developed a general framework to decide how waste streams with (legacy) SVHCs should be managed. The framework is specifically developed for the licensing process and provides guidance to license applicants and license authorities in the Netherlands. By following the framework, it will indicate whether the recycling of a specific waste stream into a specific end-product can be considered as acceptable with respect to the SVHC content. In principle, the use of this framework consists of three steps: 1) identification of SVHCs in the waste stream; 2) a basic decision scheme in order to decide whether a more elaborate risk analysis is necessary or whether the risks can be considered as acceptable; and 3) a risk analysis. Within the risk analysis, the acceptability of recycling will be assessed by weighing various aspects, including: availability and feasibility of SVHC removal possibilities, exceedance of SVHC concentration limit values, potential SVHC exposure of man and the environment, and the traceability of the material stream (including SVHC) during the whole life cycle. This framework is a first step to improve and warrant safe and sustainable recycling of waste streams. Future adjustments of this framework will be required in upcoming years based on practical experiences of licensing authorities. Furthermore, it is advised to develop a broader decision scheme that besides SVHCs also considers and weighs other risk and benefit factors of recycling, like the risk of pathogens and medicine residues and the benefits with respect to sustainability (e.g. carbon footprint). Such a development will further stimulate the transition towards a safe and sustainable economy.

323

Emissions of PFASs and alternatives from the durable water repellence layer (DWR) of textiles during use

I.v. Veen, Institute for Environmental Studies (IVM) VU University Amsterdam / Chemistry and Biology; A. Hanning, Swerea IVF AB; S. Schellenberger, Stockholm University / Department of Applied Environmental Science (ITM); J. de Boer, Vrije Universiteit Amsterdam Department Environment & Health / Environment and Health; J. Weiss, Stockholm University, ACES / Department of Aquatic Sciences and Assessment; P. Leonards, VU University, Institute for Environmental Studies / Department of Environment and Health

In the durable water repellence (DWR) layer of textiles of outdoor clothing, PFASs have been used because their perfluoroalkyl chains have the ability to repel liquids of a wide range of polarities (oleophobic and hydrophobic) due to low surface energy. Since long-chain PFASs are being phased out for their negative environmental impact, industry has started to use alternative chemicals to provide the DWR in outdoor clothing. Within the SUPFES (Substitution in Practice of Prioritized Fluorinated Chemicals to Eliminate Diffuse Sources) project alternative substances in use are assessed in comparison with the long–chain PFASs. As part of the SUPFES project we perform chemical alternative assessment including application hazards, exposure and life-cycle assessment studies. One of the studies focuses on the emissions of PFASs from outdoor clothing vs. emissions of alternative DWR chemicals such as short-chain PFAS (e.g. C4, C6) and silicones. We study the emissions using different emission scenarios which are based on real-life situations such as leaching to rain water, emission to air, weathering and washing and tumble drying. Within the SUPFES project different types of formulations, PFAS-based as well as silicon-based, have been applied to two different types of textiles, i.e. polyamide (PA) and polyester (PES). After testing the water repellence properties, a selection of four PFAS-based textiles and three silicon-based textiles have been used for assessing the emission of PFASs and silicones. For chemical alternative assessments it is highly important to include proper application tests in combination with experimental emission exposure scenarios. This information will provide valuable information to aid selection of safer alternatives. The emissions of chemicals out of the DWR-treated textiles are not only depending on the type of DWR, but also on the type of textile used. Weather conditions, like sunlight, high temperature, or humidity can have a strong effect on compounds used in DWR-treated outdoor clothing. DWR compounds, like PFASs and silicons are emitted to air, as well as to rain water and washing water. During the use phase of outdoor clothing, DWR chemicals are emitted to the environment.

324

Chemicals in plastic packaging: Prioritization of hazardous substances

K. Groh, Food Packaging Forum Foundation; T. Backhaus, University of Gothenburg / Department of Biological and Environmental Sciences; B. Carney Almroth, University of Gothenburg Sweden / Department of Biology and Environmental Sciences; B. Geueke, Food Packaging Forum Foundation; A. Lennquist, Chemsec; H. Leslie, VU University Amsterdam / Environment&Health; J. Muncke, Food Packaging Forum Foundation / General Management

Plastic packaging is increasingly used globally, causing rising concerns for the environment and human health from littering, release of microplastic and leakage of hazardous chemicals. Specifically, chemicals may migrate into foods or the environment during use, disposal, and recycling of the packaging material. Occupational exposure during plastic packaging manufacture is also relevant. One of the main obstacles to assessing the risks of chemicals originating from plastic packaging is the absence of information on the materials' exact chemical composition. In order to provide an overview of the chemicals associated with plastic packaging, we compiled the Chemicals in Plastic Packaging Database (CPP-DB), which comprises unique substances with additional substance-specific information such as toxicity data, physico-chemical properties and uses. The CPP-DB includes plastic monomers, additives, and other substances used during plastics manufacturing, such as solvents and raw materials, and the main non-intentionally added substances (NIAS) such as impurities, reaction by-products or degradation products of e.g. polymers and stabilizers. We ranked the substances in the CPP-DB according to their hazard for human health and the environment, using Classification, Labeling and Packaging (CLP) data on hazard categories, and also including endocrine disrupting properties and PBT (persistence, bioaccumulative and toxic) characteristics. Due to the lack of empirical hazard data for many of the substances in the CPP-DB, we also used in-silico tools to bridge data gaps. In this presentation we will use the CPP-DB to present an overview of chemicals associated with plastic packaging, their hazards for human health and the environment, and we will highlight priority hazardous chemicals for substitution. Finally, we will discuss the research needed to allow for a more robust hazard characterization and ranking.

325

A Safe by Design framework to support the development of sustainable nano-enabled products for the restoration of works of art

E. Giubilato, University Ca Foscari of Venice / Department of Environmental Sciences Informatics and Statistics; E. Semenzin, Ca Foscari University of Venice / Department of Environmental Sciences Informatics and Statistics; E. Badetti, University Cà Foscari Venice / Department of Environmental Sciences Informatics and Statistics; M. Picone, D. Hristozov, University Ca Foscari of Venice / Department of Environmental Sciences Informatics and Statistics; A. Brunelli, University Ca Foscari of Venice; A. Bonetto, University CaFoscari Venice / Environmental Science, Informatics and Statistics; V. Cazzagon, University Ca Foscari of Venice / Department of Environmental Sciences Informatics and Statistics; A. Volpi Ghirardini, University Cà Foscari Venice / Department of Environmental Sciences Informatics and Statistics; A. Marcomini, University of Venice / Department of Environmental Sciences Informatics and Statistics

Innovative nano-enabled products can overcome some issues of the traditional restoration techniques, especially in the case of complex and unstable materials used in contemporary artworks. However, nanomaterials have been demonstrated to be potentially hazardous to both humans and the environment, thus their application for the conservation of cultural heritage requires a proper assessment and management of potential risks. A Safe by Design (SbD) approach can support the early identification and management of uncertainties and risks during an innovation process and allows for the modification of a product design to avoid undesired properties. Within the EU H2020 "NANORESTART" project, a stepwise SbD framework for the sustainability assessment of nano-based products for restoration has been proposed, taking into account the current EU legislative context as well as the specific features of the innovation process in the restoration field. The proposed framework embraces the SbD concept proposed by NANoREG initiative, which uses the Cooper Stage Gate innovation model as backbone to incorporate SbD in structured innovation management processes for nanomaterials. Six steps build up the framework: (a) state-of-the-art assessment, (b) initial formulation, (c) screening hazard assessment (based on CLP self-classification approach for mixtures), (d) advanced hazard assessment (based on the development of an Intelligent Testing Strategy (ITS)), (e) safety assessment (including the definition of Risk Management Measures), and (f) sustainability assessment (where environmental, social, economic and technical criteria are integrated to compare new to conventional products). The framework is focused on application and post-application stages, while the manufacturing stage cannot be included until the industrial up-scale has been finalized. The SbD framework is currently being applied to NANORESTART advaced nano-based formulations for controlled cleaning and surface protection and consolidation. A specific ITS has been defined, including (i) three bioassays for acute aquatic toxicity, (ii) one bioassay for chronic aquatic toxicity, (iii) a set of tests for cytotoxicity, DNA-damage and mutagenicity. Moreover, specific leaching test protocols are being applied to investigate medium and long-term behaviour of products in post-application stages. The results of the framework application to the most promising formulations will be presented and discussed in detail.

Recent developments in environmental risk assessment for pollinators

326

Managing on the Margins: The confluence of Modern Agriculture and Apiculture

Z. Browning, Brownings Honey Co., Inc.

In the USA, beekeeping is a hobby, a sideline business, and commercial enterprise. Pollinating our backyard gardens, and local communities is made possible by backyard beekeepers with one hive or more. Beekeepers who aspire to increase their honey production, and crop pollination may have hundreds of hives serving not just their local communities, but their state or regions of the country renting their hives to pollinate specialty crops. Commercial beekeepers migrate with their tens of thousands of colonies to pollinate the nation's food supply. Once commercial bees have pollinated the majority of specialty crops they head to summer forage areas for a honey crop. The areas of conflict for bees in agriculture encompasses the urban backyard garden to the almond orchards of California. Pest and pathogens of honey bees are real challenges regardless of location. Habitat loss and pesticide exposure to bees, are greater variables, but no matter what general shared land use is considered for bee hives there are potential conflicts. In each and every case, there are also opportunities to work together with partners and stakeholders for mutually beneficial outcomes. Whether it is water issues, soil erosion and land use issues, or human health concerns with insect bourne diseases, pesticide manufacturers must address how the end user interprets the directions for use, and the cultural practices of the products. Regulatory agencies must acknowledge the pesticide end user's cultural practices of tank mixing pesticides, of fungicide and herbicide impacts upon pollinators, and to combine their agency efforts to protect the entire farm, not just each single crop from each single pest. Sustainable land management whether for food production or protection of human and animal health from disease vectors must be coordinated to ensure profitable production outputs for all stakeholders. Beekeepers can assist in the development of

scientifically supported risk assessment through participation in research development. Beekeepers know bees; researchers know research protocols. To understand how honey bees function under migratory beekeeping and crop field conditions beekeepers need to be part of designing the risk assessment research. Beekeepers, no matter the level of beekeeping or number of hives, are eager to be included in research that will help alleviate the risks to honey bees, and native pollinators. Involving beekeepers in risk assessment and research design is key to ensuring the research premise and results truly reflect the real-world of beekeepers and honey bees. In some ideal world beekeepers would be respected for the ecosystem service their honey bees provide to farmers. Beekeeper and farmer would understand their symbiosis in connection with the health of the crop, and the success of the crop's yield. Both would work to ensure a healthy crop and healthy honey bees to pollinate all crops. One begets the other; each supporting each other: beekeeper and farmer, honey bee and crop (personal examples). As such all stakeholders who rely on honey bees and native pollinators to maintain a healthy ecosystem would balance competing interests to ensure pollinators have clean, plentiful, and diverse forage, pollinators are healthy to provide appropriate pollination services to the ecosystem, and land management is facilitated to reduce soil erosion, protect water, and reduce the threat of disease vectors. (share Bee and Butterfly Fund programs an results)

327

A new multi-dimensional method for evidence synthesis and weighting in bee risk assessment

A. Ippolito, R. Sharp, C. Szentes, D. Auteri, EFSA - European Food Safety Authority / Pesticides Unit

In recent years, neonicotinoids substances have often been in the spotlight, particularly due to their effects on bees. Reporting of highly contradictory results catalysed much attention from the scientific community. The great amount of available studies requests approaches able to ensure an effective integration of the available data. To these purpose, EFSA has developed a novel approach for the most recent conclusions on imidacloprid, clothianidin, and thiamethoxam. Risk due to exposure of bees from residues in pollen and nectar of treated crops is used here as a case study to illustrate the methodology. Oral exposure was estimated by combining data on residue levels in pollen and nectar and estimation of bee food consumption. Together with exposure data, higher tier effect data were the core of the weight of evidence exercise. Each endpoint was identified by four dimensions: (I) the magnitude of the observed deviation from the control, (II) the reliability, (III) the level of exposure in the experiment, and (IV) the length of the exposure. In order to visually illustrate these four dimensions of the endpoints and in order to help the interpretation of each 'line of evidence', a tailored graphical representation was developed. The relevance of each line of evidence was established *a priori*, based on the relationship with the specific protection goals (SPGs). Integration of the lines of evidence followed a stepwise procedure, giving priority to the higher classes of relevance. Single risk assessment results are beyond the scope of this platform, which aims at communicating the features of this new approach. This was, to our knowledge, the first systematic assessment on such a large body of evidence for this specific topic. The exercise combined systematic reviews and weight or evidence, sharing many aspects with meta-analysis techniques. The approach used in this assessment addressed some issues that commonly undermine the reliability of meta-analysis such as the so-called 'file-drawer' problem. Overall, the presented approach ensured significantly more transparency than a fully qualitative expert judgment-driven assessment, but still allowed considering several dimensions in a quali-quantitative way, without oversimplifying the assessment by using fully quantitative measurements that, at present stage, are hardly capable of retaining important qualitative information.

328

PESTICIDE EXPOSURE ASSESSMENT PARADIGM FOR BUMBLE BEES

J. van der Steen, Alveus AB Consultancy; C. Cutler, Dalhousie University / Faculty of Agriculture; D. Goulson, School of Life Sciences, University of Sussex; A. Gradish, University of Guelph / School of Environmental Sciences; O. Klein, Eurofins Agroscience Services Ecotox GmbH / Ecotox Field; D. Lehmann, U.S. Environmental Protection Agency / ORD NHEERL; J. Lueckmann, Rifcon GmbH; B. O'Neill, DuPont Crop Protection; N.E. Raine, C. Scott-Dupree, University of Guelph / School of Environmental Sciences; B. Sharma, FMC Corp/ Global Regulatory Sciences / Global Regulatory Sciences; H. Thompson, Syngenta Ltd / Environmental Safety

Entomopollination is an important biological and economic factor for a number of crops in agriculture. Crop protection is an integral part of agriculture and the use of pesticides, particularly insecticides, is a potential area of conflict between economics and biology. For decades pollinator decisions in registration and re-evaluation of pesticides have been based almost exclusively on first- and higher-tier honey bee toxicity tests This approach has been challenged and regulatory agencies in the EU and USA have started to review this process in respect of non-Apis bees. In this paper we focus on bumble bees (*Bombus* spp.). The potential exposure routes and actual exposure of the bumble bee queen, workers and larvae are mapped and knowledge gaps are identified. The honey bee is used as the reference point to which the differences in biology, foraging and nursing are compared. Some significant differences in susceptibility to pesticides between *Bombus* species have also been identified. It is concluded that there are significant gaps in current knowledge for bumble bee species on both realistic levels for some key exposure routes and cumulative exposure that are not accounted for in the current *Apis* risk assessment protocols.

329

Industry research and approaches to improve the bee risk assessment scheme in Europe

E. Pilling, Dow Agrosciences / REgulatory Sciences; M. Miles, Bayer CropScience UK / Environmental Safety; A. Alix, Dow Agrosciences / Risk Management; J.C. Becker, New York State Dept. of Environmental Conservation / Biology; N. Ruddle, Syngenta Ltd / Product Safety; A. Dinter, Cheminova Deutschland GmbH & Co. KG / Global Regulatory Sciences; A. Sharples, FMC Agricultural Solutions; G. Weyman, ADAMA; L. Oger, ECPA

The crop protection industry recognizes the need to review the bee pollinator risk assessment based on scientific progress. However, the draft EFSA Bee Guidance Document is not a realistically feasible way forward. It is based on extremely conservative assumptions, its study requirements lack clarity and are not workable and guidelines for a number of studies are unavailable or not validated. Industry therefore believes that a revision of the assessment scheme for use by regulatory authorities is needed. Building on an analysis of the proposed developments in the EFSA Bee Guidance Document, we suggest proactive and practical approaches based on analysis of existing data generated thus far on honeybees Using the existing laboratory chronic data generated on adult and larval honeybees, analysis of the EFSA Guidance Tier 1 assessment showed the following: • Almost all substances and uses fail the screening step for chronic risk to larvae and chronic risk to adult honey bees for both spray and solid application types. • For bumble and solitary bees very few substances pass the acute screening step and none pass for chronic risk assessments. • Even known low-bee-toxic substances fail the risk assessment and would need higher tier refinement. In order to pass the assessment, the required doses that would have to be tested would be so high that they would not be technically (solubility) or practically (consumption by the bee) achievable. Results of the Tier 1 assessment following an industry proposed approach will be presented, together with a comparison of existing honeybee and bumblebee data, proposals for protection goals and higher tier testing methodology.\n In its present over-conservative form, the EFSA guidance will make it difficult to register any new or existing insecticide, as well as many herbicides and fungicides. Industry believes that further work and significant revision are required to build a pragmatic, applicable and consistent guidance document within the regulatory framework and has invested much time and money in developing a practical alternative based on the same science.

330

Standardization of an in vitro larval rearing method for stingless bee species Melipona scutellaris for use in toxicological bioassay studies.

A.S. Dorigo, Universidade Estadual Paulista Júlio de Mesquita Filho Unesp Rio Claro / Departamento De Biologia, Centro de Estudos de Insetos Sociais; A. Rosa-Fontana, Unesp - Institute of Biology / Departamento De Biologia, Centro de Estudos de Insetos Sociais; R. Cornelio Ferreira Nocelli, Universidade Federal de São Carlos UFSCar Araras / Ciências Biológica Departamento de Ciências da Natureza Matemática e Educação; O. Malaspina, UNESP Universidade Estadual Paulista Júlio de Mesquita Filho / Departamento De Biologia, Centro de Estudos de Insetos Sociais

Brazilhas the greater diversity of native stingless bees of the world and makes intensive use of pesticides. Thus, forager workers may collect pollen and nectar contaminated and, subsequently, to offer the resources to the brood. Studies on larval phase focus on *Apis mellifera*, since for this species the rearing method is already standardized by the OECD*. However, while in *A. mellifera* the larval food is progressively offered to the brood, in stingless bees the food consists of en masse deposition. This scenario requires the development of techniques which enable to evaluate the exposure of native bees during larval phase to pesticides, and may be used for public authorities responsible for environmental safety for studies on risk assessments. *Melipona scutellaris* is an interesting species to be used as model-organism for risk assessment, since, besides composing the native Brazilian fauna, species from the same genus are recognized as effective pollinator of important crops as eggplant, tomato and sweet pepper. Thus, the present study aimed to propose an *in vitro* larval rearing method of *M. scutellaris*. We extracted the larval food from 20 brood cells per non-parental colony (n =3), for estimating the amount of food consumed by larvae. Before the experiments, the acrylic plates with 100 brood cells were placed in glass Petri dishes containing distilled water to keep the humidity around 95% within the Petri dishes during the first five days of rearing. Each artificial cell received 130µL of larval food and, afterwards, 24-hour-old larvae were placed in the food. Then, the plates were kept in an incubator *at 30ºC and 75% of relative humidity. After the total consumption of the food, the humidity within the Petri dishes was reduced to 75%, adding NaCl This technique was carried out* five times sequentially, evaluating parameters as defecation rate, pupation, emergence, and mortality and morphometry of newly emerged workers. For the morphometric analysis we also evaluated newly emerged work from natural brood combs. The survival rates increased gradually according to the progress of the experiments, increasing from 67.1% in the first to 87.8% in the latter, and the morphometric analyzes indicated newly emerged workers *in vitro* with similar sizes to *in vivo*. The *in vitro* rearing method described showed a satisfactory survival rate, as well as produced newly emerged workers with similar to those from natural conditions, allowing its use in toxicity tests.

331
Poster spotlight: TU038, TU048, TU052

Understanding human and environmental exposure to chemicals in urban systems

332
Consumption of products - a proxy for changes in chemical flows in urban areas and to the environment?
E. Undeman, D. Bolinius, Stockholm University / Baltic Sea Centre; A. Sobek, Stockholm University / Department of Environmental Science and Analytical Chemistry ACES; M. Löf, Stockholm University / Baltic Sea Centre
Is the threat posed to the environment by harmful chemicals increasing or decreasing? Due to the extremely large number of chemicals and variety of adverse effects, it is challenging to develop indicators for the success of our management of chemical emissions. Indicators for efficiency of chemicals management can be based on a) information on production, trade and use of chemicals, b) emissions, c) concentrations in humans and the environment and c) human and wild-life health, with data on the two latter being most relevant, but also difficult and/or expensive to produce for a wide range of chemicals. In this study, we use estimated consumption of products as point of departure to analyze time trends in use and emissions of chemical substances in the urban society and ultimately in the environment. Data on trade of manufactured products available in Eurostat was combined with chemical composition of products and materials compiled in the Commodity Guide hosted by the Swedish Chemicals Agency. The total mass of manufactured products in the northern Europe decreased slightly between 2003 and 2014. Despite this decline, ca 680 substances with significantly positive time trends due to increasing consumption of many products in which they are likely to be present were identified. We conclude, however, that substantially more data on chemical content of products is needed to successfully use consumption of products as a proxy for changes in chemical flows.

333
High-throughput assessment of use-phase exposures to chemicals in building materials
L. Huang, University of Michigan / Dept of Environmental Health Sciences; V. Nguyen, University of Michigan / Department of Computational Medicine and Bioinformatics; O. Jolliet, University of Michigan
Building materials have important contribution to the chemical exposome of the US population. The NHANES data have shown that the blood levels of brominated flame retardants and phthalate plasticizers, tend to be higher in children. The present study thus aims to develop a high-throughput method to determine exposures to chemicals in building materials, which mainly happen during the use phase but are often not considered in traditional LCA. The assessment framework calculates the product intake fraction metric, PiF, to assess consumer exposures during product use, i.e the fraction of a chemical in product that is cumulatively taken in by the users. Based on the building materials Pharos database, 22 product categories for building materials and 218 chemicals were identified. We focus here on 632 unique chemical-product combinations, and assess doses based on PHAROS chemical content data. Chemical emissions from building materials are mainly determined by Dm (diffusion coefficient in building material) and Kma (material-air partition coefficient). Two simple emission models were developed for two types of behaviors: D-limited and K-limited. Use-phase exposures by inhalation, dermal contact, gaseous dermal uptake and dust ingestions were calculated using the PiF metric. Results showed that for SVOCs, the total PiF can be dominated by any of the four exposure pathways. Generally, the chemical-product combinations with low total PiFs are dominated by dust ingestion exposures, while the ones with high total PiFs are dominated by dermal contact exposures. For VOCs, Inhalation PiF always dominates the total PiF. Generally, the PiFs of VOCs are similar between 50 days and 15 years, but the daily intake doses during 50 days are much higher than those during the entire 15-year use phase. In contrast, for SVOCs, the emissions and PiFs gradually increase when the duration of use is extended from 50 days to 15 years, but the daily intake doses remain similar over time. The total intake dose, which combine the total PiFs and chemical content in building product, can range from 100 to 10^6 µg/kg-d for children. This study demonstrates the approach of high-throughput screening of use-phase exposures for chemicals in building products, which can be further integrated into characterization factors and help improve LCA and Chemical Alternatives Assessment (CAA) of consumer products.

334
OPEs - Where do they come from, where do they go? A case study from Toronto, Canada
T.F. Rodgers, University of Toronto / Chemical Engineering and Applied Chemistry; Y. Adjei-Kyereme, University of Toronto / Earth Sciences; C. Yang, University of Toronto / Department of Earth Sciences; S. Harris, Cancer Care Ontario, University of Toronto / Population Health and Prevention, Prevention and Cancer Control; L. Jantunen, Environment and Climate Change Canada; M.L. Diamond, University of Toronto / Department of Earth Sciences
Organophosphate Esters (OPEs) are used in a wide range of consumer products and building materials resulting in their ubiquitous presence in the indoor and outdoor environment. Concentrations of OPEs in indoor air can be relatively high, in the range of 10's ng/m^3 in air. Concentrations are also relatively high in urban media (e.g., low µg/L levels in urban surface waters) and OPEs are consistently found in remote Arctic air. We hypothesize that elevated levels of OPEs in the indoor environment are a source to the outdoor urban environment and then to surrounding regions. To test this hypothesis, we estimated emissions of OPEs to indoor air followed by release to outdoor air, and then compared these "bottom-up" emission estimates to "top-down" estimated aggregate emissions to outdoor urban air. We used the approach of "inverse modelling", whereby emissions are back-calculated from measured air concentrations. "Bottom up" emissions were estimated using the Multimedia Indoor Model that has been used to calculate emissions of PBDEs and PCBs to indoor air, based on indoor air concentrations measured in a study of 51 homes in Ontario, Canada. "Top down" emissions were calculated using the Multimedia Urban Model based on measured Toronto outdoor air concentrations. "Bottom up" emissions to outdoor air ranged from ~3 to 90 kg yr^{-1} for TDCPP and TPhP, respectively. The emissions factors of OPEs from indoor air to outdoor air estimated in this study were within the range estimated by Liagkouridis et al (2017) for bulk emissions to indoor air. These "bottom up" emissions from indoors to outdoors estimated here were one to two orders of magnitude lower than the "top down" estimations, which could be caused by higher emissions from commercial buildings, or through direct emissions of OPEs to outdoor air, such as from insulation. It is clear that elevated indoor and outdoor air concentrations of OPEs are due to emissions from the many products and materials to which they are added. When aggregated over a city scale, indoor air emissions vented to outdoor air were lower, but within one to two orders of magnitude of aggregate air emissions back-calculated from outdoor air concentrations. Indoors, fate is governed by partitioning between air and high sorptive-capacity materials such as PUF in upholstered furniture and carpets and ventilation rate, as with other SVOCs. Outdoors, fate is governed by air advection and water movement, because of the high solubility of OPEs.

335
Drivers of pharmaceutical exposure in urban river systems
E.E. Burns, University of York / Chemistry; L. Carter, University of York / Environment Department; J. Thomas-Oates, University of York / Chemistry Department; A. Boxall, University of York / Environment Department
Pharmaceuticals are heavily used by society and urban environments are one of the main receptors for these molecules. Pharmaceuticals can be released to the urban environment from multiple point sources within a city and can then dissipate as they move away from the point of release. Spatially referenced hydrological models such as LF2000-WQX, GREAT-ER and P*h*ATE have been developed to address this issue. Many pharmaceutical monitoring studies have indicated that temporally significant fluctuations exist in rivers receiving urban wastewater. Currently, spatial models typically overlook the temporal aspect of pharmaceutical concentrations by predicting an annual average concentration usually based on conservative flow estimates. The relevance of these estimates in comparison to real world temporal variations in pharmaceutical exposure has yet to be determined. In this study we attempt to quantify and understand the drivers behind spatiotemporal fluctuations in the concentrations of pharmaceuticals in a city system. Monitoring data for 33 pharmaceuticals were obtained monthly at 11 sites in two rivers that run through the City of York, UK. This data was compared with local monthly prescription, tourism and flow data. In the smaller River Foss, a strong relationship was found between measured concentrations and prescription amounts divided by flow. This trend was not replicated in the larger more urbanised River Ouse. Analysis of pharmaceutical loads indicate that seasonal differences exist in the Ouse, but not in the Foss. Seasonal variability in WwTP removal efficiency is expected to be a factor. In-stream losses of up to 75% were found in the River Foss during summer months and all pharmaceuticals studied followed a sinusoidal loss pattern through time. Significant losses were not identified in the River Ouse. Finally a risk assessment indicated that risk quotient ratios can vary over 3 orders of magnitude (paracetamol) throughout the year and that some of the highest risk pharmaceuticals, raloxifene and loratadine, are temporally transient. Identification of exposure drivers at this unparalleled spatiotemporal scale provides important information that may help improve the accuracy of exposure models and help ensure risks are not overlooked.

336
Past vs. recent emissions of PCBs from the city of Brescia (Italy): coupling monitoring data with a multimedia fate model to investigate PCB regional fate
E. Terzaghi, University of Insubria (Como) / Department of Science and High Technology, Como; M. Morselli, University of Insubria / Department of Science

and High Technology; G. Raspa, Sapienza University of Rome / Department of Chemical Engineering Materials and Environment; A. Di Guardo, University of Insubria / Department of Science and High Technology

Different monitoring campaigns showed higher PCB concentrations in the air of cities compared to rural areas, indicating the presence of ongoing emission sources in urban locations. This is the result of both the past usage of these chemicals in building materials and electrical equipment from which they can volatilize and/or municipal and waste incinerators, e-waste recycling sites and contaminated soils. It was estimated that cities could emit up to about 1 tons/year of PCBs (Gasic et al., 2009; Diamond et al., 2010), which can be transported to rural sites, posing adverse effects to human and ecosystems. The city of Brescia, is characterized by the presence of a factory (Caffaro s.p.a.) that produced PCBs between 1930-1984 and its surrounding areas were found to be heavily contaminated with high concentrations in soil at mg/kg levels. For this reason, about 200 ha of this city were declared National Priority Site for remediation (SIN Brescia Caffaro) by the Italian authorities, representing an important secondary source of PCBs to the atmosphere. The aim of the present study was to investigate the potential of the Brescia city in driving the PCB contamination at regional scale up to 100 km from the point source and the current effect on air concentrations, combining measured data and a multimedia mass balance model. Different sampling campaigns were organized to collect samples of soil and leaves along four 100 km transects that ran in NW, NE, SW and SE directions considering the production plant as starting point. In each sample, the following PCB congeners were determined: PCB 28, PCB 52, PCB 101, PCB 153, PCB 138, PCB 180 and PCB 209. The results were analysed to understand the presence of a spatial gradient of decreasing pollutant concentration with distance from the point source. Furthermore, a spatially resolved version of a dynamic air-vegetation-soil model (SoilPlusVeg model) was used to 1) predict a temporal emission profile from the city, 2) verify if an emission source strength previously predicted for this city justifies soil concentrations in the surrounding area and, 3) evaluate the importance of other sources and processes involved in the contamination at a regional scale. This study shows how a combined modelling approach could be used to unravel the past and recent impact of PCB emissions from a source on the surrounding areas at a regional scale.

337

Using a Dynamic, Aggregate Exposure Model to Identify Far- and Near-Field Contributions to Human PCB Exposure through Time

L. Li, University of Toronto at Scarborough / Department of Environmental Sciences; J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology; F. Wania, University of Toronto at Scarborough / Physical and Environmental Sciences

Humans are exposed to polychlorinated biphenyls (PCBs) through "far-field" sources from the ingestion of contaminated food from aquatic and terrestrial environments and "near-field" sources from building materials in indoor environments. Earlier models that consider far-field exposure only tend to underestimate historical body burdens. In this presentation, we will explore the time-dependence of the relative contribution of far- and near-field routes to aggregate human exposure to PCBs, to explain the discrepancy between previous far-field only model predictions and observations. We develop a mechanistic model that incorporates dynamic substance flow analysis, indoor-urban-rural fate modeling, and bioaccumulation and human toxicokinetic modeling, enabling a dynamic and mechanistic description of the complicated continuum from annual industrially processed amount of PCBs to the human uptake rate. The model is applied to simulate the time-variant exposure of Swedish women to PCB-28 and PCB-153 from 1930 to 2030. In terms of a female's lifetime longitudinal exposure, our modeling indicates that route-specific contributions to aggregate human exposure change with age and differ among birth cohorts: Near-field exposures are notable during childhood and teenage years, as well as for female cohorts born earlier, when the indoor environment was more highly contaminated. In terms of the exposure of individuals of different ages at the same time (i.e., the cross-sectional age-exposure relationship), the dominance of far- or near-field routes differs little among ages, but is largely dependent on the time a cross section is "sampled": Near-field routes dominate in the past (e.g., the year 1956) whereas far-field routes become predominant more recently (e.g., 1986 and 2016). This finding suggests that the dearth of PCB biomonitoring studies before 1990s has also contributed to the general belief that far-field sources dominate human PCB exposure. It further implies that there may also be a similar shift from a near-field dominance to a far-field dominance among a wide range of currently-used indoor chemicals, such as flame retardants. This work improves our understanding of the exposure dynamics, which would be beneficial for future pertinent management actions for exposure reduction and prevention.

Microplastics in freshwater and terrestrial systems - fate, monitoring and biological interactions (III)

338

Modelling of the environmental release of macro- and microplastics for seven different polymers

D. Wenger, Empa Swiss Federal Laboratories for Materials Science and Technology; B. Nowack, EMPA

Microplastic exposure is a burning topic in environmental research, but few large scale exposure studies have yet been performed in freshwaters. Assessing the emissions of plastic is possible using a life-cycle oriented approach, and permits to compare the flow magnitudes for different sources. With our ongoing study, we aim at providing large scale predictions of macroplastic and microplastic exposure in Europe using Probabilistic Material Flow Analysis (PMFA). The environmental flows of seven different commodity thermoplastics are estimated based on societal data. The polymers are chosen for their popularity of use and the frequency at which they are reported in the environment: low-density polyethylene (LDPE), high-density polyethylene, polypropylene (PP), polystyrene (PS), expanded polystyrene (EPS), polyvinyl chloride (PVC) and polyethylene terephthalate (PET). The probabilistic aspect of the PMFA framework permits one to account for the various uncertainty sources and give a quantitative estimate of the final confidence in the results. In a first step, the anthropogenic life cycle of these seven polymers is modelled, from production to end of life of a total of 35 product categories. Various trade flows are included, as well as the life cycle of textile applications. This enables us to present an accurate estimation of the European and Swiss consumptions. In a second step, every stage of the life cycle is analyzed and the voluntary or inadvertent emissions are assessed. The emission-specific pathways between the anthroposphere and the environment are described, and the resulting environmental flows are compared. Modelling the emission pathways between prime release and final discharge enables to pin-point the principal plastic pollutions sources and the possibilities for pollution mitigation.

339

Modelling Microplastics in Rivers in the US

A. Koelmans, Wageningen University / Aquatic Ecology and Water Quality Management; C.M. Holmes, Waterborne Environmental, Inc.

Pollution with nano- and microplastics (MPs; particles < 5 mm) is a topic of emerging concern and as such receives growing interest. Although measurement and monitoring data are indispensable, there also is a need for estimated concentrations to enable prospective assessments and to guide analysis of retrospective ecological analyses. Besseling et al (2017) provided the NanoDUFLOW model, a detailed MP aggregation-sedimentation model integrated in a hydrological and particle transport model. A much larger scale model potentially suitable to simulate MPs originating from WWTPs is the iSTREEM® model, which has been developed to estimate chemical concentration distributions for all rivers and streams of the USA receiving WWTP discharges. Here we merge these two riverine modeling worlds: NanoDUFLOW with iSTREEM for MPs, to simulate spreading of MPs from WWTP point sources in US waterways and to assess export to the Great Lakes for a range of particle sizes. This combines the mechanistic realism of NanoDUFLOW, accounting for formation and settling of heteroaggregates, with the US well-established iSTREEM implementation. We modeled floating as well as non-buoyant MP, for diverse sizes, from 100 nm to 10 mm, a range that incorporates the theoretical parabolic size-settling relationship reported by Besseling et al (2017). Depth dependent in-stream first order removal rate constants simulated with NanoDUFLOW were combined with standard iSTREEM output (which was used to simulate the emission, transport and water column concentrations of MP) in an Excel-based post-processing phase, without modifing the iSTREEM model directly. Simulations were spatially explicit with MP concentrations being modeled for the Sandusky River watershed in Ohio (~3500 km^2). Emissions were based on per capita usage and population served for each of the 20 WWTPs within the watershed. Modelling results show the effects of population density, MP size and density on riverine concentrations and export to Lake Erie. Buoyant as well as the smallest non-buoyant MP fractions can be transported over long distances, reaching receiving waters such as the Great Lakes. In contrast, larger non-buoyant MPs settle more locally in the vicinity of the WWTPs. Simulating depth-dependent removal as demonstrated here could be incorporated into the core iSTREEM code in order to efficiently process all US waterways impacted by WWTPs, as well as examining ultimate marine discharge proportions by particle size. \n

340

The routes to uptake and bioaccumulation of nanoplastics in freshwater sediments

R. Cross, C. Liddle, University of Exeter; T.S. Galloway, University of Exeter / Biosciences

Plastic contamination of freshwater sediments is well documented, and whilst quantitative measurements of nanoplastics in sediments are beyond current capabilities, recent models predict plastics < 1000 nm in size will be effectively retained in freshwater sediments. Current debate considers whether micro and nanoplastics can be defined as persistent organic pollutants (POPs) in their own right. In order for such a classification, four criteria must be met, one of which is for a compound to be bioaccumulative. Nanoplastics do not adhere to the classical concept of molecular bioaccumulation due to their particulate form. However, assessing their potential uptake and transfer through food chains can contribute towards the data required to inform decisions as to the suitability of classifying nanoplastics as POPs. This study provides initial insights to address this question in an ecologically relevant system, using the freshwater aquatic worm *Lumbriculus variegatus*, representing an entry point for nanoplastics from abiotic compartments of sediments, into biota. The role of surface functionalisation of fluorescently dyed nano-polystyrene (50 nm) upon their uptake is systematically examined using a combination of techniques including a novel fluorescence assay and fluorescence microscopy. A series of exposure scenarios are used to test the efficacy of different routes of uptake into the worm. Associations of nanoplastics to the worms' surface are examined in waterborne exposures, whilst dietary uptake is tested using nanoplastics associated with an algae food source. The accumulation of nanoplastics directly from contaminated sediments is also investigated, alongside the fate of these particles in sediments to assess the relationship between nanoplastic mobility and accumulation. Results indicate that pristine nanoplastics are accumulated or internalised both in waterborne exposures and from dietary uptake of a nanoplastic associated algal food source, with carboxylated and aminated plastics experiencing greater uptake than non-functionalised particles. Sediments on the other hand, reduced the availability of these particles for uptake into the worms, potentially though strong associations of the nanoplastics to solid constituents of the sediment. Ongoing work addresses the potential for formation of an "ecocorona" to alter the bioaccessibility of nanoplastics for the worms. These results will also be presented during the platform presentation.

341

Life-history and biochemical responses of Chironomus riparius exposed to different sized microplastics

C. Silva, CESAM & University of Aveiro; J. Pestana, CESAM & University of Aveiro / Biology; C. Gravato, Faculty of Sciences, University of Lisbon / department of Biology & CESAM

Freshwater basins are an integral part of microplastics life-cycle, being a repository of plastic micro-debris. In fact, the levels found so far are similar to those found in marine environment and shoreline regions. The deposition and persistence of plastic micro-debris in sediments (lakeshores and riverbanks) makes them long-time available for benthic species. The objective of this study was to investigate developmental, and physiological effects induced by the presence of polyethylene microparticles (PE) in *Chironomus riparius*, due to its key-role in the freshwater ecosystem. For that partial life cycle tests using different sized polyethylene particles (PE 40-48 µm; PE 125 µm and PE 350 µm) allowed evaluation of effects on C. riparius larval growth and emergence patterns while acute exposures were used to assess effects in parameters related to neurotransmission (AChE); antioxidant defences and biotransformation (CAT, GST total glutathione levels); oxidative damage (LPO); cellular energy allocation (CEA) and immune response (phenoloxidase). Exposure to PE 40-48 µm caused deleterious effects at lower concentrations in comparison with larger particles in several parameters: larval growth and development time of both male and female imagoes and on emergence rate. PE 40-48 µm were then selected to assess effects on physiological homeostasis. Acute exposures to PE 40-48 µm generated alterations in C. riparius larvae antioxidant and biotransformation enzymes activities (CAT, GST and total glutathione) and activation of immune response (induction of phenoloxidase). Larvae exposed to microplastics showed also a depletion in energy reserves. Our study highlights the potential deleterious effect of microplastics for aquatic invertebrate populations. Results will be discussed in terms of effects of different sized plastic particles on different levels of biological organization within freshwater invertebrates and on the needed and ongoing research aiming to address the long term and indirect effects of these particles for natural populations and ecosystem functioning.

342

The effects of rigid and flexible Polyvinyl chloride (PVC) microplastic particles on the transcriptome of Daphnia magna

B. Trotter, University of Bayreuth / Animal Ecology I; I. Schrank, J. Dummert, A. Weig, C. Laforsch, University of Bayreuth

Microplastics are ubiquitous in aquatic ecosystems, posing as a threat to biota of all trophic levels, as they have the potential to leach out incorporated additives, such as plasticizers, to the surrounding medium. Yet the question arises, if possible effects on biota are based on the polymer type alone, or if incorporated additives are responsible for the observed effects, as they might desorb from the polymer matrix. With our transcriptome analysis, which was conducted via the use of a microarray, we showed that *Daphnia* reacts substantially different to the chronic (31 days) exposure to rigid PVC or flexible PVC (with diisononyl phthalate (DINP) as a plasticizer) with changes in gene expression. Rigid PVC caused a fivefold up regulations in a total of 19 genes (15 up-regulated and 4 down-regulated) related to structural components of the cuticle, molecular functions as well as serine type endopeptidase inhibitor activity. Flexible PVC exposition lead to a fivefold change of a total of 267 genes (238 up-regulated and 29 down-regulated) related to the GO terms of proteolysis, carbohydrate and chitin metabolism, Vitelline membrane formation, yet most genes were related to immune response. Our attained results imply that flexible PVC had a more severe effect that might be attributed to the leaching of DINP or on the altered biofilm formation on these two different microplastic particles. Therefore our results highlight, for the first time that differences in additive composition (absence or presence of a plasticizer) can lead to substantial differences in effects on aquatic species.

343

Poster spotlight: TU149, TU150, TU151

When ecotoxicology meets trophic ecology

344

Does stress propagate along aquatic food chains? An experimental approach with a tri-trophic brown food chain

E.L. Fernandes, University of Koblenz Landau; M. Bundschuh, Swedish University of Agricultural Sciences / Department of Aquatic Sciences and Assessment; R. Schulz, University of Koblenz-Landau / Institute for Environmental Sciences; R. Schaefer, University Koblenz Landau / Institute for Environmental Sciences

Pollution is a major driver of ecosystem change resulting in alterations in food webs and associated ecosystem processes. Some pollutants such as systemic insecticides are taken up by terrestrial plants and may enter aquatic systems with plant parts such as leaves that are an important energy source in stream food webs. Thereby, food web interactions across the aquatic-terrestrial boundary may be affected through alterations in food quality. Here we studied the effects of a systemic insecticide, the neonicotinoid imidacloprid, and their potential propagation in a brown food chain. The model food chain consisted of imidacloprid contaminated terrestrial leaves (alder, *Alnus glutinosa* Gaertn.), aquatic merolimnic invertebrate decomposers (*Protonemura* sp.) and predators (*Isoperla* sp.). Effects of imidacloprid on survival and growth of decomposers and its leaf processing were assessed in a microcosm setting. Therefore, decomposers fed on control or contaminated leaves for 3 days on the microcosms. Every 6 hours the number of dead individuals was recorded. Potential propagation of imidacloprid effects were assessed by transferring surviving decomposers to cages containing the predator. The cages were deployed in an unpolluted stream for 9 days after which predators' growth was analysed. Imidacloprid concentrations increased within the contaminated microcosms over time. The presence of imidacloprid in the water was associated with lower survival of decomposers and leaf decomposition in contaminated microcosms compared to the control. Furthermore, decomposer's biomass and length decreased in the contaminated but not in the control microcosms. Predators hunting decomposers from contaminated microcosms decreased in body size compared to the control. Systemic insecticides in plant materials can be a relevant source of exposure for decomposers with consequences for their population dynamics (e.g. increased mortality and reduced growth) and the associated ecosystem processes (reduced leaf decomposition). The effects can propagate through food chains and result in indirect effects in predators. Future studies should elucidate the spatiotemporal dynamics of exposure and uptake given that imidacloprid leaches from plant material and may influence downstream food webs directly and indirectly.

345

Accounting for trophic relationships in fish bioconcentration models applied as emergent-pollutants risk-assessment tools

h. baveco, Wageningen Environmental Research; J. Deneer, Wageningen Environmental Research / ERA team; M. Faust, Faust & Backhaus Environmental Consulting; J. van Gils, DELTARES; C. Lindim, Stockholm University / SEAC; A. Focks, Alterra Wageningen University and Research Centre / Environmental Risk Assessment Team

In the context of the SOLUTIONS EU FP7 project, we applied non-steady state bioconcentration models to predict concentrations of organic compounds in fish. A foodweb perspective was taken, accounting for uptake from water as well as from food, and accounting for different trophic relationships for several fish species used for human consumption. The foodweb bioconcentration model will be applied for a large number of emerging pollutants and a large number of locations (around 25,000 sub-catchments in the major European catchments). Water concentrations at these locations are obtained from chemical fate modelling using the STREAM-EU model. As a case study, results for 24 WFD priority substances are presented here. Predicted concentrations will be input to human health risk assessment. The model also provides insight in how trophic relationships together with species and compound characteristics detemine bioconcentration and thus ecotoxicological risk. The core of the foodweb model is a bioconcentration model for neutral and ionisable organic compounds (Arnot & Gobas 2004; Armitage et al. 2013) underlying each fish component. It calculates for given environmental conditions (pH and temperature) the uptake and elimination rates defining the one-compartment model of the internal concentration dynamics. The considered foodweb contains fish components with different trophic relationships, representing fish species used for human consumption with different body size and lipid content, chosen to represent extreme cases with respect to expected bioconcentration. Internal concentrations in phyto- and zooplankton are assumed to be in instantaneous equilibrium with water concentration. For 24 WFD priority substances concentration timeseries per sub-catchment from the STREAM-EU model were used as input to the foodweb bioconcentration model. Results were summarized in monthly and annual maximum and mean concentrations for all foodweb components in each sub-catchment and displayed in maps covering the

EU. From these results, e.g., median concentrations can be calculated per catchment or over all catchments. Concentrations in fish depend on local exposure pattern and differ per subcatchment. They also depend on trophic position in a compound-specific way: any of the three fish components can be worst-case. To indicate risk to human health, concentrations need to be related to standards of e.g., acceptable daily intake.

346

Model-based explorations of the variability in lake trout BAFs caused by physiology and trophic relationships

S. Baskaran, University of Toronto - Scarborough / Chemistry; J.M. Armitage, University of Toronto - Scarborough / Physical and Environmental Sciences; F. Wania, University of Toronto at Scarborough / Physical and Environmental Sciences

Because dietary uptake of fish is often a major vector of human exposure to persistent organic pollutants (POPs), much effort is directed towards a quantitative understanding of fish bioaccumulation with the help of mechanistic models. Such models require the input of the growth, feeding and respiration rates of a fish. However, often little consideration is given to the interdependency of these physiological parameters. Here, we calculate the bioaccumulation factor (BAF) of hypothetical POPs, with log K_{OW} values ranging from 4.5 to 8.5, in lake trout (*Salvelinus namaycush*), with a food web bioaccumulation model that uses bioenergetic equations to ensure that the physiological parameters applied to a species are internally consistent (i.e. energetically balanced). Empirical growth rates and diets for lake trout in six Canadian lakes (Great Slave Lake, Lake Ontario, Source Lake, Happy Isle Lake, Lake Opeongo, and Lake Memphremagog) are used to determine feeding rates. Respiration rates were derived based on the routine metabolic rates and the population specific activity coefficients (multipliers). When comparing differently sized lake trout within a lake, larger fish tend to have the highest BAF, because they allocate less energy towards growth than smaller fish and have higher activity levels. When comparing fish from different lakes, diet composition and prey energy density become important in determining BAF in addition to activity and the amount of total energy allocated to growth. Specifically, fast growing Lake Ontario lake trout, feeding on slow growing alewife, have higher BAFs; while slower growing small lake trout in Happy Isle and Source Lakes have low BAFs because they feed on invertebrates, which are low in the food chain. Moreover, very large trout in Great Slave Lake with higher energy requirements feeding on an energy rich diet have lower BAFs compared to the same sized trout in Lake Memphremagog feeding on less lipid rich rainbow smelt.

347

Influence of an agriculture-associated toxicity gradient on a riparian predator-prey relationship in Romania

N. Graf, P. Dittrich, University of Koblenz Landau; M.H. Entling, University of Koblenz-Landau / Institute for Environmental Sciences; K. Frisch, M. Link, V.C. Schreiner, E. Szöcs, University of Koblenz Landau; R. Schaefer, University Koblenz Landau / Institute for Environmental Sciences

Riparian areas are complex systems and linked to streams via fluxes of material and organisms. Agricultural land use related stressors can differentially alter arthropod communities in water and on land, resulting in complex response patterns of aquatic-terrestrial predator prey relationships. Therefore complex response patterns may arise in terrestrial predators feeding amongst others on aquatic prey. While agricultural landscapes in most European countries have been intensified, resulting in the co-occurrence of pesticide use, habitat degradation and excessive nutrients, traditional low-intensity agriculture can still be found in Central Romania. We investigated the potential effects of land use related stressors including pesticides on aquatic-terrestrial predator-prey relationships using stable isotope analysis. We sampled spider communities and measured their intake of aquatic prey in 19 riparian areas around Cluj-Napoca, Romania. To investigate the spiders' diet, aquatic and terrestrial prey organisms were caught. We collected the orbweb *Tetragnatha* sp. and the ground-dwelling spider *Pardosa* sp. to analyse their stable carbon and nitrogen signals. Nutrient concentrationsin the stream were slightly positively associated with the proportion of aquatic prey of *Pardosa* sp. This may be explained by nutrients in the streams increasing productivity of primary producers and in turn resulting in a larger biomass of emerging insects. The toxicity gradient was negatively related to the number of individuals of spiders and the number of spider species. Although we found clear differences in the proportion of consumed aquatic prey of spiders, the proportion of it was not related to the toxicity gradient. Thus, potential effects of pesticides in the aquatic system did not affect the proportion of consumed aquatic prey organisms of riparian spiders. We found less individuals of *Tetagnatha* sp. when they consumed more aquatic prey. This might be due to an accumulation of toxicants in the spiders or a higher competition between the species due to resource shortage. Riparian spiders can be affected directly by agricultural land use but also indirectly via prey consumption. Changes in riparian spider communities and their diets are presumably driven by multiple stressors.

348

Migration effects on pollutants in eggs of Arctic-breeding geese

D.J. Hitchcock, University of Oslo; M.J. Loonen, University of Groningen / Arctic Center; N.A. Warner, NILU - Norwegian Institute for Air Research / Environmental Chemistry; D. Herzke, NILU - Norwegian Institute for Air Research / FRAM Centre Tromso; I.M. Tombre, NINA - Norwegian Institute for Nature Research; P. Shimmings, BirdLife Norway; L.R. Griffin, WWT Caerlaverock Wetland Centre; . Varpe, University Centre in Svalbard; T. Andersen, University of Oslo / Department of Biosciences; K. Borga, Department of Bioscences, University of Oslo / Department of Biosciences

Arctic breeding barnacle geese (*Branta leucopsis*) are a herbivorous species that migrate from the United Kingdom to the Arctic every summer to breed. Females utilise resources towards reproduction along the flyway, including distant resources (United Kingdom and Northern Norway) and local resources (Svalbard) relative to the breeding grounds. Depending on migration route, allocation of resources towards egg production may differ within a breeding goose population. Thus different energy sources may also affect how pollutants are taken up and deposited to eggs, including those which are both protein and lipid soluble. In order to examine the effect of migration on pollutants in eggs, a field study was carried out during the breeding season of 2016. Eggs (N = 60) were collected at an island breeding colony in Svalbard and several hundred grams of vegetation (N = 15 sample collections) was collected at different sites along the gooseâ_s flyway. Resightings of ringed geese also took place in Northern Norway. Egg and vegetation samples were analysed for stable isotope of carbon (δ^{13}C) and nitrogen (δ^{15}N), as well as pollutants including protein-associated poly- and perfluoroalkyl substances (PFASs), lipid soluble polychlorinated biphenyls, and hexachlorobenzene (PCBs and HCB). Stable isotope ratios in eggs could not be precisely attributed to either local or distant resources due to overlapping signal, but stable isotopes of nitrogen appeared to be fuelled by distant resources in United Kingdom and Northern Norway. When examining pollutants individually, there was no relationship found between stable isotopes and pollutant concentrations. However, when combining pollutants together as part of a multivariate analysis, it was found that egg laying date contributed to the variation in PFAS levels across eggs, but not for PCBs. Protein associated pollutants (PFASs) may be more influenced by migration strategy than lipid soluble contaminants (PCBs and HCB), due to protein stores being a more limiting energy source during migration than lipids. This knowledge furthers our understanding on how pollutants operate within Arctic terrestrial ecosystems, and the interaction between climate and pollutant bioaccumulation in highly seasonal environments.

349

Trophic Magnification of Persistent Organic Pollutants Within A Terrestrial Food-Web of An Avian Top Predator, the Cooper's Hawk (Accipiter Cooperii)

K. Fremlin, SFU / Department of Biological Sciences; J.E. Elliott, Environment Canada / Science Technology Branch; F. Gobas, Simon Fraser University / Resource & Environmental Management; K. Drouillard, Great Lakes Institute for Environmental Research University of Windsor; D. Green, Simon Fraser University

Several types of legacy persistent organic pollutants (POPs), such as PCBs and DDE, and emerging POPs like perfluorinated compounds are released from multiple sources into the ambient environment and are known to negatively impact endocrine and physiological functions within exposed wildlife. Protocols to assess bioaccumulation of these persistent chemicals within terrestrial systems are far less developed compared to aquatic systems. Presently, regulatory agencies in Canada, the USA, and the EU use only bioaccumulation information for fish to assess the bioaccumulation potential of chemicals. However, recent studies have shown that some chemicals that are not bioaccumulative in aquatic food-webs do biomagnify in terrestrial food-webs. To better understand the bioaccumulation behaviour of chemicals in terrestrial food-webs, we aim to produce a food-web model to assess the biomagnification of POPs in an apex avian predator, the Cooper’s hawk. Over 100 samples were collected from various trophic levels of the food-web including hawk eggs, songbirds, invertebrates, and berries. All samples were analyzed for a number of contaminants listed as priorities for monitoring by the Chemical Management Plan of the Canadian federal government. Stable isotope analysis of δ^{13}C and δ^{15}N signatures of hawks, songbirds, invertebrates, and berries was used to estimate the trophic position of each organism. Censored regression by maximum likelihood estimation was used to assess the relationship between the natural logarithm of each POP lipid equivalent concentration and trophic position. Trophic magnification factors (TMFs) were determined as the antilog of the regression slope. TMFs of PCBs, PBDEs, and OCPs ranged from 1.64 to 26.31, 2.87 to 14.88, and 0.61 to 38.40, respectively. Indicating that most legacy POPs are biomagnifying in this terrestrial food-web. TMFs of PFCs ranged from 11.8 to 544.6, indicating that PFCs are biomagnifying in this terrestrial system. Overall, these terrestrial TMF values for legacy POPs were comparable to or higher than TMF values determined for several aquatic systems, and the terrestrial TMF values for the PFCs were considerably higher than TMF values found in aquatic systems.

Integrated approaches in ecotoxicology: bridging the gap between experimental toxicology and mechanistic modelling

350

Toxicokinetic-toxicodynamic models as new tools for environmental risk assessment

S. Charles, University Lyon 1 / Laboratory of Biometry and Evolutionnary Biology; V. Baudrot, Université Lyon 1; A. Focks, Alterra Wageningen University and Research Centre / Environmental Risk Assessment Team

Today, the Environmental Risk Assessment (ERA) for chemicals is based on fitting standard dose-response (DR) models to quantitative data. Such data are usually collected from standard toxicity tests, from which the concentration leading to 50% lethality or effect (LC_{50} or EC_{50}) is usually estimated at the end of the exposure. In this form of evaluation, the fact that endpoints are monitored over time is not fully exploited. Standard DR models do also assume that the exposure concentration remains constant during the experiment, what makes it difficult to extrapolate the results to more realistic scenarios, for example to effects under time-variable exposure profiles. To overcome this gap at the organism level, the use of the toxicokinetics-toxicodynamics (TKTD) models is suggested, because TKTD models describe the effects of a substance by integrating the dynamics of exposure [1]. Indeed, TKTD models have many advantages in terms of mechanistic understanding of the chemical mode of action, of deriving time-independent parameters, of interpreting time-varying exposure and of making predictions for untested and time-variable exposure. Another advantage of TKTD models for ERA is that they make it possible to calculate any $LC/EC_{x,t}$, for arbitrary effect strength x and any given exposure duration t. Nevertheless, being based on differential equations their mathematical complexity makes it necessary to numerically integrate the equations when fitting the model to data, so that in practice TKTD models are still not used very often. In order to allow more users to use TKTD models for regulatory risk assessment without suffering any technicalities, the availability of a software environment for an easy handling of TKTD models would be of great value. That is the aim of the R package 'morse' in its new version 3.0.0. [2]. In this presentation, we will give an overview of TKTD models with a focus on the General Unified Threshold model for Survival (GUTS, [3]). Handling GUTS models within R will be then illustrated with one example dataset. Finally, the added-value of TKTD models for ERA will be discussed based on a number of different datasets.

351

Lethal and sublethal impacts of neonicotinoids and copper nano-pesticides on the energy budgets of an estuarine amphipod

E.B. Muller, University of California, Santa Barbara / Marine Science Institute; J. Couture, H.S. Lenihan, University of California Santa Barbara / Bren School of Environmental Science and Management; J. Means, University of California Santa Barbara; K. Tran, C. Vignardi, University of California Santa Barbara / Bren School of Environmental Science and Management

Estuaries are major recipients of run-off pesticides from agricultural and urban origin, including neonicotinoids and nano-based copper formulations. Neonicotinoids have rapidly become the most widely used insecticides globally, and have been implicated for harming pollinators and non-target species at levels below existing US EPA toxicity thresholds. With most research conducted on inland agricultural areas, there are scant data on impacts to coastal and marine environments. Advantages of nano-based copper formulations over ionic froms include better application control and slower release of copper, but the size and shape of nano formulation change the environmental behavior and toxicity profile of copper. The aim of this research is to assess the impact of those modern pesticides, in particular CuPRO, Kocide and imidacloprid, on an estuarine non-target species, the amphipod *Leptocheirus plumulosus*. The assessments are based on dynamic energy budgets. With this approach, due to its process oriented structure, toxicity assessment statistics are independent of exposure time and of choice of sublethal endpoints. Copper speciation was rapid, with little change observed in dissolved and aggregated copper after 1 day. The copper accumulation profile did not depend on the form in which copper was administered, but increased with dose. The impact of copper on respiration showed a regular dose-response pattern with little difference amon copper formulations; the no-effect body burden and the body burden at which the respiration rate doubled was estimated from pooled data at 149 and 303 µg Cu/ g DW, respectively. Similarly, survival declined in a normal dose-response manner with no difference among formulations. The no-effect body burden and LC_{50} at day 7 were estimated at 188 and 291 µg Cu/ g DW. Sublethal impacts were more pronounced than lethal ones, especially before day 7, indicating that cannibalism has an mitigating impact on the decline of the total amount of biomass in the container. We conclude that the toxicity profiles of nano-copper and ionic copper are similar for this amphipod, which indicates that this species, being a detritus feeder, readily ingests nanoparticles.

352

A biology-based model to analyze growth data of earthworms exposed to copper at different development stages

S. Bart, J. Amossé, C. Mougin, A. Péry, INRAAgroParisTech; C. PELOSI, INRA (Institut National de la Recherche Agronomiqu

Assessing effects of a contamination on populations require getting data on the whole life cycle and accounting for differences of sensitivity throughout this life cycle. Copper contamination is one of the greatest concerns in agricultural soils, especially in vineyards. This contamination may have negative effects on soil fauna, such as earthworms, which are recognized as ecosystem engineers providing very valuable ecosystemic services. To assess effects of copper on earthworm populations, we tested the impact of a commercial formulation of copper fungicide, i.e., Cuprafor Micro® (50% copper oxychloride), on different endpoints, including growth, for the earthworm species *Aporrectodea caliginosa*, one of the dominant earthworm species in agricultural fields. We performed original tests assessing the impact of copper on the growth at 3 different development stages with many times of measurements: new hatchling individuals (10-15 mg), small juveniles (90-110 mg) and large juveniles (260-340 mg). To analyze simultaneously all the data, we developed and used a biology-based model. This model is based on the DEB (Dynamic Energy Budgets) theory. A DEB model was set up and validated under controlled conditions, using different food conditions. The growth data were then analyzed with a toxicokinetics model (accounting for the influence of growth and differences between development stages) coupled with a DEB-based toxicodynamic model. Our results showed a drastic inhibition of growth once a No Effect Concentration, estimated at 65 mg kg^{-1} of copper for all stages, was exceeded. The time-profile of the effects was fully accounted for by the model. In conclusion, our model provided a relevant analysis of the toxicity data and provided understanding of the mechanisms of copper toxicity to earthworm growth, throughout development. The next step of our research will be to combine our biology-based model and reproduction data in a dynamic population model.

353

Connecting suborganismal and organismal responses using Dynamic Energy Budget Modeling and the ecological model species Fundulus heteroclitus exposed to dioxin-like chemicals

L.M. Stevenson, UCSB / Ecology, Evolution and Marine biology; E.B. Muller, University of California, Santa Barbara / Marine Science Institute; D.E. Nacci, B. Clark, U.S. EPA / Atlantic Ecology Division; A. Whitehead, University of California Davis / Environmental Toxicology; R.M. Nisbet, University of California Santa Barbara

Comprehensive and efficient management of ecological risk depends on our ability to quantitatively extrapolate the effects of stressors across levels of biological organization. Adverse Outcome Pathways (AOPs) connect sub-organismal mechanistic molecular data to organismal outcomes, while bioenergetics models, such as Dynamic Energy Budget (DEB), can extrapolate from individual-to ecological-level effects of toxicant exposure. However, the connection between the two modeling frameworks remains a challenge. The molecular mechanisms underlying Key Event (KE) relationships defined in AOPs are often poorly known, even for well-studied compounds; thus, the mechanistic linkages between KEs and effects on DEB processes are often difficult to discern. Further, AOPs whose adverse outcome is lethality may not be informative to the sublethal processes important for bioenergetics. We address these challenges through theoretical and empirical efforts. We connect AOP KEs to DEB processes through a model of damage dynamics. The model predicts regulated but increasing concentrations of damage as the result of toxicant exposure and also tipping points when damage outpaces regulatory feedbacks, leading to mortality (Klanjscek et al. 2016). The connection between damage dynamics and DEB will be influenced by empirical and theoretical observations, but potential linkages include damage causing an increased maintenance cost or specific impacts on development. Specifically, we are studying the effect of DLCs on *Fundulus heteroclitus* (Atlantic killifish). DLCs are of particular interest in this species due to the large intraspecific variability in sensitivity. There is extensive data describing the toxic effects of DLCs through activation of the AhR pathway, but the toxic mechanism is poorly understood. Further, sublethal effects of DLCs are less studied, but preliminary data indicate that sublethal PCB126 exposure leads to slower growth in larval killifish (Nacci unpublished data). Therefore, this system offers a framework to test our ability to connect effects observed at the suborganismal level to bioenergetic processes through AOP and DEB modeling. We will measure suborganismal effects of DLCs (cytochrome P450 activation & transcriptomics) along with effects on development, growth, and reproduction. We will give an overview of our objectives and methods and report preliminary findings fitting DEB models and predictions of the effect of DLCs on the bioenergetics of killifish.

354

Quantitative Adverse Outcome Pathway Modelling of Endocrine Active Toxicants in Rainbow Trout

I.R. Schultz, NOAA NWFSC / Marine Science Laboratory; L. Harding, University of Washington / Aquatic and Fishery Sciences; C. Monson, University of Washington / School of Aquatic and Fishery Sciences; K. Gillies, Pacific NW National Laboratory; J.A. Spromberg, Northwest Fisheries Science Center - NMFS / Northwest Fisheries Science Center; T. Cavileer, Unviersity of Idaho; G. Young, Advisian WorleyParsons Group / Aquatic Sciences; J. Nagler, University of idaho / Department of Biological Sciences; P. Swanson, NOAA-NWFSC

We are investigating best approaches to utilizing in vitro derived toxicity data for estimating impacts on reproduction in trout and salmon. Our goal is to facilitate development of quantitative in vitro – in vivo extrapolation (IVIVE) methods to support adverse outcome pathway (AOP) based toxicity testing. We tested a diverse suite of toxicants using cellular assays based on the female rainbow trout pituitary, ovary and liver. Each assay measures an essential reproductive endocrine function such as estrogen synthesis and secretion by ovarian follicles. In vitro results are extrapolated to metrics of reproductive performance (fecundity, fertility, egg size) in trout using a mathematical model of the trout hypothalamus-pituitary-ovary-liver (HPOL) axis. We evaluated IVIVE by comparing predicted effects against laboratory results obtained from a yearlong exposure of female trout to four different chemicals: tamoxifen (biotransformed into the anti-estrogen 4-OH-tamoxifen), prochloraz (interferes with oocyte maturation), fluoxetine (SSRI pharmaceutical largely negative in our in vitro assays) and trenbolone (potent synthetic androgen). A single water exposure level was tested for each chemical, guided by preliminary studies and a desire to use a maximum tolerated exposure that still allowed spawning to occur. Laboratory exposures began 10 d after the first spawning cycle and lasted until time of ovulation and completion of the second spawning cycle 12-14 months later. Trout were euthanized and total fecundity determined along with egg mass and diameter, fertility, hatching success and larval growth. Results indicated no effect on fecundity was observed except after the 60 ng/L trenbolone exposure, which caused regression of ovarian growth and a failure to spawn in all exposed fish. Fluoxetine had no effect on reproduction. The most significant effects on egg quality occurred after the 500 ng/L tamoxifen (30% decrease in egg mass and diameter) and 20,000 ng/L prochloraz (increase in atretic / non-fertile eggs) exposures. The tamoxifen induced decrease in egg size translated to significantly smaller larvae at 20 dph. The HPOL model, guided by in vitro testing, accurately predicted the reproductive effects of prochloraz and tamoxifen and the lack of effect by fluoxetine. Our results support the use of biologically based mathematical models of physiological systems in AOP testing. Supported by EPA-STAR grant R835167.

355

Development of a PBPK model for metal accumulation in fish infected with acanthocephalan parasites

Y.T. Le, University of Duisburg-Essen / Aquatic Ecology; M. Garcia, Spanish Council for Scientific Research; M. Nachev, D. Grabner, University of DuisburgEssen / Aquatic Ecology; E. Balsa-Canto, Spanish Council for Scientific Research; J. Hendriks, Radboud University Nijmegen; B. Sures, University of Duisburg Essen / Aquatic Ecology

Fish are affected by both exposure to metals and infection. Each of these stressors might have effects on the response of fish to the other. Some efforts have recently been made in developing kinetic models for predicting metal accumulation in fish-parasite systems. Our previous model allows for investigating the relationship between the accumulation in the whole fish and in the acanthocephalan, but does not include the mechanisms how metals are accumulated in parasites. Physiologically based pharmacokinetic (PBPK) model has been used for simulating the organ-specific accumulation of pollutants. However, the capacity of this model for simulating fish-parasite systems has not been investigated. We developed a PBPK model for simulating Ag accumulation in the host-parasite system: chub (*Squalius cephalus*) and the acanthocephalan *Pomphorhynchus tereticollis*. The acanthocephalan was considered a compartment, similar to blood, storage, gills, kidney, liver, and intestine. Metal accumulation in the system was modelled as a function of internal (i.e. exchange between different compartments) and external (i.e. exchange with water) factors. The transport from blood to other compartments depends on the diffusive exchange and the fraction of metals dissolved in blood plasma and was assumed to be independent of the infection state. The rate constants for this transport were parameterised based on published data. The model was then calibrated by MATLAB-based AMIGO modelling software for determining the rate of the transport from storage, gills, kidney, liver, and intestine to blood as well as the external exchange. Model calibration was carried out by using experimental data generated when the infected chub were exposed to Ag in 48-day exposure and 51-day depuration periods. The initial results from model calibration show potential of the PBPK model for simulating the accumulation of metals in fish-parasite systems. For example, the model could simulate the changes in the concentration of Ag in storage, gills, and intestine. The stability in the concentration of Ag in kidney was also simulated by the model. However, the model is being further calibrated to improve its capacity for modelling the accumulation in liver and in the acanthocephalan. The observed weakness of the current version in modelling the accumulation of Ag in these compartments might be related to the approaches for simulating the excretion of organo-metal complexes to intestine.

Solutions for emerging pollutants - Towards a holistic chemical quality status assessment in European freshwater resources (III)

356

High-throughput exposure and risk modelling of chemicals in European river basins

J. van Gils, DELTARES; A. Focks, Alterra Wageningen University and Research Centre / Environmental Risk Assessment Team; h. baveco, Wageningen Environmental Research; L. Posthuma, RIVM / Centre for Sustainability, Environment and Health; I. Cousins, Stockholm University / Department of Environmental Science and Analytical Chemistry (ACES); C. Lindim, Stockholm University; S.S. Kutsarova, University of Zlatarov / Laboratory of Mathematical Chemistry; S.D. Dimitrov, University of Zlatarov / Dept of Comp Inform Technologies

SOLUTIONS is a EU 7th Framework Programme R&D project that aims at strengthening the management of emerging contaminants. It developed a collection of integrated models (the “Model Train”), to increase our understanding of issues related to emerging chemicals in Europe’s river basins, to support the prioritisation of chemicals and the abatement of the problems they cause and to evaluate future scenarios. The model train consists of four key building blocks: (i) the prediction of substance properties based on their molecular structure, (ii) the simulation of emissions, (iii) the simulation of fate & transport, and (iv) the characterisation of the risk of mixtures of chemicals for human health and aquatic ecosystems. The Model Train does not rely on extensive substance-specific input data. This implies that it provides predictive power for new (truly “emerging”) chemicals and for large numbers of chemicals (“real world exposure scenario”). The approach is validated for well-studied substances and data-rich basins. On this basis we learn how accurate our model based predictions are for new substances and data poor basins. The model train operates on the scale of Europe as a whole or for one or more individual river basins. It makes use of the pre-existing Europe-wide hydrology model E-Hype. The Model Train complements lab and field based approaches, by providing information for substances and sites which are not included in monitoring and by providing full time coverage. Validation results for the Danube, Rhine, four Spanish and a series of Swedish River Basins reveal that the accuracy of the simulated concentrations of a range of chemicals is higher for substances with a single type of use (e.g. pharmaceuticals, pesticides) and lower for substances with multiple uses. The predicted risk for Europe’s aquatic ecosystems is evaluated by a correlation with the observed ecological status as EU Member States report it under the Water Framework Directive. The SOLUTIONS Model Train will offer an effective tool to screen a large number of chemicals on their impact on Europe’s aquatic ecosystems, and to do so with consideration for spatial and temporal gradients as governed by socio-economic and meteorological/hydrological patterns in combination with the chemicals’ physical and toxicological properties. The presentation will include the validation results and will highlight some of the Model Train application results from SOLUTIONS.

357

Forward-looking on possible impacts of chemical pollution: Modelling lethal and sublethal effects of chemical exposure on population viability for aquatic macroinvertebrates

A. Focks, Alterra Wageningen University and Research Centre / Environmental Risk Assessment Team; I. Cousins, Stockholm University / Department of Environmental Science and Analytical Chemistry (ACES); J. van Gils, DELTARES; S. Birk, University of Duisburg-Essen / Aquatic Ecology; e. Peeters, Wageningen University / Aquatic Ecology and Water Quality; P. van den Brink, Alterra and Wageningen University; h. baveco, Wageningen Environmental Research

One challenge in the quality assessment of water bodies in Europe is the ongoing difficulty to link the chemical and the ecological status. Currently, new approaches are being developed to align chemical and biological monitoring results, hence providing the means to elucidate possible chemical impact on the ecological status of European water bodies in a retrospective way. Ecological modelling provides an alternative approach to connect exposure information to potential impact on biota, having the advantage that such modelling can be performed in a prospective way. This presentation will show results of ongoing modelling efforts in the EU 7th framework program SOLUTIONS project, where large scale modelling approaches are applied to link exposure dynamics of a number of chemical compounds to parsimonious individual-based population models. The STREAM-EU model provides exposure concentration results at the level of subcatchments, that is at a scale of tenths of km^2. Links between exposure and effects are realised using linear or log-logistic dose-response relationships. The ecological models account for lethal and sublethal effects on the population dynamics of a number of families of aquatic macroinvertebrates. Impacts of multiple chemicals are added up in the model following basic mixture modelling rules. Results depict the simulated inhibition of population growth rates and hence the chemical impact on population viability at European scales. Ecological modelling results are for some selected parts of Europe compared with available monitoring information on the abundances of macroinvertebrate families in order to get an impression about the quality of the model predictions.

358

Eco-epidemiology of aquatic ecosystems: aligning chemical and ecological status

L. Posthuma, RIVM / Centre for Sustainability, Environment and Health; S. Birk, University of Duisburg-Essen / Aquatic Ecology; A. Burton, University of Michigan / School of Natural Resources Environment; D. De Zwart, DdZ Ecotox / Centre for Sustainability Environment and Health; S.D. Dyer, The Procter & Gamble Company / Environmental Stewardship and Sustainability Organization; A. Focks, Alterra Wageningen University and Research Centre / Environmental Risk Assessment Team; C.M. Holmes, K.E. Kapo, Waterborne Environmental, Inc.; D. van de Meent, Association of Retired Environmental Scientists ARES / Environmental Science; J. van Gils, DELTARES; M.C. Zijp, RIVM / Centre for

Sustainability Environment and Health
This paper analyses water quality assessment and management specifically looking at the diagnosis of toxic effects of mixtures in field ecosystems. We apply various methods to liaise ecosystem responses to mixture toxic pressure under natural, variable and multi-stressed conditions. We collated vast amounts of monitoring data to explore those diagnostically, in line with the Father of Epidemiology, dr. Snow, who famously provided a solution to a cholera outbreak in Soho, London, 1854, by epidemiological reasoning. So, this paper presents a set of contemporary eco-epidemiological results, the recognition of ecological impacts in surface water systems, and the diagnosis of probable causes. The paper presents the utility of that for chemical- and water quality assessment and management, thereby bridging preventive policies such as REACh via e.g. a Mixture Assessment Factor and environmental management policies such as the Water Framework Directive. We present novel results of scientific research at the nexus of chemical and water policies, connected to the European goals to reach a non-toxic environment and the good chemical and ecological status for aquatic systems. The presentation consists of the analyses of vast sets of surveillance monitoring data using a combination of techniques originating from the fields of bioassessment and ecotoxicology. It thereby bridges these – so far often disparate – scientific disciplines, to support sustainable chemical and water policies. One of the most recent examples is provided by a diagnostic analysis in which the Good Ecological Status appeared associated to the Good Chemical Status, the latter shown to be a limiting factor for reaching a good ecological status. The presentation will provide a rationale for eco-epidemiological analyses as well as various types of results, from diagnostics to prognostic use of results. The examples highlight approaches to data collection and early stress characterization of chemical mixtures up till a solution-focused approach related to ecosystem services management goals. We present specifically a body of evidence for the wide-spread occurrence of chemical mixture impacts in current water systems at the continental and national scale (Netherlands), corroborating recent findings of this kind based on chemical risk.

359
Unravelling the cocktail of stress: toxics and other stressors impacting on the ecological status of Europe's rivers
S. Birk, University of Duisburg-Essen / Aquatic Ecology; V. Bremerich, Leibniz-Institute of Freshwater Ecology and Inland Fisheries; D. De Zwart, DdZ Ecotox / Centre for Sustainability Environment and Health; M.F. Sanchez, Groundwater Management; A. Focks, Alterra Wageningen University and Research Centre / Environmental Risk Assessment Team; L. Globevnik, M. Koprivsek, University of Ljubljana / Faculty of Civil Engineering and Geodesy; J. Lemm, University of Duisburg-Essen; J. Mahnkopf, Leibniz-Institute of Freshwater Ecology and Inland Fisheries; Y. Panagopoulos, National Technical University of Athens / Laboratory of Hydrology and Water Resources Management; L. Posthuma, RIVM / Centre for Sustainability, Environment and Health; K. Stefanidis, National Institute of Public Health and the Environment; M. Venohr, Leibniz-Institute of Freshwater Ecology and Inland Fisheries
Water management requires solid understanding of how multiple stressors affect ecosystem state and services. The EU project MARS (Managing Aquatic ecosystems and water Resources under multiple Stress) recently concluded four years of in-depth research on this topic. MARS looked into multi-stressor responses from experimental water body to pan-European scale, developed tools for modeling and diagnosing multi-stressor effects and guided management of multiply stressed aquatic ecosystems. Our presentation summarizes the key conclusions of the project, with a special emphasis on pan-European multi-stressor effects on the ecological status, including river hydrology, morphology, nutrient and toxic stressors.

360
Mitigation options for chemicals of emerging concern in surface waters-operationalizing solutions-focused risk assessment
A. van Wezel, KWR Watercycle Research Institute / Chemical Water Quality and Health; A. Fischer, Utrecht University / Copernicus Institute of Sustainable Development; J. van der Hoek, Technical University Delft / Water Management
Chemicals of emerging concern (CECs) in the water cycle have been the focus of research for over a decade. To minimise concentration levels and adverse effects, removal efficiencies of various (advanced) drinking water and wastewater treatment technologies has been studied. Advanced water treatment technologies are based on sorption, (advanced) oxidation and size separation principles. The experimental settings in studies on the efficiency of these technologies are not homogeneous; technologies can be tested at bench-, pilot- or full scale, with different compounds, with different water matrices such as real or standardized surface water, ground water, drinking water or wastewater, the test chemical can be spiked or real environmental samples can be used, there can be variations in the application of the treatment e.g. dose, contact time or pore size, and variation in how all this is expressed; in mJ/cm^2 or W/m^2 in case of UV oxidation, with freundlich isotherms or removal percentage in the case of GAC etc. These variations and missing clarity therein hamper the interpretation and evaluation of the data concerning the removal efficiency of CEC of specific treatment technologies. In a previous study it was found that stakeholders within the whole urban water cycle had sufficient information on CECs and their possible mitigation options, but that the relevance of the information often was unknown. A set of criteria describing what is important to know when evaluating removal efficiency studies can be helpful in this respect, with criteria for reliability and relevance where needed made explicit for the specific technologies to be evaluated. Examples of such criteria from the field of toxicology are available and well-used, e.g. to identify studies for the derivation of environmental quality standards in a scientifically sound way. Here we aim to highlight the current knowledge of the removal efficiencies with regards to CECs of (advanced) water treatment technologies both for surface water and wastewater. This to provide decision makers with the knowledge needed to make an informed decision with regards to which technologies will be relevant for their specific needs. To be relevant to end-users in water management the treatment technologies needs to be in use and commonly available. New and developing treatment technologies can be very promising but are generally not an option for end-users in water management as they need to have been tested on large scale and to be available commercially at relatively low cost. Commonly used advanced water treatment technologies are for sorption the use of activated carbon (granular activated carbon (GAC) and powdered activated carbon (PAC)), for (advanced) oxidation the use of ozone (O_3) and UV both ± H_2O_2 and finally the use of nano- and ultrafiltration membranes for size separation. We developed an evaluation criteria set for the specified treatment technologies. We used thes criteria to evaluate removal efficiencies as collected in a dataset on removal efficiencies consisting of approximately 2000 entries, 93 compounds and 9 treatment technologies for wastewater (ozone, ozone + H2O2, conventional WWTP, UV, UV + H_2O_2, PAC, GAC, NF, UF) and drinking water treatment (ozone, ozone + H_2O_2, UV, UV + H_2O_2, PAC, GAC, UF, NF).

361
Future perspectives of chemical pollution and regulatory development
J. Munthe, IVL Swedish Environmental Research Institute Ltd; T. Skårman, IVL Swedish Environmental Research Institute; E. Brorström-Lundén, IVL Swedish Envirionmental Research Institute; M. Rahmberg, E. Westberg, IVL Swedish Environmental Research Institute; D. Bunke, Öko-Institut e.V. / Sustainable Products & Material Flows Division; K. Sackmann, OEKO Institute
Future developments in society will result in the emission of new substances to the environment which will require an adaptation of existing legislation for protection of human health and ecosystems. Scenarios for the future development of society can provide valuable indications on changes in future pollutants in river basins. Some developments are directly connected to consumption of specific substances, e.g. demographic change where a longer life expectancy will lead to changes in amounts and types of pharmaceuticals used and thus also to the related concentrations in the environment. Future technological progress may help to identify suitable alternatives for currently used additives such as phthalates, PFCs, flame retardants or nanomaterials but may also introduce new substances with negative impacts on aquatic ecosystems. Four societal sectors have been identified where major changes within the next two decades can be expected which have potential consequences for chemical use and releases: public health, food production, urbanization and technologies. With these future developments and the resulting introduction of new substances from new sources, an adaptation of current regulatory frameworks is required. However, to prepare for a future with unknown scenarios for use and emissions of potential chemical pollutants is naturally difficult but a general approach can be developed and adopted to prepare for a future where environmental pollution by chemicals is avoided or minimised. This general approach should build upon a few basic principles: (1) **The solutions-focused approach** - where the same attention is given to evaluation options for minimising risks as to quantifying risks for new substances under development or introduction; (2) **Transparency and openness of information and knowledge**. Current applied research aimed at providing solutions to identified problems of chemical contamination in e.g. water ecosystems is severely limited by a lack of information on the production and use of chemicals in society as well as emissions to water. Linkage of national databases on use volumes of industrial chemicals such as SPIN (Substances in Preparations in Nordic Countries) would allow tracking quantitatively substitution of the most problematic substances; (3) **Increased international cooperation and strengthened global agreement**s. The world is globalised and the transport of chemicals is transboundary – both via the atmosphere and via global trade.

Ecological risks under complex, multiple-stressor threat scenarios: integrating chemical effects with environmental drivers (I)

362
Towards a systematic approach for the assessment of multiple stressors: Making Aquatic Ecosystems Great Again (MAEGA)
D.J. Baird, Environment Canada; R. Schaefer, University Koblenz Landau / Institute for Environmental Sciences; C.B. Choung, Environment Canada/Canadian Rivers Institute / Department of Biology; A. Bush, Environment and Climate Change Canada; S. Bracewell, Wageningen University & Research / Department of Aquatic Ecology and Water Quality Management; A. Chariton, Macquarie University / Molecular Ecology and Toxicology; Z. Compson, Environment and

Climate Change Canada; K. Dafforn, Macquarie University / Evolution and Ecology Reserach Centre; E.L. Johnston, University of New South Wales / Evolution and Ecology Research Centre; K. Korbel, Macquarie University; D. Lapen, University of New South Wales; M. Mayer-Pinto, University of New South Wales / Evolution Ecology Research Centre School of Biological Earth and Environmental Sciences; W. Monk, Environment and Climate Change Canada; A. O'Brien, University of Melbourne; N. Rideout, Environment and Climate Change Canada; K. Sumon, R. Verdonschot, Wageningen University; P. van den Brink, Alterra/Wageningen UR / Aquatic Ecology and Water Quality Management Group b Alterra
In the Anthropocene, ecosystems are exposed to a range of stressors that if not properly managed can lead to ecosystem state shifts and significant losses in ecosystem services. We held a workshop (September 2017 in Wageningen, The Netherlands) to develop a conceptual framework to assess the effects of multiple stressors on the structure and functioning of aquatic ecosystems. This framework was subsequently applied to three ecosystem types (ditches, floodplains and harbours). The proposed framework consists of two parts: an environmental filter and a transmitting function to allow effects to propagate to higher levels of biological organisation. Applying the framework consists of the following steps: 1) Select an ecosystems of concern; 2) Identify stressors and potential interactions; 3) Identify receptors/sensitive groups for each stressor; 4) Identify stressor-response relationships and group stressors according to their mode of action; 5) Construct an ecological model that includes relevant functional groups and endpoints; 6) Predict the resultant impact of multiple stressors; 7) Confront the predictions with experimental and monitoring data and 8) Adjust the ecological model if needed. Steps 7 and 8 can be repeated until a satisfactory match between model predictions and experimental and monitoring data has been obtained. The talk will present the details of the framework and will also briefly introduce the three case studies developed during the workshop and discuss the commonalities and differences in approaches between the three case studies which all used the framework as a starting point.

363

Predicting the response of ditch ecosystems to multiple stressors

S. Bracewell, Wageningen University & Research / Department of Aquatic Ecology and Water Quality Management; R. Verdonschot, Wageningen Environmental Research (Alterra); R. Schaefer, University Koblenz Landau / Institute for Environmental Sciences; A. Bush, University of New Brunswick / Environment Canada; D. Lapen, Agriculture and Agri-Food Canada; K. Sumon, Wageningen University & Research Centre / Aquatic Ecology and Water Quality Management; P. van den Brink, Alterra/Wageningen UR / Aquatic Ecology and Water Quality Management Group b Alterra
Until recently, our knowledge of the net effects of multiple stressors on freshwater ecosystems has been limited. We still lack a general framework that can integrate known effects of individual stressors on organisms and predict how these effects propagate through higher levels of biological organisation. In light of this, a workshop was held at Wageningen University and Research, the Netherlands, (September 2017) to determine the current state of knowledge of multiple stressor effects on aquatic ecosystems and to assess how these effects can be better predicted. The workshop was attended by experts from the Netherlands, Australia, Germany, and Canada and covered a range of ecosystem types considered to be at high risk from multiple stressors. The workshop resulted in a “best-approach” conceptual framework for assessing multiple stressor effects on aquatic ecosystems. The framework was subsequently applied to three case studies: harbours, agricultural drainage ditches, and floodplains. Here, we present the application of this framework to agricultural drainage ditches. Agricultural drainage ditches are an under-appreciated and undervalued habitat for a range of aquatic and terrestrial organisms. Although these man-made features can maintain high biodiversity in agriculture landscapes, they are often ignored for their conservation value and are not protected under the EC Water Framework Directive 2000/06/EC. Using the framework developed during the Wageningen workshop, we developed a conceptual food-web model using functional groups to assess known direct effects of stressors on ditch communities. We identified the most important stressors (nutrients, pesticides, dredging and mowing, salinisation, and siltation) impacting ditch communities and performed a literature search for each stressor-functional group combination to identify sensitive and non-sensitive groups. We also reviewed the literature on experiments using at least two of the identified stressors and identified potential interactions. The conceptual food-web model was updated using this knowledge to capture interactions. Finally, the conceptual model and its predictions regarding the response to multiple stressors will be compared to large scale ditch biomonitoring data to assess the validity/predictive power of the model. We demonstrate that the framework provides a useful conceptual template to assess and predict multiple stressor impacts as well as to unravel research gaps.

364

The combined effects of nutrients and thiacloprid on macrofauna invertebrate population and community responses

H. Barmentlo, Leiden University; M. Schrama, CML Leiden University / Conservation Biology; K.J. Musters, Leiden University / Institute of Environmental Sciences; P.M. Van Bodegom, CML Leiden University / Institute of Environmental Science CML; G. de Snoo, Leiden University / Institute of Environmental Sciences; M.G. Vijver, CML Leiden University / Conservation Biology
Ditches are commonly used to control for fluctuating groundwater tables in agricultural landscapes. They provide a strong linkage between agricultural fields and adjacent water bodies as they are a common sink for agricultural chemicals such as neonicotinoid insecticides and fertilizers. As these agrochemicals are bound to co-occur in the ditches, we aimed to study their combined effects on aquatic invertebrate population and community responses. To this end, we exposed caged organisms and naturally assembled invertebrate communities to environmentally realistic thiacloprid and nutrient concentrations at the Living Lab facility. The Living Lab facility consists of 36 naturally colonized ditches of 25 cm depth in which experiments can be performed under outdoor conditions. We found adverse effects of thiacloprid on several population responses at concentrations that were comparable or far lower than laboratory derived LOECs as obtained from literature. These effects were less pronounced when organisms were exposed under nutrient enriched conditions. In addition, we observed significant dissimilarity between the naturally assembled communities under the influence of both thiacloprid and nutrients. These shifts were largely represented by a severe decrease in insect abundance under thiacloprid exposure. This decrease was not observed in ditches that received both thiacloprid and nutrient application. Thus, we showed the importance of nutrient enrichment (and the resulting increase in primary production) for coping with thiacloprid induced toxicity. This might explain the difficulties as often faced when extrapolating lab to field data and the other way around.

365

Macroinvertebrate communities across a gradient of multiple stressors from agricultural land use in Romanian streams

V.C. Schreiner, M. Link, S. Kunz, E. Szöcs, University of Koblenz Landau; B. Vogler, B. Beck, Eawag Swiss Federal Institute of Aquatic Science and Technology; K.P. Battes, M. Cimpean, Babeș-Bolyai University; E. Vermeirssen, Ecotox Centre Eawag-EPFL / Aquatic Ecotoxicology; H. Singer, Eawag, Swiss Federal Institute of Aquatic Science and Technology / Environmental Chemistry; J. Hollender, Eawag / Environmental Chemistry; R. Schaefer, University Koblenz Landau / Institute for Environmental Sciences
Pesticides from agricultural usage are one of the major drivers of biodiversity loss in freshwater ecosystems. Their entry pathways are mainly related to pesticide use agriculture. To differentiate pesticide toxicity and other agricultural stressors, we conducted a field study in Eastern Europe (Romania), where agricultural intensity varies, ranging from high to low intensity (extensive) agriculture relying largely on human or animal labour (e.g. horse ploughs). We assumed that, in contrast, to pesticide toxicity, excessive nutrient and sediment input would be unrelated to agricultural intensity. Consequently, this would allow distinguishing effects from pesticides and these other stressors. We analysed the relationships between pesticide toxicity and other agricultural stressors. Additionally, we analysed combined and individual effects of these variables on the biodiversity, as well as on the composition of stream macroinvertebrate communities. We examined 19 low-order streams across a gradient of agricultural intensity in terms of average field sizes. Pesticide concentrations were investigated using two different passive sampling methods. Firstly, we used styrene-divinylbenzene (SDB) disks to sample hydrophilic compounds, which enabled the determination of approximate time-weighted pesticide concentrations in streams during heavy rainfall events. Secondly, we used polydimethylsiloxane sheets (PDMS) focusing on the detection of lipophilic pyrethroids and organophosphates. The toxicity of the 88 detected pesticides was assessed using the sum toxic unit (sumTU). Stream macroinvertebrate communities were sampled twice, using a quantitative multi-habitat-sampling. This allowed the analysis of relationships between the community composition and diversity with a gradient of pesticide toxicity in interaction with additional agricultural stressors. The toxicity gradient originated from pesticides and nutrients (NH_4^+) showed no relationship to the intensity of agriculture expressed as the average size of the adjacent fields. This indicates that pesticides and nutrients co-occur independently of agricultural intensity. How and to which extent, in terms of effect size, the communities are affected by the pesticide gradient and the additional presence of other stressors originating from agricultural land use will be presented during the conference.

366

Daily temperature variation determines the toxicity of a pesticide mixture

V. Delnat, T.T. Tran, L. Janssens, KU Leuven / Biology; R. Stoks, University of Leuven / Department of Biology
Synergistic interactions between pesticides in mixtures and between pesticides and warming may improve the efficacy of vector control. Particularly, synergistic interactions between biopesticides and chemical pesticides would be promising as these could potentially result in the combination of efficacy of control, slowdown of resistance build-up and lower ecological damage. One understudied aspect of global warming is the increase in daily temperature variation (DTV). While DTV may increase the toxicity of chemical pesticides, it is unknown whether it also interacts synergistically with biopesticides, and magnifies the toxicity of pesticide mixtures. We tested whether DTV influences the toxicity of pesticides with a

different mode of action (the chemical pesticide Chloorpyrifos, CPF, and the biopesticide Bti) in the mosquito *Culex pipiens*. We expected that the effects of the single exposures are strengthened in the presence of DTV. In addition, we tested whether there is an interaction between CPF and Bti and whether this interaction is magnified in the presence of DTV. We crossed three DTV treatments (no DTV, a small DTV of 7°C and a large DTV of 14°C) with four pesticide treatments (a solvent control, single CPF exposure, single Bti exposure and exposure to the CPF-Bti mixture). We studied effects on a proxy for population growth rate (r') and its key components. The experiment was done in three steps: (i) 4-day exposure in L4 to DTV, (ii) 2-day exposure to DTV and the pesticide treatment and (iii) exposure to DTV until metamorphosis. The presence of a large DTV increased the toxicity (based on r') of the chemical pesticide, but not the biopesticide. Moreover, a large DTV changed the toxicity of the CPF-Bti mixture. For example, the presence of large DTV removed the antagonistic interaction effect on total mortality which was present in the absence of DTV and in the presence of small DTV. Our results underscore the importance of considering DTV as a factor shaping not only the toxicity of pesticides but also the interaction type between pesticides in mixtures. Given DTV occurs in all natural populations and may strongly differ between latitudes, DTV may be an important factor causing a mismatch between toxicity studies done in the lab at constant temperatures and the toxocity of pesticides and their mixture in the real world.

367

Warming and daily temperature fluctuations make the pesticide chlorpyrifos more toxic in Ischnura elegans damselflies

J. Verheyen, R. van Roo, KU Leuven / Biology; R. Stoks, University of Leuven / Department of Biology

Current risk assessment of pesticides fails to protect aquatic biodiversity. A key reason is the lack of realism: pesticides are tested under ideal laboratory conditions at one mean temperature. To strengthen current risk assessment it is crucial to incorporate effects of global warming on the toxicity of pesticides. Global warming studies largely overlook that climate scenarios also predict stronger daily temperature fluctuations (DTFs), which can have greater fitness effects for organisms than increases in mean temperatures. While many pesticides (like organophosphates) get more toxic at higher temperatures, it is largely unknown how DTFs influence the pesticide toxicity. We examined a multiple-stressor scenario where we quantified the single and combined effects of (i) increases in mean temperature and (ii) in DTF, and (iii) exposure to the pesticide chlorpyrifos (CPF) in larvae of high- and low-latitude populations of *Ischnura elegans* damselflies. CPF imposed mortality and more so in high-latitude compared to low-latitude larvae. Moreover, CPF was more toxic at 24°C compared to 20°C, confirming the higher toxicity of organophospates at higher temperatures. A key finding was that DTF also increased the toxicity of CPF, providing novel evidence that DTFs can amplify the toxicity of pesticides. Furthermore, the increased toxicity of CPF by DTF was more pronounced at 24°C. This novel pattern is likely general as at a higher mean temperature, DTF will expose the animals to even higher temperatures during the daily cycle, thereby increasing exposure to stressful temperatures. Also, the negative effect of CPF on larval growth strongly depended on DTF. CPF did decrease larval growth considerably, but only in the 10°C DTF treatment. Probably the higher metabolic demands for cell maintenance in the 10°C DTF treatment resulted in lower growth rates. Our results convincely show that the organophosphate pesticide chlorpyrifos is not only more toxic to damselfly larvae at the higher mean temperature (24°C) but also at higher daily temperature fluctuations (DTF of 10°C), both in terms of lethal effects (mortality) and sublethal effects (growth rate). Notably, the synergistic effect of DTF on pesticide sensitivity was higher at the high temperature. Our results highlight that incorporating higher mean temperatures and especially DTFs in ecotox testing will increase the realism of the risk assessment of pesticides under global warming.

PBT/vPvB & PMT/vPvM substances and Non-extractable residues (NER): Scientific strategies, Analytical challenges and Regulatory Issues (I)

368

RPLC-HILIC and SFC coupled with Mass Spectrometry: Polarity Extended Screening of organic molecules in the aqueous environment

S. Bieber, Technical University of Munich / Chair of Urban Water Systems Engineering; S. Grosse, T. Letzel, Technical University of Munich

Trace organic compounds are important in environmental analysis, because they impact water quality and introduce potential (eco)toxicological effects. Current analytical methods mostly rely on gas chromatography (GC) or reversed-phase liquid chromatography (RPLC) coupled with (tandem) mass spectrometry. However, neither method can easily separate very polar molecules. Two chromatographic separation strategies, a serial RPLC- hydrophilic interaction liquid chromatography (RPLC-HILIC) coupling and an analytical scale supercritical fluid chromatography (SFC) system will be presented, and their separation effectiveness as polarity-extended chromatographic methods for 274 environmentally relevant compounds were validated in a recent publication [1]. Compounds tested were grouped into three polarity classes, "very polar" log D (pH 7) below -2.5, "polar" log D (pH 7) -2.5 to +2, and "non-polar" log D (pH 7) higher than +2). Nearly all compounds could be retained in both systems with relative standard deviations of retention times (RT) (n = 6) typically between 2 and 5%. Both techniques have considerable benefits when combined with accurate mass spectrometric detection. Molecules RT and accurate mass were recorded in a database for each set up. This information was used for compound screening measurements like "hidden-target screening" in complex environmental matrices (such as wastewater treatment plant effluents). Results of both techniques are complementary and useful for all types of molecules polarity. In this study, more than 80 percent of the compounds found in wastewater treatment plant effluent samples possessed a negative log D (pH 7) value. This result highlights the basic necessity to include "very polar" compounds in water monitoring techniques and protocols. [1] S. Bieber, G. Greco, S. Grosse, T. Letzel: RPLC-HILIC and SFC with mass spectrometry: Polarity-extended organic molecule screening in environmental (water) samples. Analytical Chemistry 2017, 89 (15), 7907-7914 (DOI: 10.1021/acs.analchem.7b00859).

369

Removal options and transformations of persistent mobile organic chemicals during production of drinking water

A. TOUFFET, IC2MP CNRS; H. Gallard, IC2MP; B. Sieira, University of Santiago de Compostela; j. Chokki, b. teychene, IC2MP CNRS; R. Montes, University of Santiago de Compostela; R. Rodil, University of Santiaga de Compostela; J. Quintana, University of Santiago de Compostela

Persistent Mobile Organic chemicals (PMOCs) are well water soluble, non-volatile and are thus mobile in the water cycle. Because of their intrinsic properties, they are able to penetrate natural and artificial barriers and may constitute a threat for drinking water. Advanced treatments like activated carbon and oxidation processes can be used to limit the presence of organic micropollutants in drinking water. However, low removal by activated carbon is expected for PMOCs because of their high polarity. The behaviour of selected PMOCs identified in water resources were evaluated at lab-scale for their removal by different options including powdered activated carbon (PAC), high pressure membrane processes and transformation by ozone and chlorine. Highly polar PMOCs such as adamantan-1-amine (Log D = -2.34), trifluoromethanesulfonate (Log P = -1.35) and ?-caprolactam (Log P = 0.15) were not removed by PAC even for very high doses. Only naphtalenesulfonate (Log P = - 0.41) was fully removed for 5 mg L^{-1} PAC. The other PMOCs i.e. aromatic sulfonates, aromatic guanidines, phenols, were removed significantly for high PAC doses, but that are not compatible with drinking water production. Most of the PMOCs identified in water resources showed a very low reactivity with ozone with rate constants below 100 M^{-1} s^{-1} and thus will not be transformed during ozonation of drinking water. Two aromatic guanidines, the 1,3-diphenylguanidine and the 1,3-di-o-tolylguanidine, an olefinic sulfonate and an amine compound, the N-benzyldimethylamine, were rapidly transformed by ozone. Transformation of both guanidines occurred in few seconds at neutral pH during disinfection by chlorine. Chlorinated and hydroxylated analogues, and products of cleavage and cyclization were identified. US EPA toxicity prediction tool showed that chlorinated and hydroxylated analogues would be more toxic than the parent compound, which was confirmed by Microtox acute toxicity test for Cl_2/guanidine ratio of 1 and 10. Thus, reactions with chlorine during disinfection can be a source of new, persistent and more toxic chemicals in drinking water Some PMOCs like ?-caprolactam, halogenated methanesulfonates, adamantan-1-amine and triazine compounds will neither be removed by adsorption on activated carbon nor transformed by oxidation processes and could thus be present in drinking water. High pressure membrane processes would constitute the ultimate barrier for these compounds.

370

Removal of polar micropollutants from drinking water by reverse osmosis: a pilot scale study

V. Albergamo, University of Amsterdam/IBED Institute / IBED; E. Cornelissen, KWR Watercycle Research Institute; B. Blankert, Oasen; W. Knibbe, University of Wageningen; W. Van der Meer, Oasen & University of Twente; P. de Voogt, University of Amsterdam / IBED

The occurrence of polar micropollutants (MPs) in drinking water sources is regarded as one of the most challanging issue of our times. Polar MPs can preferentially remain in the water phase during environmental and water treatment processes, potentially reaching finished drinking water and thus raising concern over adverse effects to human health. In The Netherlands reverse osmosis (RO) has been proposed as a stand-alone treatment capable of producing impeccable drinking water from riverbank filtrate. State-of-the-art RO can be highly effective in retaining organic compounds depending on pysicochemical properties such as size, charge and polarity. The aim of this study was to assess whether riverbank filtration followed by RO can provide sufficient removal of MPs and thus be considered for further implementation. We also aimed to elucidate the transport of organic solutes through RO membranes by relating solute physicochemical properties to solute passage. A novel pilot-scale RO system capable of operating in anaerobic conditions was built for this study. Raw anaerobic riverbank filtrate was used as feed water. The feed was spiked with 30 target polar MPs selected from scientific literature and considered relevant for the quality of source waters and critical for

RO. Feed water samples were analysed by direct injection, whereas RO permeate samples where enriched by solid-phase extraction. The analysis were carried by ultrahigh-performance liquid chromatography coupled to time-of-flight high-resolution mass spectrometry. Neutral polar MPs displayed less than 5% passage, except benzotriazole, tolyltriazole and phenylurea, which displayed a passage of 25%, 17% and 10%, respectively. The data showed that removal of neutral polar MPs is mainly governed by size exclusion. For neutral and moderate polar MPs the inverse correlation between size and passage was weaker, as seen for bisphenol A, which displayed 4% passage. The higher passage of moderately polar and hydrophobic MPs could be attributed to solute-membrane hydropobic interactions followed by diffusion. All anionic MPs displayed less than 1% passage, oppossed to cations for which up to 10% passage was observed. The negative charge displayed by the membrane surface explained these results. Overall this study showed that high chemical removal rates can be achieved by RO. Tighter membranes and multi-stage RO will be investigated to improve the removal of small neutral MPs for drinking water applications.

371

Identification of transformation-derived very polar organic water contaminants and their relevance in the water cycle

D. Zahn, Hochschule Fresenius / Chemistry and Biology; P. Mucha, V. Zilles, Hochschule Fresenius, University of Applied Sciences; A. Touffet, H. Gallard, IC2MP UMR 7285 CNRS - University of Poitiers; T.P. Knepper, Hochschule Fresenius, University of Applied Sciences / Chemistry and Biology; T. Frömel, Hochschule Fresenius, University of Applied Sciences

Highly polar organic substances are well water soluble, non-volatile, and exhibit only minimal adsorption to nonpolar surfaces. Therefore, they may be able to penetrate natural and artificial barriers and are thus mobile in the water cycle[1]. If these mobile organic contaminants (MOCs) are persistent (PMOCs) against microbiological and chemical degradation, their removal during waste water treatment and drinking water purification may prove difficult. Toxic PMOCs can be classified as PMT (persistent, mobile, and toxic) substances. Since the most frequently used trace analytical method for the quantification of organic pollutants in aqueous matrices, reversed-phase high performance liquid chromatography - tandem mass spectrometry (RP-HPLC-MS/MS), is only of limited use for the analysis of very polar substances, little is known about PMOCs in the water cycle and only few (e.g. acesulfame, glyphosate) have been extensively studied and monitored[2]. PMOC may be, among others, industrial chemicals, or transformation products thereof. Most transformation processes usually result in the formation of transformation products (TPs) with increasing polarity until either mineralization is achieved or a dead end TP is formed, thus potentially resulting in persistent and highly polar water contaminants. Many PMOCs derived from transformation processes may still be unknown and thus not be represented in suspect or target screening campaigns. As a consequence, no information about their occurrence and origin is available, which severely exacerbates the sophisticated monitoring and effective regulation of their precursors. Based on the work of Arp et al.[3] and Schulze et al.[4] we selected 15 industrial chemicals with a high expected potential to form PMOCs upon transformation and studied their behaviour during hydrolysis, biotransformation, oxidation with MnO_2, and photolysis experiments. After structural elucidation of the 9 detected transformation products with high resolution mass spectrometry (HRMS) we developed a qualitative HILIC-MS/MS (Hydrophilic interaction liquid chromatography – MS/MS) method and screened 25 Hessian surface waters for the presence of these TPs. While some TPs were not detected others were present in the majority of samples. This screening data provides first information about the potential environmental relevance of the identified TPs, which can be used to prioritize them for inclusion in future quantitative screening campaigns.

372

The limited chemical application domain of regulations: An illustration using the POP screening assessment in the Stockholm Convention

M. McLachlan, Stockholm University / Environmental Science and Analytical Chemistry (ACES)

Regulatory frameworks are initiated by a societal concern and built upon the scientific knowledge existing at the time they are written. This imparts them with a chemical application domain, i.e. a chemical property space in which the frameworks can be applied in the context for which they were conceived and supported by a sound scientific foundation. As time passes, societal concerns change, and this can lead us to want to apply regulatory frameworks outside of their chemical application domain. Today we have the ambition to regulate tens of thousands of chemicals, and we are doing this with regulatory frameworks that were in some cases developed 20 years ago with a more modest level of ambition and less scientific knowledge than we have today. Are these regulations really up to the task? This question is explored using the example of the POP screening assessment in the Stockholm Convention. Using perfluorinated alkyl acids (PFAAs) and octamethylcyclotetrasiloxane (D4) as case studies, it is shown how this framework can lead to both false negative and false positive conclusions. False negative classification of PFAAs can arise because of the inclusion of bioaccumulation as a screening criterion in the framework although bioaccumulation is not a requirement for adverse effects of chemicals in remote regions. False positive classification of D4 can arise because the four screening criteria (persistence, bioaccumulation, long-range transport, and adverse effects) are not valid in the same environmental media/compartments. It is concluded that if we wish to conduct POP assessment for the broad spectrum of chemicals in modern commerce, then we will have to rely less on individual screening criteria and instead apply models that can capture and integrate the broad diversity of chemical behaviour.

373

'One for all and all for one' - Can we REACH a harmonised PBT-assessment across EU-regulatory frameworks?

C. Rauert, Umweltbundesamt / International Chemicals Management; A. Böhnhardt, German Environment Agency; J. Priegnitz, Umweltbundesamt / FG IV 1.3 - Pflanzenschutzmittel; A. Wiemann, UBA Umweltbundesamt; J. Schmidt, German Environment Agency

Persistent, bioaccumulative and toxic (PBT) substances and REACH are frequently called in one breath. However, also other European regulatory frameworks for chemicals, such as for biocides, plant protection products (PPP), veterinary and human medicinal products (VMP/HMP) stipulate the performance of a PBT-assessment during substance evaluation. As in other hazard based assessments (e.g. GHS/CLP regulation, POP), the PBT/vPvB assessment focuses on the properties of a substance only and does neither take into account the use of the substance nor its exposure. Consequently, the identification of a PBT or vPvB substance should be independent from the regulatory framework under which it is assessed. However, in our comparison of conclusions on PBT properties for a number of substances falling under more than one legislation it became apparent that the outcome of the PBT assessment does not necessarily correspond between different regulations. This stands in contrast to the goals to perform a comprehensible and consistent assessment of chemicals and to ensure a high level of protection of human health and the environment against hazardous chemicals within the EU. In order to elucidate the reasons still hampering a harmonised PBT-assessment, we did a compilation of a number of technical (amount and quality of data, acceptability of specific data, derivation of endpoints, applied guidance documents), and conceptual criteria (numerical criteria, testing strategy, assessment of transformation products) as well as of other factors (consequences of PBT-assessment, data management and publication of assessment results). Outgoing from this, we developed several proposals facilitating a harmonised PBT assessment, starting from the implementation of an overall PBT-guidance up to an inclusion of transformation products in the PBT-assessment by all regulatory frameworks. Although it cannot be denied that a harmonisation process is ongoing, we conclude that there are still some fundamental choices to be made both at the organisational level and at policy level first to achieve the goal of a standardised PBT-identification among EU-legislations.

Product benefits and positive outcomes: valuation and beyond

374

A need for a better characterisation of product benefit in life cycle sustainability assessment

T. Schaubroeck, Luxembourg Institute of Science and Technology (LIST) / Research Group EnVOC; E. Benetto, Luxembourg Institute of Science and Technology (LIST) / Environmental Research and Innovation

In life cycle assessment (LCA) the main focus is on damage assessments of production systems. These damages are conventionally characterized per so called functional unit. In practice, however, these functional units are partially descriptive, e.g. white light from a point source with 1500 lumen, and not well assessed. **In the first part of this study we therefore further elaborate the functional unit towards product benefit.** When taking a closer look at the concept of functional unit, it is imperative to define what functionality implies. Products have been created to fulfil human needs, e.g. the need for light at night provided by a light bulb. Through fulfilment of needs, human well-being is induced. The characterisation of the actual functionality hence boils down to the assessment of this well-being effect. In consequential LCA, not only the consequences related to the affected activities associated with the product life cycle should after all be considered, but also those associated with the benefit induced by the product. **In the second part, we specify how a better product benefit characterisation could improve life cycle assessment and its policy support. Three advantages are specified. First**, as these functional units are often not specifically defined and a product can have multiple functions, comparison is often impeded in practice, unless the respective products are exactly the same. A better specification of the functional unit in terms of an aggregated single score for product benefit, such as the net impact on human well-being, would permit to compare all types of products. **Second**, not only would a better characterisation of the product benefit allow for a better comparison of various production systems, it also allows one to compare the benefit of the product with the damage provided during its production. One can then derive the holistic sustainability of having the production system instead of not having it. **Third**, when an additional amount of byproduct is created by a production system, this may also induce a byproduct benefit, which can be accounted for as such. in fact, when a byproduct enters the market, a share of it can lead to a decrease in supply (substitution approach) but another share can also lead to an increased demand and thus consumption, which satisfies needs that were previously unsatisfied (production benefit approach). A consideration of both effects is needed in CLCA.

375

Assessing nutritional impacts and benefits on human health in LCA: A new midpoint impact category

K. Stylianou, University of Michigan - School of Public Health / Environmental Health Sciences; V.L. Fulgoni III, Nutrition Impact, LLC; O. Jolliet, University of Michigan

Diet is a crucial determinant of human health. According to the Global Burden of Disease (GBD), dietary risk factors are responsible for >10 million deaths/ year globally. Yet, beneficial and detrimental nutritional health effects, the dominant pathway for health in food systems and diets, are often neglected in Life Cycle Assessment (LCA). To address this, we develop 14 nutritional characterization factors (CFs) for food groups and nutrients using epidemiological evidence from the GBD, and propose a new nutritional midpoint impact category for LCA, demonstrating its application to the entire US diet. We develop marginal nutritional CFs for 8 major food groups (nuts and seeds, whole grains, fruits, vegetables, milk, sugar-sweetened beverages, red meat, and processed meat) and 6 nutrients (omega-3, calcium, fiber, polyunsaturated fats, trans fats, and sodium), identified by the GBD as dietary risk factors. CFs are estimated by coupling age- and gender-adjusted information on outcome-specific incidence rates with risk ratios (RR) and severity factors, measuring positive or detrimental effects in avoided µDALY/g. We also develop a profiling system for 6000+ food items consumed in the US that aligns publically available data from multiple databases with risk factor definitions from the GBD. Finally, for 6000+ food items we estimate the HEalth Nutritional Index (HENI), the total avoided health burden from all dietary risk factors per serving and 100 kcal. Nutritional CFs for food group and nutrient range between -8 avoided µDALY/g for sodium, up to 57 avoided µDALY/g for omega-3 from seafood. HENI score typically ranges from -80 avoided µDALY/serving for Frankfurter sandwiches to 50 avoided µDALY/serving of nuts and seeds. Absolute HENI scores and ranking of food items vary substantially when using 1 serving or 100 kcal as a functional unit. Unhealthy food groups such as mixed dishes and protein foods with the exception of seafood and nuts and seeds have negative HENI scores primarily due to detrimental effects from processed and red meat, saturated and trans fats, and sodium. Healthy food groups such as fruits, grains, and vegetables excluding starchy vegetables tend to have positive HENI scores dominated by health benefits from their respective food groups. The estimated nutritional CFs could define a new midpoint impact category in LCA that would improve human health impact assessment in LCA and allow for a comprehensive assessment of food items and diets.

376

Combining Operational Research and Life Cycle Assessment to optimize the environmental performance of Peruvian diets

I. Vázquez-Rowe, Pontifical Catholic University of Peru / Civil Engineering Environmental Science; G. Larrea-Gallegos, Pontificia Universidad Catolica del Peru / Civil Engineering Environmental Science; A. Gilardino, Pontificia Universidad Catolica del Peru / Industrial Engineering

Food production and security has been highlighted as one of the most threatened sectors worldwide by the consequences of climate change. However, food production is also responsible for an important fraction of GHG emissions. In Peru, 50% of household expenditure is destined to food purchase. In contrast, malnourishment is still rampant in many socioeconomic sectors, mainly in the Highlands in the Amazon basin. In this context, it appears as a major challenge to jointly achieve nutritional improvements in the Peruvian diet and reductions in terms of GHG emissions. Hence, the main objective of this study was to apply a methodology which allowed optimizing the environmental profile of the Peruvian diet while improving its nutritional requirements at competitive economic costs. In other words, the aim of the optimization model was to determine an optimal diet from an environmental perspective considering nutritional and economic constraints. For this, the joint combination of Operational Research and Life Cycle Assessment was performed. Based on the average diets identified for each city included in the study, an optimization was performed considering a set of criteria that respond to the three dimensions of sustainability. Nutritional aspects were included in the model through a restriction based on the minimum consumption of food types and caloric intake recommended by Peruvian authorities. Regarding economic costs, the model included a set of inequations that limited the minimum and maximum monetary changes throughout the year (i.e., 2016). Finally, environmental aspects were considered by introducing an objective function that minimizes the emissions of CO_2eq of the entire food basket. The result of the proposed linear program allows understanding the amount of each individual food product that should be consumed in each city that satisfy all the restrictions included in the model in order to attain the lowest GHG emissions possible. AMPL® was used as the programming platform, and CPLEX® as the solver. Results demonstrated that substantial reductions can be attained in GHG emissions through the optimization of diets in Peru. For instance, in Lima the reduction could reach 200 kg CO2eq per person and year (22%). These results constitute an important framework to understand the current situation of the GHG emissions of the average Peruvian diet, as well as to mitigate these emissions while improving nutritional aspects and controlling economic costs.

377

Using the first Swiss dietary survey to determine the environmental and health benefits and impacts of various dietary patterns

A. Ernstoff, Quantis / Quantitative Sustainability Assessment; S. Humbert, X. Bengoa, M. Vargas Gonzalez, Quantis; O. Jolliet, University of Michigan

Many studies compare the environmental impacts of dietary patterns such as dietary recommendations (e.g. food pyramids), vegetarian and vegan diets. Mounting evidence suggests diets high in red meat and other animal products have higher associated environmental impacts. A hypothetical non-meat diet is often considered in such assessments, which may e.g. be equi-calorie or mass to the meat containing diet. In this study we use the first Swiss National Survey (MenuCH) to determine what non-meat eaters consume in Switzerland and what potential environmental and health benefits (or impacts) may result from assessing realistic consumption. About 5% of the Swiss population self-identifies as vegetarian, and less than 1% as vegan. Meatless diets contained about the same overall mass of food consumed, generally offered environmental and health benefits through increased fruit and vegetable consumption, but vegan diets can be insufficient in certain essential vitamins if not supplemented. Nuts, seeds, and their oils were important sources of key nutrients such as vitamin E. In conclusion, using dietary surveys can help provide actual evidence as to what people consume when removing meat or other animal products from the diet. Accounting for the environmental and health benefits of realistic dietary patterns, can help support improved recommendations.

378

The cost of CO2 in Life Cycle Assessment

Y. Dong, Technical University of Denmark; R. Rousselet, Ecole Centrale de Marseille / Engineering School; H.J. Sørup, Technical University of Denmark / DTU Environment; P. Fantke, Technical University of Denmark / Quantitative Sustainability Assessment Division; M.Z. Hauschild, Technical University of Denmark / DTU Management Engineering Division for Quantitative Sustainability Assessment

Climate change has gained increasing attention over the past decade in response to the revelation that we need to maintain a viable climate for humans and the environment. The increasing emission of greenhouse gases (GHG) such as CO_2 may accelerate climate change and cause subsequent damages. Correspondingly, countries and companies actively develop strategies to minimize their GHG emissions and thus climate impacts, but which strategies that will be more beneficial is often hard to evaluate. Life Cycle Assessment (LCA) is a tool to evaluate the damages of GHG emissions from the whole life cycle of the intended strategies, taking a cradle-to-grave perspective. By monetising the impacts related to these emissions, they can be compared to the overall cost of a strategy. This secure that emissions are considered in determining the priority and benefits of the respective emission reduction strategies. Our study looks into the monetary values of GHG, represented by CO_2 (or CO_2-equivalent), and their underlying cause-effect chains in three Life Cycle Impact Assessment (LCIA) methods LIME2, EPS2015 and ReCiPe2016. The damage cost for CO_2 is in the same order of magnitude in EPS2015 and ReCiPe2016, but one order of magnitude higher than that in LIME2. Climate change-related damages on human health are well represented in all LCIA methods, and the monetised damages from this category contribute to more than 70% of the total CO_2 cost in all three methods. Social assets and ecosystem damages, on the other hand, are only counted for in two of them. Furthermore, a range of potential socio-economic damages from a changing climate are discussed in IPCC reports, including economic loss from extreme weather events, costs of potential climate-related society security and poverty, but they are not included in any of the LCIA methods. This may limited the suitable application area of the CO_2 cost evaluated by LCA, especially in studies where social and economic consequences are the major concerns. The CO_2 costs from the three LCIA methods are further evaluated in comparison with approaches from other research fields, such as Social Cost of Carbon (SCC), and discrepancies and associated uncertainties are discussed.

379

Poster spotlight: WE257, WE258, WE259

Advances in monitoring and evaluating remedy effectiveness for in situ amendments in soils and sediments

380

Assessment of Human Health Benefits and Risks of Contaminated Sediment Remediation

J. Kvasnicka, University of Michigan, Ann Arbor USA / Environmental Health Sciences; K. Stylianou, University of Michigan - School of Public Health /

Environmental Health Sciences; G. Burton, University of Michigan / School for Environment and Sustainability; J. Semrau, University of Michigan - Civil and Environmental Engineering and Program in the Environment; O. Jolliet, University of Michigan
Billions of dollars have been spent on environmental dredging projects to remediate contaminated sediments. However, the extent to which this remedy can reduce human health risks is unclear. Environmental dredging projects can also create health impacts from chemical exposures and occupational incidents. These potential adverse effects are usually not quantified in a manner conducive to comparing them to potential benefits and assessing overall remedy effectiveness. Focusing on the Hudson River PCBs Superfund Site remediation, this study demonstrates a novel approach to comprehensively evaluate the relative tradeoffs between population health benefits and risks associated with different remedial alternatives. The specific aims were to: 1) Assess health impacts on recreational anglers for a No Action scenario, due to bioaccumulation of PCBs in Hudson River fish, and exposure through fish consumption. 2) Determine and compare the reduction in health impact from reduced fish tissue PCB concentrations associated with different remedial options relative to No Action. 3) Investigate potential health impacts of the selected remedy from resuspension of sediment, air emissions of PCBs, diesel particulate matter (DPM) and NO_x, and occupational incidents. 4) Compare the avoided health impacts, *i.e.,* the health benefits, with the created health impacts. For each considered impact pathway, we derived both central and upper bound estimates, using the disability adjusted life year (DALY) as a comparative metric. For the No Action scenario, the health impacts are 11 and 78 DALY for the central estimate and upper bound, respectively. Implementing a source control action (MNA) could have achieved a 30% reduction in the total health impact, without the need for substantially more expensive and labour intensive remediation. Potential benefits of MNA are further highlighted when resuspension is an important factor. Furthermore, potential health impacts created by the selected remedy are within the same order of magnitude as potential benefits. Occupational fatal incidents are comparable to the combined benefits of MNA and the selected remedy scenario. Impacts associated with chemical inhalation exposures are less substantial, albeit not negligible. The quantitative framework of this study, when supplemented with adequate monitoring data, can provide valuable insight into the overall effectiveness of a given remediation in light of alternatives.

381

Six inches under: Remediation efficiency of activated carbon caps buried by dynamic sediment movement

S. Abel, J. Akkanen, University of Eastern Finland / Department of environmental and biological sciences
The application of activated carbon (AC) based thin layer caps is a promising and novel in-situ remediation method for contaminated sediments. The method utilizes the adsorptive capabilities of AC, allowing it to strongly bind persistent organic pollutants, thus greatly reducing the contaminants' bioavailability and sediment to water flux. Sediment capping with active materials can furthermore provide a less destructive, low-cost alternative to traditional remediation methods, such as sediment dredging. For this study, the method was evaluated under realistic conditions with the first field trial on AC based sediment remediation in Finland. At the test site in the PCB-contaminated Lake Kernaalanjärvi, a 300 m^2 plot was amended with an AC thin layer cap (1.6 kgAC/m^2). Due to the shallow nature of the lake and its large surface area, highly dynamic sediment movements occurred over the monitoring period of 14 months. As a consequence, the AC cap was buried rapidly under a layer of contaminated sediment from adjacent, untreated sites, leading to a low measurable impact of the AC amendment. Neither benthic community structure nor PCB bioaccumulation in local benthic organisms were significantly different on the AC amended plot and the surrounding reference sites. The amount of sediment deposited was high (34.2 g dry matter/m^2/day), making detailed observations on the development of the situation over time difficult. To investigate the potential for a long term remediation success of AC caps even under these unfavorable conditions, a corresponding laboratory study was conducted. The field scenario was replicated in test vessels containing a base layer of PCB contaminated sediment topped with an AC layer that was in turn buried by varying amounts of sediment (thicknesses of the overlying sediment layer ranged from >1 - 40 mm). Endpoints were the growth and PCB bioaccumulation in *Lumbriculus variegatus* and *Chironomus riparius.* The results indicate that an AC cap can remain effective even once it has been covered with contaminated sediment. This depends, however, on the intensity and depth of bioturbation. With the deeper dwelling *L. variegatus* present in the test systems, the AC layer was mixed well with the overlying sediment, allowing for a measurable remediation success. With the shallow dwelling *C. riparius*, this effect could only be observed with minimal sediment coverage of the AC (< 5 mm).

382

Ecosafe nanotechnologies for environmental remediation: the NANOBOND project

I. Corsi, University of Siena / Physical, Earth and Environmental Sciences; G. Grassi, G. Liberatori, University of Siena / Department of Physical, Earth and Environmental Sciences; c. murano, University of Siena; A. Bellingeri, University of Siena / Department of Physical, Earth and Environmental Sciences; a. fiorati, Politecnico di Milano; G. Musso, f. trotta, Università di Torino; C. Punta, Politecnico di Milano
In situ remediation of sludge and dredged harbour sediments is currently highly cost-effective despite an ever increasing number of sites requiring swift treatments to reduce contamination. Therefore there is a urgent need worldwide to identify new promising and innovative solutions ensuring a quick and efficient removal of pollutants and appropriate methods for monitoring the effectiveness of remediation strategies and any associated potential environmental risks. Although the use of engineered nanomaterials and nanoparticles (ENM/P) for environmental remediation, known as *nanoremediation*, represents a challenging and innovative solution, environmental and human risk assessment associated with the use of ENM/P are still a matter of debate. Limited *in situ* applications to water and soil/sediment remediation suffer for a lack of data regarding environmental impacts and there is a general demand for strategies aimed to fulfill such needs. As the potential and efficacy of nanotechnologies is well established, several drawbacks related to full-scale application should be overcome. In particular great efforts should be devoted to develop (nano)materials which own ecosafe features such as limited release and mobility in environmental matrices as well as no toxicity for natural ecosystems. Ecotoxicology can be thus use to develop *ecofriendly* (nano)materials for environmental application and to provide monitoring methods in a weight-of-evidence approach also to support decision-makers. The NANOBOND project (Nanomaterials for Remediation of Environmental Matrices associated to Dewatering) funded in 2015 by POR CReO FESR Toscana 2014-2020, is developing an innovative system for treating contaminated sludge and dredged sediments, by coupling the use of nanostructured *eco-friendly* materials with the classical geotextile dewatering tubes. This new solution, will enable to reduce contaminated sludge and sediments, in terms of volumes and costs of transport, but also to convert the resulting solid and liquid wastes to a renewable clean resource to be use in several other applications. The results of several preliminary trials in which ecotoxicology (bioassay and biomarkers approach) has been used to validate efficacy and to assess ecosafety (safe-by-design) of newly developed nanostructured materials as well their suitability for *in situ* application within the geotextile in terms of environmental impact will be presented.

383

Possibility of using a genotoxic tests in planning precise phytoremediation of depleted soils enriched in organic amendments

A.O. Murtaś, Czestochowa University of Technology / Department of Infrastructure and Environment; M. Jaskulak, Czestochowa University of Technology / Institute of Environmental Engineering; A. Grobelak, Czestochowa University of Technology / Department of Infrastructure and Engineering
A state of soils at many areas is getting worse due to an increase of anthropologic activities. Increasingly, human industrial activity, our irresponsibility, and impunity have a negative influence on the condition of soils. Thus, the problem of soil contamination, especially with several nanoparticles, and multiple heavy metals refers mainly to the industrial areas. Majority of those contaminations are bioavailable, and they are deposited in plant tissue as well as in edaphon. Moreover, they are included in the food chain where they pose a huge threat not only to the ecosystem but also to human health. Long-term exposition to a high concentration of pollutants (especially heavy metals and toxic nanoparticles) leads to soil depletion, and hence loss of organic fraction in the soil. Lack of organic compounds in connection with high contamination with single heavy metals, a mixture of heavy metals or toxic nanoparticles makes area impossible to any usability. Hence, a necessity to know their ecotoxicological mechanisms, and the possibility of their bioremediation, and detoxification with using various methods are justified and require increased attention among scientist. Plants diversity provides unexhausted possibilities for phytoremediation of those fields. An underestimated potential of grass for the utility to phytoremediation needs more extensive attention. Going out against this necessity, we tested *Festuca pratensis* H. for the possibility to use in phytorecultivation of exhausted wastelands in an aspect of improvement state of soil by introduction organic fraction into a soil originating from plant residues. The main aim of this experiment was to define the smallest one-time dose of organic waste fertilizer required for free plant growth for defined period of time providing organic fraction and allowing for development of fauna and flora. This definition of the minimal organic doses needed for introduction into soil as the first step of recultivation bases on testing genotoxicity of soils to *F.* pratensis L. and moreover, it allows for precise design of phytoremediation of problematic, exhausted areas.

384

Sorption of pharmaceuticals in soil systems

J. Li, Environmental Department University of York / Environment Department
Pharmaceuticals have been increasingly used worldwide to prevent or treat human and animal diseases and continuously released into the environment (Nikolaou et al., 2007). Following treatment, highly mobile and persistent pharmaceuticals tend to release into surface water and groundwater by reclaimed wastewater effluent through surface runoff and subsurface drainage, and subsequently being bioaccumulated to aquatic organisms (Kodešová et al., 2015; Tolls, 2001). Strongly adsorbed pharmaceuticals likely to be retained in the topsoil layer or leach to the groundwater, and have the potential to be taken up by plants or soil-dwelling organisms (Lin et al., 2011; Qin et al., 2015). A broad range of pharmaceuticals has

been detected at fairly high levels in aquatic systems (0.33-611 ng/L), terrestrial environments (0.53-340 µg/kg), and in the tissue of organisms (4.6-23.6 µg/kg in crop tissues, 61-127 µg/kg in terrestrial invertebrates) (Chen and Ying, 2015; Kinney et al., 2006; Pan et al., 2014). Long-term exposure to the residues pharmaceutical could pose a risk to the ecological system and exert adverse effects on human health via food chain (Carvalho et al., 2014). Adsorption processes have decisive role for the environmental behaviors and the ultimate fates of pharmaceuticals (Drillia and Lyberatos, 2005). However, relatively a few investigations of the sorption of organic compounds at the group level based on the dissociation degree of molecule in soil have been published so far (Droge and Goss, 2013; Franco and Trapp, 2008; Franco et al., 2009; Kah and Brown, 2007). The main aim of this study was to explore the effects of properties of the chemical and soil on the sorption of neutral and ionized pharmaceuticals in the soil environment. First, sorption behaviors of nineteen pharmaceuticals across four groups (neutrals, strong bases, weak bases, acids) were explored in five test soils. Using the measured sorption coefficients for each group, we evaluated the applicability and accuracy of existing predictive models that have been proposed to predict the sorption behavior of organic chemicals in soil. Finally, Pearson correlation analysis and Principal components analysis (PCA) have been carried out at the group level to systematically assess the potential factors (both soil and drug properties) influencing the sorption behavior of pharmaceuticals in soil and to get better understandings of the sorption mechanisms of different pharmaceuticals in the soil.

385
In vitro and in vivo assays to evaluate chlordecone transfer to animals: interest of soil amendment
M. DELANNOY, URAFPA-INRA / URAFPA INRA; S. Gaspard, Université de Antilles / COVACHIM; A. Razafitianamaharavo, LIEC Université de Lorraine CNRS; C. Cakir-Kiefer, Universite de Lorraine / URAFPA INRA; C. Soligot, E. Montarges-Pelletier, C. Feidt, G. Rychen, Université de Lorraine UL / URAFPA INRA
Chlordecone (Kepone) (CLD) is a highly persistent pesticide formerly used in French West Indies. Nowadays high levels of this pesticide are still found in soils which represent a subsequent source of contamination for outdoor-reared animals. In that context, sequestering matrices as activated carbons (ACs) or biochars are believed to efficiently decrease CLD transfer to animals. The present study intends to test using 2 distinct in vitro tests prior an *in* vivo assay the respective efficiency of several biochars and ACs to limit CLD transfer to animals. The Te-PBET and the ISO/DIS 16 751 availability part A protocols were used. In each test amended soils were prepared from a control one (SS) by adding 2% (mass basis) of one of the ACs or biochar. A selection of interesting matrices was realized prior the *in vivo* part of the protocol. For this final step piglets were exposed by alimentary route to contaminated soils. Only treatment groups exposed through amended soil with ACs presented significant decreases CLD availability, bioaccessibility (< 8%). Similar results were found using both *in vitro* assays. At last, concentrations of CLD in piglets liver and adipose tissue were found significantly lower after exposition to an AC amended soil ($p< 0.001$). This decrease was particularly high for a coconut shell activated carbon where relative bioavailability was found lower than 3% for both tissues. Finally, a positive correlation was found between environmental availability, bioaccessibility tests and *in vivo* results. This study leads to conclude that (i) AC introduced in CLD contaminated soil should strongly reduce CLD availability; bioaccessibility and bioavailability (ii) Tested biochars showed no reduction of transfert (iii) availability and bioaccessibility tests could be useful screening tests in order to select the appropriate biochar or AC.

Analysis and Fate of Emerging Contaminants in soils, water and plants under water scarcity (I)

386
Determination of dioxin-like polychlorinated biphenyls in land near the dumps of some settlements of the Republic of Armenia
A. Aleksandryan, Hazardous Substances & Waste Policy Division / Head of Division; A. Khachatryan, Y. Bunyatyan, Environmental Monitoring and Information Center / Division of waste inventory, classification and technology investigation
The sources of environmental pollution with polychlorinated biphenyls along with the energy production/ distribution complex include landfills, many of which do not correspond to environmental requirements and are the only attribute of the "consumer" relationship to nature, thus bringing forth a number of problems and a great danger for the normal functioning of biocenosis. In the present research we used samples of land taken on the boundary of the landfills and agricultural lands or water basins near some settlements of Armenia. The following 14 dioxin-like polychlorinated biphenyls (PCBs) were determined in soil samples: congeners NN 77, 81, 105, 114, 118, 123, 126, 156, 157, 167, 169, 170, 180, 189. Quantitative determination was carried out using chromatograph with electron capture detectors (ECD) equipped with glass capillary column with stable phase DB–5MS UI and the following parameters: 60 m x 0.250 mm x 0.25 µm. Special attention was paid to the total amounts of polychlorinated biphenyls, as the total amount of these compounds correlates with the hygienic standards, which as such are integral values. In all investigated soil samples dioxin-like PCBs were detected, however, in this case, we mainly recorded congeners NN 77, 81, 105, 114, 118, and 123, while NN 169 was determined less frequent and at insignificant quantities. Attention was drawn to the following: - out of 7 randomly selected soil sampling sites, a 2 to 3.5 times exceeding of the total/summary standard level was found at four sites; - in all cases, the excess of the standard was due to PCBs NN 81 and 114. Of special attention is the fact that at one of the soil sampling sites (Dilijan Town, Tavush Province of Armenia) along with 3.5-fold exceeding the standard, almost all dioxin-like PCBs were found.

387
Associated Health Effects of Veterinary Pharmaceutical Residues in Wastewaters around Selected Livestock Agriculture Farms in Western Cape Province
O. Fatoki, Cape Peninsula University of Technology / Chemistry; B. Opeolu, Cape Peninsula University of Technology / Faculty of Applied Sciences; B. Genthe, CSIR South Africa; O.S. Olatunji, Cape Peninsula University of Technology / Chemistry
Livestock farming is a major industry in the Western Cape Province of South Africa and livestock agricultural farms have been suggested to be a major source of pharmaceutical residues in many aqueous matrices, due to their enormous use. Pharmaceutical products such as steroids and non-steroids hormones, antibiotics and non-steroidal anti-inflammatory drugs from agriculture have been indicated to have the potential to show significant endocrine and other health effects. In this study, a High Performance Liquid Chromatography coupled to ultraviolet detector (HPLC-UV-Vis) method was optimized and validated for the detection and separation of the selected pharmaceuticals in effluents for livestock farms with major activity in the study areas. Multi-residue solid phase extraction (SPE) procedure was developed and validated for the recoveries of acetaminophen (AC), dichlophenac(DP), salicylic acid (SA), tetracycline(TC), chloramphenicol(CHR), ciprofloxacin(CP), bisphenol–A(BPA), 17β–estradiol(E2), estriol(E3), and ivermectin(IV) from agricultural wastewater using the hydrophilic-lipophilic balance(HLB)-SPE column. Recoveries of the pharmaceuticals from standard aqueous solutions containing spiked concentrations of between 2 and 10 µg/l were: E2, 76.62 – 85.47 %; AC, 78.29 - 94.34 %; TC, 88.35 – 92.15 %; CHR, 76.62 – 88.35 %; SA, 79.38 – 81.49 %; E3, 85.42 – 92.15 %; BPA, 80.27 – 89.42 %; CP, 76.58 – 90.21 %; DP, 75.46 – 87.55 % and IV, 80.27 – 84.89 %. Various levels of veterinary drugs - AC, < 0.48 – 1.07 µg/l; SA, < 1.37 – 15.49 µg/l; TC, < 3.45 – 4.57 µg/l; CP, 0.45 – 2.46 µg/l; and IV, < 1.74 – 1.63 µg/l were detected in the grab water samples. The results of the health risk assessment clearly showed mutagenic activity being observed in samples from sheep and poultry farms. It also showed high oestrogenic activity in the pig farm. The results indicated that making use of the maximum concentration of 17β Estradiol found in the samples, there was a slight risk of developing cancer through accidental ingestion via recreational activities with higher risk if the water was used for domestic purposes without treatment to remove them or if the water was used for irrigation purposes.

388
Characterization of respective contribution of agriculture and urban sources to pesticide contamination of a peri-urban river
V. Dufour, EPOC - UMR 5805 CNRS / UMR 5805 - EPOC LPTC; C. Chollet, J. Cruz, University of Bordeaux / UMR 5805 - EPOC LPTC; D. Granger, M. Capdeville, M. Chambolle, LyRE - Centre de Recherche et Développement SUEZ; H. Budzinski, University of Bordeaux
Water is nowadays a precious resource on which anthropic pressure increased drastically these last years, due to global lifestyle improvement and the population growth. Pesticides are part of the most preoccupant micropollutants in aquatic environments because of their intrinsic toxicity, even at trace-levels. They were firstly used for agricultural yield improvement but they are now used as biocides for the protection of construction materials, wood, textiles, paints, etc., or as veterinary treatment susceptible, and can be discharged in rivers via wastewater treatment plants (WWTP) or Separated Stormwater Overflow (SSO). This multiplicity of uses is linked to high concentrations in rivers, affecting aquatic ecosystems that play role of final receptacle for micropollutants in general. It is thus necessary to contain pesticide inputs to protect water resources. Treatment of pesticides can be quite expensive and inputs may not be clearly identified or collectible, and reduction at source can be considered as an interesting alternative (dose reduction, practice changes, etc.). However this approach requires first of all the identification of uses responsible of inputs, as few information is available nowadays. This study monitored for 4 years a peri-urban continuum formed by a river, SSO and a WWTP also as wastewaters to link uses and presence in environment. Water bodies presented distinct contaminations profiles: rivers were characterized by important concentration of plant protection products while WWTP effluents presented important concentrations of biocides and veterinary molecules which are among the most toxic pesticides. Flux calculation allows identifying agriculture as the major source of plant protection products while WWTP brought the most part of biocides and veterinary products, especially in low-flow period when the WWTP contributes up to 40% to the overall flow of the studied river. Storm sewers had an intermediate status, with less consequents inputs but are still significant because of lack of treatment on these effluents and a potential increase of concentration around the

discharge site. Investigation in the wastewater network identified uses responsible for introduction of some molecules like fipronil or glyphosate which is essential in order to implement actions of reduction at source.

389

Study of bioconcentration of benzophenone-3 in Gilt-head Bream and characterization of its by-products

H. Ziarrusta, L. Mijangos, University of the Basque country UPVEHU / Department of Analytical Chemistry; R. Montes, University of Santiago de Compostela; R. Rodil, University of Santiaga de Compostela; J. Quintana, University of Santiago de Compostela; E. Anakabe, University of the Basque country UPVEHU / Organic Chemistry; U. Izagirre, University of the Basque Country UPV/EHU / CBET Research Group Dept Zoology and Animal Cell Biology Faculty of Science and Technology and Research Centre for Experimental Marine Biology and Biotechnology PIE; N. Etxebarria, M. Olivares, O. Zuloaga, University of the Basque Country UPV/EHU / Plentzia Marine Station (PiE-UPV/EHU) & Dep Analytical Chemistry

Benzophenone-3 (BP-3) is a widely used organic UV filter to protect humans and materials from damage by UV irradiation, and owing to its hydrophobic properties, it could potentially bioaccumulate in aquatic biota, including fish (tens to thousands of ng/g). BP-3 can undergo both photodegradation or phase I and phase II metabolization, generating transformation products and metabolites. Environmental risk assessment approaches often require information on the free concentration in water, bioacummulation factors in aquatic organisms, by-products and their toxicity in order to determine the effect of a contaminant on ecosystems. Thus, in the present work, in order to assess the uptake, distribution in different tissues (liver, muscle and gill) and bio-fluids (plasma and bile), metabolism and elimination of BP-3 in gilt-head bream (*Sparus aurata*), a controlled dosing 14-day experiment was designed at 50 ng/mL concentration level. BP-3 was detected in all the analysed samples, with the highest concentrations at day 14. Bile concentrations were significantly higher in comparison to the rest of tissues/fluids. Since BP-3 is hydrophobic and non-ionizable compound, the lowest concentrations of BP-3 were found in plasma. Although liver tissue (highly lipidic) could be an appropriate reservoir of BP-3, the low concentration of non-metabolized BP-3 found in this tissue could indicate a high metabolization activity in liver. And on the contrary, the lack of biodegradation activity in muscle (less lipidic) can explain the second highest concentrations detected, reaching the equilibrium state in the 4th exposure day. In any case, the occurrence of BP-3 in gills suggests that at least part of the uptake occurred through the gills. To completely characterize BP-3 exposure, the analysis performed by means of liquid chromatography – high-resolution mass spectrometry allowed the identification of a broad suite of BP-3 by-products in seawater and fish tissue/biofluids (mainly in bile and liver). By the interpretation of the MS2 spectra, we identified demethylation, hydroxylation and glucuronidation as the main degradation pathways of BP-3. *Acknowledgements* - This work was financially supported by the Ministry of Economy and Competitiveness through the projects CTM2014-56628-C3-1-R and CTM2014-56628-C3-2-R, Xunta de Galicia (ED431C2017/36) and FEDER/ERDF. H. Ziarrusta is grateful to the Spanish Ministry and L. Mijangos to the Basque Government for their predoctoral fellowships.

390

Phragmites australis enantioselectively uptake, translocate and degrade the chiral pesticides tebuconazole and imazalil

L. Tao, Nottingham Trent University / School of Animal, Rural and Environmental Sciences; P. Carvalho, Aarhus University, Department of Environmental Science / Department of Environmental Science; M.E. Casas, Aarhus University / Department of Environmental Sciences; U.E. Bollmann, Aarhus University / Department of Environmental Science; C.A. Arias, H. Brix, Aarhus University / Department of Bioscience; K. Bester, Aarhus University / Department of Environmental Science

Phytoremediation is an emerging technology that utilises green plants and their associated rhizosphere microorganisms to clean polluted environmental media. However, the role of plants in removing organic pollutants is still not well understood. Phytoremediation of realistic environmental concentration (10 μg L^{-1}) of the chiral pesticides tebuconazole and imazalil by a wetland plant, *Phragmites australis*, was investigated. The experiment was carried out in a growth chamber using plants of similar initial size (6.0 ± 0.2 g fresh biomass). The plants were placed in 700 mL glass vessels containing 500 mL hydroponic solution. The pesticides were spiked separately (n=27 for each) in parallel with control samples (n=15). The experiment ran for a period of 24 days. Enantioselective fractions and transformation products (TPs) in both hydroponic growth solutions and plant tissues were measured by HPLC-MS/MS. The uptake, translocation and metabolisation of tebuconazole and imazalil inside *Phragmites australis* were documented for the first time using enantioselective analysis. The pesticides removal efficiencies from water were 96.1% and 99.8%, respectively, by the end of the experiment (day 24). Removal from the solutions could be described by first-order removal kinetics (k=0.14 d^{-1} for tebuconazole and k=0.31 d^{-1} for imazalil). Four different processes occurred simultaneously: 1) removal of the pesticides from the hydroponic solution, 2) plant uptake, 3) pesticides translocation in the plant, and 4) degradation within the plant. Tebuconazole and imazalil concentrations inside Phragmites showed a maximum level at day 10 and 5 d, respectively, followed by a decrease of both compounds concentration. Two TPs of tebuconazole could only be quantified in solution, while two imazalil TPs were quantified in both solution and plant tissue. The uptake of both pesticides by the plant was positively correlated with evapotranspiration. The removal of imazalil and tebuconazole from the hydroponic solution was not enantioselective, however, both translocation and degradation inside *Phragmites* were enantioselective. For tebuconazole, the enantioselective degradation was found in both *Phragmites* roots and shoots.

391

Effects of the non-steroidal antiinflamatory ibuprofen on growth and metabolic profiles of Vigna Unguiculata

Y. Pico, University of Valencia / Medicine Preventive; R. Alvarez-Ruiz, University of Valencia; L. Wijaya, A.H. Alfarhan, M. Alyemeni, King Saud University; D. Barcelo, IIQAB-CSIC / Department of Environmental Chemistry

The non-steroidal anti-inflammatory ibuprofen is one of the most frequently detected pharmaceuticals in wastewater treatment plants. Its metabolism has been widely studied in mammals, fungi and microbes. However, little is know on how ibuprofen is metabolized by plants, mostly due to analytical methodology gaps for determining these compounds at low concentration in complex matrices. In this study, the effects of ibuprofen treatment on the growth and its comprehensive metabolic profile in whole plant cultures and seed germinates of *Vigna unguiculata* were investigated using ultra-high performance liquid chromatography quadrupole time-of-flight mass spectrometry (UHPLC-QqTOF-MS). To this end, the developed method achieved simultaneous quantitative analysis of ibuprofen, 1 and 2-hydroxyibuprofen and carboxyibuprofen while preserving the instrument ability to get precursor and product ion mass spectra of non-target compounds. The trigger was the precursor ions to reach 100 cps intensity. Seeds of *V. unguiculata* were obtained from Gizan area of Saudi Arabia, were germinated in Petri `plates or sown in pots under graduated treatment regimen (control, 400, 800, 1200, 1600, and 2000 mg L^{-1} of ibuprofeno). Seeds and plants were incubated in a growth chamber in the dark at 26 ºC for 5 days. Forty-six metabolites of ibuprofen in *V. unguiculata* were successfully identified. The 1-hydroxy and 2-hydroxy ibuprofen were confirmed and quantified using their analytical standards. The structures of the other metabolites were proposed using high resolution mass spectrometry (HRMS) and high resolution tandem mass spectrometry (HRMS/MS) data. In particular, the combination of mass accuracy and the fragmentation patterns of metabolites and parent compounds allowed proposing plausible structures for each metabolite. Six hexosides were already reported in study on *Phragmites australis* and *Lemma gibba*. Thirty-eight of the identified metabolites were already reported in a study on cell cultures of A. thaliana and 9 of them (conjugates of ibuprofen or hydroxyibuprofen with amino acids) are, up to our knowledge, reported for first time in plants.

Prioritisation and Intelligent Testing of Pharmaceuticals in the Environment (I)

392

Environmental Risk Assessment of Active Pharmaceutical Ingredients used in Human Medicinal Products: Europe-wide Variation in Risk Quotient

J. Snape, AstraZeneca UK Ltd. / AstraZeneca Global Environment; S. Owen, AstraZeneca / Safety Health Environment; C. Tyler, University of Exeter / Biosciences College of Life and Environmental Sciences; B. Verbruggen, University of Exeter; L. Gunnarsson, University of Exeter / Biosciences

This presentation will describe the total consumption-based environmental risks posed by 130 human medicinal products and the impact of mode of action, lipophilicity and dilution on these risks. In accordance with Article 8(3) of Directive 2001/83/EC, as amended, a new marketing authorisation application shall be accompanied by the evaluation of the potential environmental risks posed by the human medicinal product. These environmental risk assessments (ERAs) estimate the potential environmental impact on a product-by-product basis rather than a substance-by-substance basis. In the cases where an active pharmaceutical ingredient (API), or substance, is used to treat multiple clinical diseases, there is the potential to under-estimate environmental impact. The European Medicines Agency (EMA) guidance for the environmental risk assessment of human medicinal products has been in place now for over 10 years. The introduction of this guidance marked a step change in the ERA requirements for human medicinal products, with a shift from short-term acute to long-term chronic environmental effects assessments, and tailored ERAs for active pharmaceuticals ingredients (APIs) with suspected or known reproductive effects. To determine the total substance or API risk, we have: (i) identified and collected definitive published no observed effect concentrations (NOECs) for available APIs (excluding anti-infectives and anti-parasitic products); (ii) collated human consumption data for each of these APIs in European Countries where these products are licenced for use; (iii) conducted a worst case exposure assessment (predicted environmental concentration; PEC) and estimated the total risk posed by each API; (iv) analysed the variability in the risk quotients (RQs) for each API across Europe and (v) looked at the impact of country-specific dilution factors applying the 5th percentile and median dilution factors and comparing risks to the EMA default dilution factor of 10.

393

Estimation and prioritization of hospital API emissions

A.M. Ragas, Radbound University / Department of Environmental Science; C. van Loon, M. Gulpen, K. Tipatet, Radboud University; R. Oldenkamp, Radboud University Nijmegen / Department of Environmental Science

Hospitals constitute an important source of APIs, particularly for substances like antineoplastics and contrast agents. Measuring these emissions and their impacts is possible, but is very time-consuming and costly. The main aim of the present study was to develop an approach for estimating hospital API emissions based on hospital purchase data and to prioritize these emissions based on potential environmental impact. A model was developed to estimate the API loads reaching the hospital sewer system. The model accounts for the return of unused APIs, route-specific excretion by patients, non-patient API use (e.g., personnel) and off-side emissions. The model was operationalized for 16 APIs emitted by two academic hospitals in the Netherlands. Model predictions were validated based on measurements of APIs in the sewer system using passive samplers with speedisk® as absorbent. The samplers were deployed over a 10-12 day period and analyzed in the laboratory using LCMS. Most of the estimated loads were within a factor of 10 of the measured loads. On average, estimations for Hospital 2 were more accurate than for Hospital 1, which was probably due to the use of monthly purchase data and some other small model improvements implemented for Hospital 2. APIs which are typical for hospitals (e.g., antineoplastics and contrast media) were relatively well predicted. The prioritization of the APIs based on environmental impact was substantially influenced by the availability and interpretation of toxicological data. Diclofenac ranked highest, but this ranking was determined by one particular toxicity study of which the validity is being disputed. Ciprofloxacin consistently ranked high, and to a lesser extent also paracetamol and metoprolol. Azithromycin and iomeprol also ranked relatively high, but only limited toxicity data were available for these substances, resulting in large safety margins.

394

Development and validation of a model to predict concentrations of human APIs in European surface waters

R. Oldenkamp, S. Hoeks, V. Barbarossa, M. Cengic, Radboud University Nijmegen / Department of Environmental Science; L. Carter, University of York / Environment Department; E.E. Burns, University of York / Chemistry; J. Wilkinson, The University of York / Natural and Built Environments; A. Boxall, University of York / Environment Department; A.M. Ragas, Radbound University / Department of Environmental Science

Active Pharmaceutical Ingredients (APIs) are consumed in large quantities, and end up in the environment as a result of not being degraded completely during passage through the human body and wastewater treatment plants (WWTPs). Although reported concentrations are generally low, adverse ecological effects caused by some human APIs are plausible considering their specific modes of action and high potency. Consequently, the issue of human APIs in the environment has been acknowledged as an emerging environmental problem requiring scientific and regulatory attention. A crucial step in environmental risk assessment of APIs is the estimation of their environmental exposure potential. Since there are currently more than 4,000 different human APIs in use, monitoring individual APIs is practically impossible. The aim of the present study was to develop and validate a model for the prediction of human API concentrations in European surface waters based on country-specific per capita consumption data. The starting point for modeling the environmental fate of APIs are country-specific per capita consumption data. Subsequently, the modeling chain follows the steps of excretion into the sewerage system, transport to and fate in WWTPs, emission into surface waters and, finally, environmental transport, partitioning and degradation. Unique features of the model include the extensive location-specific information about European WWTPs, the flexibility in modeling Europe's hydrology and accounting for ionizing properties of APIs. The model was validated using several studies reporting API concentrations in the Rhine basin. API-specific data and characteristics (e.g. physicochemical properties and consumption data) were obtained from the literature. Site-specific and API-specific measurements were directly compared to estimated water concentrations at the relevant locations in the river network. From the results shown for the Rhine basin and preliminary results of some additional basins, it can be concluded that estimations can be made with great spatial and quantitative accuracy. However, model performance depends on factors such as the allocation of country-specific consumption over relevant WWTPs, accuracy of the estimated hydrology, provided consumption data and API-specific characteristics.

395

Occurrence and fate of the antidiabetic metformin and its transformation products

S. Tisler, Environmental Analytical Chemistry, Center for Applied Geoscience, University of Tuebingen / Analytical Environmental Chemistry; C. Zwiener, Environmental Analytical Chemistry, Center for Applied Geoscience, University of Tuebingen / Geosciences

Effluents of municipal wastewater treatment are major entry pathways for pharmaceuticals and their transformation products (TPs). A wide-spread compound is the antidiabetic drug metformin (MF) with its well-known main metabolite guanyl urea (GU). GU is formed in WWTPs. So far, no other TPs of metformin are reported in the water cycle. In this study, electrochemical experiments for simulation and identification of potential new TPs of MF were performed. In addition we investigated the occurrence and fate of MF and its TPs in WWT and surface water. Analysis was performed by LC-high-resolution-mass-spectrometry (HRMS) using HILIC (hydrophilic interaction chromatography) and quadrupole-time-of-flight mass spectrometry (QTOF-MS). Four TPs of MF have been identified after electrochemical degradation. The proposed structures are 4-amino-2-imino-1-methyl-1,2-dihydro-1,3,5-triazine (4,2,1-AIMT), 2-amino-4-methylamino-1,3,5-triazine (2,4-AMT), 2,4-diamino-1,3,5-triazine (2,4-DAT) and methylbiguanide (MBG). The mass error was below 3 ppm for all 4 TPs. However, the well-known TP GU could not be formed electrochemically. The TPs found are similar to those of a former study using gamma radiolysis (Collin et al. 2004). 24-hour mixed samples of wastewater in Southwest Germany were obtained for 7 consecutive days. Elimination of MF was 92 % at an average influent concentration of 24 µg/l. GU concentrations were in the influents between 66 and 640 µg/l and in the effluents between 60 and 386 µg/l. A plausible reason for the occurrence and relatively high concentrations of GU compared to MF could be the formation of GU already in parts of the sewer system. The following oxidation products of MF have been detected for the first time: 2,4-DAT, MBG and 2,4-AMT. The concentrations of MBG ranged between 40 and 122 ng/l. For the other TPs no authentic standards were available, however TPs 2,4-DAT and 2,4-AMT showed similar increasing abundance trends from influents to effluents, which implies their formation during WWT. 3 grab samples of surface water affected by waste water showed relatively high MF (between 100 and 470 ng/l) and GU (between 3700 and 4500 ng/l) concentrations. MBG was in the range between 10 and 30 ng/l. In addition, 2,4-DAT was detected. Its response was in all three samples about 40 % of the response in the WWTP effluent samples. The study is performed within the project "Effect-Net", funded by the Ministry for Science, Research and Art, Baden-Wuerttemberg

396

Development of biotransformation half-life QSARs and PBT assessment refinement of Pharmaceuticals and Personal Care Products

E. Papa, A. Sangion, University of Insubria / Department of Theoretical and Applied Sciences (DiSTA); J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology; P. Gramatica, University of Insubria / Department of Theoretical and Applied Sciences (DiSTA)

Pharmaceuticals and Personal Care Products (PPCPs) are of particular interest for the environment since it has been demonstrated that many of them are persistent or "pseudo-persistent" due to their extensive use and continuous emission. They have potential for bioaccumulation in organisms of different trophic levels and exposures to PPCPs can result in acute and chronic effects in target and non-target organisms. Guidelines for Environmental Risk Assessment of human medicines require, in the first preliminary phase, an assessment to verify if a PPCP ingredient may be Persistent, Bioaccumulative and Toxic (PBT). Biotransformation has been recognized as a key determinant for bioaccumulation assessment for many chemicals. This study addresses the development of QSAR models for the prediction of *in vivo* whole body human biotransformation half-lives measured or empirically-derived for over 1000 PPCPs, mainly represented by pharmaceuticals. Five datasets with data for the total elimination half-lives (HL_T) in human adults and the derived whole body in vivo biotransformation half-life (HL_B) were used to develop HL-QSAR models based on theoretical molecular descriptors. The statistical parameters calculated for the models reflect the good ability to fit the data in the training sets (R^2 range: 0.77 – 0.80) the robustness (Q^2_{LOO} and Q^2_{LMO} range: 0.77 – 0.79) and the external predictivity (Q^2_{ext} range: 0.75-0.79; CCC range: 0.86-0.87). These QSARs were used, in combination with literature models for the prediction of biotransformation half-lives in fish, to refine the screening of the potential PBT behaviour of over 1300 PPCPs. Principal Component Analysis (PCA) was applied to combine experimental biotransformation half-life data and reliable QSAR-predictions to assess biotransformation process in multiple species (i.e. fish and human). This analysis was helpful to highlight the priority PPCPs according to their slow or fast potential for biotransformation, i.e. 22 slowly biotransformed compounds were highlighted as potential PBTs, 18 compounds formerly detected as PBTs resulted easily metabolized, while the PBT behavour of 59 PPCPs may have been underestimated. This study highlights the importance of biotransformation for the refinement of screening level assessments of Bioaccumulation and Bioaccumulation-related behaviour (i.e. potential PBT behaviour) of chemicals and shows how *in silico* approaches can be efficiently integrated to support these assessments

397

Predicting spatial and temporal variability in internal concentrations of amitriptyline in invertebrates within an urban catchment

A. Giorgis, University of York; A. Agatz, IBACON GmbH / Environment Department; L. Carter, University of York / Environment Department; E. Burns, University of York; M.D. Nuñez, University of York / Environment; J. Wilkinson,

The University of York / Natural and Built Environments; R. Ashauer, University of York / Environment; A. Boxall, University of York / Environment Department
The majority of active pharmaceuticals ingredients (APIs) currently in use are ionisable and may become charged at environmentally relevant pHs. Previous research has shown that the accumulation of a molecule in aquatic invertebrates depends on the ionisation state of the molecule which is driven by the pH of the surrounding medium. Recently a toxicokinetic modelling approach has been proposed to account for variation in accumulation into aquatic organisms in relation to pH. Here, we present this modelling approach to derive toxicokinetic parameters from laboratory experiments for the accumulation of amitriptyline, an antidepressant compound, in *Lumbriculus variegatus.* Toxicokinetic (TK) parameterisation and the underlying experiments involved the measurement of uptake of amitriptyline into *L. variegatus* at four medium pHs (5.5, 7, 8, 9).To simulate accumulation at the landscape scale, we used the generated toxicokinetic parameters in combination with measured monthly concentrations of amitriptyline in river water and associated water pHs obtained from a one year long monitoring study along the two rivers (rivers Ouse and Foss) in the City of York, UK. Data from the experiments at pH 5.5 and 9 were used to successfully parameterise the TK model. Data for two pH values (pH 7 and 8) were then used to predict uptake and depuration rates for the neutral and ionised species of the API. Use of the derived rate constants to simulate the accumulation at the two intermediate pHs revealed that the approach underpredicts the actual accumulation by a factor of 2-4. Predictions of internal concentrations of amitriptyline in *L. variegatus* varied by 6 times across the monitoring sites and over the year. Generally, internal concentrations were predicted to be much higher for the river Foss (which had a concentration range of 0-52.2 pmol/g and a pH range of 7.63-8.45) than the river Ouse (which had a concentration range of 0-2.95 pmol/g and a pH range of 7.41-8.44) and accumulation increased during the course of the year. Even though the model underestimated the internal concentrations by a factor of 2 and 4 in laboratory experiments conducted at pH 7 and 8 respectively, the general approach worked reasonably well to obtain internal concentrations. This study reveals important insights into the accumulation of pharmaceuticals by non-target organisms which will help to fully understand the risks posed by pharmaceuticals at the catchment scale.

Plants: predicting and assessing direct, indirect effects and recovery of plants from chemical stress

398

Waterplants in Risk Assessment - Selection of Potential Plant Species - Impact of Different Test Guidelines

G. Gonsior, Eurofins Agroscience Services Ecotox GmbH

When results of standard laboratory tests show an unavoidable high risk, aquatic higher tier tests are needed to reduce uncertainties. In case there is a high risk for aquatic plants additional species could help to reduce these uncertainties for risk assessment by performing Species Sensitivity Distribution (SSD) tests. However, it is not clear which criteria are used for selection of test species and which guideline is useful for adaption to non-standard species. Most tests were performed based on the *Lemna* guideline OECD 221, the two *Myriophyllum spicatum* guidelines OECD 238 and OECD 239, the ASTM E1913-04(2012) for *Myriophyllum sibiricum*, *Sediment contact test* with *Myriophyllum aquaticum* (ISO/DIS 16191) and the principles of the method proposed by the AMRAP (Aquatic Macrophyte Risk Assessment for Pesticides) Workgroup (MALTBY et al. 2010). Further a proposed ring test protocol for the emergent macrophyte *Glyceria maxima* was presented by Jo Davies et al. In addition a high number of forms and reactions of water plants could be observed during several years of testing. Some scenarios will be given and an introduction in the complexity of water plant testing for risk assessment will be provided. The focus will be to generate a robust test design which is applicable for most of the water plant species. A proposal for a test design in accordance to the existing guidelines and testing protocols adapted to a broad range of test species will be presented. Based on the EFSA opinion, designs will be discussed and an over view will be given how the test designs can be further adapted to provide a refined risk assessment.

399

Applying the EFSA Scientific Opinion on NTTP: Testing non-crop species and the reproductive capability of selected species under greenhouse conditions

A. Duffner, D. Ripperger, C. Groening, P. Mack, Eurofins Agroscience Services Ecotox GmbH; S. Knaebe, EAS Ecotox GmbH / Ecotox Field; T. Moser, Eurofins Agroscience Services Ecotox GmbH

Agriculture is the dominating land-use of the EU member states by covering nearly half of the surface area. Using herbicides to reduce weed competition in agricultural areas can adversely affect non-target terrestrial plants growing at field margins. According to the recent EFSA Opinion for non-target terrestrial plants (2014) one important goal is maintaining the biodiversity of plant species in the agricultural area. It is therefore recommended to include also non-crop species in the testing scheme from the list provided in OECD guidelines (OECD 208 and 227) and to assess the life-cycle with flowering and seed production. The objective of this study was to assess the viability of generative traits of non-crop species for risk assessment. For this purpose generative traits were evaluated if they provide more relevant information for the risk assessment. For this purpose they were compared with the vegetative traits, such as mortality and biomass production, which are currently assessed in the OECD guidelines 208 and 227. The selected non-crop species are included in commercially available seed mixtures for flowering strips. Our experimental design consists of one control and four different herbicide application rates, with 6 replicates. The field rate was considered as highest application rate. Assessed were mortality, biomass and phytotoxicological effects. The number of flowers and plant height were assessed for selected species to evaluate differences in development and flowering. In addition, seeds were sampled to evaluate differences in seed quantity and quality. Furthermore, the results will be compared to a non-target terrestrial plants pilot field study (Knaebe et al. 2017; Presentation at SETAC Europe 2017). References: EFSA PPR Panel (2014). Scientific Opinion addressing the state of the science on risk assessment of plant protection products for non-target terrestrial plants. EFSA Journal 2014;12(7):3800, 163 pp. OECD (2006). Test No. 208: Terrestrial Plant Test: Seedling Emergence and Seedling Growth Test. OECD Publishing, Paris.
OECD (2006). Test No. 227: Terrestrial Plant Test: Vegetative Vigour Test. OECD Publishing, Paris.

400

Predicting plant community level effects of herbicides based on monoculture dose-responses: Testing the plant community model IBC-grass with experimental data

J. Reeg, University of Potsdam / Plant Ecology and Nature Conservation; S. Heine, Bayer Ag / Effect modelling; C. Mihan, Bayer CropScience AG / Ecotoxicology; T. Preuss, Bayer Ag / Environmental Safety; F. Jeltsch, University of Potsdam

Ecological models are rarely found in terrestrial plant ecotoxicology and risk assessment. Especially on community-level, the number of suitable plant models is scarce. Existing models are often not validated with experimental data, although the validation of ecological models is an important point for their credibility for risk assessors. Nevertheless, ecological models are a suitable tool to extrapolate individual-level greenhouse experiments to the plant communities in the off-field. Especially bearing in mind that protection goals for non-target terrestrial plants as defined by EFSA are on population and community level. Reuter and Siemoneit-Gast (2007) performed an experiment that includes not only the test of monocultures but also the test of small artificial communities consisting of 6 different plant species. This study is suitable to be coupled and compared to a plant community model, since it covers the population as well as the community level. We adapted the plant community model IBC-grass to the settings of the empirical study by Reuter and Siemoneit-Gast (2007). Specifically, we analyzed to which degree the model is able to represent realistic community level effects not only by comparing visual patterns but also by calculating model adequacy and reliability as a measure for the model fit. The predicted effects of the model IBC-grass show a good agreement with the experimental data, for the monocultures as well as for the communities. Model adequacy was lower in the monocultures. However, model adequacy increases in the communities. In general, model reliability is high in the monoculture and community set up if the effect of the selective herbicide is simulated. Population level and community level effects on plant biomass predicted by the plant community model IBC-grass were in good agreement with the measured effects from the experimental study of Reuter and Siemoneit-Gast (2007). This agreement indicates the model is able to reasonably represent intra- and interspecies competition and predict community level effects based on dose-response data. Therefore, the model can serve as an important tool for predicting potential impacts on natural environments with information obtained from greenhouse studies on individual species.

401

Use in risk assessment of recovery in plants from exposure to chemicals

T.A. Springer, EAG Laboratories / Specialist Projects & Histology; H.O. Krueger, EAG Laboratories / Aquatic, Plant and Insect Toxicology; J.W. Green, DuPont / Data Science and Informatics

The plant interest group of SETAC has a committee working on the topic of recovery and this presentation concerns statistical issues arising from this work. Traditionally evaluating the risk of chemicals to plant species or communities involves assessing both lethal and nonlethal effects, but little or no consideration is given to whether the effects at the population or community level are transient or persistent. Considering the ability of plants to recover after the exposure to a chemical is important when evaluating effects on populations conducting a risk assessment. For example, a young plant in a vegetative vigor study may show leaf damage within a day after it is sprayed, but after two weeks of growth that damage may no longer be apparent as old damaged leaves have senesced and only new unaffected leaves are visible. While it is relatively easy to design studies to assess recovery of vegetative growth in terrestrial plants, this may not be indicative of recovery of the ability of a population of plants to sustain itself. In algae or lemna studies, an aliquot of cells can be transferred to untreated media at the end of a test and after several days, the growth rates of the affected groups may approach that of the controls indicating recovery. Recovery in terms of growth rate of these simple aquatic plants is likely to be indicative of a population's ability to sustain itself. In more complex mesocosm studies the concept of recovery is even further complicated by seasonality, changes in nutrients, recolonization, competition, and other factors. Terminology and methodology need to be standardized if the concept of recovery is incorporated into evaluations of chemicals. For some plant types and properties, recovery is contingent upon the timing and duration of exposure and extent of injury. In such cases, the definition of recovery must specify timing and duration of exposure in the operational definition. Examples of recovery in laboratory studies for algae, lemna, and myriophylum studies will be presented as well as results from mesocosm studies. Statistical procedures and experimental designs will be presented for these examples and interpretation of results will be discussed.

402

Aquatic primary producers and plant protection products: endpoints and level of protection achieved in the first tier of the risk assessment scheme

S. Duquesne, UBA, Federal Environment agency; L. H�nemann, S. Matezki, M. Solé, K. Swarowsky, German Environment Agency UBA; J. Wogram, German Environment Agency UBA / Department IV plant protection products

In the risk assessment for aquatic primary producers exposed to plant protection products (PPP), the endpoint (EP) corresponding to 50% inhibition of growth (EC50s) is used in the first tier. The EC50s can be expressed as inhibition of the average specific growth rate (ErC50) or as reduction in biomass, calculated from yield (EyC50) or as the integral under the growth curve (EbC50). The lowest available EP among ErC50, EbC50 or EyC50 used to be selected to derive safe concentrations of pesticides in surface water bodies. It is now recommended [1] to use ErC50s since it is a more robust endpoint. However, it is not yet clear if the protection level achieved is sufficient. This work shows that this new approach (i.e. selecting ErC50) shifts thus the level of conservatism of a factor of 6.9 and 3.5 for algae and *Lemna sp*, respectively. It also shows that the level of protection achieved for primary producers becomes insufficient in 59% of the cases, since the Tier 3 Regulatory Acceptable Concentrations (RACs) from micro-/mesocosm studies (considered as surrogate reference Tier) are lower than the Tier 1 RACs from standard toxicity tests. The results demonstrate that the intended level of protection is currently reached in only 41% of the cases versus 69% of the cases previously. In addition, this work explores which combination of E_rC_x ($E_rC_{10,\ 50}$ etc.) and assessment factor would ensure an adequate level of protection. Recommendations are provided for an optimization of the risk assessment.

403

Poster spotlight: WE152, WE153, WE154

Environmental monitoring of contaminants using terrestrial ecological biomonitors

404

Persistent Organic Pollutants in Germany: Results from the 2015/2016 moss and tree sampling

A. Dreyer, Eurofins GfA GmbH / Air Monitoring; S. Nickel, University of Vechta / 2; J. Koschorreck, Umweltbundesamt; W. Schröder, University of Vechta / 2

This presentation aims at reporting on the determination of selected POPs in eight moss samples from Germany within the framework of the 2015 European moss survey and comparing the respective results with such derived for leaves and needles collected for the German Environment Specimen Bank (ESB). The moss samples were collected in areas located in the vicinity of tree sampling sites from the ESB in fall 2016. Deciduous tree leaves and coniferous shoots were sampled in 2015 or 2016 according to the ESB protocols. Overall, 17 polychlorinated dibenzo dioxins and -furans (PCDD/F), 18 polychlorinated biphenyls (PCB), 16 polycyclic aromatic hydrocarbons (PAH), 17 perfluoroalkyl substances (PFAS), 3 isomers of hexabromocyclododecanes (HBCD), 7 polybrominated biphenyls (PBB), 24 polybrominated diphenyl ethers (PBDE), and 21 alternative halogenated flame retardants (HFR) such as Dechlorane Plus were determined. Except for PBBs and PFASs, POPs of all substance groups could be quantified, although to different extents. Concentrations of individual PAHs and HFRs were in the same order of magnitude as those observed in coniferous shoots or deciduous tree leaves from nearby located areas. Highest levels of PCDD/F, dl-PCBs, HBCD and PAH in moss were observed at sites close to the Belauer See (Northern Germany, agricultural land-use) and the Harz National Park. Concentrations of PBDEs were highest at the two sampling sites in Saarland (conurbation) and at the Harz site. Concentrations for Dechlorane Plus were highest at the Harz site followed by sites located at Solling (forestry) and Scheyern (agricultural) and were lowest at the site in the Halle-Leipzig conurbation. Thus, surrounding land-use does not seem to be the (only) driving force determining the POPs burden in moss samples. PBDE moss concentrations observed in this study were similar to those observed at background sites in Spain and lower than those of background/remote sites in Norway. Concentrations of Dechlorane Plus were more than a factor of 100 higher than moss concentrations reported for Svalbard (Arctic Norway).

405

Mapping percentile statistics of element concentrations in moss collected from 1990 to 2015 in forests throughout Germany

W. Schröder, S. Nickel, University of Vechta / 2

Monitoring and mapping of atmospheric deposition can be achieved by use of chemical transport models, technical samppling devices and bioaccumulators such as moss. Within the European moss survey programme, since 1990 every five years moss have been sampled at up to about 7300 sites in up to 36 countries. Sampling, chemical determination of heavy metals (since 1990), nitrogen (since 2005), and persistent organic pollutants (since 2010) in moss specimens, quality control and statistical evaluation was conducted according an harmonized methodology [1]. Mapping the percentile statistics of heavy metals and nitrogen concentration in moss sampled in forests accross Germany is in the focus of this paper. Thereby, element- and survey-specific as well as heavy metals and surveys integrating statistical evaluations and GIS-mapping were performed. Cr, Hg, Sb and Zn show, contrary to Fe and Pb, no constant decrease of element concentrations, but an intermediate increase between 2000 and 2005, which did not contuinue untill 2015. Al, As, Cd, Cu and V stagnated between 2000 and 2005, Hg from 2005 to 2015. Therefore, Cr, Sb and Zn will be focused in this paper together with Cd, Hg, Pb and N which are of priority according to the Convention on Long-range Transboundary Air Pollution. Survey-specific staistical analyses corroborate that the spatial patterns of element concentrations in moss are changing accross time. The long-term information on the percentile statistics of bioaccumulation of atmospheric deposition in moss is essential for further scientific evaluation as well as for measurements and reporting of nature protection and environmental management. **References** [1] Nickel S, Schröder W (2017) Reorganisation of a long-term monitoring network using moss as biomonitor for atmospheric deposition in Germany. Ecological Indicators 76:194-206. [2] Schröder W, Nickel S, Völksen B, Dreyer A. (2017) Nutzung von Bioindikationsmethoden zur Bestimmung und Regionalisierung von Schadstoffeinträgen für eine Abschätzung des atmosphärischen Beitrags zu aktuellen Belastungen von Ökosystemen. 4. Zwber., F&E UFOPLAN 3715632120, i.A. UBA, Dessau. Text: 82 S, 4 Anh.: 212 S. **Keywords**: Bioaccumulation of atmospheric deposition, European moss survey, heavy metals, nitrogen. *Acknowledgement* - The authors thank the German Environment Agency for funding.

406

Heavy metal and nutrient concentrations in different age classes of holm oak leaves and pine needles - a reference for biomonitoring and geochemistry

J. Franzaring, A. Fangmeier, University of Hohenheim / Institute of Landscape and Plant Ecology; L. Paoli, University of Siena / Dept. of Life Sciences; S. Ancora, University of Siena / Physical sciences, Earth and environment; S. Schlosser, University of Hohenheim / Core Facility Hohenheim; F. Monaci, University of Siena / Dept. of Environmental, Earth and Physical Sciences

Passive biomonitoring is being used for many years to assess changes in the state of the environment. Existing programs make use of international, national, regional and local monitoring networks addressing the effects of the widespread deposition of air pollutants and eutrophying compounds and the accumulation of these in e.g. forest and agricultural ecosystems and their effects on related food chains. However, plants differ in the uptake of essential and non-essential inorganic elements and organic pollutants and the accumulation varies over time and space mainly due to the different geochemistry and climates in different regions. Here we report on the results of a biomonitoring study using holm oak (*Quercus ilex* L.) and two pine species (*Pinus nigra* J. F. Arnold and *P. pinaster* Ait.), i.e. evergreen deciduous and coniferous species common to Mediterranean ecosystems. Leaves and needles were sampled at different locations in the Mount Amiata and Colline Metallifere region in spring 2017 and the samples consisted of three age classes. While half of the analyses were performed on unwashed samples, the other half rested on samples that were thoroughly rinsed with deionized water prior to the analyses. ICP-MS, ICP-OES and CNS elemental analyses were applied for the determination of Cu, Sb, Al, Ba, Cr, Ni, V, Fe, Hg, P, K, Mg, As, Pb, Cd, Zn, C, N and S. Apart from the significant differences in element concentrations and patterns between the tested species, regions and sites, differences between the washed and unwashed samples were less pronounced indicating that deposition of dust does not play a great role in the area. On the other hand, our preliminary analyses showed significant differences in element concentrations between different ages classes, which relate to the availability, translocation, accumulation or growth dilution of plant essential and non-essential elements. Multi-element analyses and nutrient ratios can serve to differentiate between the characteristic geochemical and species-specific patterns and the positive derivation from these patterns points to the exceedance of element levels due to pollution and eutrophication. Biomonitoring in post-mining areas serves to identify pollution hotspots and can be used as a key component in controlling the success of land reclamation for nature and agriculture.

407

Examining historical trends in diet and contaminant exposure in bats using bat guano deposits from Jamaica

L. Gallant, University of Ottawa / Department of Biology; C. Grooms, Queens University; L.E. Kimpe, University of Ottawa / Department of Biology; J.P. Smol, Queens University / Biology; W. Bogdanowicz, Museum and Institute of Zoology;

J.M. Blais, University of Ottawa / Biology
Bats are excellent ecological indicators owing to their long life span, global distribution, and predictable responses to environmental stressors. Bats play important roles in pollination, seed dispersal, and insect population control and thus it is important to determine whether bat diets change over time as a result of exposure to contaminants such as metals. Bat guano deposits are of particular use as they may serve as environmental archives as the cave environment preserves stable isotopes and metals which allows for the determination of historical exposure to contaminants as well as any fluctuations in diet. This research provides the rare opportunity to examine two bat guano deposits from Jamaica: bat guano was heavily mined for gun powder and fertilizer and as such, there are few bat guano deposits that have been unaltered by human exploration or exploitation. The objective of this research is to reconstruct historical changes in diet and contaminant exposure to bats in order to better understand how anthropogenic activity affects these high trophic level mammals. We constructed the ^{210}Pb, ^{137}Cs, and ^{14}C dating profiles in both bat guano deposits: this revealed that one of the deposits is over 3,000-years-old. We constructed the δ^{15}N, δ^{13}C, and δ^{34}S profiles in order to determine the long-term dietary trends in the bat guano deposits. Preliminary results suggest that the 3,000-year-old bat guano deposit is tracking a change in stable isotopes associated with the agricultural history of Jamaica, specifically, the introduction of: nitrogen fertilizers, the Bordeaux mixture, and sugarcane. We also examined the sterol profiles in the bat guano deposits for the purpose of determining more specific dietary information. Recent peaks in cholesterol and stigmastanol, for example, could be evidence of fluctuations in feeding habits (or bat colony composition) over the past 3,000 years. We also present the long-term increase in metals such as Cd, Hg, Pb, and Zn within the bat guano deposits associated with contaminant exposure in relation to the onset of the Industrial Revolution, a period characterized by an increase in atmospheric emissions of metals owing to increased mining and production. Lastly, we present the decrease in ^{206}Pb/^{207}Pb within the bat guano deposit in association with the introduction of leaded gasoline.

408
Perfluoroalkyl substances and metallic elements in South African dragonflies
H. Bouwman, North-West University / Unit for Environmental Science and Management; V. Lesch, North-West University; Y. Shibata, National Institute for Environmental Studies / Fellow; A. Kinoshita, National Institute for Environmental Studies
Adult dragonflies are aerial predatory arthropods that occur globally. However, no research on adult dragonflies as potential indicators of environmental metallic elements or perfluorinated substances (PFASs) pollution have been done. PFAS and elevated concentrations of environmental metallic elements are toxic to organisms and can cause disruption of biological processes. Adult dragonflies were collected and analysed for PFASs and metallic elements. The results indicated that dragonflies from farming areas had significantly lower ΣPFASs concentrations than sites located closer to industrial areas (median ΣPFASs of 0.32 ng/g wm (wet mass) for North, and 9.3 ng/g wm for South). All substances, except perfluorooctanesulfonic acid (PFOS) occurred at similar concentrations at all six sites, when quantifiable, but PFOS dominated in the Southern sites. The highest median concentration was from Bloemhof Dam (ΣPFASs = 21 ng/g wm), which is known to be polluted by PFOS. The results also indicated that all species of dragonflies, regardless of body size and habitat type preference are suitable indicators of environmental metallic elements. Sites located near wastewater treatment plants had elevated concentrations associated with mining and industries. Dragonflies from sampling sites near potential pollution sources that seemed to have isolated water sources, showed lower metallic element concentrations when compared with other sites. Based on these results we conclude that dragonflies would be excellent indicators of environmental metallic elements and PFAs.

409
Bioavailability of Arsenic and Antimony co-contamination to vegetable crops in agricultural soils
L.P. Egodawatta, University of Wollongong / School of Chemistry; A. Holland, La Trobe University; D.F. Jolley, University of Wollongong / School of Chemistry
Antimony (Sb) is an emerging contaminant that is assumed to behave in a similar way to arsenic (As). Sb and As often co-occur because of mining. Bioaccumulation and phytotoxicity of As is well studied, but there is little evidence on Sb and its interactive effects with As. Metalloid accumulation in agricultural soils may present health risks and hazards to humans and ecosystems through direct ingestion or contact with contaminant soil and food, a reduction in food quality (safety and marketability) via phytotoxicity and reduction in land usability for agricultural production. Plant bioassays allow inferences regarding the potential toxicity of contaminants. The phytotoxicity in the contaminated soils is governed by the bioavailability of the contaminant, which in turn is influenced by soil physical and chemical characteristics, contaminant speciation and the species of plant. However, it is still unclear the impacts of ageing of agricultural lands have on the co-contamination of As and Sb on alternation of crops. Our study evaluated the potential use of vegetable crops to identify and assess the bioavailability and toxicity of As and Sb in co-contaminated soils using bioassays. Water spinach and choy sum are herbaceous leafy vegetable belonging to the morning glory (*Convolvulaceae*) and mustard (*Brassicaceae*) families, respectively. Our study compares the soil characteristics in terms of total and bioavailable metal fractions with plant accumulation and toxicity data. Plant toxicity parameters (tissue biomass and lengths) were used to evaluate impacts of contaminant exposure on plant productivity. This information was used to understand the tolerance of plants grown in As and Sb contaminated soils, and the risks associated with As and Sb co-contaminated soils. This was done as single element and mixed metal exposures. Test soils were silty sand and slightly acidic. Bioavailable As and Sb in soils increased proportionally with total metal concentration. A clear increase in the tissue accumulation of As and Sb was observed with increasing bioavailable metal fraction for both individual (As and Sb) and combined (As+Sb) treatments. Vegetable productivity decreased when grown in As only and As+Sb combined contaminated soils. Sb contamination in agricultural soils poses a greater human health risk and hazard than As only and As+Sb co-contamination, because Sb accumulates in edible crops with no observed phytotoxicity or reduction in the vegetable productivity.

Systems ecotoxicology: application of OMICS data across multiple level of biological organization in research and risk assessment (I)

410
Transcriptomic responses of the endangered freshwater mussel Margaritifera margaritifera to trace metal contamination
A. Bertucci, F. Pierron, Université de Bordeaux / UMR EPOC CNRS 5805; J. Thébault, Université de Brest / LEMAR UMR 6539 CNRS/UBO/IRD/Ifremer; C. Klopp, INRA Institut National de la Recherche Agronomique / Plate-forme bio-informatique Genotoul, Mathématiques et Informatique Appliquées de Toulouse; J. Bellec, Université de Brest / LEMAR UMR 6539 CNRS/UBO/IRD/Ifremer; P. Gonzalez, University of Bordeaux / UMR EPOC CNRS 5805; M. Baudrimont, Université de Bordeaux / UMR EPOC CNRS 5805
The freshwater pearl mussel *Margaritifera margaritifera* is one of the most threatened freshwater bivalves worldwide. In this study, we aimed (i) to study the processes by which water quality might affect freshwater mussels *in situ* and (ii) to provide insights into the ecotoxicological significance of water pollution to natural populations in order to provide necessary information to enhance conservation strategies. *M. margaritifera* specimens were sampled in two close sites located upstream or downstream from an illegal dumping site. The renal transcriptome of these animals was assembled and gene transcription determined by RNA-seq. Correlations between transcription levels of each single transcript and the bioaccumulation of 9 trace metals, age (estimated by sclerochronology) and condition index were determined in order to identify genes likely to respond to a specific factor. Amongst the studied metals, Cr, Zn, Cd and Ni were the main factors correlated with transcription levels, with effects on translation, apoptosis, immune response, response to stimulus and transport pathways. However, the main factor explaining changes in gene transcription appeared to be the age of individuals with a negative correlation with the transcription of retrotransposons-related genes. To investigate this effect further, mussels were classified into 3 age classes. In young, middle-aged and old animals, transcription levels were mainly explained by Cu, Zn and age, respectively. This suggests differences in the molecular responses of this species to metals during its lifetime that must be better assessed in future ecotoxicology studies.

411
LC-HRMS based-metabolomics to highlight biotransformation products and effects of diclofenac in Mytilus galloprovincialis
F. Courant, Université de Montpellier - UMR 5569 Hydrosciences / UMR Hydrosciences; B. Bonnefille, H. Fenet, Université de Montpellier / UMR Hydrosciences; E. Gomez, Université de Montpellier, Hydrosciences Montpellier UMR 5569 / UMR Hydrosciences
Diclofenac (DCF) has become a major contaminant of interest as shown by its inclusion in the EU Water Framework Directive (2015/495/EC). However, relatively little is known regarding its biotransformation and effects in the Mediterranean mussel. Environmental metabolomics affords several advantages to study both topics. The metabolome refers essentially to i) the “endometabolome”, constituted by endogenous metabolites, and to ii) the “xenometabolome”, in reference to xenobiotics and their biotransformation products [1]. Metabolomics profiles acquired through mass spectrometric techniques may reveal the exposure by direct detection of xenobiotics and their metabolites (xenometabolome investigation) and effects by the detection of endogenous metabolites which concentrations may differ from physiological levels following the exposure (endometabolome investigation). To demonstrate the approach feasibility, an experiment was carried out whereby marine mussels were exposed for 7 days to ethanol (< 1°/oo, vehicle) or to 100 µg/L DCF. Analytical methods relying on Liquid Chromatography-High Resolution Mass Spectrometry were developed to generate metabolic profiles from mussel’s tissues. The obtained profiles for both groups (controls and exposed) were compared. We highlighted DCF and 13 DCF metabolites in exposed mussels. Three of them were phase I metabolites such as

4'-hydroxy-diclofenac, and 10 were phase II metabolites such as amino acids conjugates. Five were reported for the first time in an aquatic organism. Regarding the effects, two main metabolic pathways were found to be impacted by diclofenac exposure. The tyrosine metabolism was mostly down-modulated and the tryptophan metabolism was mostly up-modulated. To our knowledge, such DCF effects on mussels have never been described despite being of concern for these organisms: catecholamines and serotonin are involved in osmoregulation, and in gamete release in mollusks [2-4]. Our results highlighted potential impairment of mussel osmoregulation and reproduction following a DCF exposure in agreement with recent publications that have shown reproductive disturbance following DCF exposure in other aquatic organisms such as xenopus [5] or fish [6]. [1] Holmes et al., Anal. Chem. **79**, 2629 (2007) [2] Wang & Croll, Aquaculture **256**, 423 (2006) [3] Fong et al., J. Exp. Zool. **267**, 475 (1993) [4] Fong et al., J. Exp. Zool. **266**, 79 (1993) [5] Efosa et al., Chemosphere **173**, 69 (2017) [6] Gröner et al., Chemosphere **166**, 473 (2017)

412

Metabolomics used to link affected molecular pathways with behaviour outcomes after a single dose of pesticide exposure in mice

P. Leonards, VU University, Institute for Environmental Studies / Department of Environment and Health; H. Viberg, I. Lee, S. Buratovic, P. Eriksson, Uppsala University

Worldwide, serious concern has arisen about the increased incidence of learning and developmental disorders in children. From a scientific point of view, there is no doubt that exposure to neurotoxic chemicals during early brain development can adversely affect learning and development. Various recent epidemiological studies have indicated that exposure to low doses of environmental biologically active contaminants (e.g. pesticides) during human development can have deleterious effects on cognitive development in childhood. The European commission-funded project DENAMIC "Developmental Neurotoxicity Assessment of Mixtures in Children" investigates neurotoxic effects (e.g. learning and developmental disorders) of low-concentration mixtures of pesticides and a number of common environmental pollutants in children. We focus on (subclinical) effects on learning (cognitive skills) and developmental disorders in children (e.g. ADHD, autism spectrum disorders and anxiety disorders). The aims are to develop better and sophisticated tools, procedures and testing methods to screen compounds for (developmental) neurotoxicity, and to improve our understanding of chemical exposures and the observed effects (www.denamic-project.eu). As part of the project, a new alternative assessment strategy based on a combination of *in vitro, in vivo* assays, omics, and human exposure assessment is under development in order to prioritize compounds, and to further investigate the pathways and mechanism involved in disorders and diseases. The final aim of DENAMIC is to reduce effects of environmental contamination on learning and developmental disorders in children. In the current study metabolomic pathway analysis was used to improve our understanding of the underlying molecular mechanisms of observed effects on behaviour and cognitive function after various pesticide and other contaminant exposures in mice.

413

Relationships Between Persistent Pollutant and Metabolomics Profiles in Tissues of Polar Bears From Hudson Bay, Canada

A.D. Morris, Environment and Climate Change Canada / Ecotoxicology and Wildlife Health; R.J. Letcher, Environment and Climate Change Canada / Ecotoxicology and Wildlife Health Division; M. Dyck, Government Of Nunavut / Nunavut Department of the Environment; B. Chandramouli, J. Cosgrove, SGS AXYS

Metabolomics profiles are comprised of targeted endogenous metabolites (< 1 kDa) such as amino acids (AAs), fatty acids (FAs), and membrane lipids such as phosphatidylcholines (PCs) to identify how changes in the metabolome relates to extrinsic factors, including e.g. exposure to persistent organic pollutants (POPs) and metals. Polar bears (*Ursus maritimus*) from Hudson Bay (Canada) are differentially exposed to complex mixtures of POPs and metals including total mercury (THg = inorganic + methyl-mercury), and legacy and new POPs. In the present study, quantifiable profiles of 295 organic POPs and THg in fat and liver (respectively) as well as 156 endogenous metabolites in liver of Southern (SHB; n = 14) and Western Hudson Bay (WHB; n = 15) male polar bears were combined for multivariate and univariate statistical analyses. Correlated compounds and significantly different or impacted physiological pathways were identified that may be related to differences in POP exposure or other environmental factors. Partial least squares discriminant analysis (PLS-DA) and variable importance in projection (VIP) were applied to the combined metabolite-contaminant profiles of these polar bears, and Spearman correlation analyses were used to establish relationships between metabolites and contaminants, as well as with other biological factors. Forty-one metabolites [membrane lipids, acylcarnitines (ACs), and symmetric dimethyl arginine (SDMA)], and 21 POP discriminated the subpopulations. Perfluorinated alkyl substances (PFASs), polybrominated diphenyl ethers (PBDEs), *p, p'*-dichlorodiphenyldichloroethylene (DDE) and some *ortho*-polychlorinated biphenyls (PCBs) were greater in the SHB bears and changes in the metabolite concentrations had some consistency with previous laboratory studies. Arachidonic acid (ARA), glycerophospholipid and amino acid pathways were identified as being differently enriched or impacted between the subpopulations. Greater ARA in SHB bears may be related to differences in chronic exposure to POPs such as the hepatotoxic PFASs, as ARA is part of the inflammatory response in liver. SDMA and most legacy organochlorine compounds were greater in the WHB bears, which may be indicative of differences in renal function. Consistency of relationships between metabolites, POPs biomarkers from laboratory studies suggests linkages between POP concentrations and differences in the hepatic metabolome of SHB and WHB polar bears.

414

Integrative Omics linkage to reproduction effects of a fungicide in the soil invertebrate Folsomia candida

T.F. Simoes, S.C. Novais, Polytechnic Institute of Leiria / MARE IPLeiria; T. Natal da Luz, University of Coimbra / Department of Life Sciences, University of Coimbra; J. Sousa, University of Coimbra / Department of Life Sciences; B. Devreese, Ghent University / Laboratory for Protein Biochemistry and Biomolecular Engineering; T. de Boer, Vrije Universiteit; D. Roelofs, Vrije Universiteit / Department of Ecological Science; N. van Straalen, Association of Retired Environmental Scientists ARES / Department of Ecological Sciences; M.F. Lemos, Instituto Politécnico de Leiria / MARE IPLeiria

Due to high complexity of ecosystems, environmental risk assessment can be a challenging task and there is the need to develop and validate innovative and reliable tools and integrated approaches for fast detection of changes in population and community structures that can be applied by regulatory agencies. *Folsomia candida* is among the most sensitive representatives of its taxon and has been selected as a genomic model organism for soil toxicology studies on non-target soil arthropods. This work aimed to determine the toxicity mechanisms of a widely applied fungicide formulation (Bravo500®), with active compound chlorothalonil (CHT), in *F. candida*, by linking effects at different levels of biological organization like reproduction, gene expression and protein levels, following a time series exposure. Therefore, organisms were exposed in laboratory to a natural agricultural soil. To find the reproduction EC50, several dilutions of the formulation were spiked according to nominal concentrations of the active ingredient. For the mechanistic assessment of effects, and to better understand the correlations between omics information through time, organisms were then exposed to the estimated EC50 of the formulation (plus control) and sampled at consecutive time points (2, 4, 7, and 10 days). Four replicates per treatment and time point were used (32 in total). CHT exerts its toxic effects primarily through binding to thiol-rich molecules (ex. glutathione), exhibiting often a multi-site activity and the results with the formulation were very indicative of these mechanism of toxic activity. Also in this study, results point for similar effects of the fungicide formulation, affecting mainly mechanisms involved in detoxification and excretion (also involving glutathione), normal cellular respiration and protein metabolism, leading to impairment in development and reproduction. The datasets presented highly significant positive correlations between the gene expression levels at a certain time-point and the correspondent protein products from the consecutive time-point, thus highlighting the importance of considering a time series analysis when integrating geno- and phenotypic data. Integrated omics could thus provide useful insights, exhibiting their relevance in toxicological studies and proving the importance of a time-series analysis in correlations between these datasets.

415

Using functional genomics to find mechanisms of herbicide toxicity in Chlamydomonas reinhardtii

A. Zupanic, Eawag Swiss Federal Institute of Aquatic Science and Technology / Environmental Toxicology; A. Betz, Eawag / UTOX

At present, environmental risk assessment of chemicals is limited to measuring physiological endpoints in model species. To test all chemicals that require testing, a shift to mechanistic-based testing is needed. However neither direct molecular targets nor the stress pathways that lead to adaptation to chemical exposure are usually known. Finding the genes encoding sensitivity or tolerance to chemicals is one of the highest priorities of the (eco)toxicological research community. One of the best method for gene function discovery is functional genomics based screening, in which libraries of single-gene mutants are exposed to interventions (such as chemical exposure) of interest and mutants which are the most susceptible to the intervention and those that are most tolerant are found. We exposed a pooled library of loss-of-function mutants of *Chlamydomonas reinhardtii* to three herbicides that target photosynthesis: atrazin and diuron which target photosystem II, and which disrupts the transition of electrons from photosystem I to photosystem II which leads to production of reactive oxygen species. The pooled library was exposed to the EC20 concentration of each herbicide for 3-4 days, at which time samples from the library were taken for DNA sequencing to find mutants that have become enriched or have disappeared from the culture after exposure (compared with non-exposed control). The expectation was that the functional genomic profiles of atrazin and diuron would be similar, as the target of both chemicals is the same, while a different profile would be obtained for paraquat. This was indeed the case, with the profiles for both diuron and atrazine enriched for mutant of genes involved in the photosynthesis. The profiles of paraquat also included photosynthetic genes, but also several genes involved in defence against oxidative stress and lipid metabolism. Finally, there were also several genes that were among the enriched separately for diuron and atrazin, which points to possible different secondary modes of action for both herbicides. While we are currently still analyzing the obtained profiles and individual genes, our results demonstrate that functional genomics is a useful method for discovery of molecular mechanisms of chemical toxicity.

Harmful effects of plastic litter and mitigation strategies in the Mediterranean Sea

416

Harmful effects of plastic litter on Mediterranean Biodiversity: what and what's new?

M. Fossi, M. Baini, C. Panti, University of Siena / Department of Physical Sciences, Earth and Environment

Concern about the occurrence, quantity and effects of marine litter in the world's ocean and seas has grown rapidly in recent years, attracting interest from a wide range of stakeholders: governments, environmental NGO, the scientific community, the media and the general public. Mediterranean Sea, which is a crucial biodiversity hotspot and a critically polluted area, has been also described as one of the areas most affected by marine litter, including microplastics, in the world. Recent studies in the different regions of the basin suggested that some areas are affected by important concentration of microplastics and plastic additives, representing a potential risk for endangered species (baleen whales, sea turtles, filter feeders sharks) and for the all Mediterranean biodiversity. To cover the current knowledge gaps on this issue a harmonised methodological approach for the assessment of the marine debris impact on Mediterranean biodiversity is needed. The quantification of marine debris/microplastics in the marine environment can depend on several environmental factors and change according to multiple oceanographic features, and therefore, cannot reflect the potential impact on organisms and ecosystems. The information obtained by biondicator species could better integrate the spatial and temporal presence of marine litter/microplastics in the marine environment. In addition, the use of biondicators can allow to measure not only the occurrence of marine litter in the species and its environment but also the threat posed to organisms by the evaluation of contaminants accumulation and any related biological effect. A new integrated monitoring tool that would provide the information necessary to design and implement future mitigation actions in the Mediterranean basin is proposed. Applying ecological and biological criteria to the most threatened species obtained by statistical analysis, bioindicator species for different habitats and monitoring scale were selected. To assess the harm by marine debris ingestion a threefold approach, simultaneously measuring the presence and effects (accumulation of plastic associated contaminants and biomarker responses), can provide the harm and the sub-lethal effects to organisms related marine litter ingestion. The gaps pointed out by this research and the bioindicators species selected could represent a step forward for the risk assessment and the implementation of future mitigation measure for the Mediterranean area, habitat and species affected by marine litter ingestion.

417

Impact of marine litter in the Mediterranean Sea: monitoring and specific reduction measures within MSFD

F. Galgani, IFREMER

Periodic assessments of the state of the marine environment, monitoring and the formulation of environmental targets are perceived as part of the continuous management process within the MSFD. Of the 11 descriptors listed in Annex I of the MSFD for determining GES, Descriptor 10 has been defined as "Properties and quantities of marine litter do not cause harm to the coastal and marine environment". In 2016, the Revised Commission Decision identified four indicators for Descriptor 10, of which two are focusing on harm considering (i) The amount of litter and micro-litter ingested by marine animals is at a level that does not adversely affect the health of the species concerned (indicator 10DC3), and (ii) The number of individuals of each species which are adversely affected due to litter, such as by entanglement, other types of injury or mortality, or health effects (indicator 10DC4). For these two indicators, Member States shall establish that list of species to be assessed and thresholds values for these levels through regional or sub regional cooperation. In the context of the Mediterranean Sea, we discuss the ongoing work that is focusing on the implementation of monitoring and reduction measures, defining constraints, protocols, better defining harm and research needs to support monitoring efforts and reduction measures.The analysis of existing data will reveal (i) the suitability of some approaches for better monitoring the adverse effects of litter, and (ii) the potential of visual observations of the sea floor for the measurement of interactions between litter and invertebrates as an approach for evaluating entanglement. Strategies for the implementation of monitoring are discussed, as well as risk assessment and the possible associated measures within MSFD.

418

Addressing the growing threat of marine litter: NGOs essential role in strengthening the science-policy-society interface

T. Vlachogianni, Mediterranean Information Office for Environment, Culture and Sustainable Development (MIO-ECSDE)

The growing urgency and complexity of interconnected societal challenges, such as marine litter, demand that they be addressed through the strengthening of the science-policy-society interface so as to provide the necessary conditions for translating research-based knowledge into effective action. NGOs are essential partners in promoting environmental protection and achieving sustainable development. Their active participation at local, national and transboundary level in all phases of projects and processes, from their design, implementation in the field, operationalization, monitoring and evaluation, contributes not only to increased transparency, wide visibility and outreach of the project or process, but also to enhanced overall quality and increased ownership of the outcomes, as well as amplified possibilities for replication of its activities. In full acknowledgement of the prominent role of NGOs in the realm of environmental governance, MIO-ECSDE, a Federation of some 130 Mediterranean NGOs working on Environment and Sustainable Development, in fulfilling its vision and mission, has developed and implemented a number of actions on the science-policy-society interface that address the growing threat of marine litter in the Mediterranean, ranging from the monitoring and influencing of relevant policy, all the way to hands on and pilot activities (e.g. within the framework of the IPA-Adriatic DeFishGear, the FP7 MARLISCO, the Interreg Med ACT4LITTER, the EU SWIM-H2020 SM, etc.). How marine litter and its inherent environmental, economic, social, political and cultural dimensions have been tackled by MIO-ECSDE illustrates the broad extent of involvement and interventions required for the protection of the marine and coastal environment. Furthermore, it highlights the diversity, added value and particular strengths that the NGO community brings to environmental governance, such as leadership, creativity, flexibility, entrepreneurship and capacity for vision and long-term thinking. As scientists call for more research on global environmental changes in an effort to gain a better understanding of the human induced implications for all of life on Earth, it remains an inconvenient truth that if the world had acted upon the knowledge that the scientific community already produced, the state of many ecosystems would be different today. The NGO community has an essential role to play in terms of strengthening the science-policy-society interface towards an effective response from society on the issue of marine litter and other societal challenges.

419

Biodegradable plastics: potential application in aquaculture and other applications at high risk of dispersion

F. Degli Innocenti, Novamont SpA

The problem of plastic marine debris is caused by inadequate waste management and entails investments in consciousness, prevention, and recovery programs. The bioplastics industry does not consider biodegradability as a license for littering in the environment. All packaging and consumer products must be recovered in some way at their end of use and, in certain contexts, biodegradability allows recovery through organic recycling. This option is contemplated by the European Directive on Packaging and it is beneficial whenever packaging is mixed with food waste (biowaste). The term "biodegradable" could be misunderstood and induce the consumer to littering. In order to avoid such problem, the biodegradable packaging is labelled "compostable" or "biodegradable and compostable". The term "biodegradable" is only used in business-to-business communications (e.g. "biodegradable" mulch films are used by professionals who are well aware of the meaning of the term). In agriculture, tests specific to soil define mulch film biodegradation because this depositional environment is microbiologically different from composting. Similarly, tests specific to the marine environment are now under development at ASTM and ISO level. Some biodegradable plastics showed biodegradation levels (as CO2 evolution) comparable to cellulose in less than 1 year using these test methods. Generally speaking, the environmental risk depends on the concentration of the environmental stressor and on its residence time in the environment. The lower the concentration and the shorter the residence time, the better. Bioplastics do not immediately disappear upon exposure to the sea. However, biodegradability reduces the risk by reducing the stressor's residence time. Concluding, the idea of solving the problem of plastics in the ocean just by shifting to bioplastics is groundless (bioplastics does not disappear "by magic"). However, for those applications where accidental release is certain or very probable, biodegradability decreases the environmental risk. Materials that show full and relatively fast biodegradation may be suitable for plastic products known to wear down or become stranded (for example, fishing gear) and scatter into the sea. Bioplastics hold promise for aquaculture professional applications (e.g. nets for mussels farming) where the disposal of plastic waste is an inevitable outcome. Bioplastics can be the right solution for specific products, if properly applied.

420

Marine Litter Governance in the Mediterranean through the implementation of the Regional Action Plan on Marine Litter Management in the Mediterranean

T. Hema, UN Environment / Mediterranean Action Plan Coordinating Unit; G. Leone, UNEP/Mediterranean Action Plan

The UN Environment/ MAP Barcelona Convention was the first Regional Sea Programme to approve a legally-binding Regional Plan on Marine Litter

Management (RPML) in December 2013, providing for a set of programmes of measures and implementation timetables to prevent and reduce the adverse effects of marine litter on the marine and coastal environment. It includes innovative and traditional measures of a policy, regulatory and technical nature, addressing different aspects of marine litter prevention and management from land and sea based sources. The Regional Plan measures impose clear obligations regarding the waste management hierarchy, closure of illegal dumping/dumpsites, shift to sustainable consumption and production patterns, removal of existing marine litter using environmental sound practices e.g. fishing for litter, clean up campaigns, port reception facilities at possibly no special fees, and monitoring, assessment and reporting on implementation of measures as well as enforcement of national legislation. Significant effort has been made on marine litter at regional and national levels, since the adoption of the RPML. The Mediterranean countries have included marine litter in their updated National Action Plans (NAPs) and the ambitious and novel Integrated Monitoring and Assessment Programme (IMAP) of the Mediterranean Sea and Coast and Related Assessment Criteria has been adopted since 2016 by the Mediterranean countries including two common and one candidate indicators on marine litter. Furthermore, with the support of the EU-funded Marine Litter MED project, UN Environment/MAP is implementing key reduction and prevention measures on marine litter in the Southern Mediterranean. At the invitation of the UN Environment/MAP, the Regional Cooperation Platform on Marine Litter in the Mediterranean was established in September 2016, consisting of more than 20 international and regional partners with a clear mandate on marine litter management. The aim of the platform is to assist the implementation of the RPML, enhance cooperation and maximize results, towards a more effective marine litter management at regional level. One of the latest developments of the UN Environment/MAP is the 2017 Mediterranean Quality Status Report (QSR) that dedicates two chapters on marine litter related to beach, floating, and seafloor marine litter.

421
Science and awareness: a Mediterranean Connection Against Marine Litter. First Results of the Commitment Presented at UN Ocean Conference
G. Zampetti, Legambiente
“Science and awareness: a mediterranean connection against marine litter” is the title of the voluntary commitment that Legambiente and the University of Siena presented at the last UN Ocean Conference in New York, in June 2017. There was a connection between scientific research and raising awareness built to tackle marine litter in the Mediterranean Sea by sharing experiences and developing new integrated action. In 2013, Legambiente started the monitoring of floating macro litter within Goletta Verde, one of the most popular campaigns of analysis and information about sea pollution. In the last few years, there has been an increase in the marine-litter-related activities, including surveys using citizen science and awareness raising projects. Following the Scientific Environmentalism purpose Legambiente applied official methods and protocols to contribute to the estimation of the marine litter amount in seas and along the coastline, cooperating also with national research institutes, universities and other research organizations. Now, thanks to the cooperation with the University of Siena, a further step has been added carrying out studies and research on the presence of contaminants adsorbed by floating plastics and their potential effects on biodiversity. The first results of this research will be presented in this meeting.

422
Discussion

423
Final Remarks
G. Leone, UNEP/Mediterranean Action Plan

Ecological risks under complex, multiple-stressor threat scenarios: integrating chemical effects with environmental drivers (II)

424
Biodiversity patterns in the GLOBAQUA basins and their relationships with multiple stressors
N. De Castro-Català, Universitat de Barcelona / Department of Evolutionary Biology, Ecology and Environmental Sciences; I. Muñoz, University of Barcelona / Department of Evolutionary Biology, Ecology and Environmental Sciences; E. Kalogianni, I. Karaouzas, A. Vourka, E. Smeti, L. Vardakas, Hellenic Centre for Marine Research, Institute of Marine Biological Resources & Inland Waters (HCMR); M. Paunovic, University of Belgrade, Institute for Biological Research Siniša Stanković; C. Borrego, M. Petrovic, Catalan Institute for Water Research ICRA; S. Sabater, ICRA Catalan Institute for Water Research; S. Diaz-Cruz, M. Farre, IDAEA-CSIC / Environmental Chemistry; S. Monllor, N. Guillem, Institute of Environmental Assessment and Water Research IDAEA CSIC / Department of Environmental Chemistry; M. Lopez de Alda, Institute of Environmental Assessment and Water Research; S. Lutz, Hemholtz Centre for Environmental Research UFZ; A. Bellin, E. Stella, University of Trento / Department of Civil, Environmental and Mechanical Engineering
Rivers suffer from an important decrease in species diversity compared to other aquatic and terrestrial ecosystems due to a variety of stressors related to human activities. Species play different roles in the functioning of the ecosystem, and their species loss may reduce the response capacity of the ecosystems to a stressor. The effects on diversity will obviously differ based on the type of stressors and their combination and severity, as well as on the characteristics of the local community composition, and the community tolerance to the type of stressor affecting the system. This study presents the results of the analyses of the biodiversity patterns for bacteria, algae, macrophytes, macroinvertebrate and fish communities related to environmental pressures. The data obtained from the field work conducted in three of the Globaqua case study basins (Adige, Sava and Evrotas) has been evaluated according to structural biological community parameters (species composition and abundance). The most evident relationships between changes in species richness and diversity were explained by changes in hydrology (e.g. mean discharge, intermittency) and morphological changes in the basins (e.g. land uses, channel transformation). The presence of pharmaceutical products (urban pollution) and pesticides was related to lower insect richness. Also emerging compounds, despite their low concentrations, were related with a reduction in macrophyte diversity. These results open the way to compare responses in the studied basins as representative of the European reality of the combined effects of multiple stressors on biological diversity.

425
Changes in pCO2 alter the reproductive toxicity of common active pharmaceutical ingredients
C.M. Hird, N. Wichman, C. Lewis, K. Smith, A. Wilson McNeal, University of Exeter / Department of Biosciences; J. Snape, AstraZeneca UK Ltd. / AstraZeneca Global Environment; T.S. Galloway, University of Exeter / Biosciences
Increasing pCO_2 in aquatic environments is occurring as a consequence of the release of anthropogenic carbon dioxide in to the atmosphere, which is absorbed by surface waters. Traditionally this stressor has been studied in isolation, however environmental variation such as changes in pCO_2 or pH can alter the ionisation and consequently the effects of contaminant compounds. A notable group of compounds susceptible to these changes include the active pharmaceutical ingredients (APIs), which often have pH-specific biological effects and are increasingly detected in sewage effluent and receiving waters. The aim of this study is to investigate the hypothesis that changes in pCO_2 alter the effects of active pharmaceutical ingredients on sperm swimming parameters and fertilisation success. The species chosen to explore these effects were the lugworm *Arenicola marina* and the purple sea urchin *Paracentrotus lividus* due to them being keystone coastal species in areas where API contamination is occurring, and them being established model species for artificial spawning in controlled laboratory conditions. We used a range of neonicotinoid pesticides and non-steroidal anti-inflammatory drugs (NSAIDs) at both environmentally relevant and mechanistic concentrations to test this relationship due to them having chemical properties identified as making them pCO_2-sensitive. pCO_2 conditions equating to current (8.10 ± 0.1) and future (7.75 ± 0.1) pH conditions were selected for this study. Endpoints measured included a range of sperm motility parameters, using computer-assisted sperm analysis (CASA) software and fertilisation success. Our findings indicate that pCO_2 conditions may play a vital role in determining the toxicity of common chemical pollutants through changes in sperm swimming parameters and consequently fertilisation success. Our results also indicate that this combination of stressors is compound-specific between contaminants with multidirectional effects dependent on conditions of the exposure. Our findings provide novel evidence that future environmental conditions may substantially alter the role which contaminants such as APIs play in the environment. API contamination is likely to increase following anthropogenic pressures such as population growth and healthcare advances. As a result, we believe it necessary to consider future conditions such as increased pCO_2 conditions when accurately assessing the environmental risks of such compounds.

426
From individual traits to ecosystem functioning: natural phytoplankton community responses under combined environmental stress and chemical pollution
D. Baho, Nowegian Institute for water research; E. Leu, Akvaplanniva AS; F. Pomati, Eawag Swiss Federal Institute of Aquatic Science and Technology / Aquatic Ecology; J. Moe, Norwegian Institute for Water Research (NIVA) / Section for Catchment Processes; D.O. Hessen, University of Oslo / Department of Biosciences; J. Norberg, Stockholm Resilience Centre; L. Nizzetto, NIVA
Phytoplankton are crucial for lentic ecosystems productivity and foodwebs, but facing multiple anthropic challenges that may lead to complex alterations of their ecology and function. Climate change is expected to decrease the stability of lentic ecosystems and enhance fluctuations in environmental conditions. More frequently occurring storm events will potentially disrupt the normal stratification patterns in boreal lakes, thereby dispersing algae from the depth layers they are optimally

acclimated to. Many species of algae are highly adaptive and can respond by growing rapidly after disturbance. However, the diffuse burden of chemical pollutants concomitantly present in freshwater ecosystems, can favour species of algae that are more tolerant to chemical pollution to the detriment of more adaptive ones. We carried out a field experiment, using a non-invasive mesocosm approach, to study the response of phytoplankton communities to combined physical and chemical anthropogenic stressors. We aimed at investigating if chemical pollution can prevent the ecosystem to promptly re-gain structures and functions after extreme events. An extreme metrological event was mimicked by sampling and mixing phytoplankton communities over the entire water depth of the lake. The chemical stressors comprised of a mixture of chemical pollutants added to the mesocosms at five increasing concentrations (typically considerably below the EC_{50} of individual substances). The mixture included 12 pharmaceuticals and personal care products commonly detected in lakes and rivers in Europe. In addition, a mixture isolated from treated waste water effluents was used as a treatment of its own. Individual level traits (cell size, pigments), community parameters (biomass, functional diversity, species composition and photosynthetic efficiency), chemical concentrations and nutrients were routinely monitored during the 3-weeks-experiment. Our results, showed that diffuse contaminants at environmentally relevant concentrations have a drastic persistent impact on the different levels of organisation of the phytoplankton community. At community level, contaminant decreased the photosynthetic yield. At higher concentration levels these effects persisted throughout the duration of the experiment, resulting in lower productivity and communities with contrasting. This suggests that diffuse chemical pollution can disrupt the capacity of natural communities to handle environmental changes.

427

The role of multiple stressors in an Alpine river and the response of the macroinvertebrate community.

M. Giulivo, Universita Cattolica del Sacro Cuore / Institute of Agricultural and Environmental Chemistry; E. Stella, University of Trento / Department of Civil, Environmental and Mechanical Engineering; E. Capri, Universitá Catolica del Sacre Cuore / Institute of Agricultural and Environmental Chemistry; I. Muñoz, University of Barcelona / Department of Evolutionary Biology, Ecology and Environmental Sciences; A. Bellin, University of Trento / Department of Civil, Environmental and Mechanical Engineering

In this study the combined effects of hydrological and chemical stressors on benthic macroinvertebrates are evaluated in order to explore the response of the biological community in relation multiple pressure factors. The Adige River, located in the Southeastern Alps, was selected as a case study because representative of a variety of stressors that characterize the Alpine region. As expected, streamflow showed a seasonal pattern, with higher values in the spring-summer period; however, in some sites the natural hydrological regime was altered by the presence of hydropower plants, which management affected most low values of streamflow. Statistical analysis showed a clear seasonal and spatial pattern in both chemical and hydrological parameters; in detail higher concentrations of nitrate, Personal Care and Pharmaceutical products were found in winter season associated with lower streamflow. Changes in richness, diversity and composition of macroinvertebrate community are related with inputs of urban pollution along the river, and with hydrology, chiefly downstream hydropower plants. Pollution (nitrates and other compounds such as PhACs and PCP) favor higher invertebrate densities but lower diversity, changes in thermal natural regime affects Plecoptera, and *Gammarus* sp density is significantly correlated with flow variability. This study lies in giving a comprehensive and general explanation of the response of biological communities to multiple stressors investigated in an Alpine environment; in particular the analyses performed allow to distinguish the main pressures that impact macroinvertebrates in the Adige river.

428

Coping with antidepressants in a changing ocean: tissue bioaccumulation and behavioural implications in juvenile Argyrosomus regius exposed to venlafaxine

A. Maulvault, Instituto Português do Mar e da Atmosfera / Division of Aquaculture and Seafood Upgrading.; L.H. Santos, Catalan Institute for Water Research ICRA; S. Rodriguez-Mozaz, Institute for Water Research (ICRA) / Water Quality; P. Anacleto, Portuguese Institute for the Sea and Atmosphere / Division of Aquaculture and Upgrading; V.F. Barbosa, Portuguese Institute of Sea and Atmosphere IPMA / Division of Aquaculture and Seafood Upgrading; R. Alves, IPMA, I.P.; J.R. Paula, Faculdade de Ciências da Universidade de Lisboa / MARE Marine and Environmental Sciences Centre; r. rosa, MARE Marine and Environmental Sciences Centre; M. Diniz, UCIBIO, REQUIMTE / UCIBIO, REQUIMTE; P. Pousão, IPMA Portuguese Institute for the Sea and Atmosphere; A. Marques, Portuguese Institute of Sea and Atmosphere IPMA / Division of Aquaculture and Upgrading

Anthropogenic activities have contributed to great environmental challenges: remarkable chemical contamination and dramatic climate change. Both factors strongly affect marine ecosystems and are expected to worsen in the future, threatening marine species' welfare and survival. Yet, information on how fish will cope with the presence of chemical contaminants in the future is still extremely limited. Emerging and non-regulated pharmaceutical and personal care products (PPCPs) have recently become a great environmental concern, since these compounds are often discharged into the aquatic environments, but their elimination through conventional wastewater treatments is rather limited. Within pharmaceuticals of human use, venlafaxine (VFX) is one of the most ubiquitous in the aquatic environment, often reaching higher concentrations than other well-known psycho-active drugs, such as fluoxetine (i.e. Prozac). In this context, the aim of this study was to assess, for the first time, the effect of seawater warming and acidification on VFX bioaccumulation in fish tissues, as well as the behavioural implications resulting from the exposure to these stressors (alone or combined), using juvenile meagre (*Argyrosomus regius*) as model organism. Overall, data evidenced that seawater temperature and pCO_2 levels can strongly affect the bioaccumulation patterns of antidepressants in marine organisms. Furthermore, the distinct behavioural patterns observed when VFX contamination, acidification and warming acted alone or in combination evidenced that multiple environmental stressors should be considered when assessing fish behaviour under a future changing ocean. The results here gathered further strengthen the need to carry out greater research efforts to understand how multiple environmental stressors interact with each other.

429

A modelling approach to assess present and future land use pressures on a salmonid river: a case study in the River Tamar catchment (UK)

M. Assuncao, Cefas Lowestoft Laboratory; P.E. Posen, Centre for Environment Fisheries and Aquaculture Science Cefas; M.G. Hutchins, Centre for Ecology Hydrology

A linked-model approach was applied to the River Tamar catchment (Southwest, UK) to assess current and likely future impacts of land use practices on salmonid populations; Atlantic salmon (*Salmo salar*) and brown trout (*Salmo trutta*). Land use data were incorporated into a validated water quality model (QUESTOR) at the sub-catchment scale and a baseline generated for the period of 2000 to 2012. Future scenarios of water quality were also generated based on land use practices recommended under ongoing catchment initiatives. Overall, the baseline water quality parameters found to be non-compliant with "Good Status" under the Water Framework Directive, or outside the freshwater requirements for salmonids, corresponded with reported land use pressures in the Tamar, namely, catchment-wide frequent elevated levels of inorganic phosphorus and, less frequently, suspended sediments. Tested future land use scenarios bringing improvements in inorganic phosphorus levels included upgrading technology at sewage treatment plants and the implementation of riparian buffer strips, combined with a corresponding reduction in livestock density. These improvements however, were marginal therefore the tested land use scenarios should be adjusted and/or new scenarios explored. Baseline seasonal average values for water quality parameters in different areas of the catchment explained 68% of salmon and trout fry density variation, and showed how different parameters might be affecting the density of these two species. Our results suggest that catchment pressures are contributing to the regulation of salmonid fry densities in some tributaries and upper catchment reaches. Moreover, they can be used to inform local and seasonal targeted measures, aimed at improving those water parameters most influential on fry densities. These types of measures are likely to bring the highest benefits to salmonid productivity in the catchment.

PBT/vPvB & PMT/vPvM substances and Non-extractable residues (NER): Scientific strategies, Analytical challenges and Regulatory Issues (II)

430

Evaluation of PBT and vPvB substances based on exposure dynamics, use-specific impacts and costs for emission reduction or abatement in the context of REACH

S. Gabbert, Wageningen University / Social Sciences; F. Oosterhuis, Vrije Universiteit Amsterdam / Institute for Environmental Studies; S. Hahn, Fraunhofer ITEM / Chemical Risk Assessment; M. Klein, Fraunhofer Institute for Molecular Biology and Applied Ecology IME; M. Nendza, Analytisches Laboratorium

A key objective of the European chemicals legislation REACH is to ensure that the risks caused by substances of very high concern (SVHC) are adequately controlled. The two regulatory procedures adopted in REACH to control the risks arising from SVHCs are authorisation and restriction. Both regulatory instruments make use of socio-economic analysis (SEA), which is generally defined to be a tool for assessing all relevant positive and negative impacts from substances' use or non-use, and for comparing these impacts across different scenarios. Impacts arising from chemicals' use, including PBTs/vPvBs, are use-specific. Furthermore, due to stock pollution properties of PBTs/vPvBs, impacts may last for long periods and even long after emissions have ceased. In addition, information about (long-term) impacts needs to be balanced with costs of emission reduction and abatement. Acknowledging that monetary valuation of impacts using stated or revealed preference methods is not possible for a broader set of PBT/vPvB substances, the evaluation of PBT/vPvB substances in a SEA has to rely on cost-effectiveness analysis (CEA). This requires specifying benchmark values, i.e.

target values (standards) by means of which it can be determined whether or not the costs of a control measure are excessive. This paper suggests an approach for the evaluation of PBT/vPvB substances by means of CEA that accounts for the complex environmental distribution patterns, and that allows balancing (long-term) impacts from PBT/vPvB use against costs for emission reduction and abatement. The approach proceeds along a sequence of steps and uses different analytic tools and data. Starting with a grouping and ranking of PBT/vPvB substances, exposure dynamics are analysed with a multimedia stock pollution approach. Based on the assessment of exposure dynamics in different compartments, impacts arising from the stock can be evaluated via different routes. To assess the cost-effectiveness and proportionality of possible (policy-) measures for PBT/vPvB control, the routes to impact evaluation are linked to assessments of costs for restricting or stopping a specific or multiple uses of a PBT/vPvB substance, and to benchmarks, being standard values of a specific parameter to which the actual/estimated value of that parameter can be compared. As illustrative case study, the approach is applied to perfluorooctane sulfonate (PFOS).

431

Grouping and relative ranking of the impact potential of PBT/vPvB substances for comparative assessments in the context of socio-economic analysis under REACH

M. Nendza, Analytisches Laboratorium; S. Hahn, Fraunhofer ITEM / Chemical Risk Assessment; M. Klein, Fraunhofer Institute for Molecular Biology and Applied Ecology IME; F. Oosterhuis, Vrije Universiteit Amsterdam / Institute for Environmental Studies; S. Gabbert, Wageningen University / Social Sciences

The assessment of PBT/vPvB substances under REACH may evaluate long-term exposure dynamics and impact potential. Grouping and relative ranking of PBT/vPvB substances can support comparative assessments of either several substances with the same use, or for a particular substance with different uses (emission patterns) in the context of socio-economic analysis (SEA). The aim of the grouping of PBT/vPvB substances is to classify a target chemical with respect to its similarity of properties/behaviour with chemicals with known impacts, serving as points of reference for the impact evaluation. The relative ranking of PBT/vPvB substances is based on an impact score, which captures diverse properties and effects of target substances. The impact score consists of individual scores assigned to expected environmental stocks, possible effects of PBTs/vPvBs on the environment and on human health via the environment (toxicological benchmarks), and to other specific concerns. The resulting fingerprints of concern (pattern of the individual scores) and the overall scores for impact potential can be used for comparative assessments. For example, if similar fingerprints are observed for two substances with a similar use, the total score gives an indication which substance might be less (or more) critical. For 17 case study chemicals, data on partitioning properties (log Kow, log Koa), stocks in water/sediment and soil, long range transport potential (LRTP), overall persistence (Pov), toxicological benchmark values for water and soil, CMR properties (H-phrases), endocrine disruption (ED) potential, production/emission volume, and use pattern have been collected from REACH dossiers or estimated with suitable tools. The examples illustrate the possibilities and present limitations of the grouping and relative ranking to describe and evaluate differences of PBTs/vPvB substances with regard to impact potential. Based on current knowledge, this grouping and relative ranking can guide the formation of concern-based categories for a possible read-across or comparative evaluation of impact potential of PBT/vPvB substances. *Acknowledgement* – This work was funded by the European Commission, Directorate-General for Internal Market, Industry, Entrepreneurship and SMEs under Contract number: 30-CE-0830972/00-26 ‘Approach for evaluation of PBTs subject to authorisation and restriction procedures in context of socio-economic analysis’.

432

Interpretation of non-extractable residues (NERs) in the persistence assessment

U. Jöhncke, Federal Environment Agency (UBA) / IV 2.3 Chemicals; V. Bonnomet, European Chemicals Agency ECHA; I. Doyle, Environment Agency / Evidence Directorate; R. Hornek-Gausterer, Environment Agency Austria; A. Kapanen, European Chemicals Agency - ECHA; M. Kästner, Helmholtz centre for environmental research - UFZ / Department of Environmental Biotechnology; J.R. Peltola-Thies, ECHA-European Chemicals Agency; L. Ribeiro; A. Schäffer, Institute for Environmental Research RWTH Aachen University; M. Telscher, Bayer AG Division CropScience/Environmental Fate / Development Environmental Safety; S. Trapp, Technical University of Denmark DTU / DTU Environment; P. Van Elsacker, FPS Health Food Chain Safety and Environment; E. Verbruggen, RIVM Expertise Centre for Substance / Centre for Safety of Substances and Products

Abstract: The aim of this ECHA project on Non-Extractable Residues (NERs) is to improve the interpretation of NERs in the persistence (P) assessment of substances, in particular biocides and REACH substances. The project outcome will be a discussion paper containing an approach proposal and a review of state-of-science on the role of NERs in the degradation assessment in soil, sediment and water with suspended solids. The work will be based on the results of scientific work carried out by the Member States, academia and Cefic/ECETOC in the last few years. The discussion paper will serve as background document for the development of the assessment of NERs under REACH and the Biocides Products Regulation (BPR). It will also be used for updating ECHA Guidance, where appropriate. Different NER fractions will be defined with regard to their potential for binding, remobilisation and hazard. Different extraction methodologies will also be presented that could be used for identifying and quantifying those different NER fractions. The applicability, limitations and potential technical challenges of those extraction methodologies will be discussed. Preliminary results have already indicated that further research on the topic is needed. The project will include recommendations for further research needs and for addressing the limitations identified in the current testing guidelines. The main outcome of the project will be presented at the SETAC meeting.

433

Quantification of different NER fractions in soil - Extraction matters

J. Hogeback, Federal Institute of Hydrology; D. Loeffler, D. Albrecht, A. Martin, M. Fligg, German Federal Institute of Hydrology; A. Wiemann, UBA Umweltbundesamt; G. Speichert, German Environment Agency UBA; A. Hoellrigl-Rosta, Umweltbundesamt / Section Plant Protection Products; T. Ternes, German Federal Institute of Hydrology

The formation of non-extractable residues (NER) of chemicals in soils and sediments is a critical issue for the environmental risk assessment of these compounds, as they may potentially be remobilised as parent or transformation product. However, a standardised or commonly accepted methodology for the characterisation and evaluation of NER does not yet exist. In scientific literature different types of NER are described [1,2]. One type includes the heavily sorbed and the sequestered fraction which are considered to be possibly remobilised into the environment. Therefore, it is neccessary to determine at least this fraction for an adaequate risk assessment. Other types encompass residues which are covalently bound to the soil and those residues where the test substance or its breakdown products were incorporated into the biomolecules. These residues are considered to be irreversibly bound to the soil or transformed into biomass and therefore no risk for the environment can be anticipated from these fractions. The comprehensive scientific assessment of this classification and the analytical accessibility of these NER types will be discussed and supported by experimental data. Therefore, incubation experiments were carried out following the OECD 307 test guideline, which allows a mass balance of the spiked ^{14}C-labelled compound. Three well characterised standard soils were used and with acetaminophen, triclosan and fenoxycarb, three radiolabelled test compounds were selected representing a pharmaceutical, a biocide and a pesticide. The substances are already well described with regard to their degradation kinetics and the formation of different NER types. Different mild to harsh extraction procedures like shake flask extraction and pressurised liquid solvent extraction are comprehensively discussed. Furthermore, different soil matrix destabilising and destroying procedures are evaluated in order to characterize the four NER types proposed by Eschenbach et al. [2]. Finally, a refined extraction scheme shall be proposed with respect to the general applicability for an adequate risk assessment of NER. [1] Kästner et al.. 2014. Classification and Modelling of Nonextractable Residue (NER) Formation of Xenobiotics. In Soil – A Synthesis, Critical Reviews in Environmental Science and Technology, 44:19, 2107-2171. [2] Eschenbach et al.. Sequential extraction procedures to characterize non-extractable residues (NER). 2013. Poster at SETAC 2013, Glasgow.

434

Elucidation of the nature of soil bound non extractable residues

M. Telscher, Bayer AG Division CropScience/Environmental Fate / Development Environmental Safety; F. Schmidt, C. Leake, Bayer Ag

Non extractable residues (NER) so called “bound residues” of plant protection products are formed in soil as a result of degradation processes. Due to their inherent nature, analysis and further assessments of bound residues are challenging. In a recent publication (Possberg et al. 2016) a distinct analysis of NER has been reported. The method relies on the determination of natural amino acids as the main part of biogenic residues in soil. The amino acids were liberated via a digestion of the soil with 6 N HCl at 110°C. Within this presentation we focus on the utility and validity of this analytical method using 14C bromoxynil and an agricultural soil from Germany. As a main result above 55% of bound residues could not be liberated and remained bound to the soil even after such a harsh digestion step. During further clean-up of amino acids further losses of radioactivity of approximately 40% of those liberated bound residues has been observed. Further analyses elucidated up to 50% of those unidentified losses, however, in total approximately 75% of bound residues stuck to the soil and therefore could not be identified or unambiguously assigned. However, 16% of the generated NER was extractable and could be assigned to amino acids. Both results indicate that bound residues will not contribute to future risks or hazards because they are on one hand not bioavailable or on the other hand they have been converted to natural biogenic residues like natural amino acids.

435

A tool to establish the role of Non-Extractable Residues (NER) in soil on toxicity

J. Harmsen, Wageningen Environmental Research / CALM; D. Hennecke,

Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Ecological chemistry; K. Hund-Rinke, Fraunhofer IME / Department of Ecotoxicology; J. Lahr, Wageningen Environmental Research; J. Deneer, Wageningen Environmental Research / ERA team
There is already a long discussion around the bioavailability and ecotoxicological relevance of Non Extractable Residues (NER) in soil. Is NER formation a detoxification process or should it be considered to be a hidden hazard? NER can only be established using labelled chemicals (e.g. ^{14}C) and cannot be measured with conventional chemical analytics. But even using labelled compounds uncertainty exists about the identity of measured radioactivity. Do we measure: 1) Strong adsorption or association of the parent chemical or breakdown product with mineral and/or organic matter, 2) Mineralisation and incorporation of carbon into microbial biomass and carbonates. Regulations ask for understandable and measurable parameters. The approach of Ortega-Calvo et al. (2015) has been followed, because this approach defines clear and measurable fractions. The only not measurable fraction is NER, but can be considered as a residual fraction if all others are measured. Considered are: Chemical present in the water phase, actual available, (Passive sampling or $CaCl_2$-extraction) A potentially available fraction in equilibrium with the water phase. (Tenax, ISO TS-16751); The total extractable amount, measured with a (standard) method NER is considered, but mentioned as non-measurable and also non-bioavailable. We studied three NER-forming chemicals and followed their fate in a period of 6 months after addition. An important part of the study were experiments using ^{14}C chemicals. In first experiments formation of Non-Extractable ^{14}C was observed for all chemicals. For the chemical Tri-NitroToluene (TNT), NER-formation was reproducible and NER formation during aging removed toxicity. By removing the bioavailable fractions directly after spiking and after aging it was also possible to remove toxicity. The experiments with and also without labelled TNT clearly showed that toxicity was caused by the bioavailable chemical and not by NER. The tool developed can be used if the fate of the chemical including NER formation is obvious With the other selected chemicals, Cypermethrin and Carbendazim, results were less clear, because there was a large uncertainty in NER-formation. The degree of biodegradation was not reproducible for Cypermethrin and unexpected losses occurred with Carbendazim. This gave a very large uncertainty about NER using non-labelled compounds. For these compounds it is not possible to draw conclusions from only a non-labelled experiments.

LCA and beyond - integrating sustainability and/or other dimensions to improve decision support (I)

436
How to make LCA fit for purpose as decision making tool
E. Mieras, PRe Sustainability; A. Gaasbeek, PRe Consultants / Consultancy; J. Coustillas, PRe Consultants
To understand how Life Cycle Thinking (LCT) and Life Cycle Assessment (LCA) can be improved to support decision making we first have to look at how decisions are made. One of the first distinctions that are made when it comes to decision making is between normative and descriptive decision making. The latter describes how people actually make decisions and that can be quite irrational, as also Daniel Kahnemann, the Nobel Prize Winner, argues in his book “Thinking Fast and Slow”. The first tries to define how to come to the best option, assuming that decision makers are fully rational and that the world can be modeled accurately. These are quite opposite ways of how decisions are made. The second distinction to be made is between what type of decisions are made, by whom and what the objectives are. It’s a big difference if you want to make a decision about different options for new products, determine the long term company strategy or select the most sustainable supplier. Even more so, the objectives will differ per organisation or even situation and thus the criteria will also be different. In other words: the context of the decision determines what support is needed and what’s the relevance of the outcomes. Results can have a different meaning in different context. Therefore, it’s important to assess first which methods are fit for purpose to support decisions in a specific context. To enable this, we want to introduce an intermediate step to determine whether LCA, LCT or any other assessment is best suited to answer the questions that are relevant in a specific decision making situation. For this goal we developed a tool to map between decision making objectives and LCA indicators. We will show examples of how this tool is applied for strategic goal setting as well as policy objectives. Furthermore, to support decisions it’s important that the insights are accessible for decision makers in a way that they understand, that speaks to their heart and mind and that is relevant for their job at hand. This requires that LCA is integrated in their daily (decision making) work. We will share examples of how a harmonized approach, benchmark models and simplified (but not simplistic) tools can play in instrumental role in bringing these insights closer to the decision making process. *Keywords: fit for purpose, decision making, LCA, Sustainability Assessment; Presentation preference: platform presentation*

437
Using Life Cycle Assessment (LCA) to Evaluate Global 6-Aminopenicillanic Acid (6-APA) Manufacture and Make Recommendations for Future Developments in the Biopharmaceutical
C. Chau, University Colleege London / Department of Biochemical Engineering; N. Titchener-Hooker, University College London / Department of Biochemical Engineering; P. Lettieri, University College London / Chemical Engineering
6-Aminopenicillanic acid (6-APA) is the beta-lactam nuclei of penicillin and is the intermediate to most semisynthetic antibiotics. Manufacturing of the nuclei represents one of the largest production scale processes within the biopharmaceutical industry. Whilst the environmental impacts associated with the industry are poorly understood, due to limited life cycle assessment (LCA) studies in the literature, the paper presents a LCA of 6-APA production to illustrate the burdens manufacture places on the environment as a function of manufacturing location. We make recommendations for future development of large scale biopharmaceutical manufacture by drawing on our 6-APA analysis where necessary. A typical manufacturing plant producing 2000 tonnes of 6–APA per annum has been modelled under USA operating conditions and a LCA hot-spot analysis was carried out. A process at this scale has a global warming potential (GWP) of 143,000 $tonnes_{CO2-Equiv}$/yr. which is largely caused by the high annual fossil fuel usage. The energy mix selected for the model is critical. Choosing a USA mix comprising mostly non-renewable resources provided the base case. Switching of the assumed energy mix to a Brazil mix (constituting a higher proportion of renewable resources), the contribution to climate change was reduced by 15%. Manufacture in China and India where coal combustion is the main source for electricity; the emissions were significantly higher (20%). Other location dependant variables were inputted into the model in conjunction with the switch of energy mix. Depending on the location’s water scarcity, the burden of 6-APA on this resource varies greatly. A 66% increase in water burden was seen when switching production from US to China. This is due to the higher use of hydroelectric power in the national energy mix and lower abundance of water in China. Production itself is water intensive due to high volume required for fermentation media and cleaning. Thus, overuse of local freshwater may compete with other sectors, e.g. agriculture, accessing this resource. This suggests that location of production is a critical factor in the planning of biopharmaceutical manufacturing.

438
A SUSTAINABILITY PERFORMANCE-BASED METHODOLOGY AND TOOL FOR ECODESIGN: the case of transport infrastructures
D. Lo Presti, The University of Nottingham / Nottingham Transportation Engineering Centre NTEC; J. Oliveira Dos Santos, IFSTTAR; S. Bressi, University of Palermo; S. Brodie, The University of Nottingham; J. Bryce, AMEC Foster Wheeler; V. Cerezo, IFSTTAR; T. Parry, The University of Nottingham; G. Di Mino, The University of Palermo
The importance of sustainability in transportation infrastructure has raised in response to the link between anthropogenic activity and global challenges, such as climate change, as well as in consequence of the ongoing development of models quantifying the social and economic impacts resulting from infrastructure development. Therefore, addressing the sustainability of transportation infrastructures requires exploring the environmental, social, and economic impacts of technological options while balancing the often-conflicting priorities of different stakeholders, at an early design phase of the infrastructure delivery process. That is a typical multi-criteria decision-making (MCDM) problem, in which the decision-makers need to measure the sustainability through a set of meaningful, representative and quantifiable criteria, balance the relative importance of those criteria and determine the sustainability sequence of multiple alternative technologies for fostering transportation sustainability. In order to help the decisions makers to efficiently address this challenging task, a decision support toll (DST) was developed in the scope of the training-through-research programme Sustainable Pavements & Railways Initial Training Network (www.superitn.eu). It consists of a computational platform that implements a conceptual framework developed to quantify sustainability. It comes with a set of sustainability indicators tailored to both road and railway systems as well as several objective and subjective weighting methods. Amongst those belonging to the last category, the DST includes a set of default weights derived from an Analytical Hierarchy Process (AHP)-based survey that engaged stakeholders from different sectors and from several European countries. At last, the Preference Ranking Organization Methodology of Enrichment Evaluation II (PROMETHEE-II) MCDM method is employed for prioritizing alternative road pavement and railway tracks solutions at the design stage. The DST will be freely available and can be used at professional level, by professionals interested in advancing sustainability in transportation, as well as for educational purposes, to provide knowledge and educate on the use sustainability concepts and on what are the important issues to consider during the sustainable transportation decision-making process.

439
Influence diagrams and scoping for Life Cycle and Sustainability Assessment, an example from sustainable mining
A. Ciroth, GreenDelta; C. Di Noi, GreenDelta GmbH; D. Bizarro, GreenDelta; H. Wessman-Jääskeläinen, VTT Technical Research Centre of Finland
Life Cycle Assessment is a technique typically intended to provide a holistic assessment of environmental and possibly also social impacts over the entire supply chain and life cycle. However, LCA has limitations, for a variety of reasons. In this situation, it is interesting to investigate, for a given issue, the ideal portfolio of tools

to be used, including LCA, but not necessarily limited to it. Moreover, in every LCA, it is in a first step important to specify goal and scope for the further analysis, and it is worthwhile to be aware of aspects which have an influence on the overall environmental impacts of an investigated product. So far, goal and scope in LCA is conducted typically without a diagram or visualization of relations between different aspects to be decided about in goal and scope. We introduce influence diagrams and advanced hot spot analysis as a means to both “tailor” the approaches to be applied for assessing the sustainability of a given situation, and also to shape goal and scope of an LCA, where LCA is part of said portfolio. As an application, we develop and present a causal loop diagram for sustainability assessment of mining in general, and apply this to specific mine sites in Finland, Portugal, and South Africa, where this approach is currently applied, led by GreenDelta, in the European H2020 research project ITERAMS. In the presentation, the developed causal loop diagram and the approach for obtaining the diagram for the case will be explained, with results from the ITERAMS project. Results are quite promising and we believe that using causal loop diagrams in sustainability and life cycle assessments helps to clarify selection of the (combination of) appropriate tools for the assessment, and help to structure the goal and scope setting in LCA.

440

Life Cycle Sustainability Assessment for Improved Space Mission Design

A.R. Wilson, M. Vasile, University of Strathclyde / Department of Mechanical & Aerospace Engineering; K. Baker, Glasgow Caledonian University / School of Engineering & Built Environment

The adoption of the Paris Agreement and Sustainable Development Goals in 2015 has been the driver for a more coordinated global approach towards achieving environmental sustainability. However, to be successful, this vision must run through every sector of society and the space industry is no exception. In the context of renewed global awareness on environmental sustainability, Life Cycle Assessment (LCA) is an important environmental management technique increasingly applied within the space industry to assess the environmental impacts of products over their entire life cycle. The European Space Agency (ESA) began work on this topic in 2009, employing an internal concurrent design study called ECOSAT to consider the life cycle impact of the design, manufacturing, launch and operations of a satellite. Since then ESA has continued to develop LCA methodology for the space sector, creating the first set of LCA guidelines for space systems in 2016 and now intend to integrate LCA into the concurrent design process. Whilst space-based LCA is still in its early stages, its further development relies on it being increasingly employed within the broader space sector to give parity across the industries. For this reason, moving towards space-based Life Cycle Sustainability Assessment (LCSA) is a logical next step which allows for the three pillars of sustainability (environment, society, economy) to be addressed within one assessment. Tailoring this integration for space systems will allow the industry to become more accountable and responsible for their operations by taking into account the full spectrum of life cycle sustainability issues associated with the operation of space systems. This paper will present the LCSA methodology used in an open-source platform under development at the University of Strathclyde, outlining the integration of social and economic aspects with environmental LCA to evaluate the life cycle impacts of space systems. As adverse impacts are more difficult to modify the later into the design process that they are identified, the integration of LCSA into the concurrent design process is essential for the early mitigation of sustainability issues. As such, the intention of this platform is to help decision-makers choose sustainable technologies and products at the design stage by determining those that are not only cost-efficient, eco-efficient or socially responsible, but also ones that can easily justify and evidence their sustainability.

441

How can Agent-based Modeling improve decision making in Life Cycle Assessment?

A. Micolier, University of Bordeaux / The Life Cycle Group CyVi; P. Loubet, University of Bordeaux / ISM CyVi; F. Taillandier, University of Bordeaux / I2M GCE; G. Sonnemann, Université Bordeaux / ISM CyVi

Life cycle assessment (LCA) is now acknowledged as the worthiest methodology to evaluate environmental performance of whole system in a holistic way. Thus, it has been tempting to extend LCA to support both design and implementation of sustainable complex system. Notwithstanding the high potential of using LCA as a decision-making tool, some limitations have been identified: (a)uncertainties due to social and economic context and (b)system complexity. Agent-based models (ABM) are computational models for complex systems simulation. ABM is a bottom-up approach in which agents interacting between them can be defined, driven by behavior established on simple rules. Coupling of ABM with LCA has a high potential to supplement LCA in some of its methodological weaknesses for better decision support. We carried out a literature review of papers combining LCA and ABM based on a set of criteria in order to understand to what extend can ABM enhance LCA at different stages. This review suggests that ABM has the capacity to (a) measure phenomena not only driven by economic or rational factors and (b) forecast emerging dynamics not analytically predictable. Therefore, coupling LCA and ABM is a promising approach to guide the design of products exhibiting dynamics mainly driven by human behavior and to support consequential analysis through its capacity to explore the effectiveness of different sustainable policy scenario. Coupling ABM&LCA can be done with different strategies (extension of LCA, hybrid analysis and complementary use) depending on the expected trade-off between consistency and flexibility. Hybrid analysis is adapted to most of situation since both methods can exchange data externally without impacting the other one. Extension of LCA with ABM leads to a consistent model in which LCA is embedded in ABM, which is particularly relevant for study requiring to take into account dynamic effects in the technosphere. Different degrees of coupling (hard-coupling, tight-coupling and soft-coupling) are defined according to (a)data flow direction and (b)coupling dynamic. Higher the degree of coupling, higher the computational time; therefore the use of hard-coupling should be limited to studies integrating feedback in adaptive decision-making process. This paper addresses some methodological guidelines on the way of creating a dynamic LCA on the inventory phase. Future research in this field should now address temporal dynamics in the life cycle impact assessment.

Environmental risk assessment and management of the spoil material produced in tunnelling excavation

442

Characterization and management of excavated soil and rock

G. Mininni, CNRIRSA; A. Sciotti, F. Martelli, Italferr SpA

This paper describes some case studies relevant to management of excavated soil and rock (ESR) produced when tunnelling is performed using Tunnel Boring Machine with Earth Pressure Balance system (EPBs-TBM). ESR can be alternatively qualified as waste or as by-products depending on utilization conditions (in particular, certainly of further use) and on their characteristics (especially in terms of environmental and health protection requirements). Tunnels construction entails production of large quantities of ESR, up to several millions of tons. Their management as a waste certainly requires a huge amount of financial resources that, in some cases, will dramatically impair the feasibility of civil work realization. In this paper the legislation framework of Italy, United Kingdom and France, regarding management of ESR is presented and compared, with attention to the characterization protocol used to distinguish waste from by-product. Moreover, some Italian and international case studies will be presented showing validated data, courtesy provided by important construction companies. Case studies will include information on management model of ESR, adopted treatments before final use (considered as normal industrial practice) and final destination. Different approaches clearly appear from this study: in the Northern part of Italy authorities have allowed to use the ESR as by-products while in the area of Rome management as waste prevails. Other cases such as Crossrail (the railway tunnel crossing London), Cityringen, (Copenhagen underground) and Le Grand Paris (Paris metro) will be discussed. The new circular economy package of the European Commission will push all the member states to move from linear to circular economy with consequent reduction of natural resource exploitation. ESR can be one of the important sector where the challenging objectives may be reached. Now in Europe does not exist a clear understanding of the legislative measures and technical rules needed to harmonize the ESR management. Protocols are needed for their characterization, including biotests, limit values of some contaminants should be fixed to consider ESR as by-products, the allowed treatments as normal industrial practice should be clearly stated. Current legislation may have a non-unique interpretation and therefore the operators are exposed to uncertainties. The general principles of fair competition inside Europe are totally disregarded.

443

REALIZZATION OF ROAD GALLERY: ADVANTAGES, CRITICALITY AND FUTURE PERSPECTIVES

A. Selleri, Autostrade per lItalia / direzione tecnica; S. Frisiani, Spea Engineering S.p.A.

For those who carry out public works at the service of the territory, which are the motorway routes, it is essential that in all phases, from approval to the realization of the work, the respect of the pre-established times in the project and the relevant regulations is guaranteed. In fact, time is a factor that directly affects the costs of execution and, above all, the costs borne by the community, which, in the event of bureaucratic red tape, cannnot benefit from the availability of a more efficient, safer and less impactful service. Even though the regulatory framework tends to introduce simplifications in procedures with the latest updates, there are rare cases where there are no burdens that often negate the positive effects of investments in technologies put in place to improve construction techniques. and increase the speed of completion. To the complexity inherent in the realization of linear works, which involve the excavation of tunnels and the consequent management of the excavated soil, is added the paradox that, at equal environmental conditions, the same lands can be considered by-products or waste, even if deriving from the same pile and if produced with the same excavation system within the same work. A case that represents this situation well is the mechanized excavation technique, whose adoption often involves an excess of provisions that substantially increase the size of the characterization areas inside the building sites, and the observation times, against a null environmental advantage.

444

Management of the spoil material produced by EPB-TBM: from experimental design to the excavation phase

S. Padulosi, A. Martino, D. Putzu, ITALFERR S.p.A

The use of foaming agents and additives is one of the fundamental factors allowing the correct use of the EPB-TBM (*Earth Pressure Balance-Tunnel Boring Machine*) for the excavation of underground works. On the other hand, their use must be carefully assessed in environmental terms, starting from the initial planning stages, in order to meet the requirements for the current legislation, but also because the subsequent use of spoil materials must not pose risks for the environment and human health. During the environmental design of the project, it is therefore essential the developing of a site-specific conditioning study for the management of excavation material. In fact, on the basis of the results obtained from the conditioning tests, it is possible to hypothesize a maximum dose of the required conditioning agents, which will constitute the reference for the environmental risk assessment in the context of a sustainable management of spoil materials. Anyway, the management of spoil materials produced from excavation by EPB-TBM is extremely complex and is based on numerous design assumptions, characterized by high levels of uncertainty: from the definition of the commercial products to be used and of the most suitable soil conditioning parameters, to the choice of analysis and tests to be performed (biodegradation and/or ecotoxicological), up to the methods of interpreting the results. The uncertainties connected to the experiments carried out during the design phase can be reflected in: differences in the operational protocols to be adopted during construction, unavailability of the approved project, consequent increase in time and costs to reiterate the site-specific experiments and the following approval process. The environmental legislation on this field, clarifies only partially how to control and manage the substances contained in foaming agents and additives used for the excavation process by EPB-TBM. To overcome the design problem related to the interpretation or absence of regulatory reference limit values for the main substances contained in the additives, site-specific experiments were carried out in some projects, aimed at evaluating the eco-compatibility of the conditioned soils through biodegradation and ecotoxicological studies of the foaming agents.

445

Environmental effect of chemicals injected into the soil in mechanized tunnelling applications

G. Vilardi, DICMA La Sapienza / Department of Chemical Engineering Materials Environment; D. Sebastiani, I. Bavasso, S. Miliziano, L. Di Palma, Università La Sapienza; F. Carriero, R. Sorge, Astaldi

In recent years the management of the soils and rocks resulting from the mechanized tunnel excavation process, raised increasing concern in Europe. The notable amount of chemically treated soil generated during tunnel excavation activities has lead companies, governments and research groups to face economical, technical, logistical and environmental issues. The most widely used tecnique in mechanized tunnelling, named Earth Pressure Balance (EPB), requires the continuous injection of chemicals during the advancement of the machines to enhance the excavation activity. Depending on the excavated materials features, various possibilities are technically feasible for the material re-utilization as aggregate for concrete, for filling in civil engineering, fills for road, railway embankments or breakwater harbour dam, stabilization of slopes affected by landslide phenomena or through projects of landscape management. The lack of information about the environmental impact of these chemicals, introduced during the tunnelling process, may lead to remarkable economical impact of the spoil management process, due to the necessity to store (see Figure 1), transport and dispose of the treated material. In the worst case, the uncontrolled chemical injection could lead to the production of several tons of hazardous waste, whose management might be significantly onerous in terms of cost and time. The University of Rome *Sapienza* and Astaldi started a joint research program with the aim of acquiring aknowledge, data and expertise in the use of chemicals currently used in the soil conditioning processes. This research program has led inevitably to deal with the environmental impact of different products. Preliminary experimental studies started to be performed, a large number of different product were considered, preliminar screenings on the chemical structure and properties of each compounds has been necessary and the physical and chemical features of pure products and their aqueous solutions were analyzed. Particularly referring to the biodegradation phenomena, the influence of key-parameters, such as additive composition, concentration and bacterial typology was studied. Using data collected so far it was possible to obtain a fairly clear picture of the properties of the chemicals used in tunnelling applications. Comparative evaluations among several chemicals or dosages have been possible basis on results obtained performing tests in standard environment.

446

Site-specific protocol to assess the environmental compatibility of spoil materials produced by EPB-TBM

A. Barra Caracciolo, National Research Council / Water Research Institute; P. Grenni, National Research Council of Italy (CNR) / Water Research Institute; E. Beccaloni, National Health Research Institute / Health and Environment Department; L. Patrolecco, Water Research Institute-National Research Council / Water Research Institute

The increasing use of Earth Pressure Balanced Shields (EPB-TBMs) in the tunnelling industry has been due to their advantages over conventional excavation methods, such as continuous operation, safer working conditions, reduced damage at surface level and higher tunnelling speed. The performance of EPB-TBMs relies on the use of appropriate soil conditioning foaming agents containing water solutions of surfactants, mainly sodium lauryl ether sulphate (SLES) and in smaller concentrations other additives. In accordance with the Italian legislation, spoil material from excavation processes can be re-used as by-products if the chemical thresholds for organic and inorganic contaminants (e.g. heavy metals, hydrocarbons C>12; Italian Decree 120/2017) are not exceeded. However, there are currently neither SLES soil threshold limits in European and Italian legislation (Annex 4 of the Italian Decree 120/2017), nor comprehensive studies on its possible ecotoxicological effects on soil and water organisms. The ecological approach here reported, consisting of site-specific studies together with ecotoxicological tests performed on the real excavated soils, aims to fill the gap between the lack of threshold limits in soil and water for these multicomponent commercial products and the environmental protection. The studies are planned following a preliminary evaluation of the foaming agent treatment ratios to be used for the specific lithological characteristics of the excavated soils. Here we report the main steps of the environmental studies useful for producing a "Protocol for the assessment of environmental compatibility of the spoil material during the tunnelling in the construction site". The aim of the protocol is to address engineering contractors and stakeholders (e.g. Railway and Motorway operators) on how to verify the environmental compatibility of excavated soil before putting it in the destination site. It is very important to highlight that the protocol (e.g. the ecotoxicological test selection) has taken into account the site-specific characteristics and the possible environmental exposure scenarios in order to protect ecosystems and human health.

447

Mineral-based soil conditioner for EPB TBMs: An environmentally friendly alternative

M. Greenhill-Hooper, Imerys / Performance Additives; H. Spengler, Imerys / Imerys Metalcasting; G. Collard, Imerys / Performance Additives; C. Egerton, Consultant / Tunnelling

A novel product, based on a natural mineral has been developed for use as a foaming and soil conditioning agent with earth pressure balance (EPB) tunnel boring machines (TBMs). It is available as readily water dispersible granules or pre-prepared aqueous suspension form and can be dispersed and diluted readily in water with low shear mixing. The resulting dilute suspension can be converted into a foam using an industrial foam generator, or by other mixing methods. It can be used with existing equipment found on EPB TBMs, without the need for further modifications and investment. The major component of this new product is a natural mineral widely distributed in the earth's crust. It is virtually insoluble in water and has no known ecotoxicity. Specifically, there is an absence of toxic effects on two aquatic organisms (*Danio rerio* and *Daphnia magna*) and a demonstrated low risk to arthropods, earthworms and soil bacteria. In a recent study commissioned with an environmental consultancy, it was considered that excavated soil conditioned with the product and deposited in a licenced waste facility would not pose a risk to the surrounding water environment. Compared with existing products it contains very low levels of synthetic chemicals, relying instead on the natural mineral component to stabilise foams by a completely different mechanism; one that is potentially less susceptible to the degrading influence of soils that can compete for the surfactants that are present in, and that stabilise conventional foams. Tests at an independent geotechnical laboratory have demonstrated the good stability of the foams produced using the product (half-life measurements of water drainage), and confirmed that mixtures of the foam with fine and coarse grained soils have the desired consistency and cone slump behaviour for EPB tunnelling. A series of specifically developed tests reveal that the addition of foam substantially reduces adhesion of the finer grained soils to metal surfaces. In practice this will translate to a substantial reduction in the clogging potential of excavated clay in the TBM cutter head and spoil conveyors, crucial for maintaining good advance rates. The new product will be attractive to those seeking to minimise the environmental impact of tunnelling projects.

Analysis and Fate of Emerging Contaminants in soils, water and plants under water scarcity (II)

448

Quantification of Carbon Nanotubes in Complex Matrices: Possibilities of Electron Microscopy

R. Kaegi, A. Gogos, Eawag Swiss Federal Institute of Aquatic Science and Technology

Fascinating properties of Carbon nanotubes (CNTs) allow the development of novel materials with increased functionalities (e.g. reduced weight, increased strength). Nevertheless, CNTs do pose potential environmental and human health risks and reliable methods to quantify CNTs at low concentration in complex matrices are still lacking. We therefore developed a method, based on the unique shape of the CNTs to quantify these materials in complex matrices. Multiwalled CNTs (IRMM 382) suspended in either ultrahigh quality (UHQ) water or in soil extracts were directly centrifuged on transmission electron microscopy (TEM) grids, resulting in an even distribution of the CNTs on the grids. Samples were investigated with a scanning electron microscope (SEM, Magellan XHR 400, FEI) operated at an acceleration voltage of 20 kV in emersion mode and using a bright field transmission electron detector. A ridge detection algorithm implemented in the image processing software Fiji was used to detect and characterize individual CNTs on recorded images. The concentration of the CNTs in the suspension was calculated based on the total length of all CNTs (provided by the ridge detection algorithm) detected on the images in combination with their thickness (20 nm), their density (1.4 gcm^{-3}) and the well-defined volume of suspension that was centrifuged on the TEM grids. CNTs were well separated on the TEM grids and an increasing number of CNTs was observed on images with increasing concentrations of CNTs in suspension. Plotting the calculated concentration of CNTs in suspension against the nominal CNT concentrations (10 μgL^{-1} – 100 μgL^{-1}) resulted in a linear relationship. The calculated and the nominal CNT concentrations were in good agreement at low CNT concentrations, but at high concentrations, the calculated concentrations underestimated the nominal values by a factor of ~2. Almost identical results were obtained from CNTs in UHQ water and in soil extracts (5 mgL^{-1}) indicating that the detection of the CNTs was not compromised by the presence of soil particles. Future experiments will focus on a selective removal of the soil particles by an additional treatment with diluted hydrofluoric acid. Initial experiments are promising and suggest that the detection limit of the methods can be lowered to 1 mg(CNT) / kg(soil), which would represent huge step forward in detecting of CNTs in complex matrices.

449

Monitoring for perfluorinated compounds, insecticides, and brominated flame retardants in the water of Daechung lake and Geum river basin

H. LEE, National Institute of Environmental Research (NIER / Geum River Water Environment Research Center; Y. Cho, J. Khan, National Institute of Environmental Research NIER / Geum river environment research center; B. Lee, National Institute of Environmental Research NIER / Han river water environment research center; B. Seol, M. Chae, S. Cheon, National Institute of Environmental Research NIER / Geum River Water Environment Research Center

A multiresidue analytical method using LC-MS/MS was developed for perfluorinated compounds (PFCs), insecticides, and brominated flame retardants (BFRs) in water samples with the simultaneous SPE method. The ranges of recoveries were 19.7 ~ 135.0 % (PFCs), 95.0 ~ 117.2 % (Insecticides), and 72.5 ~ 86.4% (BFRs), with coefficients of variation of less than 15%. Method detection limit (MDLs) of PFCs, insecticides, and BFRs were 0.3 ~ 7.1 ng/L, 3.0 ~ 3.7 ng/L, and 5.1 ~ 11.7 ng/L, respectively while limit of quantifications (LOQs) were 0.9 ~ 21.4 ng/L (PFCs), 9.0 ~ 11.0 ng/L (Insecticides), and 15.4 ~ 35.0 ng/L (BFRs). For understanding the background levels of PFCs, insecticides, and BFRs in the river water, those compounds were monitored in Geum river main stream, So-ok stream, Juwon stream, and Daechung Lake (Dam) every month (March to December) utilizing the developed method. The compounds of the highest detection frequency were PFOA, PFHxA, and dinotefuran (Insecticide), whereas BFRs were detected only in March and December, except for main stream. In conclusion, the trends were not observed on periodical and spatial characteristics and the background levels were secured for PFCs, insecticides, and BFRs in Geum river basin.

450

Impacts of Contaminants of Emerging Concern on Terrestrial Organisms

S. Dudley, UC Riverside / Environmental Toxicology; M. Pennington, UCR / Environmental Toxicology; C. Sun, UC Riverside / Environmental Science; J. Trumble, University of California, Riverside / Entomology; J. Gan, University of California, Riverside / Department of Environmental Sciences

Impacts of Contaminants of Emerging Concern on Terrestrial Organisms

Stacia Dudley[1], Marcus Pennington[1], Chenliang Sun[2], John Trumble[3], Jay Gan[2]
[1]Environmental Toxicology Graduate Program, University of California, Riverside, CA [2]Department of Environmental Sciences, University of California, Riverside, CA [3]Department of Entomology, University of California, Riverside, CA

Reclaimed water is a historically underutilized resource. However, with increased population growth and global climate change placing increased pressure on fresh water resources, reclaimed water is evolving into an economical and sustainable means to meet the needs of citizens, industries, and agriculture. The use of recycled water for agriculture comes with the potential risk of environmental and food contamination by contaminants of emerging concern (CECs). These compounds pose a potential threat to the health of ecosystems because they are designed to be biologically active at low concentrations and are considered "pseudo-persistent" due to their continuous release into the environment. Using high resolution mass spectrometry, ^{14}C tracing, enzyme extraction and Illumnia sequencing techniques we evaluated a wide range of biological effects in terrestrial organism caused by exposure to CECs. Organisms in these studies included, the cabbage looper (*Trichoplusia ni*), an earthworm (*Eisiena fetida*), a model plant (*Arabidopsis thaliana*) and cucumbers (*Cucumis sativus), radishes (Raphanus raphanistrum sativus)* and tomatoes *(Solanum lycopersicum*). These studies have revealed a multitude of effects including increased mortality and development time, reduction in weight, changes to the microbiome and up-regulation of enzymes associated with oxidative stress. Further, the study has highlighted the potential for higher plants to take up, translocate and detoxify CECs.

451

Occurrence of pharmaceuticals and their metabolites in Euthynnus alletteratus bile

J.M. Peña Herrera, Institute of Environmental Assessment and Water Research (IDAEA-CSIC) / Environmental Chemistry; N. Montemurro, IDAEA CSIC Barcelona / Dipartimento di scienze agro-ambientali e territoriali; E. Navarro, CSIC - Spanish National Research Council / Dept. Recursos marins renovables; J. Navarro, ICM-CSIC / Dept. Recursos marins renovables; M. Solé, ICM-CSIC; S. Perez, IDAEA CSIC / Environmental Chemistry; D. Barcelo, IIQAB-CSIC / Department of Environmental Chemistry

The primary source of pharmaceuticals in surface waters has been attributed to the effluents of WWTP among others. The widespread occurrence of pharmaceuticals in the aquatic environment has raised concerns about their potential adverse effects on exposed wildlife. Little is currently known on exposure levels of drugs in fish, but some studies reported the detection of pharmaceuticals and endocrine disrupting compounds in this type of biota. Due to possible accumulation processes, pharmaceuticals and metabolites could be thousand times more concentrated in fish than in polluted living waters. By other hand, fish are known to possess a hepatic detoxification system which are likely capable of metabolizing pharmaceuticals taken up from polluted waters. Some studies proposed the analysis of bile from fish to evaluate pharmaceuticals exposure including the identification of metabolites by UPLC-HRMS. In this context, we propose the evaluation of the metabolism of frequently detected drugs in fish, performing a rapid screening of bile by HR-MS for the presence of stable intermediates. Fish were collected from different regions in the Mediterranean coast of Spain. Afterwards, their bile was isolated from the fish and analyzed for the detection of parent drugs and some metabolites listed in an in-house suspected list. A sampling campaign was planned to collect tuna fish, *Euthynnus alletteratus,* from the Mediterranean coast of Spain: Tarragona, Cartagena and Ceuta catched by spearfishing between 2015-2017. Their bile samples were analyzed directly by UPLC-HRMS after a protein precipitation. The HRMS data allowed screening for suspected pharmaceuticals and their metabolites and provided plausible chemical formulae. The comparison of MS/MS spectra of the parent compounds and their metabolites allowed to propose chemical structures for possible metabolites in fish bile. With this analytical methodology some metabolites, corresponding to different reactions that includes products of hidroxylation, glucuronide conjugates were identified. The suspect analysis of bile samples allowed the detection of several pharmaceuticals. Psycho-active drugs were one of the most commonly detected drugs. Their identities were proposed by matching their accurate MS and MS/MS data against different libraries. Finally, authentic standards were employed to confirm the proposed drug identities and to determine analyte concentrations in the fish samples.

452

Accumulation and fate of 12 human drugs through the soil-root-leaf system

N. Montemurro, IDAEA CSIC Barcelona / Dipartimento di scienze agro-ambientali e territoriali; C. Postigo, IDAEA, CID-CSIC / Environmental Chemistry; S. Perez, IDAEA CSIC / Environmental Chemistry; D. Barcelo, IIQAB-CSIC / Department of Environmental Chemistry

Crop irrigation with reclaimed water has become an extended practice in many countries worldwide where the water scarcity and excessive exploitation of agriculture are forcing local authorities to look for alternative resources. Despite this practice increases local water resources and contributes to nutrient recycling, using reclaimed water for irrigation, however, represents a primary source of emerging organic contaminants resilient to wastewater treatment processes, such as some pharmaceuticals and personal care products [1]. These pollutants can be retained in the soil, directly uptaken by crops or translocated from soil to plant tissues above the ground [2,3]. The present work aimed to evaluate the transfer and the bioaccumulation of organic contaminants of emerging concern (mainly pharmaceuticals) in lettuce tissues and soil. The distribution of twelve relevant wastewater-derived pollutants was evaluated in lettuce tissues (leaves and root system) and soil. This list included nine prescription drugs (diclofenac, trimethoprim, carbamazepine, oxcarbazepine, lamotrigine, cis-diltiazem, valsartan, midazolam, and methadone), an illegal drug (cocaine) and two transformation products (acridone and valsartan acid). Lettuce plants were grown in pots in a controlled environment and irrigated with artificial spiked water containing the 12 compounds during the entire growing period (60 days). Control was irrigated with tap water. Afterwards, a set of new lettuce plants were grown in the same soil pots and irrigated with rainwater or with tap water, if necessary. At the end of each experiment, leaves, roots and soil samples have been collected for each plot. All pharmaceutically active compounds were extracted by ultrasonic liquid extraction (USE) [4]. The detection of the target analytes was performed using liquid chromatography coupled to tandem mass spectrometry (LC-MS/MS). The results from the first growing season evidenced the presence of all analytes in all investigated matrices. Carbamazepine was the analyte that accumulated the most in lettuce plants (leaves and root system), whereas cis-diltiazem, methadone, and midazolam were preferentially accumulated in the plant root system and the soil. Concentrations of the target analytes in the plant-root-soil system after the second growing season were significantly lower than those measured after the first growing season, but still detectable for most of the compounds.

453

Root-uptake and dissipation of atenolol, sulfamethoxazole and carbamazepine applied as a single compound solution or in mixture of all compounds in three soils and five plants

R. Kodesova, A. Klement, Czech University of Life Sciences Prague / Dept of Soil Science and Soil Protection; O. Golovko, University of South Bohemia in Ceske Budejovice / South Bohemian Research Center of Aquaculture and Biodiversity of Hydrocenoses; M. Fer, Czech University of Life Sciences Prague / Dept of Soil Science and Soil Protection; O. Koba, University of South Bohemia in Ceske Budejovice / South Bohemian Research Center of Aquaculture and Biodiversity of Hydrocenoses; M. Kocarek, A. Nikodem, Czech University of Life Sciences Prague / Dept of Soil Science and Soil Protection; R. Grabic, University of South Bohemia in Ceske Budejovice / South Bohemian Research Center of Aquaculture and Biodiversity of Hydrocenoses

This study was focused on a root uptake of carbamazepine, atenolol and sulfamethoxazole from 3 soils: Haplic Chernozem, Haplic Cambisol and Arenosol Epieutric. Five plants (radish, arugula, lettuce, spinach and green peas) planted in those soil were initially irrigated by fresh water and next with water contaminated by a single compound or their mixture. After 3 or 4 weeks, each plant was divided into separate parts: roots (including bulbs of radish), leaves, stems (green peas) and pods (green peas). Plant parts and soils were freeze-dried and dry-masses and concentrations of pharmaceuticals and their metabolites were measured. Despite that atenolol and sulfamethoxazole relatively rapidly dissipate from soils, they and metabolites of atenolol were detected in all plants. Carbamazepine is very stable in soils and fractions of its metabolites are usually low. However, very high concentrations in all plants were measured not only for carbamazepine but also for its metabolites. The degree of compounds' transformation depended on a plant family. Considerably higher concentrations of atenolol, sulfamethoxazole and metabolites of atenolol were measured in roots in comparison to those in leaves and soils. In the case of carbamazepine, the highest concentrations were measured in leaves followed by roots and soils. Both indicate a high potential of plants to accumulate studied pharmaceuticals in their bodies and a high ability to transform studied compounds. Particularly in the case of carbamazepine, the considerably higher concentrations of metabolites were measured in leaves in comparison to concentrations in roots and very low or negligible concentrations in soils. Transformation of compounds in plant bodies is attributed to enzymes CYP450. Larger concentrations of carbamazepine metabolites were measured in leaves of lettuce, spinach and green peas than in leaves of radish and arugula (Order – Brassicales, Family – Brassicaceae). Oxcarbazepine was detected only in plants (not in soils). The impact of soil type on compound's uptake was not proven for all tested plants. The impact of application (single compound versus compounds' mixture) differed for different plants. Antibiotic sulfamethoxazole likely reduced dissipation of other two compounds in soils, which increased relative concentrations of compounds in plants (i.e., concentrations of compound in plant divided by compound loads in soils that is a total amount of applied solute divided by a dry mass of soil).

Prioritisation and Intelligent Testing of Pharmaceuticals in the Environment (II)

454

Inter-individual variation in the bioavailability and effects of NSAIDS in fish

A. Brown, Exeter University / Biosciences; L. Gunnarsson, University of Exeter / Biosciences; A. Lange; D. Rowe, The University of Exeter; M. Trznadel, University of Exeter / Biosciences; M. Linder-Nording, S. Gouveia, University of Umea; J. Wu, Umea University; C. Tyler, University of Exeter / Biosciences College of Life and Environmental Sciences

A basic tenet in the environmental risk assessment of pharmaceuticals is that pharmacological effects will occur in advance of any adverse effects, if their molecular targets are conserved in wildlife and if circulating blood plasma concentrations approach therapeutic concentrations established in humans. Fish generally display high levels of conservation of human drug targets and may be exposed continuously to pharmaceuticals via discharges from wastewater treatment plants. The Non-Steroidal Anti-inflammatory Drugs (NSAIDs) ibuprofen and diclofenac are present in effluents, resulting in low mg/L concentrations in surface waters and fish blood plasma below or bordering on "therapeutic" concentrations. However, some studies suggest that diclofenac and ibuprofen can induce harmful effects in fish at measured environmental concentrations. Here we seek to refine the environmental risk of NSAIDs by gaining greater understanding of their bioavailability, pharmacologically effective concentrations and inter-individual variations in fish. We quantified plasma prostaglandin (PG) and plasma NSAID concentrations in individual female rainbow trout during and after 12 days continuous flow-through exposures to ibuprofen (0, 10, 200 mg/L) or diclofenac (0, 5, 100 mg/L). High-level NSAID exposures significantly reduced plasma PGE2 concentrations, but pharmacological effects were not detectable for low-level exposures, due in part to considerable inter-individual variation in plasma PGE2: 2.6-143 pM for ibuprofen; 0.8-188 pM for diclofenac; versus 0.8-316 pM in control fish. There was no significant correlation between plasma PG and plasma NSAID concentrations within exposure treatments; plasma NSAID concentrations exhibited much lower inter-individual variation, with blood plasma: water partition coefficients ranging from 1-3 for ibuprofen and 1-9 for diclofenac. To identify factors affecting PG levels in individual fish we measured plasma lipid content and plasma protein binding influencing partitioning and bioavailability, haematocrit and plasma C-reactive protein concentration quantifying baseline immune system status, and plasma cortisol concentrations as a measure of stress in fish, potentially affecting plasma NSAID and PG concentrations. From our analyses, no single factor could explain the observed variations in NSAID uptake and pharmacological response. Our data highlight some of the complexities in interpreting biological exposure and effects data for NSAIDs.

455

Environmental effect assessment of human pharmaceuticals - the regulatory way forward

J. Bachmann, German Environment Agency (UBA) / Section IV2.2 Environmental Risk Assessment of Pharmaceuticals; S. Schwarz, German Environment Agency UBA / Section IV 2.2 Pharmaceuticals; U. Brandt, German Environment Agency UBA / Section IV Environmental Risk Assessment of Pharmaceuticals; I. Rönnefahrt, German Environment Agency - UBA / Section IV 2.2 Pharmaceuticals

Human pharmaceuticals are extensively studied and assessed before marketing approval. The EMA guideline for environmental risk assessment of human pharmaceuticals (EMEA/CHMP/SWP/4447/00 corr 2) was adopted in 2006 and is currently under revision. Input has been provided by several stakeholders from academia, industry and government. In this context, the UBA experiences with effect based assessment of human pharmaceuticals will be evaluated and presented. The basis for the evaluation are double quality checked effect data of algae, aquatic invertebrates and fish provided within several European authorization procedures. Because of owner protection, most of the data sets can only be presented in an anonymous and encoded form. One discussion point will be the question whether the current base of data is sufficient to draw general conclusions. Although the results are based on more than 10 years of experience with environmental risk assessment within the authorization of new human medicinal products, the data basis is still lower than desired. So for some pharmaceutical ingredients detected in surface waters environmental effect data are lacking, because they entered the market before implementation of the EMA guideline. Furthermore, the tailored assessment approach for substances with very specific mode of action will be addressed, especially regarding the remaining uncertainties for protection of biodiversity and the environment. This does apply e.g. for endocrine active substances, like contraceptives or anti-cancer drugs. Furthermore, the evaluated data allow discussion about effect sensitivity of several taxonomic groups, as well as about the PEC action limit (10 ng/L) for effect data justification. The quotient between the effect values of most sensitive and most insensitive species in more than 20% of the evaluated cases is greater than 100. Fish are the most sensitive organism group in more than half of the cases. In this context, the proposed approach to replace long-term by acute date coupled with changed assessment factors as applied usually for chemicals without any specific mode of action will be analyzed.

456

Prioritising human health risk of environmental residues of pharmaceuticals and personal care products in use in southern Nigeria

U. Agusiegbe, Environment Department, University of York / Environment; C. Eze, UNIVERSITY OF NIGERIA NSUKKA NIGERIA; A. Boxall, University of York / Environment Department

Pharmaceutical and personal care products (PPCPs) are used worldwide for medical treatment and personal hygiene. PPCP residues are usually discharged into the environment during wastewater treatment. The environmental fate of these chemicals is poorly understood, leading to concerns about potential health effects and ecological impact posed by their use and release. In many developing countries, information regarding the consumption patterns of PPCPs is often limited, even though consumer product usage data are crucial for more realistic exposure estimates that are needed for risk assessment. To address this need, the following study was performed to estimate PPCP use in Nigerian households to prioritise the potential for PPCPs to enter source water. Using questionnaires as the survey instrument, we elicited information from 350 participants, concerning the most frequently used PPCPs, duration and amount of use in households. Drug usage was limited to over-the counter(OTC) medicines and was estimated by application of the World Health Organization defined daily dose methodology. Annual consumption of personal care products (PCPs) was calculated by multiplying the quantity of products used by the frequency of use. To prioritise PPCPs, a risk index was developed to rank chemicals according to their potential to enter source water. Consumption of PPCPs varied considerably. Analgesics were the most consumed OTC medicines and highest use was observed for paracetamol. Household cleaning products were the most consumed PCPs and highest use was observed for detergent powder and dishwashing liquids. Overall, 12 PPCPs were identified as having the greatest potential to reach source water and pose adverse effects to human health. These include 8 active pharmaceutical ingredients (acetaminophen, tetracycline,

ciprofloxacin, ampicillin, cloxacillin, sulfamethoxazole, trimethoprim and pseudoephedrine) and 4 PCP ingredients (sodium lauryl ether sulphate, alcohol ethoxylates, ammonium thioglycolate and dichlorvos). This is the first attempt to prioritize PPCPs in Nigeria and it provides a useful priority set of chemicals for source water monitoring in the region. Future work will focus on evaluating the results of the prioritisation approach against real world monitoring data for Nigeria.

457

Aquatic toxicity related to pharmacological or secondary targets of human pharmaceuticals

A. Coors, ECT Oekotoxicologie GmbH; A. Falkenhain, C. Brüggemann, ECT Oekotoxikologie GmbH; M. Scheurer, DVGW Water Technology Center / Analysis and Water Quality

Human pharmaceuticals target specific biological structures to exhibit their intended therapeutic effect. The presence of the anticipated biological target of a pharmaceutical in a non-target species may lead to specific effects in that organism, while in the absence of the target non-specific baseline toxicity such as narcosis would prevail. Yet, pharmaceuticals often do not only interact with the anticipated pharmacological target in patients, but can also interact with secondary targets. Hence, specific toxicity could occur in non-target species also in the absence of a conserved pharmacological target simply because the secondary target is conserved in that species. The present study explored this hypothesis testing anti-histamines as model substances in *Daphnia magna* and the green algae *Raphidocelis subcapitata*. *Acknowledgement* - The research leading to these results has received support from the Innovative Medicines Initiative Joint Undertaking under iPiE grant agreement n° 115735, resources of which are composed of financial contribution from the European Union's Seventh Framework Programme (FP7/2007-2013) and EFPIA companies' in kind contribution.

458

Neurotoxicity testing approach to investigate venlafaxine and oxazepam modulation of transcriptomics and behavioral profiles in zebrafish embryos and larvae

C. Di Paolo, RWTH Aachen University / Ecosystem Analysis (ESA); M. Gundlach, RWTH Aachen University / Department of Ecosystem Analysis ESA; A. Haigis, Institute for Environmental Research, RWTH Aachen / Department of Ecosystem Analysis ESA; R. Hamann, Fraunhofer IME; S. Wuester, Institute for Environmental Research RWTH Aachen; M. Krauss, Helmholtz centre for environmental research - UFZ / Effect-Directed Analysis; M. Fenske, Fraunhofer Gesellschaft / Translational Medicine and Pharmacology; I. Werner, Ecotox Centre Eawag-EPFL / Department of Anatomy Physiology and Cell Biology; H. Hollert, RWTH Aachen University / Institute for Environmental Research

Neuroactive pharmaceuticals are of growing concern as aquatic contaminants due to environmental and human health risks. Even low concentrations can interfere with molecular pathways and population-relevant behaviors. At the same time there is no EU regulatory framework for environmental neurotoxicity assessment. This project aimed to contribute for establishing a neurotoxicity testing approach by investigating molecular (transcriptome) events involved with behavioural alterations in zebrafish embryos and larvae exposed to neuroactive pharmaceuticals. *Danio rerio* up to 5 days post fertilization (dpf) were statically exposed to venlafaxine (serotonin norepinephrine reuptake inhibitor antidepressant) or oxazepam (benzodiazepine derivative anxiolytic) at the µg/L range (1 nM to 10 or 100 µM). Solution concentrations were measured at the start and end of exposures by LC-HRMS. Assessed behavioral endpoints were embryonic spontaneous movement (1 dpf), touch-evoked escape response (3 dpf), and phototaxis and thigmotaxis reactions (5 dpf). RNA was extracted from pooled embryos or larvae (n=20-50) and submitted either to RNA sequencing with Illumina Next Generation Sequencing System (RNAseq) or Sybr Green based quantitative real-time PCR (qPCR). qPCR target genes were selected with basis on RNAseq results, but also a few targets proposed as markers of exposure or modulation by neuroactive compounds were selected from literature studies (e.g. *fkbp5*, *cfos*, *per3*). Reference genes were *ef-1α*, *rpl13*, *rpl8*. Chemical analysis indicated that solution concentrations were stable along exposure periods and in general accordance with nominal values. Oxazepam caused behavioral alterations mainly at 1 and 3 dpf stages, while venlafaxine affected prevalently larval behavioral endpoints. RNAseq of embryos exposed to 100 nM oxazepam indicated gene ontology enrichment for notochord morphogenesis. Larvae exposed to 1 nM venlafaxine presented differential modulation of response to abiotic stimulus, while 100 nM venlafaxine affected mainly muscle processes and to a minor extent circadian rhythm modulation. Confirmatory qPCR is being conduted. Zebrafish embryo-larval assays supported the elucidation of molecular mechanisms at the transcriptome level that occured concurrently with organism-level behavioral effects. Our results are expected to contribute in the future for AOP annotation and for the setup of a regulatory assessment approach to evaluate neurotoxic environmental contaminants.

459

Virtual fish tales: Liver, Intestinal and Gill Organoids as an in vitro alternative to live fish for prioritising pharmaceuticals and other compounds of highest concern in the environment.

S. Owen, AstraZeneca / Safety Health Environment; L.M. Langan, Plymouth University / Biological and Marine Sciences; R.J. Maunder, Plymouth University / Biological Sciences; M. Baron, Plymouth University; A.N. Jha, Plymouth University / Biological Sciences

Pharmaceuticals enter the aquatic environment largely through patient use, and result in a pseudo- persistent background in the aquatic environment. The risk of these compounds is assessed for new products registered since 2006, but there is a legacy of essential medicines for which we need to understand more. Since most of the thousand or so pharmaceuticals used by patients are likely to pose little environmental risk, it is important to identify those of most concern in order to prioritise effort and resources; it is vital to be able to predict internal concentrations in aquatic organisms. One method to assess uptake potential is to expose the animals of most concern. However, there are potentially thousands of compounds to be prioritised. Factoring that we already know the uptake rate can be influenced by the concentration of the compound in the water, there are enormous ethical implications for conducting this work with live animals (fish), and significant cost of resources to practically conduct the work. Alternatives are clearly required. Significant efforts to better predict environmental exposure are underway as part of the iPIE project (IMI grant no.115735—iPiE). Those *in silico* methods provide a first tier of screening, but we are likely still faced with hundreds of compounds to assess at multiple concentrations. We have been developing *in vitro* tissue micro-organs (organoids) that replicate the *in vivo* tissue. These can be used to build a virtual fish that will allow the screening of pharmaceuticals (or other compounds of concern or even metals) without testing live fish (BBSRC/NERC grant BB/L01016X/1). By building fish tissue cultures that better represent the complexity of the *in vivo* situation, we are able to offer *in vitro* models that can simulate live fish. Water exposure to the gill model can now be tolerated (without compromise) for several weeks, intestinal models are similarly robust. Since both methods employ a permeable barrier culture, rates of flux can be measured that provide not only simple information such as a compound may be taken up, but also rates of uptake and excretion. These data can be used to build kinetic models. The liver spheroids provide a metabolic tissue that when used in co-culture with the gill or gut provide a simple virtual fish alternative to live fish. These methods offer a critical step between predicting compounds of highest concern and prioritising which require further testing.

Emergence and multidimensional interactions of engineered nanoparticles in toxicology

460

Effects of fullerene C60 increasing concentrations in Mytilus galloprovincialis: role of mTOR in cellular/tissue alterations

S. Sforzini, Universita Del Piemonte Orientale Amadeo Avogadro / Department of Sciences and Technological Innovation (DiSIT); C. Oliveri, University of Piemonte Orientale / Department of Sciences and Technological Innovation DiSIT; A. Barranger, University of Plymouth / School of Biological Sciences; J.W. Readman, University of Plymouth / Biogeochemistry Research Centre; Y. Aminot, University of Plymouth; A.N. Jha, Plymouth University / Biological Sciences; M. Banni, Laboratory of Biochemical and Environmental Toxicology; A. Viarengo, IRCCS Istituto di Ricerche Farmacologiche Mario Negri / Laboratory of Environmental Chemistry and Toxicology

Little is known about the effects at cellular, tissue and individual levels of emergent contaminants such as fullerene C60; in particular, the mechanisms of action are poorly investigated. In this research, the effects of C60 on mTOR (mechanistic Target of Rapamycin) activity in mussel digestive gland were studied. mTOR is an evolutionarily-conserved serine/threonine protein kinase that senses and integrates a variety of cellular physiological and environmental signals to regulate cell growth. mTOR is found in two functionally distinct complexes, mTORC1 and mTORC2. In particular, the phosphorylated active form of mTORC1 mediates temporal control of cell growth by activating anabolic processes (such as transcription, ribosome biogenesis, protein synthesis), and by inhibiting catabolic processes (such as autophagy); mTORC2 is primarily involved in actin cytoskeleton reorganization. Mussels were exposed to C60 (0.01, 0.1 and 1 mg/L) for 72h. Tissue C60 accumulation was evaluated by immunofluorescence using a specific antibody as well as by chemical analysis. Immunohistochemical analysis revealed the presence and cellular distribution of C60 in mussel tissues, already at the lowest concentration. Our data demonstrated that the changes of the phosphorylation of mTORC1 and mTORC2 may explain most of C60 effects studied at cellular and tissue level. Indeed, the C60 induced dephosphorylation of mTORC1 contributed to increase autophagy and to decrease protein synthesis as highlighted by the reduction of lysosomal membrane stability and the enhancement of lysosomal/cytoplasmic volume ratio of the digestive gland cells; and mTORC2 to affect cytoskeleton organisation as revealed by the changes of actin/tubulin structures. Transcriptomic data are important to understand the cellular adaptive responses to the chemical. For this purpose, a novel low density oligo microarray (470 genes, suitable to follow 15 stress response pathways) was used. Transcriptomic analysis identified a number of DEGs showing a bell-shape trend with a maximum in animals exposed to 0.1 mg/L C60. In terms of processes related to the DEGs depicted in all conditions, the most affected are associated to translation, cytoskeleton organization and mitochondrial activity. Transcription of selected genes was verified by RT-qPCR. These represent the first data on C60 tissue subcellular distribution and on the possible involvement of mTOR in the physiological alterations due to nanoparticle accumulation.

461

Proteomic responses to nanoparticulate and ionic silver in freshwater microbes with different background

D. Barros, Universidade do Minho / Centre of Molecular and Environmental Biology, Department of Biology; A. Pradhan, University of Minho / Department of Biology; P.M. Santos, C. Pascoal, F. Cassio, University of Minho / Centre of Molecular and Environmental Biology CBMA Department of Biology

Enhanced use of silver nanoparticles (AgNPs) has inevitably resulted in their release into freshwaters raising concern about the risk to non-target biota and related ecological functions. Functional proteomics is an emerging technology that provides high-throughput analyses augmenting measurements of direct and highly sensitive responses at the cellular and sub-cellular levels. The impacts of AgNPs and ionic Ag at EC_{20} (effective concentration) were assessed based on the variations in the overall proteome in i) two aquatic fungal strains of *Articulospora tetracladia,* one isolated from a non-polluted stream (At72) and the other from a metal-polluted stream (At61), and ii) the bacterial strain *Pseudomonas* sp. M1 (PsM1) isolated from a metal-polluted stream. At72 was the most sensitive to AgNPs, whereas PsM1 was the most tolerant one. Characterization of AgNPs showed increased particle stability and lesser agglomeration with time in At72 while for At61 and PsM1 there was an increase in AgNPs agglomeration explaining its lower impacts on their growth. In fungi, ≈40% of the total quantified proteins were significantly altered after exposure to AgNPs and/or Ag^+ whereas for PsM1 this percentage was lower (≈20%). At72 and At61 shared only 20% of the proteins suggesting that the biological pathways involved in Ag^+ and AgNPs exposure were different. At61 had ≈25% more proteins induced by both Ag forms (compared to At72), suggesting higher response which is consistent with the background of this fungal strain. In PsM1, 32% of the proteins increased under exposure to AgNPs whereas the percentage for Ag^+ was higher (68%) indicating different responses to Ag^+ and/or AgNPs. In At72, Ag^+ increased the content of proteins involved in protein homeostasis while AgNPs increased the content of proteins related to DNA repair, the transport of substances and energy production. In At61, AgNPs increased the content of proteins involved in protein synthesis and energy production while both forms of Ag increased the content of proteins related to cell-redox and protein homeostasis, biomass and spores production and also to nucleic acids metabolism. Both Ag forms induced stress-responsive proteins which was consistent with the responses of enzymes involved in oxidative stress. Overall, functional proteomics can be useful to get a mechanistic insight on the stress induced by AgNPs and/or Ag^+ in microbes that play key roles in freshwater ecossystems.

462

Hazard assessment of seven different commercial silica nanoparticles on a battery of test species: bacteria, algae and fish cell lines

F. Book, University of Gothenburg / Department of Biological and Environmental Sciences; J. Sturve, Goteborg University / Departmen of Biological and Environmental sciences; T. Backhaus, University of Gothenburg / Department of Biological and Environmental Sciences

Manufactured silica nanomaterials are widely used in numerous applications in society such as paints, coatings, cosmetics, textiles and food and its release into the environment is inevitable. This has raised a global concern regarding the risk of silica nanomaterials and was in 2010 selected as one of the priority substances by the OECD Working Party on Manufactured Nanomaterials. In order to thoroughly examine the toxicity of silica nanomaterials to destruents, primary producers and fish, a panel of seven well characterized (with different size, coating and charge), biocide free, silica nanomaterials, were tested on bacteria, algae and fish cell lines. Based on the result, the current study also examined the selection of an appropriate exposure metric comparing mass (mg/L), number of particles (No/L) and surface area (m^2/L) against the observed toxicity. The results show that gill cell lines were the most sensitive test model with the lowest reported EC20 value of 5.1mg/L after exposure to the smallest particle at a concentration range of 12.5-100mg/L. Toxicity to fish cells was determined to be surface dependant, except for particles coated with ethoxy silane, which did not show any toxicity. For bacteria and algae, the cell wall seems to play a major role in the uptake and toxicity of silica nanoparticles. Keywords: hazard assessment, silica nanoparticles.

463

Toxicity Assessment of Engineered Titanium Dioxide Nanoparticles

S. Bitragunta, Birla Institute of Technology & Science Pilani, Hyderabad Campus / Biological Sciences; S. Palani, Birla Institute of Technology & Science, Hyderabad Campus / Biological Sciences

Titanium dioxide engineered nanoparticles (TiO_2-ENP) are extensively employed in manufacturing of cosmeceuticals, pharmaceuticals and health care products. As a result, TiO_2-ENP can reach the ultimate sink such as soil in the environment during their life cycle. In this context, investigations to understand environmental implications of nanoparticles including TiO_2-ENPs are gaining prominence across the globe. In the backdrop of assessment toxicity of rutile TiO_2-ENP (r-TiO_2-ENP) in soil sentinels, present study is aimed at evaluating their toxicity as per OECD-207 guidelines on earthworm, *Eisenia fetida*. Physicochemical characterization of r-TiO_2-ENP using dynamic light scattering revealed their tendency to form agglomerates (330-480 d.nm) in water. Soil exposure of earthworms to r-TiO_2-ENP (0.1, 0.15, 0.2 and 0.25 mg/kg) showed no mortality after 48 h. Increased specific activities of antioxidant enzymes including catalase, superoxide dismutase and glutathione reductase as well as lipid peroxidation indicate the potential of r-TiO_2-ENP to induce oxidative stress in the sentinel organism. Interpretations of the study can serve as cues to design a comprehensive approach for developing invertebrate based biomarkers and indicators as early warnings for assessing environment and health impacts of engineered nanoparticles.

464

Combination effects of chlorpyriphos and ZnO on oxidative stress and reproduction of the earthworm Dendrobaena veneta

D. Hackenberger, Department f Biology, University of Osijek / Department of Biology; . Lonńarî, University of Osijek / Department of Biology; D. MarkoviDŽ, University of Rijeka / Department of Biotechnology; J. ObhoDŽax, Rudjer Boskovic Institute; B. Hackenberger, Department f Biology, University of Osijek / Department of Biology

When assessing the toxicity of pesticides and other chemicals to non-target organisms the most common experimental set-up is an exposure to a single compound. However, it is more likely for organisms under the environmental conditions to be exposed to a combination of chemicals that have different modes of action and potentially can interact with each other. The aim of this research was to investigate the effects of a mixture of ZnO and chlorpyriphos (CHP) on biochemical biomarkers and reproductive success of the earthworm *Dendrobaena veneta* with possible differences in effects of ZnO in nano and bulk form, and the difference of effects in various soils. EC50 values for reproduction of *D. veneta* after exposure to CHP and ZnO were calculated and used in the binary toxicity experiment. Concentrations were as follows, with EC50 being 100%: 100% CHP, 75% CHP/25% ZnO, 50% CHP/50% ZnO, 25% CHP/ 75% ZnO, 100% ZnO. ZnO was separately tested as bulk and nano sized particles and CHP was tested as a commercial preparations. The entire experiment was conducted according to the OECD earthworm reproduction test. At the end of the experiment the number of juveniles and activities of AChE, CAT, GST, TBARS, MT and PC were measured. The concentrations of pesticides in soil samples were measured with UPLC and the concentrations of Zn was measured with the energy dispersive x-ray fluorescence technique (EDXRF). CHP had a clear effect on AChE activity with almost 50% inhibition after 28 days of exposure in artificial soil. CAT activity did not change on any concentration, for both nZnO and bZnO, while GST activity decreased. The number of juveniles was significantly reduced with all mixture ratios. In natural soil CAT activity was also unaffected with bZn, and slightly inducted with nZnO/CHP combination. The inhibition of AChE was present at all applied combinations. In natural soil the number of juveniles was reduced with nZnO, while it remained the same as control after bZnO exposure. The effects of combined exposure of CHP and ZnO varied depending on the form of ZnO applied as well as on the characteristics of the soil in which the earthworms were exposed. More biomarkers should be employed to elucidate which low-level biomarker can be linked with the effects on the higher (reproductive) level.

465

Poster spotlight: WE305, WE323, WE324

Improving the environmental risk assessment of the aquaculture 'Blue Revolution'

466

Tools for Assessment and Planning of Aquaculture Sustainability (TAPAS)

P. van den Brink, Alterra and Wageningen University; A. Lillicrap, NIVA Norwegian Institute for Water Research / Ecotoxicology; A.L. Macken, Norwegian Institute for Water Research, NIVA; A. Rico, IMDEA Water Institute / Aquatic Ecotoxicology; T.C. Telfer, University of Stirling

Aquaculture is a major food production subsector which will play a major role in filling the growing seafood supply gap, estimated to be of the order of 47.5 million tons by 2050. However, aquaculture cannot be practiced everywhere; it requires a particular set of natural, social and economic resources which must be used wisely if the development of the sector is to be sustainable. Appropriate environmental characteristics, good water quality, well-understood social interactions and use of inland and coastal resources are essential to maintaining in existing and future aquaculture systems. Consequently, suitable zoning, selection of sites and application of carrying capacity are among the most important issues for the future success of European aquaculture, and also predicate the need for sustainability, resilience and best practice guidelines, as provided by the Ecosystem Approach to Aquaculture. The four-year Horizon 2020 TAPAS research project, which started in March 2016, aims to consolidate the environmental sustainability of European aquaculture by developing tools, approaches and frameworks to support EU Member States in establishing a coherent and efficient regulatory framework, implementing the Strategic Guidelines for the sustainable development of European aquaculture and delivering a technology and decision framework for sustainable growth. The ultimate goal of the project is to create cost-efficient management tools and practices for the European aquaculture sector to investigate the scope of fish farming activity, social interactions, potential environmental impacts and any future risks.

467

Preliminary investigation on the occurrence of multifunctional organic micropollutants in offshore seawater and fish farm

L. You, National University of Singapore; K. Gin, National University of Singapore / Civil & Environmental Engineering

Limited research has been conducted on the occurrence and distribution of antibiotics, pharmaceuticals, personal care products, endocrine disrupting chemicals and artificial sweeteners in the marine environment despite being increasingly impacted by these micropollutants (MPs). In this study, the presence and distribution patterns of 53 multifunctional organic micropollutants belonging to 14 different groups were investigated in offshore seawaters and fish farms of Singapore. The sampling area is affected by various anthropogenic pressures including treated effluents, fish farming, shipping and port activities. A total of 23 MPs were found in offshore seawaters, 9 of them with detection frequencies higher than 50%. The highest detected values corresponded to cyclamate, salicylic acid and sucralose, with concentration range of

468

Perspectives on Urbanization, Water Reuse, and Aquaculture Product Quality

B.W. Brooks, Baylor University / Dept of Environmental Science; J.L. Conkle, Texas A&M University Corpus Christi / Physical and Environmental Sciencees

By 2050, it is estimated that global food production must increase by 50%. Aquaculture will play an important role to meet these needs. For example, in 2014 aquaculture surpassed global fisheries in providing fish for human consumption. It is important to note that global aquaculture activities can in urban and periurban regions with differential waste management capacity. Yet high population densities in urbanizing regions result in concentration of food, energy, water and other resource consumption. Urbanization also leads to concentration of chemical use, which inherently results in exposures to human populations and ecosystems receiving waste streams within and from these urban centers. In developing nations, where many of the megacities will continue to emerge over the next few decades, access to chemical products is occurring faster than public health interventions and environmental management systems are being implemented. Unfortunately, 80% of the global sewage production is not treated, but returned to the environment and thus reused for various purposes. These non-traditional reused waters are being recycled for agriculture, including aquaculture in areas experiencing rapid urbanization, yet implications for water security, food safety and international trade are not routinely examined to manage more sustainable aquaculture practices. In the current presentation we draw from our ongoing efforts in Asia and North America to understand bioaccumulation of organic contaminants of emerging concern in common fish and shellfish used for aquaculture. For example, we have observed aquacultured bivalves to accumulate diverse contaminants of concern (e.g., pharmaceuticals, pesticides, flame retardants), apparently from landfill leachates and effluent discharges of marginal quality, in Hong Kong. Our findings from laboratory uptake and depuration studies with channel catfish and tilapia focus on contaminants with diverse physico-chemical properties (e.g., weak base medicine, phosphorus-based flame retardant, perfluorinated compound, cyanotoxin) and provide an approach to improve aquaculture practice and to support bioaccumulation assessments for chemicals falling outside of applicability domains for nonionizable organic contaminants. In North America we are examining intersections among water reuse practices and aquaculture for various products. Such efforts apparent warranted at the global scale.

469

Bioaccumulation of selected veterinary medicines in the blue mussel (Mytilus edulis)

S. Brooks, NIVA / Ecotoxicology and Risk Assessment; B. Beylich, NIVA; A. Ruus, NIVA / NIVA; J. Rundberget, NIVA; A. Lillicrap, NIVA / Ecotoxicology and Risk Assessment

Veterinary medicines are widely used within the fish farming industry for the control of sea lice infestation. In 2016, over 10 tonnes of veterinary medicines were used by Norwegian fish farms for the control of sea lice. The impact on non-target species has raised increasing concern. For instance, do wild and farmed mussels in the vicinity to these fish farms have the potential to bioaccumulate these chemicals and thereby pose a threat to human health? On the other hand, mussels may be the most suitable biomonitoring species for the presence of veterinary medicines in the environment. To better understand these scenarios, and the fate of these chemicals in the environment, a series of laboratory controlled exposures were performed to determine the bioaccumulation and depuration of selected veterinary medicines in the blue mussel (*Mytilus edulis*). The veterinary medicines included teflubenzuron, emamectin, deltamethrin and azamethiphos. Due to the low solubility of teflubenzuron and deltamethrin a saturation column was employed within a flow-through system to deliver a stable concentration of test chemical over a 14-day uptake phase. Water and mussel samples were collected at time intervals during the 14-day uptake phase, and again following transfer of the mussels into clean flowing seawater during the 7 to 14 day depuration phase. The effects of salinity on the bioaccumulation of teflubenzuron was also investigated to see whether mussels in brackish waters show different bioaccumulation dynamics. So far, the results have shown a clear uptake of teflubenzuron over 14 days, reaching maximum concentrations (~1500 ng/g) after 10 days. Depuration of teflubenzuron was fast for the first 2 days, although still present at approximately 250 ng/g after 7 days depuration. Salinity had no apparent effect on the bioaccumulation of teflubenzuron. In contrast, emamectin showed lower bioaccumulation, with maximum concentrations of 45 ng/g after 6 days. No significant depuration of emamectin was observed after 7 days in clean flowing seawater. The results suggest that mussels are a suitable biomonitoring species for the presence of veterinary medicines in the environment. Additionally, mussel farms in close proximity to fish farms have the potential to bioaccumulate these chemicals in their tissues and is subsequently recommended for monitoring.

470

Contribution of nuclear applications to better understand bioaccumulation of contaminants in aquaculture species

M. Metian, IAEA-EL / Radioecology Lab; S. Pouil, F. Oberhänsli, International Atomic Energy Agency / Environment laboratories; P. Bustamante, Université de La Rochelle / LIttoral ENvironnement et Sociétés LIENSs; P. Swarzenski, International Atomic Energy Agency / Radioecology Lab

Environmental pollution from aquaculture is often seen as a major concern, but today, increasingly is the potential exposure of aquaculture to contaminants. In order to fully understand the contamination risk of farm-raised species, nuclear applications can be used. This is a very powerful approach that allows identifying the susceptibility of economically important species (fish and seafood) to be contaminated. Marine fish farming is regarded as the future of aquaculture and thus, the safety of these farm-raised fish is paramount particularly as 50 % of fish consumed are now farm-raised. Therefore, a better understanding bioaccumulation processes of such contaminants with current aquafarming practices is essential. Such work will attempt to better understand the role the fish food or key environmental parameters on contamination of fish that may affect the health of the farmed species and/or the human consumer. This has been commonly done in a natural setting but is now beginning to be examined for fish farming practices. Major advantages of radiotracer techniques over conventional techniques are their very high sensitivity and discrimination capacity: it permits the measurement of bioaccumulation kinetics of several elements at realistic (viz. low) environmental concentrations in a single experiment. Furthermore, some radiotracer permits the non-destructive analyses of contaminant levels in living organisms. This paper identifies present and future threats on farm-raised fish from a contamination point of view, and presents a synthesis of experimental results completed on farm-raised fish exposed to contaminants in realistic environmental conditions. It revealed, for example, the various effects that food, water salinity and temperature can have on the Assimilation efficiencies of trace elements and radionuclides in farmed fish. Such findings paving the way for further investigations on the potential use of nuclear techniques in aquaculture and food safety.

471

Effects of antibiotic´s medicated fish feed in the marine environment

B. Gonzalez-Gaya, IMDEA Water (G84912732) / Environmental Chemistry; N. García Bueno, I. Gomez, B. Martinez-Lopez, P. Franco, University of Murcia / Ecology and Hydrology; E. Buelow, Limoges University / Medicine Faculty, Inserm Umr 1092; A. Marin, University of Murcia / Ecology and Hydrology; A. Rico, IMDEA Water Institute / Aquatic Ecotoxicology

Intensive aquaculture is considered to be an important source of antibiotics into the marine environment. Antibiotics used in aquaculture have been reported to accumulate on sediments and non target aquatic organisms, modifying the biodiversity and the environmental conditions in areas close to fish farms. Moreover, recent studies show an increasing ocurrence of resistance genes in environmental bacteria next to fish farms, which indicates the assembly, selection and dissemination of antimicrobial resistance through open aquaculture installations and might thereby compromise environnmental and human health. A field experiment was performed in a moderatelly impacted bay in the south east coast of Spain (Aguilas, Murcia), which consisted of a series of sediment traps (covered by a net vs. uncovered) filled with local sediment and fish feed; non medicated or medicated with three antibiotics (oxytetracycline, florfenicol and flumequine). Fish feeds were applied simulating fish farm loses for a period of 3 weeks. Measured antibiotic concentrations in the sediment were 2700 – 8000 ng/g (average 1% of the applied ammount) for oxytetracycline, and 19000 – 54000 ng/g (average 10% of applied amount) for flumequine. Florfenicol was not detected. Different accumulation rates were found in covered/uncovered traps due to wild fish influences in the availability of feed and bioturbation. Physico-chemical characteristics of the sediment also changed; with a higher S and lower N content and a larger percentage of fine material in feed affected treatments. Invertebrate

presence was also correlated with the food availability, although no evident effects of the antibiotics were found over the analyzed samples. Bioaccumulation of the target antibiotics in the invertebrate community and evaluation of the antibiotic impacts over the microbiome and resistome of the sediment bacteria is still ongoing. This is one of the first studies describing fish feed and antibiotic impacts produced by aquaculture under Mediterranean conditions.

Systems ecotoxicology: application of OMICS data across multiple level of biological organization in research and risk assessment (II)

472

Systems toxicology approach for the assessment of zebrafish cardiac and neurotoxicity

R. Li, M. Talikka, Philip Morris International; S. Madan, Fraunhofer Institute for Algorithms and Scientific Computing; J. Doerpinghaus, Fraunhofer Institute for Algorithms and Scientific Computing; A. Zupanic, Eawag Swiss Federal Institute of Aquatic Science and Technology / Environmental Toxicology; J. Fluck, Fraunhofer Institute for Algorithms and Scientific Computing; C.M. vom Berg, Eawag Swiss Federal Institute of Aquatic Science and Technology / Environmental Toxicology; J. Szostak, F. Martin, M. Peitsch, J. Hoeng, Philip Morris International

A major goal in the field of toxicology is to predict long term animal health risks and/or environmental hazards associated with a particular substance(s). Traditionally utilised classical toxicology methods involve animal exposure over a relatively short period and recording adverse outcomes. These data are then extrapolated to long term effects and to other species. The accuracy of such extrapolation would benefit from mechanistic understanding of toxicity. However, molecular basis for adverse outcomes is not easily interpreted from classical toxicology methods. Here we present our systems toxicology approach that focuses on deciphering biological mechanisms responsible for adverse outcomes. The underlying structure of this approach is a computable biological network model. We have developed two models describing molecular pathways that lead to cardiotoxicity and neurotoxicity in zebrafish larvae based on the knowledge curated from scientific literature. Key signalling nodes in the model are linked to information about downstream gene expression. Differential expression of downstream genes can be used to infer activity of the upstream protein – a process termed network scoring. Scoring of the network highlights the most affected nodes, which leads to mechanistic hypothesis generation and gives a quantifiable measure of network perturbation. In parallel to network scoring, we utilize classical toxicology methods to detect adverse outcomes. We present the acute toxicity results for selected chemicals (e.g. acrylamide, arsenic, citalopram, imidacloprid) according to the OECD fish embryo acute toxicity test (OECD test guideline 236). We then report results from chemically exposed larvae in functional cardiac and behavioural assays, and transcriptomics analyses. Finally, we describe the utility of the network model in interpreting transcriptomics analyses to gain mechanistic insight into the molecular events initiated by a given chemical. Cardiac and neural apical endpoints together with computational network scoring provide a comprehensive method for linking molecular events to organ toxicity. This approach will enable more accurate toxicity predictions over long exposures and in different species.

473

Time response relationship between gene expression and life history in a Daphnia population exposed to heavy metals

J. Asselman, I. Semmouri, Ghent University / Laboratory for Environmental Toxicology and Aquatic Ecology GhEnToxLab unit; K. De Schamphelaere, Ghent University (UGent) / Applied Ecology and Environmental Biology

Over the last decade, molecular technologies have evolved into robust high throughput platforms available to many scientists in a wide variety of disciplines. Implementation of these technologies in ecotoxicology and risk assessments have focused on mechanisms of toxicity and stress response on the gene level to explain effects at the organism level. However, current studies remain focused at the individual level and rarely include population level molecular responses. Population level molecular responses may provide a better insight into the potential mechanisms at play at the population level while at the same time avoid focusing on gene expression patterns that are the cause of clonal or interindividual variation. Furthermore, most studies select an arbitrary timepoint to measure gene expression responses without any prior knowledge. Here, we focus on population level responses of a *Daphnia magna* population to arsenic and copper and their binary mixture. The population was exposed to low chronic toxicity concentrations of arsenic and copper resulting primarily in effects on reproduction rather than survival. Rather than focusing on a single arbitrary timepoint, gene expression data and life history data were both recorded at multiple time points. As such, these datasets will provide a first basis on how exposure duration may affect the conclusions and decisions made about the toxicity of chemicals. In addition, by collecting both molecular data and life history data, we will be able to better understand the time response relationship in populations under stress both at the life history level and the molecular level. This will allow us to better integrate these two data types and identify potential causal relationships between the molecular level and the life history level. The identification of such causal relationships will play an integral part of incorporating omics data in environmental risk assessment.

474

How to implement functional responses of microalgae in risk assessment processing?

F. Larras, Helmholtz Center for Environmental Research - UFZ GmbH; E. Billoir, Université de Lorraine, CNRS UMR 7360; S. Scholz, Helmholtz Centre for Environmental Research / Department Bioanalytical Ecotoxicology; M. Delignette-Muller, VetAgro Sup / Laboratory of Biometry and Evolutionnary Biology; M. Schmitt-Jansen, UFZ - Helmholz Ctre Environm. Research / Department of Bioanalytical Ecotoxicology

Microorganisms (e.g. bacteria, fungi and algae) are involved in various ecosystem functions such as biogeochemical cycles or pollutants degradation meaning that they are crucial for ecosystem functioning. In the environment, organisms are exposed to anthropogenic pressures which are known to potentially induce structural and functional changes. If such causal links are identified, little is known about the involved biochemical pathways supporting specific functions. Moreover, most of *a priori* ecological risk assessment (ERA) tools are based on structural endpoints and do not necessarily ensure the protection of these functions. The recent raise of OMICs approaches (e.g. transcriptomics and metabolomics) opens the perspective in ecotoxicology to explore pathways involved in ecological functions. The main aim of this study was to provide a new and innovative risk assessment tool based on functional responses (captured via OMICs approaches) of periphytic communities in order to protect the functions that they ensured. The rationale to use OMICs in such context is to provide more protective and early warning thresholds. The transcriptomic and the metabolomic responses of *Scenedesmus vacuolatus* to triclosan were explored after exposure of 14 hours along an increasing gradient of 5 concentrations (from 0.69 to 6.63 µg/L, 5 replicates). Within a dedicated workflow, we selected the responsive molecular items (metabolites/transcripts), we built concentration response curves for each of them and we derived a sensitivity value from each curve (even the non-monotonic one). Molecular items showed mainly non-sigmoid and even non-monotonic responses to triclosan exposure. For example, the transcripts data were mainly best described by an exponential model for more than half of the curves and a Gaussian or log-Gaussian model for more than a quarter of the curves. Then, the molecular items were linked (when possible) to the pathways they are involved in. From that information, we built a SSD-like tool based on functional responses captured at the community level in order to protect functions and integrating two levels of OMICs responses. The next step consists to build such tool from the periphytic community level.

475

Sex, drugs and Daphnia magna. A multi-omics approach suggests conserved mechanisms of interaction between metallohormones and endocrine disruptors

E. Caamano-Gutierrez, University of Liverpool / Computational Biology Facility; P. Antczak, University of Liverpool / Institute of Integrative Biology; L. Mirbahai, The University of Birmingham / School of Biosciences; K. Grintzalis, The University of Birmingham; M.R. Viant, University of Birmingham / School of Biosciences; F. Falciani, University of Liverpool / Institute of Integrative Biology

The assessment of environmental exposure to toxic chemicals released by human activity as well as their impact on biological systems is key to protect the biosphere. Current environmental monitoring protocols are based on chemical analysis and an assessment of biodiversity. Although this has been a very effective strategy, it has some shortcomings. These include the fact that a relatively limited number of compounds can be measured and linked to biologically relevant organism-level responses. The issue is particularly challenging in chronic exposures and in complex mixtures scenarios. This project aims at identifying the molecular networks linked to single and mixture exposures and to use these to infer the effects of chemical mixtures. We approached this important challenge by applying a systems biology approach to integrate expression profiling, metabolomics and phenotypic data (respiration and feeding rates), representing the response of *Daphnia magna* to a panel of environmentally relevant chemicals and their mixtures. Firstly, it was exposed to a battery of single compounds with known mode of action (MoA) i.e. estrogen disruptors and acetylcholine esterase (AChE) inhibitors as well as metals with unclassified MoA. We have been able to model the differences between the two main MoA studied and linked them to biological activities within *Daphnia*. Furthermore, we have found that as expected, metals do not show a common MoA, with some of them clustering closer to either endocrine disruptors or AChE inhibitors. Cadmium (Cd), which clustered with endocrine disruptors, has already been shown to play a role with the estrogen receptor in humans but its role in *D. magna* is still under surveillance. To further study this finding we exposed *D. magna* to complex mixtures of Cd and ethinylestradiol. While the individual exposures triggered the alteration of expression of a relatively large number of genes, the exposure to the mixture showed little or no effect. These results indicated that both compounds share a complex interaction at a molecular level suggesting that the degree of conservation of the regulatory pathways underlying response to endocrine disruptors may be higher than previously thought. Overall, our work shows that it is possible to predict a compound MoA from its

molecular state and also predict additive or synergistic effects of mixture exposure.

476

Data-driven systems biology approach gives insight into a complex process of water remediation

J. Kronberg-Guzman, The University of Birmingham / School of Biosciences; T.D. Williams, University of Birmingham / School of Biosciences; A. Murk, Wageningen Agricultural University / Dept of Toxicology; E. Foekema, Wageningen IMARES; R. van der Oost, Waternet / Onderzoek en Advies; K. Chipman, University of Birmingham; F. Falciani, University of Liverpool / Institute of Integrative Biology

Introduction. Increasing population and industrial production put strain on clean water resources. Even in highly developed countries with advanced waste water treatment plants, water quality could be improved further before releasing it. Constructed wetlands have been used for water treatment for decades and are a low-cost natural option. Waterharmonica is an example of such additional treatment, consisting of a sedimentation pond, reed bed and a wetland forest.In this work, we have used three-spined stickleback living in mesocosms containing water from various stages of additional remediation from three different sites in the Netherlands. **Aim.** The aim of this work was to understand the effects of additional steps of water remediation. **Results.**We have used a data-driven systems biology approach to understand the relationship between the environment (chemical concentrations), molecular high-throughput measurements (stickleback liver gene expression), physiological parameters and more traditional measures of toxicity. We first integrated all different measurements into static similarity networks and modularised these so that in each module, genes are responding in a similar way during different stages of remediation. We see that some chemicals with high chemical risk (aldicarb, chlorpyriphos, fluoranthene, pirimiphos methyl) decrease in all sites and are also correlated with gene expression in both male and female stickleback. However, some chemicals are only correlated with gene expression in only male of female stickleback. We also see that some chemicals for which predicted no-effect concentration (PNEC) is not known (such as PCB-s and mineral oils) are associated with modules containing several high-risk chemicals. Functional annotation reveals further insights. For example a module of the male stickleback network correlated with liver weight and several chemicals including triclosan and phthalates has a statistically significant number of genes from the KEGG pathway „metabolism of xenobiotics by cytochrome P450“. However, a module in the male-specific network that is not correlated with any chemicals, is enriched in KEGG functions related to immune functions („antigen processing and presentation“, „type I diabetes mellitus“ and „cytokine-cytokine receptor interaction“). **Conclusion.** We have showed the potential of data-driven systems biology approach integrationg various types of data to give insight into the effects of a complex remediation system.

477

Co-expression network analysis of massive proteogenomic data: applications in ecotoxicology.

D. Degli Esposti, Irstea / UR RIVERLY Laboratoire Ecotoxicologie; C. ALMUNIA, CEA Paris-Saclay; D.D. Gouveia, Irstea Lyon / UR MALY Laboratoire Ecotoxicologie; J. Trapp, Irstea Lyon; J. Gaillard, CEA / Laboratoire de Biochimie des Systemes Perturbés; O. Pible, CEA; a. chaumot, O. Geffard, Irstea / UR MALY Laboratoire Ecotoxicologie; J. Armengaud, CEA / Laboratory Innovative technologies for Detection and Diagnostics

Data mining of high throughput omics data acquired in test species under contaminant exposure promises the possibility to gain insights into the mode of action of chemical compounds and molecular pathways involved in toxic responses. Intuitive network concepts (e.g. connectivity and modularity) have been found useful for analyzing complex interactions and successfully applied to study gene-gene and protein-protein interactions. Currently, a majority of protein networks are constructed using protein-protein interaction (PPI) databases. However, manually curated PPI databases are typically heterogeneous, documented for few model species, and often characterized by uncomplete coverage, and selection or detection biases. *De novo* (or no *a priori*) approaches based on observed data offer an alternative under which prior knowledge of protein interaction is not necessary but rather advantageously replaced by direct measurements and pair-wise correlation analysis of their abundance. This approach may be particularly powerful to identify signaling pathways which proteins with unknown function belong to or to identify novel, pertinent biomarkers of toxicant exposure. Here we present a network analysis method applied to shotgun high-throughput proteomic data we produced for the aquatic sentinel organism *Gammarus fossarum*. In particular, shotgun proteomics was used to identify the molecular key players involved in different physiological states linked to reproduction and in case of exposure to insecticides potentially inducing endocrine disruption in this crustacean. We identified protein modules significantly associated to morphologically well-characterized physiological states and to pesticide exposure. Moreover, the identification of crucial hub proteins could allow proposing exposure-related or toxicological functional biomarkers. This new data mining procedure opens interesting perspectives for the development of a novel generation of molecular diagnostic biomarkers in ecotoxicology.

Towards a shared understanding of science and risk communication in the context of the inevitability of chemicals and the hazard they may represent (I)

478

How researchers can work in alliance with citizens to fight misinformation and improve public debates

S. Vanthournout, Sense About Science EU

Public resistance against glyphosate, GMOs, animal testing, vaccination and numerous other scientific innovations has made many scientists defensive and paranoid about the public. Recent discussion about a post-truth society and anti-intellectualism have increased this perception of a hostile and ignorant public. With concrete examples, Sofie will illustrate a different, more effective approach for both researchers and non-researchers to bring back reason into emotional debates. This approach, called public-led, expert-fed – in which scientists respond directly to real, unedited questions from the public – breaks through polarised and difficult debates because conversation is led by the questions and issues people raise. It allows researchers to identify gaps and misunderstandings in the public debate and to respond to them. With this approach, rather than fighting the public resistance against new technologies, researchers can work in an alliance with citizens to fight misinformation and improve the public debates.

479

Discussion: the need to promote good science and evidence in public debates

480

How to communicate the risks posed by endocrine disrupting chemicals? (I)

J. Legler, Utrecht University / Institute for Environmental Studies

There is emerging evidence that some of the increasing occurrences of endocrine-related disorders in humans and wildlife are linked to the exposure to endocrine disrupting chemicals (EDCs). Because of the potential of significant impacts on organismal and population health that can result from disruption of endocrine homeostasis, numerous governments have established legislations that regulate chemicals that have the potential to interact with the endocrine system of humans and wildlife. However, while there is agreement on the need for regulation of EDCs, the frameworks to assess and regulate candidate EDCs differ significantly among regions. Furthermore, the mixed messages delivered by the media to the public with regard to the risks EDCs may pose add to the confusion currently existing within society, and which has split opinions on how to address this issue. This presentation will review the issue of endocrine disruption from a human health and environmental perspective, and discuss current approaches to the assessment of the risk/hazard of EDCs in Europe and North America. Through discussions among the presenters and the audience we aim to explore a roadmap on how to address the risks posed by EDCs and where the priorities for future research should lie.

481

How to communicate the risks posed by endocrine disrupting chemicals? (II)

M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre

There is emerging evidence that some of the increasing occurrences of endocrine-related disorders in humans and wildlife are linked to the exposure to endocrine disrupting chemicals (EDCs). Because of the potential of significant impacts on organismal and population health that can result from disruption of endocrine homeostasis, numerous governments have established legislations that regulate chemicals that have the potential to interact with the endocrine system of humans and wildlife. However, while there is agreement on the need for regulation of EDCs, the frameworks to assess and regulate candidate EDCs differ significantly among regions. Furthermore, the mixed messages delivered by the media to the public with regard to the risks EDCs may pose add to the confusion currently existing within society, and which has split opinions on how to address this issue. This presentation will review the issue of endocrine disruption from a human health and environmental perspective, and discuss current approaches to the assessment of the risk/hazard of EDCs in Europe and North America. Through discussions among the presenters and the audience we aim to explore a roadmap on how to address the risks posed by EDCs and where the priorities for future research should lie.

482

Discussion Endocrine Disrupting Chemicals

483

A regulator's perspective in involving stakeholders and the public in the regulation of a substance

C. Ajao, ECHA-European Chemicals Agency; W. de Wolf, ECHA / Product Safety & Regulatory Compliance

The European Chemicals Agency (ECHA) was established in June 2007 through the REACH Regulation, and the registration of all substances already on the market above 1 ton per annum will be completed in 2018. Since its start the areas of responsibility expanded from industrial chemicals to biocides, capturing as well the communication of chemical hazards to workers and the public through the Classification, Labelling and Packaging Regulation (CLP), and the regulation of international trade of hazardous chemicals. The latter includes support for the protection of human health and the environment by providing developing countries with information on how to store, transport, use and dispose of hazardous chemicals safely through the Prior Informed Consent Regulation (PIC). \nIn its decision making and opinion forming, ECHA uses the scientific information provided by academia and industry and applies them within the regulatory framework that it operates. Transparency is one of the values that is driving ECHA in its interactions with its different stakeholders, and the ECHA Scientific Committees invite ECHA's accredited stakeholders as regular observers and contributors to its meetings. \nThis presentation will look at regulatory science communication by describing the different stakeholders that ECHA interacts with, the forms of communication used and their timeframes. It will also explain the regulatory boundaries ECHA has to abide to, which influence the uptake of the latest science developments, and their communication with a special focus on the decision making and opinion forming at the Member State Committee. DISCLAIMER: 'The views expressed in this abstract are solely those of the authors and the content of the paper does not represent the views or position of the European Chemicals Agency'.

484
Questions/Discussion

485
General Discussion with panel of Sofie Vanthournout, Juliette Legler and Markus Hecker

486
Concluding remarks part I and a teaser for part II!
A. Leopold, Calidris Environment BV / Calidris Environment BV; T. Seiler, RWTH Aachen University / Ecosystem Analysis; C. Ajao, ECHA-European Chemicals Agency

Ecological risks under complex, multiple-stressor threat scenarios: integrating chemical effects with environmental drivers (III)

487
The impact of chemical pollution on the resilience of soils under multiple stress
A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics; W. Amelung, University of Bonn; H. Hollert, RWTH Aachen University / Institute for Environmental Research; M. Kaestner, Helmholtz Centre for Environmental Research UFZ / Dept Environmental Biotechnology; E. Kandeler, University of Hohenheim; J. Kruse, University of Bonn; A. Miltner, Helmholtz Centre for Environmental Research UFZ / Department of Environmental Biotechnoloy; R. Ottermanns, RWTH Aachen University / Institute for Environmental Research; H. Pagel, University of Hohenheim; S. Peth, University of Kassel; C. Poll, University of Hohenheim; G. Rambold, University of Bayreuth; M. Schloter, Helmholtz Zentrum Munchen / Research Unit Environmental Genomics; S. Schulz, Helmholtz Zentrum Munchen; T. Streck, University of Hohenheim; M. Roß-Nickoll, RWTH Aachen University / Institute for Environmental Research
Soils are faced with man-made chemical stress, such as the input of organic or metal-containing pesticides, in combination with non-chemical stressors like soil compaction due to agricultural traffic and natural disturbance like drought. Although multiple stress factors are typically co-occurring in the environment, research in soil sciences on this aspect is limited and focuses mostly on single structural or functional endpoints. A mechanistic understanding of the reaction of soils to multiple stressors is currently lacking. Based on a review of resilience theory, we introduce a concept for research on the ability of soil polluted by xenobiotics or other chemicals as one stressor to resist further natural or anthropogenic stress and to retain its functions and structure. There is strong indication that pollution as a primary stressor will change the system reaction of soil, i.e., its resilience, stability and resistance. It can be expected that pollution affects the physiological adaption of organisms and the functional redundancy of the soil to further stress. We hypothesize that the recovery of organisms and chemical-physical properties after impact of a follow-up stressor in polluted soil differ from that in non-polluted soil, i.e., polluted soil has a different dynamical stability, and resilience of the contaminated soil is lower compared to that of not or less contaminated soil. Thus, a polluted soil might more easily change into another system regime after occurrence of further stress. We highlight this issue by compiling the literature exemplarily for the effects of Cu contamination and compaction on soil functions and structure. However, examples of further co-occuring stress scenarios will be described as well. In this discussion paper, we propose to intensify research on effects of combined stresses involving a multidisciplinary team of experts and provide suggestions for corresponding experiments. Our concept offers thus a framework for system level analysis of soils paving the way to enhance ecological theory.

488
Combined effects of temperature and metal exposure on cell membrane fatty acid composition, lipid peroxidation, antioxidant capacities and desaturase and elongase transcription in freshwater fish
M. Fadhlaoui, INRS - Eau, Terre et Environnement / Centre Eau Terre Environnement; F. Pierron, Université de Bordeaux / UMR EPOC CNRS 5805; P. Couture, INRS / Centre Eau Terre Environnement
In this project, two freshwater fish commonly found in areas affected by metal contamination were acclimated to different temperatures (9 and 28°C for yellow perch (*Perca flavescens*) and 15, 25 and 30°C for fathead minnow (*Pimephales promelas*)) and exposed either to Cd or Ni during 8 weeks. At the end of exposures, we measured cell membrane phospholipid fatty acid composition, the activities of superoxide dismutase, catalase, glutathione-S-transferase, glutathione peroxidase (enzyme indicators of antioxidant capacities), the concentrations of glutathione (antioxidant) and malondialdehyde (indicator of lipid peroxidation (LPO)) as well as the transcription levels of desaturases (*fads2*, *degs2*, *scd2*) and elongases (*elovl2*, *elovl5*, *elovl6*). Both yellow perch and fathead minnow counteracted the effects of changes in acclimation temperature on cell membrane properties by remodelling their phospholipid fatty acid composition. Specifically, in the muscle of both species, polyunsaturated fatty acids increased in cold-acclimated fish compared to warm-acclimated fish, in agreement with the theory of homeoviscous adaptation. However, brain cell membrane composition was more conservative, especially in fathead minnows. Polyunsaturated fatty acids are more vulnerable to LPO than saturated fatty acids and metal contamination leads to oxidative stress. We therefore tested the hypothesis that temperature-induced changes in cell membrane polyunsaturation are accompanied by variations in LPO in metal-exposed fish. Unexpectedly, in both species, metal exposure itself affected membrane fatty acid composition. In yellow perch, the normal response of cell membrane composition to thermal acclimation was reversed by exposure to both metals. Yet, in spite of the high polyunsaturation level in warm-acclimated fish under Ni exposure, MDA concentration was the lowest, suggesting a massive response of the antioxidant system to fight against LPO. In fathead minnow, metal exposure also affected the membrane fatty acid composition of both tissues, but the response was subtler than for yellow perch. We observed a mismatch between desaturase and elongase gene transcription and membrane composition. Overall, our results suggest that levels of control of cell membrane fatty acid composition other than gene transcription may be affected by temperature and metal exposure, such as post-transcriptional regulation of gene transcription and *de novo* phospholipid biosynthesis.

489
The effect of water chemistry on cadmium induced olfactory impairment in juvenile rainbow trout (Oncorhynchus mykiss)
S. Volz, RWTH Aachen University / Department of Ecosystem Analysis; S. Bogart, A. Macdonald Wilson, University of Lethbridge / Department of Biological Sciences; H. Hollert, RWTH Aachen University / Institute for Environmental Research; G.G. Pyle, University of Lethbridge / Biological Sciences
Fish are dependent on olfaction since a variety of essential behaviours, such as foraging, predator avoidance and mate selection, are mediated by the olfactory system. Metals are well known to affect the olfactory system of fishes at environmentally-relevant concentrations. As metal toxicity varies with water chemistry in a predictable manner, modelling approaches, such as the Biotic Ligand Model (BLM), are powerful tools to predict site-specific effect concentrations. To date, the BLM used in risk assessment for fish only predicts gill-based metal toxicity. However, metal-binding dynamics at the olfactory epithelium may be different than for gills. For this reason, the present study investigated the impact of water chemistry on cadmium(Cd)-induced olfactory impairment. In order to assess the effect of Cd on the olfactory system, fish were exposed to 45-720 µg/L Cd for 24 h. Subsequently, olfactory responses to two odorants were measured via electro-olfactography (EOG). To investigate the impact of water chemistry on Cd-induced olfactory impairment, fish were exposed to the EOG-based 24-h IC50 of Cd (210 µg/L) in reconstituted water with varying hardness, pH, and dissolved organic carbon (DOC) concentrations for 24 h. Cd inhibited the EOG response of rainbow trout in a concentration dependant manner. Fish exposed to 210 µg/L Cd for 24 h showed reduced olfactory response to TCA by 50%. Changes in water chemistry had a significant impact on Cd-induced olfactory impairment. Decreasing water hardness from 150 to 40 mg/L as $CaCO_3$ increased the inhibitory effect of Cd on the EOG response from 55% to more than 95%, respectively. Hence, hardness ions ameliorated Cd-induced olfactory impairment. By contrast, Cd-induced olfactory inhibition increased with rising pH, which may be due to a difference in metal speciation. DOC had a protective effect against Cd-induced olfactory impairment, l ikely by forming complexes with Cd ions and reducing their

bioavailability. In conclusion, water chemistry is an important modulator of metal toxicity, not only for acute lethality but also for sub-lethal effects, such as olfactory impairment. In order to enable the prediction of site-specific olfactory toxicity, the development of a BLM parametrized to the olfactory system of fish would be very beneficial. However, more data on the effect of water chemistry on metal-induced olfactory impairment is required to be able to determine affinity constants and maximal binding capacities.

490

Physiological and biochemical responses of polychaetes: interplay of elements contaminated sediments and salinity changes

A. Pires, Universidade de Aveiro / Biologia; R. Freitas, University of Aveiro / Departamento de Biologia CESAM; C. Patinha, Universidade de Aveiro; E.F. Silva, University of Aveiro / Geosciences; E. Figueira, University of Aveiro / Biology CESAM

Coastal systems often serve as sinks for toxic elements, and changes in salinity, predicted to occur due to global climate changeare expected to influence elements geochemistry in aquatic systems. The effects of these changes can also alter biota sensitivity and elements uptake rates, leading to important changes in estuarine communities, mainly on those organisms living in sediments, such as polychaetes, which also support much of the diversity at higher trophic levels. So, this work examines the interactions of elements contamination and change in salinity and polychaetes performance, highlighting modifications that coastal systems may undergo due to alterations driven by salinity change. Polychaetes were exposed to elements contaminated sediments (median sand and fine sand sediments, collected from contaminated areas from ria de Aveiro lagoon, Portugal) and salinities 21, 28 and 40, for 28 days. This study aimed to evaluate physiological (regenerative capacity of *Diopatra neapolitana*), behaviour (burrowing tests with *Hediste diversicolor* and *Arenicola marina*) and biochemical responses: indicators of cell damage (LPO) antioxidant (SOD and CAT), and biotransformation (GSTs) enzymes. After exposure, both types of sediments had lower total elements concentration (TEC), when compared to original sediments, mainly in sediments with *A. marina* and *H. diversicolor*. Sediments exposed to salinity 40, mainly those containing *H. diversicolor* had evenless TEC than remaining sediments, which seems that salinity changes may influence elements availability. LPO levels were higher at salinity 40 for *H. diversicolor* and *A. marina*, and at salinities 21 and 40 for *D. neapolitana*. Although polychaetes were able to increase the activity of SOD, CAT and GSTs, these defense mechanisms were not sufficiently efficient to fight against the excess of ROS, leading to LPO.Furthermore, in *H. diversicolor*, the burrowing behaviour was impaired in polychaetes in fine sand sediments and salinity 40. For *A. marina* exposure to median sand sediment for all salinities and to fine sand sediment at salinity 40 led to a decrease of burrowing kinetics. *D. neapolitana* individuals exposed at salinities 21 and 40, for both sediments, exhibited lower capacity to regenerate their body when compared to salinity 28 (control). Overall, this study demonstrates that variations in salinity can strongly affect elements availability. Interaction of both variables impacted polychaetes responses differently.

491

Do trace metal contamination and parasitism infestation influence the activity of the bioturbator Upogebia pusilla?

a. dairain, EPOC, University of Bordeaux / UMR EPOC CNRS 5805; X. de Montaudouin, A. Ciutat, P. Gonzalez, University of Bordeaux / UMR EPOC CNRS 5805; M. Baudrimont, Université de Bordeaux / UMR EPOC CNRS 5805; O. Maire, P. Gourves, G. Daffe, A. LEGEAY, University of Bordeaux / UMR EPOC CNRS 5805

In marine environment, bioturbating species are considered as important ecosystem engineer species. Bioturbators are mainly benthic organisms living in the sediment. Their fossorial life style deeply alters the physical and biochemical properties of sediments. In marine soft-bottom environments, mud shrimp are considered as among the most important bioturbators. Because of their intense burrowing activities, they exert a major influence on solute and porewater exchanges, habitat structuration and benthic community composition. The influence of mud shrimp on their environment is related to the intensity of their behavior. Several factors could interfere with bioturbators fitness and therefore modify their influence as ecosystem engineer species. Regarding mud shrimp, only a few factors have been studied. Among them, the role of trace metals on mud shrimp fitness and bioturbation activities has never been investigated yet. Besides, mud shrimp are frequently parasitized by bopyrid isopods, known to have a deleterious effect on their host. Indeed, they deeply impair mud shrimp physiological state and potentially alter their bioturbation intensity. The aim of this study was to examine the role of a common trace metal (cadmium) and of bopyrid parasites on the physiology and the bioturbation activity of the mud shrimp *Upogebia pusilla*. We performed a 14-days *ex-situ* experiment evaluating the influence of Cd and/or parasite on the mud shrimp *U. pusilla*. Cadmium bioaccumulation and potential genetic responses to stress exposure were determined after 3, 7 and 14 days to trace metal exposition. The influence of both stressors on *U. pusilla* bioturbation activity was determined by evaluating sediment reworking rates of the mud shrimp after 3, 7 and 14 days to trace metal exposure.

492

Integrating ecotoxicology and ecology to advance understanding and prediction in multiple stressor research

R. Schaefer, University Koblenz Landau / Institute for Environmental Sciences; J. Piggott, Trinity College Dublin / Zoology

Global environmental change is driven by multiple anthropogenic stressors. Conservation and restoration requires understanding the individual and joint action of these stressors to evaluate and prioritize management measures. To date, most studies on multiple stressor effects have sought to identify potential stressor interactions, defined as deviations from null models, and related meta-analyses have focused on quantifying the relative proportion of stressor interactions across studies. These studies have provided valuable insights about the complexity of multiple stressor effects, but remain largely devoid of a theoretical framework for prediction of effects and null model selection. We suggest that multiple stressor research would benefit by 1) integrating additional null models from ecotoxicology and 2) selecting null models based on their mechanistic assumptions of the stressor mode of action and organism sensitivities as well as stressor-effect relationships. We present a range of null models and outline their underlying assumptions and application in multiple stressor research. Moving beyond mere description requires multiple stressor research to shift its focus from identifying statistically significant interactions to the use and development of mechanistic (null) models. We discuss how ecotoxicological and ecological concepts will aid in achieving this.

Improving the Quality of Ecotoxicological Testing and Assessment

493

Updating the OECD Guidance Document 23 on aquatic toxicity testing of difficult substances and mixtures to include state-of-the-science approaches

W.S. Hunter, U.S. Food and Drug Administration / Center for Veterinary Medicine; G. Stoddart, C. Faßbender, PETA International Science Consortium Ltd.; M. Halder, European Commission Joint Research Centre / DG Joint Research Centre IHCP EURL ECVAM; E. Salinas, BASF SE / Eperimental Toxicology and Ecology

The Organisation for Economic Cooperation and Development (OECD) Guidance Document (GD) on Aquatic Toxicity Testing of Difficult Substances and Mixtures (GD 23) was first published in 2000 and provides crucial guidance that supplements OECD Test Guidelines. Since its release much experience has been gained in handling difficult-to-test chemicals in aquatic exposures as well as progress made in developing methods for testing difficult test chemicals. The GD was revised as recently as 2016 to include state-of-the-science approaches. We provide an overview of the updated GD 23. One significant revision was the expansion of the guidance on testing of poorly water soluble test chemicals. Attention was paid to updating exposure methods that do not employ a solvent in order to eliminate the need for a solvent control, and thus, reducing the number of animals used in aquatic toxicity tests. Another major revision was the addition of more detailed guidance for substances of unknown or variable composition, complex reaction products, and biological materials (UVCBs). The presentation also briefly describes other aspects of the updated GD of interest to those involved in aquatic toxicity testing. The updated GD 23 will help government agencies, industry, and contract research organisations conduct valid and reliable aquatic toxicity studies on difficult-to-test chemicals while minimising both the number of animals used and the need to repeat studies. The views, conclusions and recommendations expressed in this presentation are those of the authors and do not necessarily represent the policies or positions of the United States Food and Drug Administration, the PETA International Science Consortium Ltd., the International Council on Animal Protection in OECD programmes, the European Commission or the OECD.

494

Calibrating Non-Target Arthropod (NTA) Lower Tier Assessment Factors

F.M. Bakker, Eurofins-Mitox; S. Aldershof, Bioresearch and Evaluation; A. Dinter, Cheminova Deutschland GmbH & Co. KG / Global Regulatory Sciences; C. Elston, Syngenta Ltd; C. Mayer, BASF SE / Ecotoxicology; E. Pilling, Dow Agrosciences / REgulatory Sciences; G. Weyman, ADAMA; P. Neumann, Bayer Ag

Assessment factors for Tier 1 and Tier 2 non-target arthropod (NTA) studies were calibrated against NTA full fauna field studies for a large array of species, 20 active ingredients (23 products) and wide geographic coverage in the EU. It was investigated whether the current assessment endpoints at lower testing tiers are sufficiently protective and whether the array of test systems is sufficiently comprehensive. Lower tier studies, both Tier 1 and Tier 2, with several test species were available for calibration against 46 field studies. Both in-field and off-field studies were included. For each product a Hazard Quotient (HQ) was calculated based on the most sensitive lower tier test result, both lethal and sublethal (only tier 2), and the test rate applied in the field study against which the HQ was calibrated. Thus, multi-rate studies could yield more than one HQ. Values obtained were related to the longest duration of adverse effect observed in the field. With this information we derived limit values for assessment factors based on different risk tolerance criteria, for example in-field recovery periods varying from 1 to 12 months. Phytophagous taxa were analyzed separately, but as no differences in outcome with other taxa were observed, these were considered jointly. As expected Tier 1 studies had the most sensitive endpoint and consequently the largest HQ. Using the recovery endpoint, it was found that for the off-field HQ's of 1, 6 and 250 delimited recovery ranges of 0 weeks (no effects), 4 weeks and 8 weeks, respectively in the off-field situation (hay meadow paradigm). For the in-field situation recovery intervals of 0-1, 1-2, 2-6, 6-12 and >12 months were delimited by HQ-values of 40, 375, 620 and 2590. Tier 2 studies could have lethal and sublethal endpoints. Using the most sensitive of the two and including a Vegetation Distribution Factor (VDF) of 5 the following HQ-values were derived for the off-field: HQ=1.7 for a no effect level. These HQ's also correspond to 1 and 2 month recovery periods. For the in-field, using VDF=1, HQ's are 1.3 for no-effect and 6.6, 15, 60 and 560 for 1, 2, 6 and 12 months respectively.

495

The unforeseen consequences for animal welfare of the OECD TG 240 (MEOGRT) biological validity criteria

E. Salinas, BASF SE / Eperimental Toxicology and Ecology; L. Weltje, BASF SE / Crop Protection Ecotoxicology

The Medaka Extended One Generation Reproduction Test (MEOGRT) was established in 2015 as OECD test guideline (TG) 240; a level 5 investigation under the OECD conceptual framework for endocrine disruption assessment. The MEOGRT brings together all aspects of the OECD chronic fish TGs 210, 234, and 229 into one test. OECD TGs include validity criteria as minimum standards for acceptable performance and particularly the biological control performance criteria are critical to assure relevant effects of a test chemical are detectable. Validity criteria assist regulators in determining study quality and reliability; studies that do not comply may be rejected and/or repeated. Compared to other fish TGs, the quantity of validity criteria in the MEOGRT TG have increased and are more stringent, thus elevating the potential for failure and repetition. Other investigators have already noted a high incidence of study repetition following well established OECD fish TGs. However, little data is available for the MEOGRT as currently very few laboratories can implement this highly complex TG. The MEOGRT arose from an international validation effort and recently the data from 9 validation studies were published. We compared control performance in those studies against the existing MEOGRT validity criteria to evaluate the compliance rate. Only 3 studies reported the control parameters corresponding to all biological control criteria and only 1 out of the 9 studies demonstrated successful compliance. The most prevalent deviation from the validity criteria was in the fecundity performance (4 out of 9 studies). Although some deviations from the validity criteria were minor, the failure to meet the fecundity criterion cannot be dismissed in a reproduction test where high fecundity is directly related to statistical power. The MEOGRT fecundity validity criterion is in principle achievable; however, given the available data, nearly 50% of all studies will need at least one repetition to meet this standard. The high likelihood of study repetition and consequent excessive vertebrate use must be considered when conducting a MEOGRT. The biology of the medaka has advantages, but also poses large hurdles to achieving reliable and valid test results. Therefore, alternative species and/or study designs should be considered to generate the data required to address protection goals, which also reduce the potential for excessive animal use.

496

Variability in Non-Target Terrestrial Plant Studies Should Inform Endpoint Selection

J.P. Staveley, Exponent; J.W. Green, DuPont / Data Science and Informatics; J. Nusz, Exponent, Inc.; D.E. Edwards, BASF Corporation / Ecotoxicology; K. Henry, NovaSource / Tessenderlo Kerley, Inc. / Ecological Sciences; M.E. Kern, Waterborne Environmental, Inc. / Ecotoxicology Risk Assessment; A. Deines, Exponent; R. Brain, Syngenta Crop Protection, Inc. / Department of Environmental Risk Characterization; B. Glenn, Bayer CropScience / graduate student; N. Ehresman, Nufarm; T.S. Kung, FMC Corporation/Global Regulatory Sciences / Global Regulatory Sciences, Department of Biochemistry and Microbiology; F. Kee, FMC Corporation; K. Ralston-Hooper, Dow Agrosciences; S. McMaster, Industry Task Force II on 2,4-D Research Data

Inherent variability in Non-Target Terrestrial Plant (NTTP) guideline testing of pesticides creates challenges for using and interpreting these data for risk assessment. Standardized NTTP testing protocols were initially designed to calculate the application rate causing a 25% effect (ER25, used in the U.S.) or a 50% effect (ER50, used in Europe) for various growth measures based on an observed dose-response relationship. The requirement to generate a no-observed-effect rate (NOER), or, in the absence of a NOER, the rate causing a 5% effect (ER05), from these studies has raised questions about the inherent variability in, and statistical detectability of, these tests. Statistically significant differences observed between test and control groups may be a product of inherent variability and may not represent biological relevance. Attempting to derive an ER05 and the associated risk assessment conclusions drawn from these values can overestimate risk. To address these concerns, we evaluated historical data from approximately 100 seedling emergence and vegetative vigor guideline studies on pesticides to assess the variability of control results across studies for each plant species, examine potential causes for the variation in control results, and define the percent effect that can be estimated or the minimum percent effect that can be reliably detected statistically. The results indicate that with current test design and implementation, the ER05 cannot be reliably estimated.

497

An avian reproduction study historical control database: A tool for data interpretation

J. Wheeler, Dow Agrosciences; P. Valverde-Garcia, Dow Agro Sciences LLC; T.A. Springer, EAG Laboratories / Specialist Projects & Histology; V.J. Kramer, Dow AgroSciences LLC / Ecotoxicology; M. Foudoulakis, Dow Agrosciences / RSRA ERS; I. Barber, Dow Agrosciences

Avian reproduction studies are a regulatory requirement for pesticides in many regions. The data often require careful interpretation due to the nature of the study design. Here we present the historical control dataset for bobwhite quail and mallard duck reproduction studies performed at the Evans Analytical Group LLC avian toxicology laboratory over the period 1985 - 2016. The analysis demonstrates the stability of reproductive parameters over time and good agreement to normal control ranges as required by the regulatory test guidelines. The major source of variation is shown to be within study variation. Recommendations for the use of historical control data for the interpretation of avian reproduction studies are made. We believe the analysis and evaluation presented here can facilitate the development of practical guidance that can be implemented in regulatory programmes requiring this test.

498

Experimental Design and Model Selection for Ecotox Risk Assessment

J.W. Green, DuPont / Data Science and Informatics

Recent experience with regulatory requests for re-analysis of older studies using newer statistical methodology has resurrected an old statistical issue of designing a study to fit its objectives and the dangers of imposing a statistical structure on data not fit for purpose. There is a continual need to update statistical methodology as new ideas arise and software to implement these methods become available. Problems can arise when new methods are imposed on old experimental designs. Imagine buying a plot of land with a small cottage. If we tear down the cottage, but leave the cellar and foundation, and then build a mansion in its place but based on the existing foundation, the resulting structure can be unstable and severely restricted in functionality. This presentation will explore the relationship between experimental design and the type of statistical model that can be fit to the resulting data and endpoints that can be estimated or determined from the model. In some instances, newer methods can be applied without problem to existing data. In other cases, existing data cannot support newer methods. It is important to understand the data requirements of the methods or models we intend to use. The size effect that can be estimated or detected is critically important and is strongly related to experimental design and biological variability. There is a model underlying every statistical test used to derive a NOEC or estimate an ECx. The basic statistical model for a simple toxicity experiment is given by $Y_{ij}=\mu_i + e_{ij}$, where μ_i is the expected mean response in the i^{th} concentration, and the e_{ij} are independent random errors, usually assumed to be identically distributed. What distinguishes one model from another are what distribution is assumed for the errors or responses and what restrictions or assumptions are placed on the treatment means, μ_i. It is possible to determine the size effect that can be estimated or detected from a given dataset and it depends largely on experimental design and response variability. Statistical models used for hypothesis testing or regression estimates have data requirements. Model assessment tools are well established and should be used in fitting models to ecotoxicity data. Ignoring these tools or model requirements can lead to poorly estimated effects and misleading results. Understanding these concepts enables the scientist to make sound assessments of the data collected.

LCA and beyond - integrating sustainability and/or other dimensions to improve decision support (II)

499

Integration of Risk Assessment and Life Cycle Assessment in the context of recycling wood waste into particleboard

S. Huysveld, R.A. Alvarenga, J. Dewulf, Ghent University / Department of Sustainable Organic Chemistry and Technology

Recycling of wood waste into particleboard has some environmental advantages, for instance, by giving second life to wood waste, (i) the use of fresh wood is avoided and (ii) it allows an extended carbon storage, which is beneficial in terms of climate change. However, wood waste sources have a varying chemical quality, therefore, recycling of post-consumer wood waste requires special attention to the potential presence and downstream fate of contaminants. Demolition wood waste, for example, may contain high concentrations of heavy metals such as arsenic and copper. Heavy metal toxicity is a threat to the environment and is associated with adverse health effects. In the particleboard industry, heavy metals may be discharged into the air when dust from wood waste is incinerated to supply heat for dryers. Moreover, downstream industrial customers of particleboard (e.g. furniture manufacturers), who incinerate cuttings and dust from particleboards for internal heat supply, are of concern as well. Local human health effects due to reduced local air quality may question the overall benefit of recycling contaminated wood waste into particleboard. A need exists to investigate the local human health risks associated with recycling contaminated wood waste, while simultaneously considering other impacts on human health and the environment throughout the entire life cycle traditionally modelled with Life Cycle Assessment (LCA). The objective of this study was the combined use of local Risk Assessment (RA) and LCA to achieve a broader assessment of the sustainability of recycling contaminated wood waste into particleboard. The current scenario, in which the use of contaminated wood waste in particleboard is limited by Flemish government's standards, is compared to a future scenario with a higher use of more contaminated wood waste. As a consequence, in the future scenario, a lower proportion of the contaminated wood waste will be incinerated with electricity (and heat) recovery. Modeling of the local air pollution is performed with the Immission Prognosis Air Concentration Tool (IMPACT) of the Flemish government. The LCA scope includes the particleboard industry, relevant upstream and downstream processes of the particleboard industry, and the incineration of wood waste. To integrate RA and LCA results on human health effects the concept of disability-adjusted life years (DALYs) is adequate as a common metric. Results will be presented at the conference.

500

Development of non-conventional LCA indicators for circular characteristics of bio-based products

K. Lokesh, University of York / Department of Chemistry; X. Bengoa, Quantis; S. Majer, DBFZ, Deutsches Biomasseforschungszentrum gemeinnützige GmbH / Bioenergy Systems; A. Ernstoff, F. Sessa, V. Rossi, Quantis

This paper is dedicated to the identification of key "un-conventional" indicators that demonstrate the sustainability and circular characteristics of promising bio-based products, complementing conventional life cycle analysis. Some of the new LCA complementary indicators proposed as a part of this study emphasise on resource efficiency and material circularity of bio-based value chain and include (but not restricted to) waste circularity, critical material circularity, land-use efficiency and output quality services. The proposed indicators were drawn out of a comprehensive evaluation of more than 45 certification labels, schemes and initiatives associated to bio-based products, relevant international and national standards and 80+ scientific articles encompassing bio-product life cycle assessment. A current need for a harmonised sustainability certification protocol, coupled with an aim to develop an indicator-led assessment framework lead to the identification of potential gaps in criteria and indicators. Adoption of bio-based products has been identified as the one of the pathways to reach a sustainable economy. Some of the many advantages conceived from adopting bio-based value chains include development of waste-management infrastructure, job creation, SME's and other environmental opportunities, contributing directly to seven out of 17 *UN Sustainable Development Goals*. The benefits of such systems approach can be realised only via quantitative and qualitative evaluation of the embedded environmental, techno-economic and societal impacts, all of which are a function of a product's variables like feedstock type, technology-route, product's functionality and application [1]. Life cycle assessment, a robust impact-led sustainability analysis tool that enables one envisage these impacts via use of holistic indicator also has insufficiencies, mainly the limitations in addressing the circular product characteristics. This work is a part of the EU-H2020 funded project, Sustainable Transition Assessment and Research of Bio-based products, the ultimate aim of which is to expand existing tools and methodologies for sustainability certification of bio-based products and for their speedy commercial uptake.

501

Toward a more sustainable biochemical industry - Early stage assessments and methodological overlaps between life cycle- and techno-economic assessments of biochemicals

. Ãgmundarson, DTU (Technical University of Denmark) / Division for Quantitative Sustainability Assessment DTU Management Engineering and DTU Biosustain; S. Sukumara, DTU Technical University of Denmark / DTU Biosustain; P. Fantke, Technical University of Denmark / Quantitative Sustainability Assessment Division

Existing Life cycle assessment (LCA) studies of biochemicals reveal that there are challenges that need to be overcome in order to reach an overall high sustainability performance. While in some cases biochemicals have lower global warming impacts compared to fossil-based chemicals, other impacts may become higher, like eutrophication. One of the major sources of environmental impacts of biochemicals is the growing of biomass, which in most cases today is corn. This has led to investment in assessing opportunities of using side streams, like leftover agricultural lignocellulosic biomass, or non-cultivated biomass like algae. Macro-algae is one such potential source given that they grow without being farmed, while simultaneously being an important sink for CO2. The objective of this study is to identify trade-offs between assessed environmental impacts and possible burden shifting between macro-algae compared to more conventional feedstockıs like maize and lignocellulose. While it is imperative that any change in process configuration reflects in Techno-Economic assessment (TEA) and LCA, there are very few studies which couples these two assessment demonstrating the trade-offs for improved decision support. The focus of this contribution is to explore methodological overlap between the two assessments and develop a framework, supported by a proof-of-concept. When contrasting current results from the TEA and LCA cradle-to-gate study, some interesting trends were observed. The TEA show that itďıs biggest hot-spots are identified as feedstock cost which is a function of growing, transportation of biomass and if drying is taking place at the refinery site or closer to the harvesting sites of the feedstock. Whereas, the LCA shows the biggest environmental hot-spots occur in relations to growing of biomass, if it requires external application of nutrients and intensity of chemical pretreatment. Today decisions on if chemicals are further developed companies mostly rely of results from TEAs. Our results show that the methodological overlap between TEA and LCA are of that magnitude that justifies the appraisal of this integrated methodology. Introducing LCA as a decision support tool would integrate sustainability requirements in development of technology and solutions. All technologies and products have a life cycle, and by analyzing their impacts, we put numbers on sustainability and benchmark the solutions.

502

A risk evaluation approach for indirect land use change associated to biobased products

D. Marazza, University of Bologna; S. Majer, DBFZ, Deutsches Biomasseforschungszentrum gemeinnützige GmbH / Bioenergy Systems; V. Rossi, Quantis; J. Golaszewski, UniwersytetWarminskoMazurski W Olsztynie

Biobased products include a vast range of traditional and innovative materials and substances for purposes other than food and energy such as wood and composite materials, bio-plastics, adhesives, lubricants, paints and many other material categories feeding large economic activities. There is international recognition that developing a climate-smart bio-based economy is essential to the continuation of economic development, reduction of greenhouse gas emissions, and adaptation to climatic change. However, as biobased products are ultimately obtained from land or sea, a specific attention has to be payed when considering additional exploitation. Changes of land/sea uses can rebound and cancel out environmental performances and the original purpose of sustainability. Indirect land use change (ILUC) has been defined as an unintentional, negative, displacement effect of commodities in the primary sector such as agriculture causing additional land use changes. Provided that ILUC depends on specific legacy effects stemming from land condition prior and after land use changes, overall indirect effects are connected to the 1.1 billion tons of greenhouse gases per year generated because of land use changes. However the application of ILUC provisions as for biofuels has been and stays controversial. The Project STAR-ProBio is a multi-actor collaborative research and innovation action and supports the European Commission in the full implementation of European policy initiatives, including the Lead Market Initiative in bio-based products, the industrial policy and the European Bio-economy Strategy. One of the specific goals calls for identifying and mitigating the risks of negative ILUC effects associated to production routes for bio-based products. In this contribution the authors present the conceptual model and the results of the identification of risk factors obtained from the analysis of economic models and a sensitivity analysis performed over one selected case study.

503

How to find sustainable applications for new materials and how to overcome the relativity of LCA

C. Som, EMPA Technology & Society Lab; R. Hischier, EMPA / Technology and Society Lab; F. Piccinno, EMPA

The Multi-Perspective Material Selection (MPAS) method has been developed as a decision support tool to identify the most sustainable application fields for new materials that are still under development. This selection includes a 3-step method considering technical, economic as well as environmental criteria. So far, the method was best used for new materials that are replacing existing materials in a given application. But applying the MPAS in the case of a completely new kind of material or application field with no clear and existing competitor for comparison, the method revealed its limitations. Especially, the environmental assessment, that uses simplified LCA studies, is a relative approach. Another difficulty for the simplified LCA studies is when the production data of the material and, at the same time, the knowledge about the properties of the end-product are unknown and highly speculative. This is a common problem since the MPAS method is intended to support decision making at an early development stage. Here, we present an expanded and further developed MPAS method that mitigates exactly these limitations meaning that the environmental assessment can be perfomed without a comparison case and also without the necessity of a lot of data. The development and expansions of the MPAS method are applied to each of its three steps. However, the main development of the method is made to Step 3, the environmental evaluation of the material. Our solution here uses a highly flexible set of criteria that are specifically adapted to the various cases and that are mainly LCA based. This means that the environmental score can now be obtained regardless of the ability to estimate the production data of the material and of the knowledge about the exact properties of the end-product. This evaluation can be applied absolutely or relatively/comparatively. Furthermore, the criteria are expanded with other criteria that go beyond only LCA relevant aspects and also include aspects like circular economy. The method is illustrated with a case study on nanoporous carbonaceous material. As a result, the most sustainable applications for this nanoporous carbonaceous material are identified and used to set parameters to be achieved for the developers of the material. The new independent environmental assessment part in Step 3 overcomes the necessity of a comparison case while also reducing of the required amount of LCA data. This makes the method universally applicable.

504
Consumption and consumer footprint: LCA as pivotal methodology for assessing consumption patterns and ecoinnovations
S. Sala, A. Cerutti, European Commission Joint Research Centre / Bioeconomy unit; V. Castellani, EC-JRC; M. Secchi, European Commission Joint Research Centre / Bioeconomy unit
The European Commission has been developing an assessment framework to monitor the evolution of environmental impacts associated to the EU consumption. The assessment framework aims at supporting a wide array of policies, such as those related to bioeconomy, resource efficiency, ecoinnovation and circular economy. The assessment framework is composed of two sets of consumption-based indicators: the Consumption footprint and the Consumer footprint. The Consumption footprint assesses the potential environmental impact of apparent consumption, focusing on a territorial scale and accounting for trade, assigning the impact to the country where the final consumer is located. The Consumer footprint assesses the potential environmental impact of consumption, based on the results of life cycle assessment (LCA) of representative products purchased and used in one year by an EU citizen. The Consumer footprint allow assessing environmental impacts along the products life cycle (raw material extraction, production, use phase, re-use/recycling and disposal). For the calculation of the Consumer footprint, the consumption of European citizens is split into five key areas (food, housing, mobility, household goods and electric/electronic appliances). For each area, a respective Basket of representative Products (BoP) has been built based on statistics on consumption and stock of product. For each of the five BoPs, a baseline scenario is defined, taking as reference the consumption of an average EU citizen in the baseline year 2010. For this purpose, more than 100 LCAs have been conducted. The baseline models allow for identifying the environmental hotspots along the product lifecycle and within the consumption area of each specific BoP. The results of the hotspot analysis are then used as a basis for the selection of actions towards environmental burden reduction, covering either consumption pattern, behavioral changes, implementation of eco-solutions, or a combination of the previous. For each of the actions, a scenario has been developed, by acting on the baseline model and simulating the changes associated to the specific intervention. The LCA results of each scenario are then compared to the results of the baseline, to identify potential benefits or impacts coming from the implementation of the solution tested, as well as to unveil possible trade-offs.

Environmental Risk Assessment in Sediments

505
Assessment of risk from historic contaminants in sediments of the Elbe flood plain, using a multiple line of evidence approach
S. Heise, Hamburg University of Applied Sciences / Life Sciences; U. Rieth, Institut für Hygiene und Umwelt
The Elbe river has one of the largest catchment areas in Europe (150 000 km^2) of which two thirds lie in Germany. While it was once considered to be among the most polluted rivers in Europe, water quality has largely improved since the fall of the Iron Curtain in 1989. Sediments, however, still carry the memory of an industrial past and are often “on the move”, transported by the current and especially during flood events to downstream sites. The question, where they originate, what chemicals they carry and how much of it may still be around, has been in the focus of several previous studies (e.g. Heise et al. 2008, Hillebrand et al. 2015). Little attention, however, had been paid for a long time to sediments in those ca. 1000 backwaters and flood plain lakes along the Elbe with regard to their contamination, their ecotoxicity and their mobility in times of high water discharges and flood plain submergence. Two studies, carried out in 2013 and 2014, were dedicated to this kind of structures with the task of evaluating a potential risk from these sites. Over a stretch of 230 km along the Middle Elbe, sediments from 25 backwaters were sampled, analyzed for heavy metals and for Elbe-typical historic contaminants (HCH, HCB, PCB, PAHs, DDX). Additional lines of evidence in an assessment of risk comprised the thickness of the sediment layer and sediment mobility during flood events. Dating of sediment cores by ^{137}Cs analyses facilitated interpretation of the results. Samples were ecotoxicologically tested for inhibitory effects in the bacterial sediment contact test (*Arthrobacter globiformis*), the luminescence bacteria test (*Allivibrio fischeri*) with elutriates and methanol extracts and the algae growth inhibition test (*Raphidocelis subcapitata*) with elutriates. The studies showed that - more than 75 % of all sampled sites were contaminated with heavy metals and organics well beyond the threshold values of the Elbe River Commission. - ecotoxicological effects provided a distinct line of evidence and could not be simply related to analyzed contaminant concentrations. - when integrating chemical, ecotoxicological and erosion stability data into a weight of evidence approach, high risks could be identified for 50 % of the sampled sites in 2013. - dating of sediment cores from 2014 pointed towards a strong impact of the 4 extreme flood events between 2002 and 2013 on the erosion of highly contaminated sediments from backwaters into the Elbe river.

506
Multiple lines of evidence for risk assessment of old sea deposits for ilmenite mine tailings in SW Norway
M. Schaanning, H. Trannum, K. Ndungu, S. Øxnevad, NIVA Norwegian Institute for Water Research
Annually this mine disposes up to 3 mill. tons of tailings contaminated with trace amounts of Ni and Cu sulfides. During 1960-94 the tailings were placed in two sea deposits, first in a sheltered fjord and then in a more exposed basin. After 1994 the tailings have been placed in a land-deposit. To protect the downstream watershed area, some of the metal contaminated drainage water is recycled, mixed with other discharge and fed into the water column at the site of the fjord deposit. In 2015 the deposit and reference sites were sampled for studies of macrobenthic communities, biogeochemical fluxes, metal mobilization and metal uptake in gastropods and DGT probes. O_2 and pH in the sediments were measured using micro-electrodes. The tailings were easily traced in the sediments by high concentrations of fine fractions, Fe, Ni, Cu and Co. Tailings were still abundant in the top 0-1 cm of the sediments at both deposit sites, but clearly less abundant at the sediment surface than in deep deposit layers and also less in the outer basin deposit than in the fjord deposit. Compared to Norwegian and European quality standards [1], Cu exceeded MAC-EQS (“Maximum Admissible Concentration”) for coastal sediments indicating a “risk of acute toxic effect” on marine organisms. The DGT-profiles showed that Fe and Mn was recycled within the sediments, whereas Ni and Cu leaked to the overlying water from mobilization centers consistently located in the oxidising environment between 5 and 20 mm depth. The total leakage of Ni from the sea deposits corresponded to 5% of the leakage from the land deposit and 8% of the current discharge to the water-column at the fjord deposit site. Organic carbon (TOC) and fluxes of O_2 and nutrient species were low throughout the investigated area, and the macrobenthic communities showed reduced number of species at the inner fjord site, but ecological status was classified as “good” at all sites and “very good” at the reference station. The multivariate statistical test (DistLM marginal test) showed that in addition to depth, fine fractions(< 63 μm) and Cu were the only significant environmental parameters explaining the variance in the benthic community data. We conclude that both the current discharge to the water column and the leaching of Cu and Ni from the sea deposits are likely to contribute to the moderate reduction of benthic biodiversity at the old deposit sites. [1] Guideline M-608, 2016. Norwegian Environmental Agency. 24 pp.

507
In situ metal fluxes for the assessment of metal bioavailability in sediments
E.D. Amato, University of Antwerp / Department of Biology; S.L. Simpson, CSIRO Land and Water / Centre for Environmental Contaminants Research; D.F. Jolley, University of Wollongong / School of Chemistry; C.P. Marasinghe Wadige, University of Canberra / Institute for Applied Ecology; W.A. Maher, University of Canberra / Institute of Applied Ecology; A. Taylor, University of Canberra / Ecochemistry Laboratory, Institute for Applied Ecology
Analysis of pore waters, dilute-acid extractable metal (AEM), acid volatile sulfide (AVS), and organic carbon (OC) concentrations are frequently used to evaluate and predict metal bioavailability in sediments.[1] Where concentrations of bioavailable contaminants are determined to exceed sediment quality guideline levels, bioassays are usually performed to evaluate toxicity effects resulting from contaminant exposure. Although the chemical analyses used for bioavailability assessment have been shown to be useful for predicting metal toxicity in sediments,[1] the predictions for more oxidized surface sediments can be quite poor, frequently owing to a broader range of factors influencing metal bioavailability including variability in phases that are easily oxidized or reduced (e.g. AVS and Fe(II)). In addition, laboratory-based bioassays may provide inadequate predictions of metal bioavailability and toxicity due to their inability to adequately replicate field exposure conditions. A comprehensive series of studies combining laboratory and field experiments were carried out to evaluate the performance of the diffusive gradients in thin films (DGT) technique for predicting metal bioavailability in sediments.[2] The DGT device uses an ion-exchange resin (Chelex) which selectively accumulates divalent metal present in the sediment porewater and weakly-bound to the sediment particulate phase.[3] The DGT metal flux measured at the sediment water interface (SWI) was compared to biological responses of organisms exposed to sediments contaminated with mixtures of metals, in the laboratory (amphipods and bivalves) and in the field (bivalves). To assist in the analysis of effects from the mixtures of the metals (Cd, Cu, Ni, Pb, Zn), DGT metal fluxes were normalised using water quality guideline values to account for predicted differences in the toxicity of the different metals. Strong dose-response relationships were found between the normalised DGT metal flux measured at the SWI (±0.5 cm) and adverse effects to reproducion and survival of the amphipod exposed to laboratory conditions. Useful predictions of bioaccumulation were obtained for both marine and freshwater bivalves in laboratory and field set-ups. Differences in bioaccumulation between organisms exposed to identical sediments in laboratory and field set-ups highlighted the importance of including in sediment quality assessments lines of evidence based on *in situ* evaluations of metal bioavailability.

508
An Overview of the Refinements and Improvements to the USEPA's Sediment

Toxicity Methods for Freshwater Sediment

T.J. Norberg-King, U.S. EPA / NHEERL/Mid-Continent Ecology Division; J. Hockett, U.S. EPA-Duluth, MN / ORD NHEERL Mid-Continent Ecology Division-Duluth, MN; C. Ingersoll, Retired; D.R. Mount, USEPA-Duluth, MN / ORD, NHEERL, Mid-Continent Ecology Division-Duluth, MN

Sediment toxicity tests are used for contaminated sediments, chemical registration, and water quality criteria evaluations and can be a core component of ecological risk assessments at contaminated sediments sites. Standard methods for conducting sediment toxicity tests have been established by USEPA, ASTM, Environment Canada and OECD. Revisions to USEPA's *Methods for Measuring the Toxicity and Bioaccumulation of Sediment-Associated Contaminants with Freshwater Invertebrates* is planned for 2018. USEPA's manual describes toxicity and bioaccumulation testing of freshwater sediments with 3 freshwater species, *Hyalella azteca* (amphipod), *Chironomus dilutus* (midge) and *Lumbriculus variegatus* (oligochaete) and 5 sediment toxicity test methods: 10-d tests with *H. azteca* and *C. dilutus*; a 42-d life-cycle test with *H. azteca*; a 50-d life-cycle test with *C. dilutus* and a 28-d bioaccumulation test with *L. variegatus*. While laboratories routinely met test acceptability criteria (TAC) for short-term sediment toxicity exposures (10-d control survival and ash-free-dry weight), laboratories reported variable biological performance with the longer exposures. With input from both midge and amphipod workgroups, each test method has updated guidance for the starting size/age of organisms, diets and rations, reconstituted water changes, modifications to the test acceptability control survival, weight and other endpoints. Control waters needs to have a minimum level of chloride and bromide and use of a sand substrate control to evaluate water and diet used in sediment testing included. While designed for sediment evaluations, the methods are readily adapted for waterborne exposures. For the oligochaete bioaccumulation test, the recommendation for the maximum loading rate is increased to 1 g dry tissue:100 g of sediment organic carbon (previously 1:50). Use of laboratory proficiency criteria is incorporated in addition to test acceptability criteria for individual tests for each species and method, laboratories would report their averages for the test endpoints/measures. These proficiency criteria would not be used to accept or reject individual tests, but serve as a broad indicator of laboratory performance and possibly provide insight where refinements are needed. In this talk, we will focus on the testing methods and improvements that have been made in each method for USEPA and ASTM methods. *This abstract does not necessarily reflect the views or the policies of the USEPA.*\n

509

Sediment-spiked toxicity tests with benthic macro-invertebrates and the fungicide fludioxonil

I. Roessink, Alterra / Environmental Risk Assessment; J. Romão, University of Aveiro; X.H. Yin, Zhe Jiang Agriculture and Forestry University; T. Brock, Alterra, Wageningen University and Research Centre / Environmental Risk Assessment Team

In the EFSA scientific opinion on sediments, one of the oligochaete worms *Lumbriculus* spp. or *Tubifex tubifex*, supplemented with a second standard test species of a different taxonomic group, such as the midge *Chironomus riparius* or the amphipod *Hyalella azteca*, are proposed as Tier-1 test organisms for fungicides with a biocidal mode-of-action. To investigate i) the potential difference between the use of field-collected and artificial sediments and ii) whether the proposed Tier-1 approach is protective, 28-d tests with fludioxonil-spiked field-collected and artificial sediment were performed with all sediment-dwelling invertebrate taxa mentioned above. In a previously performed spiked-sediment outdoor microcosm experiment with fludioxonil the most sensitive sediment-dwelling organism was the oligochaete *Dero digitata*. The test systems used were 1.5L glass vessels containing approximately 2 cm sediment and 1 L aerated spring water. Tests were considered valid if control mortality did not exceed 20% or, in the case of the larvae of the midge *C. riparius*, if more than 70% emergence had occurred. Fludioxonil concentrations were measured at the start and end of the testing. Endpoints for both worms comprised surviving animals and their weight, including yield and growth rate. For *Hyalella* both survival, weight and length were assessed, while for *Chironomus* emergence and total survival was monitored. All tests met the validity criteria of less than 20 percent control mortality or more than 70 percent emergence, with the exception of the *C. riparius* test on artificial sediment. Overall, tests with field-collected sediment gave more sensitive responses. Apparently growth of the animals was better here, allowing for a better expression of effects. Sometimes a factor of 10 difference between the estimated 28d-EC_{10} value and its upper or lower confidence limit was present, indicating a high associated uncertainty. The confidence intervals were considerably smaller in corresponding 28d-EC_{50} values, indicating that these were more reliable. In both lab as the outdoor microcosm testing the most sensitive organism was an oligochaete. In general, biomass reacted more sensitive than length and survival endpoints. Using either field-collected or artificial sediment data to derive a Tier-1 RAC_{sed} seems to be suffiently protective when compared to the outdoor microcosm response.

510

Spatio-temporal exposure of Plant Protection Products in OECD 219 sediment test systems - Comparison of model results with measurements

K. Hammel, Bayer AG, Crop Science Division / Environmental Safety; A. Dorn, Hochschule Niederrhein / Department of Chemistry; P. Dalkmann, Bayer AG Crop Science Division; E. Bruns, Bayer AG, Division Bayer CropScience / Ecotoxicology; D. Faber, Bayer AG, Crop Science Division / BCS D ETX Ecotoxicology

Sediment toxicity testing among other ecotoxicologic tests is currently revised under the premise to improve quality and consistency of regulatory environmental risk assessment. In 2015, the European Food Safety Authority (EFSA) has published a scientific opinion on environmental risk assessment for sediment organisms where a water-spiked test system (OECD 219) is considered to study chronic effects on sediment organisms. Prominent test organisms are Chironomids, aquatic insects which live in and on soft sediments. Due to the design of this study initially large gradients between the exposure in the overlying water and in the sediment layer are established. As a consequence, substantial temporal and spatial dynamics of local concentrations have to be expected, especially in the vicinity of the interface beween water and sediment where the Chironomids are supposed to stay. To describe local concentrations in such water-sediment test systems we simulated the transport and the redistribution of two moderately mobile (K_{OC} 200 to 300) plant protection products with the mechanistic model TOXSWA. The results of the simulation are compared with measured sediment concentrations in three depths (see contribution submitted by Dorn et al.). The compound properties were parameterised using values derived independently in standard tests ($K_{OC,}$ $DT50_{water/sediment}$) or from literature (diffusion coefficients). Other parameters were derived from OECD 219 experimental design information. The simulations matched the measured concentrations spatially and temporally well. The simulated concentration depth profiles averaged for the layers which were measured lay almost always within the range of single measurements. Also the concentrations in the overlying water were reproduced well. The main findings are that the concentrations in the sediment show a pronounced temporal pattern and that the concentrations in the sediment are strongly depth-dependent. The dominant transport process in the sediment is obviously diffusion which however did not lead to homogeneous penetration of the sediment. Presuming that chironomids live in the upper three millimetres of the sediment, they are exposed to approximately four times higher test compound concentration (total and liquid) than the average concentration in the sediment. This has important consequences for the derivation of effect endpoints of chironomids from OECD 219 study data, which should consider the local exposure of the organisms.

Wastewater effluents: How research can improve risk assessment and regulation

511

Effects of untreated wastewater dilution in surface waters on pharmaceuticals natural attenuation and on the community genomics: Implications for ERA

S. Bagnis, M. Fitzsimons, Plymouth University; J. Snape, AstraZeneca UK Ltd. / AstraZeneca Global Environment; A. Tappin, Plymouth University; S. Comber, Plymouth University / Environmental Science

The increasing consumption and production of active pharmaceutical ingredients (APIs) in low and low-middle income countries (LLMICs) is of growing environmental concern owing to their possible ecotoxicological effects. This is related to the practice of direct discharge of untreated wastewater (DUW), which creates a heavily polluted area, named the “impact zone”. Little is known about the environmental fate of APIs in this area. Nevertheless, a few available measured environmental concentrations (MECs) of LLMICs show higher concentrations than for high-income countries with developed wastewater treatment infrastructures. Globally, the MECs of APIs in the “impact zone” are typically above 0.01 $\mu g\ L^{-1}$, which, if predicted, would trigger the environmental fate refinement of the environmental risk assessment (ERA). In the ERA PEC calculation, a default dilution factor (DF) of 10 is used, but in at least 53 countries worldwide, the local predicted median DF is lower than 10. There is no information available in the literature about the effects of low dilutions on the natural attenuation of APIs nor impacts of DUW. Furthermore, information on the effects of low dilution on mixtures of APIs is missing, hence necessitating the requirement for evaluation of biological endpoints for the impact zone ERA. This information is pivotal for the development of an impact zone ERA approach, and we are proposing an original attempt to expand this area of essential research. The biodegradation of a set of APIs was studied in batch tests at several levels of dilution. Nevirapine shows persistency across the experimental period and only the dilution is controlling the observed concentrations. Acebutolol and Diclofenac show a decrease in concentration of up to 90% as a result of a combination of dilution and biodegradation. The biodegradation at no dilution shows a behaviour consistent with previous reported studies. Also, Amitriptyline shows persistency but the sorption alone is responsible for the 70% of the removal. The TOC analyses do not show significant consumption rate changes caused by dilution. The results regarding the effects of the APIs mixture and the dilution on the microbial composition are been analysed through bioinformatic statistics, and will be presented if significant.

512

Active Pharmaceutical Ingredients Entering the Aquatic Environment From

Wastewater Treatment Works: Measurement, Prediction, Risk - A Cause for Concern?

S. Comber, Plymouth University / Environmental Science; M. Gardner, Atkins Ltd.; P. Sorme, AstraZeneca / AstraZeneca Patient Safety; B. Ellor, UKWIR

This work reports on the ability for wastewater treatment works (WwTW) to remove active pharmaceutical ingredients (APIs), the variations within and between works, the effectiveness of trying to model removal and the risk of exceeding predicted no effect concentrations (PNEC) in the environment. The research is based on data generated from two large UK-wide WwTW monitoring programmes. Taking account of removal of parent compound from the aqueous phase during treatment in combination with estimates of dilution available it is possible to prioritise the APIs of greatest risk of exceeding estimates of their PNEC in receiving waters for all WwTW in the UK. The majority of substances studied were removed to a high degree, although with significant variation, both within and between WwTW. Poorer removal (between influent and effluent) was observed for ethinyloestradiol, diclofenac, propranolol, the macrolide antibiotics, fluoxetine, tamoxifen and carbamazepine. All except the last two of these substances were present in effluents at concentrations higher than their respective estimated PNEC (based on measurement of effluents from 45 WwTW on 20 occasions). The application of models to predict removal efficiencies are reported. Based on available dilution data as many as 890 WwTW in the UK (approximately 13% of all WwTW) may cause exceedances of estimated riverine PNECs after mixing of their effluents with receiving waters. The overall degree of risk is driven by the toxicity value selected, which in itself is controlled by the availability of reliable and relevant ecotoxicological data and consequently the safety factors applied. The dataset and discussion, provides information to assist in the future management of these types of chemicals.

513

Impact of a wastewater treatment plant upgrade on amphipods and other macroinvertebrates: individual and community responses

K. Peschke, Tübingen University / Animal Physiological Ecology; S.A. Kohler, University of Portsmouth / Animal Physiological Ecology; K. Wurm, GÖL Water Ecology Laboratory Starzach; R. Triebskorn, University of Tuebingen / Animal Physiological Ecology

Conventional wastewater treatment plants (WWTPs) equipped with secondary and tertiary treatment steps do not or only partially remove micropollutants which makes them important point sources for the release of these substances in the water cycle. Micropollutants can cause short- or long-term adverse effects in aquatic organisms even at low concentration levels. One possibility to reduce the input of micropollutants into the water cycle is the upgrading of WWTPs with an additional purification stage using e.g. ozonation or powdered activated carbon. The current work is part of the joint research BMBF project "SchussenAktiv*plus*" funded by the German Federal Ministry of Education and Research (BMBF) and the Ministry of Environment, Baden-Württemberg, Germany. In this project, the efficiency of an additional wastewater treatment based on powdered activated carbon for the ecosystem of the Schussen river, a major tributary of Lake Constance, Southern Germany, has been investigated. Our part of the project focuses on assessing the health status of gammarids and the macrozoobenthos community in the Schussen river. Samples were taken up- und downstream of the WWTP, as well as before and after the upgrading of the WWTP. Gammarid populations from all sites were investigated with respect to sex ratio and fecundity of breeding females. In addition, analyses of heat shock protein (hsp70) levels and lipidperoxides allowed us to draw conclusions about proteotoxic and oxidative stress in gammarids. Macrozoobenthos community integrity was determined by means of the saprobic index as well as by the number of sensitive taxa (EPT index). Prior to the WWTP upgrade, the health status of gammarids as well as the integrity of the macrozoobenthos community was negatively influenced by the WWTPs effluent. After the upgrading of the WWTP, gammarids from the downstream site did not differ any longer from those collected upstream of the WWTP with respect to the investigated health parameters. Furthermore, the overall number of taxa and particularly the number of EPT taxa within the macrozoobenthos community downstream of the WWTP increased distinctly after the upgrade of the WWTP with the additional activated carbon step. We conclude that the efficiency of the activated carbon step to eliminate toxic and endocrine active chemicals from the effluent can plausibly be related to the improved integrity of macroinvertebrate health and community structure in the connected river Schussen.

514

Effects of full-scale ozonation of treated effluent - Environmental impact in a receiving river

J. Fick, Umea University / Department of Chemistry; T. Brodin, Umea University / Department of Ecology and Environmental Science; J.D. Larsson, University of Gothenburg, Sweden / Department of Infectious Diseases; L. Norrgren, Swedish University of Agricultural Sciences / Department of Biomedical Sciences and Veterinary Public Health; B. Björlenius, KTH Royal Institute of Technology / Industrial Biotechnology Division; S. Örn, Swedish University of Agricultural Sciences / Department of Biomedical Sciences and Veterinary Public Health; J. Pohl, Swedish University of Agricultural Sciences (SLU) / Department of Biomedical Sciences and Veterinary Public Health; J. Jutkina, C. Flach, A. Karkman, C. Rutgersson, University of Gothenburg Sweden

Pharmaceuticals have been found in aquatic systems globally, due to a combination of worldwide usage and low removal efficiency in wastewater treatment plants (WWTPs), or a complete lack of WWTPs (1). In surface waters, concentrations of pharmaceuticals usually range from low $\mu g\ l^{-1}$ close to point sources to low $ng\ l^{-1}$, and are correlated to human population density in the drainage area, volume of the receiving water body and technologies used in WWTPs. One technique to increase the removal of pharmaceuticals in WWTPs is to add a tertiary treatment step based on the addition of ozone. Ozonation is a cost efficient way to degrade chemicals and several studies have shown that most pharmaceuticals are readily degraded in the presence of ozone (2). However, several oxidized degradation products are formed during ozonation and the environmental impact of these are largely unknown. The aim with this study was to evaluate the removal of pharmaceuticals in a WWTP, when adding ozonation as an additional tertiary treatment step and also to investigate the environmental impact of this effluent on the receiving river. All treated effluent from a minor WWTP (10000 PE) were treated by an addition of 8 $mg\ h^{-1}$ ozone during 6 months. Removal rates in the WWTP as well as levels of pharmaceuticals in the receiving river (both in water and biota) were monitored. Surface water data from 10 sampling sites and 10 sampling occasions, before, during and after ozonation, will be presented. Ecological status and levels of pharmaceuticals in exposed biota (n=5) at each site and sampling occasion will also be presented. Several additional methods to evaluate the impact of ozonation was used including impact on microbial community composition, presence of antibiotic resistance genes as well as studies to detect endocrine, reproductive and behavioral effects in fish and its progeny.

515

Dreissena polymorpha as purifier tool of protozoa in wastewater treatment plant effluent

E. Géba, University of Reims ChampagneArdenne / UMRI SEBIO; D. Aubert, Laboratoire de Parasitologie-Mycologie / EA3800; S. Betoulle, URCA / UMRI SEBIO; O. Dedourge-Geffard, University of Reims Champagne-Ardenne / UMRI SEBIO; D. RIOULT, UMRI 02 INERIS-URCA-ULH SEBIO / MOBICYTE flow cytometry core facility; L. Durand, S. La Carbona, ACTALIA; I. Villena, Laboratoire de Parasitologie-Mycologie / EA 3800; A. Geffard, Université de Reims Champagne Ardenne; A. Bigot-Clivot, University of Reims Champagne-Ardenne / UMRI SEBIO

Aquatic environments are subject to discharges of multiple contaminants (chemical and biological compounds). Wastewater treatment plants (WWTPs) are ineffective to remove environmental forms of protozoa such as *Toxoplasma gondii* and *Cryptosporidium parvum* oocysts or *Giardia duodenalis* cysts because of their resistance to chemical and physical treatments. These protozoa are clearly identified as a public health priority since they are major parasites of waterborne outbreaks. Many studies underline the interest of using of freshwater bivalve *Dreissena polymorpha* for biomonitoring. Indeed, this bivalve has a huge filtration capacity leading to an accumulation of chemical and biological contaminants in its tissues. The DROPPE (The dreissene as purifier tool of protozoa in WWTP effluent) project aims to test the depurative capacity of the zebra mussel in terms of protozoa's contamination in WWTP effluents. To answer of this issue, it's necessary to determine if *D. polymorpha* is able to live in good health in the multi-contaminated conditions in WWTPs effluent and *D. polymoroha* is able under these conditions, to bioaccumulate protozoa. For this purpose, two experiments were performed: 1- Zebra mussels were caged in the WWTP's outlet channel (Charleville-Mézières, France) for 28 days. We studied morphometric parameters, filtration capacity, energetic reserves, enzymes related to oxidative stress (Superoxide dismutase, Catalase, Glutathione S-Transferase and Glutathione Peroxidase) at biochemical and molecular levels. The results suggest that *D. polymorpha* can maintain itself in effluent for 21 days. 2- *D. polymorpha* was exposed to different concentrations of protozoa (100, 1000 and 10000 protozoa per bivalve per day) for 21 days followed by 21 days of depuration in laboratory conditions. Detection of oocysts and cysts in tissues and heamolymph of bivalves was carried out by molecular biology techniques. The results highlight a time-dependent and dose-dependent bioaccumulation of protozoa by *D. polymorpha*. Moreover, the parasite load remains stable during the 21 days of depuration, suggesting that zebra mussels could integrate this biological contamination Considering these results, *Dreissena polymorpha* seems to be a promising tool for protozoa depuration. Keys words: protozoa, wastewater treatment plant, bivalve, depuration

516

Aquatic macrophytes potential for the removal of water contaminants - The Green Liver Application

S. Calado, Universidade Federal do Paraná / Ecologia e Conservação; M. Esterhuizen-Londt, Technical University of Berlin; H. Silva de Assis, UFPR / Pharmacology; S. Pflugmacher, University of Helsinki

Reservoirs are aquatic environments that are impacted by anthropogenic activities. The mainly activities around reservoirs in Brazil are agriculture and settlements. Agriculture and increase of nutrients can results in cyanobacterial blooms and cyanotoxins contamination; and settlements can results in inputs of several contaminants such as pharmaceuticals. Iraí Reservoir, located in South of Brazil, is used to water supply and has been reported as contaminated by cyanotoxins and pharmaceuticals. Therefore, this contamination increases costs to the water treatment and can cause toxic effects to the aquatic organisms and human health. The aim of this study was to test Green Liver System to remove the contaminants, at the same concentrations that were found in the reservoir, using aquatic macrophytes. *Egeria densa*, *Ceratophyllum demersum* and *Myriophyllum aquaticum* were exposed to concentrations of paracetamol, diclofenac and microcystin-LR using a laboratory model of the Green Liver System for 14 days. Water samples were collected in 0, 1, 3, 7 and 14 days and plants samples were collected at the end of the experiment. Two control experiments were carried out in parallel. Water and plants samples were used to quantify the contaminants and plants samples were also used to evaluate the catalase and glutathione S-transferase activities. Plant species took up the contaminants and the removals of compounds were 93% for diclofenac and 100% for microcystin-LR. Our results showed that the plants antioxidant system was not activated and the Green Liver System was a suitable methodology to clean the water and to implement in phytoremediation programs. **Keywords:** Green Liver System, Reservoir, aquatic macrophytes, phytoremediation.

Antibiotics and Antibiotic Resistance in the Environment: Fate and Ecological Effects, Resistance Development and Implications for Human Health

517

Identifying hotspots of Antimicrobial Resistance Selection in the Natural Environment

J.B. Sallach, University of York / Environment; A. Boxall, University of York / Environment Department

Releases of antimicrobials into the environment increase selective pressures on environmental microbes contributing to the proliferation of antimicrobial resistance (AMR) and perhaps the inevitable 'post antibiotic era'. One of many challenges in understanding environmental AMR is the high cost and organization required to provide widespread environmental monitoring. In lieu of this monitoring data, predicted environmental concentration (PEC) modelling based on pharmaceutical usage data has been demonstrated to be a useful tool in approximating antimicrobial exposures to the environment. Recently, attempts have been made to predict no-effect concentrations (PNEC) for selective pressures in the development of AMR. Coupling PEC and PNEC values provides a powerful tool to estimate the risks associated with a particular compound or class of compounds relating to AMR proliferation. Here we use this approach to identify hotspots where antibiotic exposure may be contributing to AMR selection for a range of different scenarios. Antibiotic usage data, data on metabolism, wastewater treatment and dilution data were used to determine PEC values, which were compared with reported PNECs to determine AMR hotspots for 56 compounds used in Wales as well as 9 chemical classes of antimicrobials in European Countries. Finally, using daily flow data, the approach was applied to a single wastewater treatment utility serving a population of approximately 18,600 persons with effluent discharge into the River Foss, UK to highlight the variation patterns in daily risk associated with AMR selection. Having illustrated the utility of the approach for a range of spatial and temporal scenarios, we believe that these results will be invaluable in informing future monitoring of antibiotics and AMR in the environment.

518

Urban and rural antibiotic resistance

C. McCann, M. Cooke, Newcastle University; C. Knapp, University of Strathclyde / Civil and Environmental Engineering; J. Su, Y. Zhu, Chinese Academy of Science; D.W. Graham, Newcastle University / School of Civil Engineering and Geosciences

Soils are both a source and a sink for antimicrobial resistance (AMR). Despite growing awareness of AMR in the soil resistome, debate continues over responsibility for increased AMR dissemination in this important environmental reservoir. While soil AMR is innate, the relative abundance of antibiotic resistance genes (ARGs) in soil has significantly increased over the last 60 years since the industrialisation of antibiotics. The reasons (e.g., antibiotic misuse, agriculture) for this rapid emergence continues to be debated. It is known that soil pollution is inherently linked to co-selection for ARGs yet limited information exists on large scale and multi-contaminated sites. This study examined 24 locations across the North East of England to evaluate AMR in urban and rural soils with low and high pollution levels. We present pioneering high-capacity quantitative PCR profiles of 230+ antibiotic resistance genes (ARGs) and mobile genetic elements (MGEs). The project has coupled ARGs and MGEs to comprehensive geochemical datasets including 12 metals (total and bioavailable), 16 PAHs, and nutrient conditions to understand the evolution and dissemination of modern antibiotic resistance due to pollution.

519

Dissemination of extending-spectra β-lactamase E. coli carrying multidrug resistance and virulence factors in tropical rivers receiving hospital effluents

A. Laffite, University of Geneva / Institut Forel; V. Slaveykova, University of Geneva / Département F.-A. Forel des sciences de lenvironnement et de leau; J. Poté, University of Geneva / Department F.A. Forel of environmental and aquatic sciences

The occurrence and dissemination of antibiotic resistant bacteria and their resistance genes from clinical settings to environmental compartment have become a major concern because of serious threat human health worldwide. Given the serious clinical threat of Extended-spectrum β-lactamases (ESBL) and carbapenem-resistant Enterobacteriaceae (CRE), studies are available in many countries from clinical settings. However, there is the dearth of studies in environmental compartments for the presence of these high threat gram-negative bacteria. This situation is particularly alarming in developing countries in which the freshwater resources receive urban and hospital effluent water without previous treatments. Additionally, data concerning the occurrence and dissemination of ESBL and CRE in sub-Saharan African Countries are very limited. The aim of this research is to assess the role of untreated hospital and urban wastewaters on the biological contamination of urban rivers receiving systems in the city of Kinshasa, Republic Democratic of the Congo. 147 *E. coli* strains resistant to 3^{rd} generation of β-lactams (ESBL) were isolated from water samples issued along 5 rivers receiving hospital effluents. They were analysed for their clonality and the carriage of multidrug resistance and virulence genes. The results highlight a high level of clonality in strains (67 clones) and an important level of multidrug resistance regardless the sampling point. 53% of *E. coli* resistant to the 3^{rd} generation of β-lactams were also resistant between 6 to 8 antibiotics. 14% of ESBL producers also carried virulence genes factors linked to *E. coli* pathotype determination. The genes *eaeA*, ST1, LT1 and *aggR* were carried by 1.3%, 5.4%, 2.7% and 6.8% of the strains. These results indicate the human and environmental potential risk of tropical urban rivers. Indeed, ESBL strains carried by urban rivers are associated to resistance against numerous antibiotic classes (i.e. inhibitors combination, 4^{th} generation of β-lactams, monobactams, carbapenems, aminoglycosides, tetracyclin, quinolones and phenicol classes) and may also carry virulence genes factors. The presence of multi-drug resistant *E. coli* are not intimately linked to untreated hospital wastewater discharge in urban receiving system and are widely distributed along the river, thus highlighting the risk of surface water use.

520

Methods for determining selective endpoints of antimicrobials

A. Murray, University of York; L. Zhang, I. Stanton, University of Exeter; J. Snape, AstraZeneca Global Environment / Medical School; W. Gaze, University of Exeter / Medical School

Antimicrobial resistance is one of the most significant threats to modern society. Use, misuse and overuse of antibiotics clinically and in the community; in agriculture and in aquaculture results in antibiotics and antibiotic resistant bacteria being released into the natural environment. Environmental concentrations of antibiotics are very low (ng/L range), but recent studies have shown that these concentrations may be sufficient to select for antimicrobial resistance. Currently, antibiotics are not risk assessed in terms selection for antimicrobial resistance *in situ*. This is largely because there is no standardised ecotoxicological assay which can determine such selective endpoints. This work compares previously published methods for determining and predicting selective concentrations of antibiotics to two novel methods developed in this study. The first method tracks resistance gene prevalence over time in a complex community using qPCR, and the other is based on reduction in growth of a complex community. Results show that predicted no effect concentrations (PNECs) derived using standard ecotoxicological assays are not always protective against resistance selection. Currently, no single published method for selective endpoint determination is always protective of the other; though there is good agreement between $PNEC^R$s (PNECs for resistance) published previously and $PNEC^R$s determined in this study. A novel method, based on growth of a complex community, is proposed for environmental risk assessment as it can be easily standardised, can rapidly generate selective endpoint data, and results show good agreement with more indepth data which tracks resistance gene prevalence over time. Results show that continued data generation and method optimisation is required to develop a reliable assay for determining $PNEC^R$s for environmental risk assessment of antimicrobials.

521

Determining the minimal selective concentrations of macrolides in a complex microbial community

I. Stanton, University of Exeter / Medical School; A. Murray, University of York; L. Zhang, University of Exeter; J. Snape, AstraZeneca UK Ltd. / AstraZeneca Global Environment; W. Gaze, University of Exeter / Medical School

Antibiotic resistant bacteria are found throughout the environment. Continuous release of antibiotics from human activity can and does lead to measureable concentrations in surface waters (ng/L - µg/L), however these are lower than minimum inhibitory concentrations (MICs) and concentrations used in the clinic. Due to these relatively low concentrations, until recently it was thought that selection for resistant bacteria did not occur within the environment. Research published in 2011 and 2014 by Gullberg *et al.* showed selection at low, environmental concentrations using single species assays. The macrolide antibiotics, azithromycin, clarithromycin and erythromycin, were added to the European Commission's Water Framework Directive's priority substances

watchlist in 2015 due to their measured environmental concentrations (MECs) and predicited environmental concentrations (PECs) being higher than their predicted no effect concentrations (PNECs). The aims of this study were to investigate the selective potential of these three compounds in a complex microbial community and to determine minimal selective concentrations (MSCs) for each. A number of week-long evolution experiments were conducted at a range of macrolide concentrations. QPCR determined the prevalence of a variety of macrolide resistance genes (*ermF*, *ermB*, *mphA*, *msrD* and *mef* family) and *intI1* within the community. Change in prevalence of resistance genes, when in the presence of antibiotic, was compared to change in prevalence when no antibiotic was present. Out of all of the genes tested, the *ermF* gene shows a selective response at the lowest concentration for all three macrolide antibiotics. No significant selection is seen for *ermF* at 500µg/L but we do see significant selection at 750µg/L for all three compounds. The highest current MEC for any of these macrolide compounds is 4µg/L (erythromycin-H_2O). Our data suggests, therefore, that current environmental concentrations of the macrolide compounds do not select for resistance genes in a complex microbial community.

522

Impact of multi-year exposure of agricultural soils to antibiotics on the soil resistome and mobilome.

C.H. Lau, Y. Tien, Agriculture and Agri-Food Canada; E. Topp, Agriculture and Agri-Food Canada (AAFC)

Antibiotics are entrained into agricultural soil through the application of animal manures and sewage sludge. In order to understand the potential long term effects of antibiotics on soil microorganisms, field plots were established in 1999 that have since received annual applications of a mixture of tylosin, chlortetracycline and sulfamethazine, and a second series of plots was established in 2010 that receive a mixture of erythromycin, clarithromycin and azithromycin. Antibiotics have been applied every spring at concentrations ranging from 0.1 to 10 mg kg soil^{-1}, and plots have been continuously cropped to soybeans. A library of large cloned fragments was constructed using DNA sampled in 2014 from plots receiving the high application rates, or no antibiotics. The library was cloned into antibiotic-sensitive *Escherichia coli*, and antibiotic resistance genes (ARGs) in the library were discovered by identifying E. coli clones that grew upon plating on solid growth media containing various antibiotics. Genes encoding resistance to many classes of antibiotics including the sulfonamides, tetracyclines, macrolides and β-lactams were identified, and sequence analysis revealed some to be entirely novel (Lau et al. 2017 Appl. Environ. Microbiol. 83 no. 16 e00989-17). A key question was whether the abundance of these genes increased in response to antibiotic exposure, evidence that would be consistent with functional importance *in situ*. The abundance of the novel targets as well as previously known ARGs, integrons and plasmids in soil DNA was quantified by real time PCR or with the WaferGen Biosystems high throughput qPCR instrument. Some gene targets (eg. *int*1, *sul*1, *str*A) were much more abundant in soils exposed to antibiotics whereas the vast majority of targets were not detectably increased in abundance. Overall, these results suggest that genes associated with integron cassettes are amplified in soil following repeated exposure to antibiotics.

Distribution, transformations and biological effects of incidental nanoparticles and nanoplastics in the environment from a more realistic point of view

523

Inter-annual monitoring of microplastics in marine intertidal sediments of the Firth of Forth

M. Hartl, Heriot-Watt University / Centre for Marine Biodiversity & Biotechnology, Institute of Life and Earth Sciences; Z. Lawrecne, Heriot-Watt University / Institute of Life and Earth Sciences, Centre for Marine Biodiversity & Biotechnology; C. Holmes, A. Deery, Heriot Watt University / Centre for Marine Biodiversity Biotechnology Institute of Life and Earth Sciences; J. Blumenröder, Heriot Watt University / Institute of Life and Earth Sciences, Centre for Marine Biodiversity & Biotechnology; P. Sechet, Heriot Watt University / Centre for Marine Biodiversity Biotechnology Institute of Life and Earth Sciences; R. Wood, Heriot Watt University / Institute of Life and Earth Sciences, Centre for Marine Biodiversity & Biotechnology; N. Mearns, Heriot Watt University / Centre for Marine Biodiversity and Biotechnology School of Life Sciences; S. Paterson, Heriot Watt University / Centre for Marine Biodiversity Biotechnology Institute of Life and Earth Sciences; M. Viguiaud, H. Walker, F. Kinsley-Willis, J. Mccreton, Heriot Watt University / Institute of Life and Earth Sciences, Centre for Marine Biodiversity & Biotechnology

Microplastics (MP), defined as pieces of plastic smaller < 5mm are commonly found in the marine environment and originate either consumer care products and plastic production plants or from the disintegration of larger pieces. MPs need to be monitored in order to evaluate the effectiveness of Government initiatives to reduce plastic debris in the environment. The aim of the present study, therefore, was to contribute to the development of a hitherto lacking quantitative long-term marine MP database. We present the results of a three-year pilot project in the Firth of Forth, point to innovations in sampling and contamination prevention, as well as the limitations. Sediment samples were obtained in triplicate from intertidal sites in May2014, May & Sept2015, May & Sept2016, 2017 using glass bijoux tubes as miniature cores and sealed with metal screw caps, processed using a density separation procedure and the polymer types determined using FT-IR spectroscopy. The results showed that there are high numbers of plastic particles (34-4,800 kg^{-1}) and fibres (1,700-4,300 kg^{-1}) along both shores of the Firth of Forth. The number of Fibres was generally higher than particles. There was no apparent pattern of spatial distribution. Although a spike in MP particles was observed in Sept2015 and May2016, there was no significant difference in MP particle concentrations between May 2014 and May 2017. There was also no significant difference in MP fibre concentrations during the same three-year period. There was also no evidence of seasonal fluctuations in MP concentrations. The results show that, for intertidal sediments in the Firth of Forth, the MP concentration has remained stable. This is significant baseline information and will be instrumental in assessing the effectiveness of Government policy regulating industry and consumer behaviour towards the production and use of particularly single-use plastic products. However, in order to compare results between countries and laboratories, for the purpose of gaining a more global insight into the microplastics contamination issue, more standardized sampling and extraction procedures need to be developed.

524

Do nanoparticles cause stress effects on microalgae ? An infrared spectroscopy study.

M. Déniel, Institute of molecules and materials of Le Mans / Physique des Interfaces et des MésoStructures; N. Errien, Institute of molecules and materials of Le Mans; A. Caruso, laboratory Mer Molécule Santé; f. lagarde, Institute of molecules and materials of Le Mans / Institute of Materials and Molecules of Le Mans IMMM UMR CNRS

Nanoparticles are constantly used at world level leading to their presence in the aquatic environment lead to possible particles interaction with living organisms. The potential impacts of these interactions are essential to evaluate for a better understood of the induced mechanisms. Microalgae are the base of aquatic trophic chain and different possible pathways of interaction between microalgae and nanoparticles are described in literature [1]. Moreover, infrared spectroscopy is known to monitoring material chemical composition. In this work, infrared spectroscopy is used to monitor the interaction between different types of nanoparticles and a model freshwater microalga (*Chlamydomonas reinhardtii*). The aim is to understand the effect induced by nanoparticles and discriminate the responses of the microalgae in comparison with known stress mechanisms. Different stresses are realised in this goal: nutriment deficiency, light deficiency, metallic stress, gold and polystyrene nanoparticles. After 48h or 72h of interaction, microalgae are collected for infrared analysis. In parallel, biological parameters (growth and genes expression) are relieved to indicate if the induced stresses imply cytotoxic effects or molecular effects on the organism. The multivariate analysis highlight that microalgae responses are stress dependent. Thus, infrared spectroscopy could be a new method to analyse stress effect on microalgae and particularly nanoparticles. Interaction with nanoparticles seems to induce an over-expression of astaxanthin biosynthesis pathway genes.In conclusion, infrared and biological data relationships could explain interaction mechanisms between nanoparticles and microalgae. Keywords: nanoparticles, infrared microspectroscopy, effects monitoring [1] von Moos N & Slaveykova VI. 2014. Oxidative stress induced by inorganic nanoparticles in bacteria and aquatic microalgae–state of the art and knowledge gaps. Nanotoxicology. *8*(6): 605-630.

525

Ecotoxicological evaluation of high-generation cationic PAMAM dendrimers towards a representative organism of aquatic ecosystems

G. Pulido-Reyes, M. Tamayo-Belda, M.G. Pleiter, Universidad Autónoma de Madrid; k.m. betancor, F. Leganes, Universidad Autónoma de Madrid / Biology; R. Rosal, University of Alcala; F. Fernandez-Piñas, Universidad Autónoma de Madrid / Biology

Nowadays, nanomaterials are extensively used worldwide in many different fields and their potentially serious effects in aquatic ecosystems have become a global concern [1]. Poly(amidoamine) (PAMAM) dendrimers are polymeric nanoparticles, radially symmetric molecules with well-defined, homogeneous, and monodisperse structures that have a typically symmetric core, an inner shell, and an outer shell. Due to these characteristics, their use is being tested in the implementation of targeted therapies in biomedicine so that they might end up in environment [2]. In this study, we have investigated the effect of high-generation cationic G5-NH_2 and G7-NH_2 PAMAM dendrimers in a prokaryotic primary producer of aquatic ecosystems, the filamentous cyanobacterium *Anabaena* sp. PCC7120 (*Anabaena)*. Dendrimers significantly decreased the growth of the cyanobacterium and both dendrimers induced morphological alterations of both filaments and individual cells. Furthermore, cyanobacteria exposure to dendrimers resulted in significant alteration of physiological parameters: increase in the formation of intracellular reactive oxygen species, damage in membrane integrity, membrane potential depolarization, increase of metabolic activity, acidification of intracellular pH and alteration of intracellular free Ca^{2+} homeostasis. Dendrimers also induced alterations in the photosynthetic responses of *Anabaena*. In conclusion, high-generation cationic dendrimers exhibited high toxicity towards

Anabaena and several physiological, morphological and photosynthetic parameters. [1] Navarro E, Baun A, Behra R, Hartmann NB, Filser J, Miao AJ, Sigg L. 2008. Environmental behavior and ecotoxicity of engineered nanoparticles to algae, plants, and fungi. Ecotoxicology 17(5): 372-386 [2] Gonzalo S, Rodea-Palomares I, Leganés F, García-Calvo E, Rosal R, Fernández-Piñas F. 2015. First evidences of PAMAM dendrimer internalization in microorganisms of w relevance: A linkage with toxicity and oxidative stress. Nanotoxicology 9(6): 706-718 *Acknowledgement* - This research was supported by CTM2013-45775-C2-2-R and CGL2010-15675 grants from MINECO/FEDER EU.

526

Interactive effects of carbon nanoparticles and benzo(α)pyrene on marine mussels, Mytilus galloprovianciallis

A. Barranger, University of Plymouth / School of Biological Sciences; Y. Aminot, University of Plymouth; M. Banni, Laboratory of Biochemical and Environmental Toxicology; S. Sforzini, Universita Del Piemonte Orientale Amadeo Avogadro / Department of Sciences and Technological Innovation (DiSIT); V.M. Arlt, Kings College London; G. Rance, A. Khlobystov, University of Nottingham / School of Chemistry; A. Viarengo, University of Piemonte Orientale / Department of Sciences and Technological Innovation DiSIT; J.W. Readman, University of Plymouth / Biogeochemistry Research Centre; A.N. Jha, Plymouth University / Biological Sciences

The production and discharge of nanoparticles has grown extensively over the last few years, raising concerns over their potential impact on environmental health, either alone or in combination with other anthropogenic contaminants. The study, funded by Natural Environment Research Council (NERC), UK aims to test the hypothesis that environmentally relevant carbon based nanoparticles (CNPs) and polycyclic aromatic hydrocarbons (PAHs) can interact with each other to differentially modify their potential toxicity. To probe this hypothesis, marine mussels were exposed for 3 days to benzo(a)pyrene (BaP) and to two different types of carbon nanoparticles, [C_{60} fullerenes and multi-walled carbon nanotubes (MWNTs)], both alone and in combination with BaP. Tissue specific distributions and concentrations of CNPs and BaP were determined in exposed mussels. To enhance the analytical traceability of these CNPs in biological systems, some nanoparticles were labelled with rare elements. CNP uptake was followed by ICP-MS and/or HPLC-UV, with the BaP uptake tracked by GC/MS. CNP uptake was also investigated by electron microscopy. The genotoxic effects were characterised by the level of DNA strand breaks (comet assay), micronuclei and DNA adduct analyses. Global gene expression profiles were analysed using microarray technologies targeting 15 stress response pathways. Contrasting results were obtained according to the type of carbon nanoparticles used. Co-exposure of mussels to MWNTs and BaP seems to reduce the uptake and genotoxic effects of BaP in the digestive gland. Conversely, co-exposure to C_{60} and BaP does not seem to affect the uptake and genotoxic effects of BaP. Different responses were also observed with the transcriptomic studies. Microarray analysis identified several key biological processes (e.g. DNA metabolism, cytoskeleton, oxidative stress and heat shock response). In order to have a better understanding of the effects of these CNPs, further biological analysis (e.g. DNA oxidation and proteomics) are currently in progress. This study opens new questions highlighting the importance of studying the potential interaction between nanomaterials and environmentally important pollutants.

527

Trophic transfer of CuO NPs and Aqueous Cu: from worms to fish - a proof of concept study

T. Lammel, University of Gothenburg / Dep of Biological and Environmental Sciences; A. Thit Jensen, Roskilde University / ENSPAC; C. Mouneyrac, Université Catholique de lOuest / Université Catholique de lOuest; A. Baun, Technical University of Denmark / DTU Environment; J. Sturve, University of Gothenburg / Department of Biological and Environmental Sciences; H. Selck, Roskilde University / Dept Science and Environment

Nanoparticles (NPs) will inevitably end up in the aquatic environment, where they will settle out and accumulate in the sediment. Therefore benthic fauna is at particular risk of NP exposure. Uptake of both dissolved and nanoparticulate metals have been reported for different benthic invertebrates, which serve as foraging organisms of fish. Here we examine if transfer of copper (II) oxide (CuO) NPs and dissolved copper (administered as $CuCl_2$) can occur from sediment to worms (*tubifex tubifex*) and further from worms to fish (*Gasterosteus aculeatus*). CuO NPs (< 50 nm; Sigma) were characterized with regard to primary particle size, shape, hydrodynamic diameter and dissolution at different experimental conditions using TEM, DLS, PALS and ultrafiltration followed by ICP-MS analysis, respectively. Worms were exposed to sediment amended with CuO NPs or $CuCl_2$. Cu concentrations in sediment, overlaying water and worm tissue were determined using ICP-MS. In addition, the metal binding protein metallothionein (MT) was quantified with DPP (differential pulse polarography). Fish were exposed for up to 7 days to worm-shaped CuO NP and $CuCl_2$-spiked food packages produced from uncontaminated tubifex homogenates (2 µg Cu/g fish/day). Cu concentrations were measured in intestine, liver and carcass using ICP-MS. In addition, intestinal and hepatic mRNA expression levels of genes relevant for Cu uptake, storage and toxicity including metallothionein A (*mta*) were measured using RT-qPCR. The total Cu body burden of tubifex increased by 3 and 3.5 µg Cu/dw tissue after 7 days of exposure in CuO NP- and $CuCl_2$ spiked sediment, respectively, suggesting that NP uptake into the organism occurred. Cu accumulation was also observed in fish receiving CuO NP and $CuCl_2$-spiked food packages, in particular in intestine, and was concomitant with upregulation of *mta* transcription. The increase in the intestinal Cu concentration and *mta* expression in CuO NP-exposed fish was higher than in the control, but did not reach levels measured in $CuCl_2$-exposed fish. At the same time the amount of Cu egested with the faeces was significantly higher than in the $CuCl_2$-treatment. These results suggest that transfer of CuO NP along the food chain may be limited compared to dissolved Cu. We suggest future studies on how NP bioavailability and accumulation in fish is influenced by other important factors, such as exposure dose, time and NP properties upon biotransformation by the foraging organism.

528

Corbicula fluminea exposure to copper oxide nanoparticles: an integrated mesocosm study

V. Koehle-Divo, LIEC - Université de Lorraine - CNRS / LIEC, CNRS; S. Pain-Devin, Université de Lorraine - UL / LIEC - CNRS - UMR 7360; B. Sohm, University of Lorraine / LIEC, CNRS; S. Devin, LIEC, CNRS UMR 7360, Université de Lorraine / LIEC, CNRS; M. Auffan, CEREGE / International Consortium for the Environmental Implications of Nanotechnology; C. Mouneyrac, Université Catholique de lOuest / UBL, Mer Molécules Santé; L. Giamberini, Université de Lorraine, CNRS UMR 7360 / LIEC, CNRS

Engineered nanoparticles (ENP) are now part of our daily life because of their introduction in a wide variety of products. Their concentrations in environment are not yet known but release during their life cycle is obvious. Copper oxide nanoparticles (CuO ENP) are well known for their antimicrobial properties allowing their use in numerous products as in wood-paints, textiles or food packaging. Since aquatic compartments are the ultimate sink of contamination, they should be impacted by release of ENP. Some studies highlighted the ability of CuO ENP to induce stress responses in several levels of biological organisation in aquatic organisms, indicating their toxic potential. Most studies were nonetheless made using simplified exposures, thus maximizing ENP dispersion and contact with the studied organisms. The fate and consequently the toxic potential of CuO ENP differ depending on the complexity of the exposure media that can considerably modify ENP physico-chemical properties and consequently, their bioavailibility to living organisms. Thereby, setting up more complex design of exposure may help to gain in environmental realism. The aim of this work was to evaluate the fate and effects of different CuO ENP on a widespread endobenthic freshwater bivalve, *Corbicula fluminea*. In order to improve environmental realism, *C. fluminea* were exposed in indoor mesocosm containing sediment, water and food. A cumulative contamination was applied until reaching a final concentration of 50 µg CuO/L at the end of the exposure period (28 d). Behavioral, physiological, biochemical and molecular parameters were quantified in order to assess CuO ENP impacts on *C. fluminea*. Results of this study allow to conclude that CuO ENP affected *C. fluminea* behavior by increasing burrowing, suggesting an avoidance reaction. CuO ENP also induced significant impacts at the biochemical and molecular levels. However, the detected changes were low and did not show a clear and constant pattern. Further studies are needed to better understand whether detected effects may induce other effects at higher biological level (such as affecting behavior) or whether the avoidance behavior may have protected organisms from exposure, then lowering the effects that we were able to measure.

Luminescent biomonitoring via bioassays of different complexity - from cells trough enzyme reactions to proteins

529

Applications of Luminous Bacteria Enzymes in Toxicology and Ecology

V. Kratasyuk, Siberian Federal University / Biophysical; E. Simbekova, Siberian Federal University / Biophysics

A new approach in developing bacterial bioluminescent enzymatic biosensors, application to toxicity bioassays, and the needed reagents has been developed. To solve the problem of how to detect, identify, and measure the numerous chemical compounds in environmental monitoring, food product contamination, and medical diagnostics, bioluminescent enzymatic toxicity assays were proposed, wherein the bacterial coupled enzyme system NAD(P)H:FMN-oxidoreductase-luciferase substitutes for older methods using living organisms. The immobilized reagent Enzymolum was used to facilitate and accelerate the development of the bioluminescent enzymatic systems as biological part of biosensors for toxicological assays. The reagent is easy to use and convenient to be applied not only in toxicology studies but also in education, mainly in ecological and enzymological practical courses. Prototype biosensors offer cost advantages, versatility, high sensitivity, rapid response, extended shelf-life and flexible storage conditions. This study was supported by the Russian Science Foundation (project no. 16-14-10115).

530

Toxic and adaptive effects via luminescent assay systems of different

complexity: bacterial cells, enzyme reactions, and fluorescent proteins

N. Kudryasheva, R. Aleva, Institute of Biophysics SB RAS; T.V. Rozhko, Siberian Federal University; A.S. Petrova, Insitute of Biophysics SB RAS; A. Lukonina, Siberian Federal University / Biphysics; E. Kovel, Siberian Federal University; A. Sachkova, National Research Tomsk Polytechnic University

Luminescence of living systems is a convenient parameter to monitor environmental toxicity. Luminescent systems of different complexity – luminous marine bacteria, their enzyme reactions, and coelenteramide-containing fluorescent proteins (CLM-CFPs) were used as bioassays to monitor toxicity of water solutions under model conditions; toxic effects were compared at cellular, biochemical and protein levels, respectively. Organic compounds, metallic salts, and radioactive elements (under conditions of low-dose irradiation) were applied to vary toxicity of media. Luminescence inhibition (toxic) and activation (adaptive response) effects were evaluated and discussed. Application of CLM-CFPs as toxicity bioassays of a new type is justified, they can serve as a proper tool for study efficiency of primary physicochemical processes in organisms under external exposures. Coelenteramide (CLM), fluorophore of CLM-CFPs, is a photochemically active molecule; it acts as a proton donor in its electron-excited states, generating several forms of different fluorescent state energy and, hence, different fluorescence color, from violet to green. Contributions of the forms to the visible fluorescence depend on the CLM microenvironment in proteins. Hence, CLM-CFPs can serve as fluorescence biomarkers with color differentiation to monitor results of destructive biomolecule exposures. Variations of spectral-luminescent and photochemical properties of CLM-CFPs under different exposures – chemicals, temperature (1), and ionizing radiation (2) is considered. Application of the luminescent bioassays of different complexity for detoxification efficiency evaluation is discussed. Natural and artificial bioactive compounds, humic substances (3) and fullerenols (4-5), are used as detoxifying agents. Detoxification mechanisms were revealed to be complex, with chemical, biochemical, and cellular aspects conditioning those. Active role of the bioassay systems in the detoxification processes was demonstrated. 1. Alieva R., Kudryasheva N. *Talanta,* 2017, 170, 425. 2. Petrova A., et al. *Anal&Bioanal. Chem,* 2017. DOI: 10.1007/s00216-017-0404-9 3. Kudryasheva N., Tarasova A. *Environ. Sci. Pollut. Res.,* 2015, 22 (1), 155. 4. Kudryasheva N., et al. *Photochemistry and Photobiology.* 2017, 93(2), 536 5. Sachkova A., et al. *Biochemistry and Biophysics Reports*. 2017, 9, 1-8

531

Assaying the prooxidant and antioxidant potentials of nicotine products: Tobacco versus electronic cigarettes

A. Trofimov, V. Menshov, O. Yablonsky, Emanuel Institute of Biochemical Physics Russian Academy of Sciences

Cellular oxidative stress, derived from imbalance between the production of reactive oxygen species and the efficacy of the antioxidant defense, can be a consequence of using the nicotine products. Prooxidant properties of the tobacco smoke are accounted for by the abundance of the smoke oxidants. The antioxidant potential of the smoke is scantily addressed in the literature. However, one should take into account that any reactant in oxidation process may exhibit both oxidant and antioxidant propensities depending on the reaction conditions. And we have shown that smoke constituents indeed exhibit at the same time both prooxidant and antioxidant activities. Such smoke-borne antioxidants may be assessed through both the direct chemiluminescence (CL) derived from the smoke samples as a function of the smoke tar content and using the probe CL preparations of hydrocarbon substrates being oxidized. In addition to exogenous antioxidants derived from the smoke, we have studied the possibility of the antioxidants generation directly in smokers. For that purpose, we have developed a novel assay based on the CL of luminol, which involves tobacco-smoke extracts, peroxidase and amino acids. Using such a system, we have demonstrated that under physiological conditions the oxidation of the smoke tar and its individual components, e.g. catechol, in the presence of H_2O_2, peroxidase, and glycine affords the products (first of all, catechol-glycine adducts) whose antioxidant potential is much higher than that of initial, unoxidized, chemicals. Conversely, we have not observed any significant antioxidant activity of aerosols derived from electronic cigarettes (ECs). For ECs, the following feature is noteworthy. We have found for the first time that all ECs, regardless of their technical complexity, generate in their emissions hydroperoxides (ROOH) of propylene glycol (the main component of e-liquids served as solvent for nicotine), which are potential prooxidants (ROS sources), whose physiological significance still requires elucidation. The content of these products depend on the type of EC and on the mode of its use, which makes possible to minimize the ROOH generation. The CL methodology has proved to be the most useful tool in assessing the oxidant and antioxidant potentials of nicotine-containing aerosols, i.e. tobacco smoke and emissions from ECs, which exhibit essentially different impacts on oxidative developments in a smoker's organism.

532

The possibilities of using fungal fluorophores to assess responses of filamentous fungi to external stimuli

E.V. Fedoseeva, Pirogov Russian National Research Medical University / Pediatric faculty; D. Khundzhua, Lomonosov Moscow State University / Depament of General Physics; V. Terekhova, Lomonosov Moscow State University / Lab of Ecotoxicological Soil Analysis; M. Freidkin, Lomonosov Moscow State University / Department of General Physics; S. Patsaeva, Lomonosov Moscow State University / Depament of General Physics

The interest to functional and structural indicators of mycobiota with a respective to use them in biogiagnostics is currently rising because fungi represent as essential component among environmental decomposers of organic material in ecosystems. The complexity of utilization of fungi in biogiagnostics is explained by the variability of reactions to external stimuli due to their physiological and biochemical plasticity. In this regard, it seems very important to find the reactions of fungal cultures adequately reflecting their response to external stimuli in different conditions. The essential biogenic fluorophores such as NADH, tryptophan, melanin, ergosterol, pyridoxine, riboflavin, FAD, and FMN can be monitored instrumentally by spectroscopic techniques. The aim of the present study was to investigate the features of fluorescence spectra of fulamentous fungi cultivated under different concentrations of source of bioavailable and nor readily bioavailable carbon in the growth medium. The research objects were strains of *Alternaria alternata*, *Cladosporium cladosporioides,* and *Trichoderma harzianum.* The strains were kindly provided by O.E. Marfenina and A.E. Ivaniva, Soil Science Faculty of MSU. The filamentous fungi were grown on liquid and agar Czapek medium contained a varying level of sucrose (0.3 and 3%) and humic substances (0.02 and 0.1%). Fluorescence spectra were measured using a luminescence spectrometer Solar CM2203 at several wavelengths of the exciting radiation (250, 280, 310, 350, and 450 nm). Fluorescence excitation spectra were registered for emission at 350, 440, and 460 nm. Typical fluorescence spectra of fungal samples (spore suspensions and supernatant liquids) with the UV-excitation consist of two overlapping bands. The UV-band with maximum at 300-350 nm under excitation at 280 nm is a protein-like fluorescence, and the band in the blue region with the maximum at 400-450 nm under excitation at 310-370 nm is emission of fungal chromophores like NADH and or melanins. We suggest using the intensity of the first fluorescence band in the fungal samples for rapid evaluation of spore biomass. Measured fluorescence characteristics were found correlating with saturation of growth medium by source of bioavailable and not readily bioavailable carbon. Therefore we consider this research as promising on the way of using fungal fluorophores to assess responses of filamentous fungi to external stimuli.

533

Effect of surface functionality on Fe3O4 nanoparticles toxicity

L. Kulyabko, Moscow Aviation Institute; P. Uchanov, Institute of Ecology and Evolution RAS / Laboratory for soil ecological functions; S. Patsaeva, Lomonosov Moscow State University / Depament of General Physics; V. Terekhova, Lomonosov Moscow State University / Lab of Ecotoxicological Soil Analysis; K. Kydralieva, Institute of Chemistry and Chemical Technology

Surface characteristics stands out as one of the most important, if not the main, determinants of biological performance, as the nanoparticles (NPs) surface is the most prominent and earliest point of exposure. Complexity of this issue is that the coordination of ligands on the surface of NPs can significantly enhance subsequent cytotoxicity. The principal challenges that have to be addressed are a detailed understanding of how the NP's original "engineered" surface chemistry influences subsequent NP interactions with biosystems. In this work results of a complex study of Fe_3O_4 functionalized by humic acids (HA) were described. We hypothesized that, along with the NPs size, the surface functionalization was a major factor contributing to sorbent toxicity mitigation. The average particle size calculated by the Scherrer equation tended to decrease from 8.4 nm for Fe_3O_4-HA20 to 4.5 nm for Fe_3O_4-HA80. Optical spectroscopy indicated that the fluorescence quantum yield depended on the HA content in the nanocomposite and confirmed that the humic component interacted with ferric ions. Biosafety of Fe_3O_4-HA NPs was investigated in laboratory biotest systems using algae, infusorians, and higher plants as test-cultures. Concentration limits for using the Fe_3O_4-HA NPs suspended in water under controlled artificial conditions were found experimentally by ecotoxicological tests. Experiments with this "battery" of three biotests showed that, in controlled chemical conditions, water suspensions of the preparation can be safely used for biota given a certain concentration limit. It was found that samples of bare Fe_3O_4 and the Fe_3O_4-HA80, 0.01 (%), were remarkably more toxic than water suspensions Fe_3O_4-HA20 and Fe_3O_4-HA50 in this concentration in biotests with algae and higher plants. Effect of 10, 100 and 1000 times dilution revealed that the effective concentration (EC_{20}) for *Sinapis alba* did not exceed 0.1%Fe_3O_4. With that, algae appeared more sensitive: EC_{20} was 0.01%in the biotest with *Scenedesmus quadricauda* for Fe_3O_4, Fe_3O_4-HA50, and Fe_3O_4-HA20. Humic substances in natural conditions are likely to increase the permissible concentration limit and to mitigate harmful impact of the NPs. Obviously, before applying such remediation agent in specific biotope conditions, its biosafety should be additionally assessed by the bioindication method based on response of biotope inhabitants.

534

Poster spotlight: WE209, WE210, WE211

Obesogens and lipid disruptors

535

The evolution of obesogen-induced phenotypes in vertebrates

A. Capitão, M. Lopes-Marques, R. Ruivo, E. Fonseca, R. Jorge, M. Barbosa, CIIMAR University of Porto; Y. Hiromori, Y. Ishi, T. Miyagi, T. Nakanishi, Gifu Pharmaceutical University; R. Santos, Hepia University of Applied Sciences Western Switzerland; F. Castro, CIIMAR - University of Porto

Global obesity is an escalating pandemic in western societies. Triggered by nutritional variations and overconsumption, this condition is also influenced by individual and environmental cues. Of note are the globally persistent man-made chemicals, with ever-growing ecosystemic consequences, a hallmark of the Anthropocene epoch. A striking example highlights the role of a group of compounds known as "*obesogens*". In mammals, most examples involve the modulation of the peroxisome proliferator-activated receptor g (PPARg) nuclear receptor. To decipher the pattern of PPARg exploitation by a model obesogen, tributyltin (TBT), we employed an extensive analysis from comparative genomics to transactivation assays, site-directed-mutagenesis, and homology modeling, to unfold the structural and biological determinants of PPAR exploitation by TBT.Our findings endorse the modulatory ability of man-made chemicals and suggest an evolutionary diverse setting of "*obesogenic*" responses to TBT, with impacts for human health risk assessment.

536

Aging Extension and Modifications of Lipid Metabolism in the Monogonont Rotifer Brachionus koreanus under Chronic Caloric Restriction

M. Lee, J. Park, J. Lee, Sungkyunkwan University

To examine the interrelationship of the aging extension and modification of lipid metabolism under chronic caloric restriction (CCR; concentration from 0 to 100% of the diatom *Tetraselmis suecica*) in the monogonont rotifer *Brachionus koreanus*, we assessed the life cycle parameters, fatty acid composition, and *sirtuin* and lipid metabolism-related genes expression. As a result, in the 5% exposed group, *B. koreanus* showed the increased life span but the decreased reproduction. Based on this finding, we chose 5% of *T. suecica* and performed the rest of the experiment compared to 100%. As a result, up-regulation of *sirtuin* genes expression was observed. In addition, despite the reduction in the amount of total fatty acid (FA) and the area of triacylglycerol, the increase in the ratio of saturated fatty acid and monounsaturated fatty acid (MUFA) among the total FA in 5%-exposed *B. koreanus* were observed. Furthermore, the mRNA expression of *Δ9 desaturase* confirmed that CCR promoted the synthesis of MUFA through *Δ9 desaturase*. Moreover, the expression of docosahexaenoic acid (DHA) synthesizing gene, *Δ4 desaturase*, has also been up-regulated along with DHA content. These data suggest that CCR modified histone acetylation and lipid metabolism, leading to decrease in reproduction, consequently resulting in life span extension.

537

Lipidomic and transcriptomic changes induced by compounds enhancing accumulation of storage lipids in Daphnia magna.

I. Fuertes, Institute of Environmental Assessment and Water Research IDAEA CSIC; R. Jordão, Institute of Environmental Assessment and Water Research IDAEA CSIC / Department of Environmental Chemistry; B. Pina, IDAEA-CSIC / Department of Environmental Chemistry; C. Barata, CSIC / Environmental Chemistry

The analysis of lipid disruptive effects in invertebrates is limited by our poor knowledge of the lipid metabolic pathways and of their complete lipidoma. Recent studies showed that tributyltin and juvenoids activated the ecdysteroid, juvenile hormone and retinoic X receptor signalling pathways, and disrupted the dynamics of triacylglycerols in lipid droplets in the crustacean *Daphnia magna*. This study aimed to explore how ecdysteroids, juvenoids and bisphenol A disrupt the dynamics of phospholipids and neutral glycerolipids in adult daphnias during the reproductive cycle from both lipidomic and transcriptomic points of view. Comparison of the lipidomic profile between treatments and controls revealed relative abundance changes for 194 out of 235 individual lipids detected, corresponding to three classes of neutral glycerolipids (TAGs, DAGs, MAGs) and nine of phospholipids (PCs, LPCs, PEAs, LPEAs, PSs, LPSs, PGs, LPGs, SMs). Cluster analysis defined two major clusters, one corresponding to control, BPA and 20E samples, with low levels of TAGs but higher levels of PCs; and another one corresponding to juvenoid-treated samples (PP and MF), with higher levels of TAGs and lower levels of PCs. In addition, subclusters corresponding to lower and higher exposure time were also observed. Transcriptomic analyses identified 1,964 de-regulated genes that were clustered in three groups corresponding to up-regulated gene transcription after either 8 or 24h of TBT exposure, and to up- and down-regulated genes after 24 h of exposure to BPA, PP, or TBT. Gene ontology analysis indicated an enrichment of gene signalling pathways involved in lipid metabolism, specifically in lipid catabolic process, triglyceride homeostasis, glycolipid biosynthesis or fatty acid beta-oxidation. This work as supported by the Spanish Research Project EMRISK Code CTM2014-51985-R, (2015-2017). Inmaculada Fuertes acknowledges the Ministry of Economy, Industry and Competitiveness for her fellowship (FPI-MICINN BES-2015-075023).

538

Lipidomics profiling of wild fish to identify patterns associated with pollution exposure

C. Porte, IDAEA-CSIC / Department of Environmental Chemistry; M. Blanco, IDAEA -CSIC-; A. Maceda-Veiga, University of Barcelona / Department of Animal Biology

New developments of analytical techniques have allowed the effective identification and characterization of lipids and the development of lipidomics, which has recently emerged as a key technology for human disease research and discovery of biomarkers. However, on an environmental toxicology context, studies are still few, in spite of lipids being considered key molecules for the bioaccumulation of chemicals. This work applies ultra-high performance liquid chromatography coupled to high resolution mass spectrometry (UPLC-HRMS) to characterize the lipidome of the muscular tissue of two fish species (*Barbus meridionalis*, *Squalius laietanus*) collected along the Ripoll River. Sampling sites included upstream (reference) and downstream (urban and industrial discharges) areas. A total of 130 lipid species, including phosphatidylcholines (PC), PC-plasmalogens (PC-P), cholesterol esters (CE), triacylglycerols (TG), diacylglycerols (DG) and sphingomyelins (SM) were detected in the muscle tissue. Partial least squares discriminant analysis (PLS-DA) allowed a clear separation of the lipidome of fish from polluted and reference sites. Specifically, a relative increase of CEs (18:1, 20:4, 22:5, 22:6) and some PC-Ps (32:0, 36:4, 36:5, 38:6) was detected in the muscle of *B. meridionalis* sampled in polluted sites. In contrast, the lipidome of *S. laietanus* from polluted areas was characterized by a significant increase of TGs and PC-Ps and a concomitant decrease of PCs with a high number of double bounds (36:5, 36:6, 38:6, 40:6, 40:7). The results suggest potential lipid oxidation of highly unsaturated PCs, particularly in *S. laietanus* living in polluted sites together with a concomitant increase in neutral lipids (TGs, CEs), possibly due to an increase in the energy demand to respond to stress in polluted sites.

539

Lipidomics profiles distinguish fish from organochlorine pesticide contaminated lakes compared to control lakes

N.D. Denslow, M. Nouri, University of Florida / Physiological Sciences; K.J. Kroll, University of Florida / Physiological Science; C.J. Martyniuk, University of Florida / Physiological Sciences; V. Dang, Iowa State University

The organochlorine pesticide (OCP) contamination of Lake Apopka largely derived from high application use in the muck farms on the North Shore. These practices were discontinued in the 1970's but fish in Lake Apopka continue to have relatively high body burdens of organochlorine contaminants. Previous transcriptomics experiments have indicated that the OCPs alter endocrine related endpoints in ovary and liver of exposed fish. In addition, changes in lipid transport and metabolic pathways are affected. Current work explores changes in the lipidome of largemouth bass caught in Lake Apopka compared to fish from a relatively clean lake in the Ocala National Forest. We used both a shotgun approach and a targeted approach to quantify perturbations in phospholipids in liver of largemouth bass from Lake Apopka compared to a relatively clean lake. Follow up experiments with fish exposed in the laboratory support the changes seen in the field. Cholesterol was decreased and cholesterol esters were elevated in the livers of fish from Lake Apopka compared to Wild Cat Lake. This finding corroborates reduced hormone biosynthesis in organochlorine contaminated fish. Other changes in the lipidome are consistent with predicted changes that are related to immune dysfunction. Enrichment in Lake Apopka fish was observed in short chain length free fatty acids, such as palmitic acid and in ceramides, phosphatidic acids and phosphatidylinositols. But decreases were observed in sphingomyelins, phosphatidyl-ethanolamines and other phospholipids. These changes are consistent with lipids that are changed due to inflammation and other immune responses. We postulate changes in the lipidome are important biomarkers of OCP contamination.

540

Poster spotlight: WE027, WE028, WE029

Towards a shared understanding of science and risk communication in the context of the inevitability of chemicals and the hazard they may represent (II)

541

Nanotechnology: When shading effects through agglomeration of carbon nanotubes (CNT's) are confused with toxicity by media and the public - a case example revisited

F. Schwab, Adolphe Merkle Institute / Materials Science

Engineered nanomaterials are relatively new contaminants with that can enter the environment *via* an increasing variety and number of waste streams. The long-term toxicity of nanomaterials is not well understood, and these materials are therefore of emerging public concern. In 2011, we published a press release about our scientific publication on the effects of carbon nanotubes on green algae [1]. We found that the nanoparticles under investigation did not directly affect the algal viability, but indirectly reduced the algal growth *via* shading and agglomeration. To our surprise,

this press release led to a cascade of secondary articles and events. On the one hand, some online newspapers used our article to produce alarming articles about the dangers of nanoparticles for the environment (example translated from German: “Nanoparticles Identified as Potential Environmental Killers” [2]). On the other hand, some individuals used the press release to draw the oversimplified conclusion that all engineered nanomaterials will eventually agglomerate and therefore be harmless. Nevertheless, most of the media took over the message with no or minor modifications. The press release also triggered surprising responses from within the research institutions. In this presentation, I was invited to briefly summarize the different responses that we got to this press release, and re-iterate the short- and long-term lessons learned from this case study. Most importantly, the ‘real’ work for a scientist writing a press release starts after its publication. News on topics of public concern such as the toxicity of engineered nanomaterials are very closely watched and instrumentalized both by the pro- and the anti-nano community for their respective intentions. Reactions on press releases concerning these topics have to be monitored closely, wrong quotations must be corrected and biased interpretations must be adjusted, in order to provide correct scientific information for the common public. The reward for this work is an overall more balanced communication of the results. [1] Schwab F, Bucheli TD, Lukhele LP, Magrez A, Nowack B, Sigg L, Knauer K. 2011. Are carbon nanotube effects on green algae caused by shading and agglomeration? Environ Sci Technol 45:6136-6144. [3] Georgescu V. 2011. Nanopartikel als potenzielle Umweltkiller ausgemacht. www.lifegen.de/newsip/shownews.php4?getnews=m2011-11-09-3109&pc=s02. Accessed 22 Nov 2017 *Acknowledgements and Disclaimer - Schwab,* F was supported by an Ambizione fellowship of the Swiss National Science Foundation (grant number 168187). Any opinions, findings, conclusions or recommendations expressed in this material are those of the author and do not necessarily reflect the views of the Adolphe Merkle Institute or the SNSF. This work has not been subjected to their review and no official endorsement should be inferred. The author reports no other conflicts of interest and is responsible for the content of the abstract and presentation.

542
Nanotechnology: Communicating scientific findings through media – what could possibly go wrong? Lessons learned from Schwab’s nanotubes
G. Oberg, UBC / IRES; A. Seal, University of British Columbia / School of Journalism
There is no single effective method for scientists to communicate their findings with the media. Unfortunately, Dr. Fabienne Schwab found this out after publishing a press release about the effects carbon nanotubes (CNTs) have on green algae. When the story hit the press, mainly through an article in Der Spiegel, many readers understood CNTs to be toxic. As a result, Dr. Schwab and her colleagues were accused of fear-mongering. Things escalated to the point that Der Spiegel had to shut down the article’s comments section. Where did things go wrong? How can scientists make sure the media presents their work accurately, but also in a way the general public can understand? We will discuss the diverging roles and realities of science and media, particularly the considerations that scientists and editors need to take into account when they decide to write/publish something.

543
Discussion Nanotechnology

544
Microplastics: The risks of plastics – perceived or real?
M. Kotterman, IMARES / Fish
Plastic has not only become a major research topic, it is also broadly covered in popular news. As result the general public knows about the plastic soup and how dangerous it is, supposingly, for wildlife and ultimately for human health. However, the history of plastic research is peculiar. Some of it was straightforward; wildlife choking in plastic does not need much additional proof or QC QA. But, as with many new research topics, the first articles about the dangers of plastic were soon followed by others. The focus was on the presence of small plastics particles everywhere; from seafood, honey to even drinking water. Any quality control seemed absent, as most of the published articles did not have proper controls. And if they did, it became apparent that many of the plastic fibres observed in the samples were a result of cross contamination by air. Secondly, while plastic particles do not behave very differently from other particulate matter with respect to absorption of organic contaminants, all known equilibrium processes of contaminants between particulate matter, biota and water were blatantly ignored. Contaminants in open seas would first sorb strongly to plastics, to desorb readily in the gastrointestinal tract of fish, leading to higher bioaccumulation of pollutants like PCBs in the food chain. The fact that the amount of ingested plastic is still almost negligible compared to the natural food intake makes these claims even more difficult to uphold. Therefore, it was disappointing that even Science published an article about the dangers of plastic microparticles for fish larvae, while the manuscript did not comply to the journals own quality standards. And as it seems now, the described research has not even been performed. So, besides the obvious and clear detrimental effects of plastic debris in the environment, an important concern of plastic may be that research on the environmental impact of plastics is not always conducted following proper scientific guidelines. In this presentation I will also discuss shortly the more recent progress in plastic research, such as the exposure of humans to plastic particles.

545
Lost in translation: Do we communicate the risks of (micro)plastics in the right way?
M. Wagner, Norwegian University of Science and Technology / Department of Biology
While research on the environmental and health risks of microplastics is still in its infancy, the public has already concluded there are unacceptable risks and, consequently, demands for action. This puts environmental toxicologists and chemists in an uncommon position, namely that public awareness of a potential environmental issue is way ahead of an evidence-based assessment of the actual risks. To further complicate the matter, researchers face a fundamental dilemma: Current narratives on the negative implications of (micro)plastic pollution create public awareness and promote change towards more sustainable economic practices, e.g., with regard to circular economy. However, these narratives are in many cases not backed by scientific evidence. The question is now: How can we promote positive societal change and at the same time stay true to the scientific principles? In my talks, I will not present final answers to this question but rather provide a diagnosis of the recent microplastics debate. I will argue that plastic pollution represents a challenge to our disciplines with regard to the following fundamental aspects: 1) absence of a common risk understanding, 2) bias when dealing with information-scarce situations, 3) lack of mechanisms to prioritize environmental issues, 4) lack of mechanisms for consensus-building regarding the risk of environmental stressors. I will further argue that the field of plastics pollution represents an ideal playing ground to explore, discuss and advance these aspects. This will be crucial to get our disciplines fit to deal with the wicked problems, we face in the Anthropocene.

546
Ocean Literacy – changing attitudes and behaviour of society in the face of the problems of the oceans
A. Borja, Azti-Tecnalia / Marine and Coastal Environmental Management
The H2020 project ResponSEAble (www.responseable.eu) is trying to raise awareness around six key-stories (fishing, eutrophication, renewable energies, coastal tourism, microplastics, and ballast waters), within the four European regional seas. Under the DAPSIWRM framework (Drivers, Activities, Pressures, State, Impact, Wellbeing, Responses, Measures) we are developing products to change attitudes and behaviour of different actors related to each story, but also to the society at large, from children to adults, trying to test the changes in those attitudes. The products include videos, cartoons, serious games, interactive tools, specialized courses, etc. Our aim is that if scientists and society have a shared understanding of science and risk communication regarding the problems of the oceans, these can be solved through the individual and collective changes in our attitudes towards the oceans in our daily lives.

547
Discussion Microplastics

548
General discussion with panel of all speakers about topics emerging from the session

549
Wrap-up and closing
A. Leopold, Calidris Environment BV / Calidris Environment BV; T. Seiler, RWTH Aachen University / Ecosystem Analysis; C. Ajao, ECHA-European Chemicals Agency

Hazard and exposure assessment of chemical mixtures: steps towards increasing the realism of chemical risk assessment (I)

550
Development of a diagnostic toolbox for ecological effects of pollutant mixtures and application to evaluate results from the third Joint Danube survey
A. Focks, Alterra Wageningen University and Research Centre / Environmental Risk Assessment Team; T. Seiler, RWTH Aachen University / Ecosystem Analysis; P. van den Brink, Alterra/Wageningen UR / Aquatic Ecology and Water Quality Management Group b Alterra; B. Deutschmann, RWTH Aachen University

/ Institute for Environmental Research Biology V; H. Hollert, RWTH Aachen University / Institute for Environmental Research; S. Kaišarević, Faculty of Sciences University of Novi Sad / Department of Biology and Ecology, Laboratory of Ecotoxicology (LECOTOX); I. Teodorovic, University of Novi Sad / Department of Biology and Ecology, Laboratory of Ecotoxicology (LECOTOX); T. Backhaus, University of Gothenburg / Department of Biology and Environmental Sciences

Toxic chemicals from point and diffuse sources might impact the ecological status of aquatic ecosystems. Appropriate strategies are needed to identify impacted sites, quantify impacts, or evaluate the causative involvement of chemical contaminants. Since environmental compartments usually contain mixtures of chemicals with low, possibly non-toxic concentrations of the individual compounds, any approach to identify casual links between ecological impacts and chemical contamination has to involve concepts for mixture toxicity. However, in addition to toxic chemicals, other non-chemical stressors such as habitat degradation, nutrient pollution, oxygen depletion, pH shifts, hydromorphological changes or others, may also cause a site to fail achieving good ecological status. Since the EU Water Framework Directive (WFD) aims at a good ecological status of all European water bodies through addressing water pollution, for water quality monitoring and assessment under WFD it is necessary to discriminate the impact of such non-chemical stressors from the effects of toxic chemicals. This is challenging, and no single "one size fits all" strategy exists. Therefore, multiparametric approaches, so-called "toolboxes", are often used. This presentation will show a toolbox for the detection of the ecological impact of chemicals that was developed within the Solutions EU project. It uses a statistically supported, transparent and formalized weight of evidence (WOE) approach. The developed toolbox was applied to the Danube case study, to facilitate evaluation of the very comprehensive data set from Joint Danube Survey 3. The toolbox concept proved to be practical, simple and promising for further studies, with fairly high diagnostic power

551

How can we identify "drivers of mixture risks"?

T. Backhaus, University of Gothenburg / Department of Biological and Environmental Sciences; R. Altenburger, UFC Centre for Environmental Research / Department Bioanalytical Ecotoxicology; W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; M. Faust, Faust & Backhaus Environmental Consulting; A. Kortenkamp, Brunel University London / Institute of Environment, Health, and Societies; L. Posthuma, RIVM / Centre for Sustainability, Environment and Health

Mixtures relevant for human health or the environment can easily contain dozens or even hundreds of chemicals. However, those components do not contribute equally to the mixture risk. In fact, empirical evidence seems to point to the fact that often only a very few chemicals dominate the mixture risk. The European Commission has therefore emphasized in its communication the need to "*identify chemical substances that are the main drivers of mixture toxicity*". This could tremendously help to steer future chemical monitoring efforts and risk mitigation measures. However, it is currently unclear how a common definition of the term "driver of mixture risk" for human health and environmental assessments could look like, and how the concept could be operationalized. In the presentation, we will provide the background of existing approaches to define "mixture risk drivers" and explore the consequences of their application to a real-world dataset (Swedish pesticide monitoring data). In particular, we will demonstrate that the use of Concentration Addition, which is common in all approaches, might not always be justifiable for the ranking of mixture components. Additional we will discuss: (i) the sensitivity of the various methods to data gaps, (ii) the challenge of tiering the various approaches for risk driver identification, and (iii) the question whether a risk-based ranking is the optimum approach, or whether hazard- or exposure-based methods can be suitable alternatives. In summary, we conclude that, although the identification of drivers of mixture risk would constitute a major step forward to systematize and simplify the seemingly overwhelming complexity of chemical exposures encountered during human health and environmental risk assessments, the concept is currently not yet fit for purpose. Important steps remain to be taken, in order to better conceptualize and operationalize the concept. However, more and more empirical data become available from monitoring studies that provide ample material to explore the applicability of the concept and its consequences for risk management.

552

Application of new statistical distribution approaches for mixture risk assessment

A. KIENZLER, JRC-EC / F3-Chemical Safety and Alternative Methods Unit-EURL ECVAM; M.G. Barron, U.S. EPA / Gulf Ecology Division; A. Beasley, Down; S.E. Belanger, The Procter & Gamble Company / Environmental Stewardship and Sustainability Organization; S. Bopp, EC-JRC / F3-Chemical Safety and Alternative Methods Unit-EURL ECVAM; M.R. Embry, ILSI Health and Environmental Sciences Institute (HESI)

Wildlife is exposed to an infinite number of different combinations of chemicals. There is evidence that single substances that are present below their individual thresholds of effect can still be of concern and contribute to combined effects. A framework for environmental mixture risk assessment (MRA) has been suggested by Backhaus and Faust (2012), however MRA is often hampered by limited data availability. An ecological Threshold for Toxicological Concern(ecoTTC) approach has been recently developed based on a database of more than 100 000 acute and chronic aquatic toxicity data. The tool allows for the calculation of predicted no effect concentrations (PNEC) derived from the underlying data to which an assessment factor (AF) is applied depending on the comprehensiveness of the selected dataset and the geographical area considered. The result is a PNEC distribution, from which the ecoTTC value is derived by calculating the fifth percentile. Other types of chemical toxicity distributions (CTD) are also possible i.e. distribution of acute (LC50) or chronic (NOEC) ecotoxicological data without applying any AF. It is common practice to predict combined effects and risks based on information of the mixture components, most of the time based on the concept of concentration addition (CA). For this case study the sum of risk quotients has been used as a surrogate for CA based predictions. The risk quotient for the mixture (RQ_{mix}) is based on the summation of the risk quotients of the individual substances. This approach is conceptually different from CA because the involved PNECs might be based on different groups of species and using different AF. However, it can be used as a screening level approach. If a $RQ_{mix}>1$ is identified, the MRA can be refined by using the sum of toxic units, based on LC50 data. The case study is based on chemical monitoring data in European rivers, which give realistic environmental concentrations and co-exposure scenarios to a relevant number of chemicals. Available ecotoxicological values have been gathered for the identified chemicals from regulatory sources when available, or from the literature and existing database. The possible use of the ecoTTC approach and other type of acute and/or chronic CTD for screening level and data gap filling is explored in this case study, within the framework for environmental MRA previously mentioned.

Backhaus, T. and Faust, M., 2012. Environ ScTechol 46 (5), 2564-2573

553

Towards the development of a framework for applying non-target chemical analysis data within exposure and risk assessment

T. Gouin, TG Environmental Research / Safety and Environmental Assurance Centre; R. Parmar, ARC Arnot Research Consulting; J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology

There is an increasing trend towards multi-targeted and non-target analysis (NTA) screening methods to increase the number of analytes monitored in biomonitoring and environmental samples. While the opportunities from advances in chemical analytical capabilities have shown substantive development over recent years, application of information related to data reported from NTA represents a challenge to the field of exposure modelling. For instance, there is no framework for interpreting and using data reported from studies involving NTA to inform exposure and risk. The absence of guidance may consequently lead to difficulties in prioritizing risk assessment activities. In this study we examine the state of the science with respect to NTA, and present a summary of the merits and limitations for exposure assessment and risk assessment. These preliminary observations are then used to propose an initial framework for the appropriate use of NTA data within exposure assessment. Recommendations include a number of suggestions regarding how these data can be better gathered and reported in order to strengthen their applications for chemical exposure and risk assessment, including emerging contaminants.

554

A common framework for the assessment of human and ecological risks from pollutant mixtures in European surface waters - case study with > 300 chemicals co-occurring in the Danube

A. Kortenkamp, Brunel University London; M. Faust, Faust & Backhaus Environmental Consulting; S. Ermler, Brunel University London / Institute of Environment, Health and Societies; D. De Zwart, DdZ Ecotox / Centre for Sustainability Environment and Health; L. Posthuma, RIVM / Centre for Sustainability, Environment and Health; T. Backhaus, University of Gothenburg / Department of Biology and Environmental Sciences

Experimental mixture studies have shown that the toxicity of a mixture is usually greater than that of the most toxic component and that substantial mixture effects can occur even though all components in the mixture are present at levels that individually are without observable effects. These observations have lent urgency to the need of evaluating the risks from multiple pollutants both to humans and wildlife. Here, we present a common decision tree and tiered work flow scheme for performing human and ecological mixture risk assessments (MRA) in the context of assessments of multiple pollutants in European rivers. The scheme focuses on MRAs for humans and aquatic species groups. It uses measured concentrations of chemicals co-occurring in water and builds on the principle of a tiered approach, where unnecessary expenditure of resources is avoided by discontinuing the analysis when cumulative exposures are judged to be acceptable on the basis of crude and simple worst-case assumptions. The analysis is refined when previous tiers reveal clearly unacceptable exposures, with refinements based on best-case assumptions of minimum expectable risks. The workflow is divided into three main tiers in which the distorting influence of different assessment factors present in regulatory values is successively removed, and increasingly sophisticated assumptions about modes of action are introduced. We tested the utility of the scheme by using data on the levels of more than 300 chemicals that occur together in the river Danube, from the Joint Danube Survey 3 (JDS3). For each of the 54 sites along the river Danube we ranked the chemicals in terms of their contribution to a mixture effect, separately for algae, daphnia and fish. We found that the overall mixture toxicity was driven by only approximately 10 chemicals. Substances not yet defined as priority substances under the EU Water Framework Directive made a substantial contribution to combined exposures. We also assessed possible combined risks to humans by evaluating whether water drawn from the Danube would be fit for human consumption. Overall, exposures of concern for humans could not be detected at higher tiers of the assessment. We conclude that the protection goals defined in the Water Framework Directive for freshwater aquatic communities are not achieved for combined exposures at many sites along the Danube.

555

Pesticides do rarely come alone, except in risk assessment - Risk indices of ranked spray series of the project COMBITOX

B. Scholz-Starke, RWTH Aachen University, Institute for Environmental Research / Institute for Environmental Research; S. Bär, German Federal Environment Agency UBA / Section Plant Protection Products; B. Daniels, RWTH Aachen University / Institute for Environmental Research; T. Frische, Federal Environment Agency UBA / Section Plant Protection Products; M. Liess, UFZ Center for Environmental Research / System-Ecotoxicology; R. Ottermanns, M. Roß-Nickoll, RWTH Aachen University / Institute for Environmental Research; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics; A. Sybertz, RWTH Aachen University / Institute for Environmental Research; C. Ullrich, German Federal Environment Agency UBA / Section Plant Protection Products; S. Knillmann, Helmholtz Centre for Environmental Research UFZ / System-Ecotoxicology

In the European Union, legislation so far strictly regulates plant protection by means of chemical pesticides on the level of single products. Common agricultural practice and manifold pest pressures lead to the exposure of non-target organisms to complex mixtures in a spray series. In the on-going COMBITOX project, we collated a dataset for the actual application patterns of pesticides in 12 representative crops in terms of acreage and pesticide application rates from the years between 2007 and 2015. The data was used for classification of sprayseries by typical mixture patterns, sequences and toxic pressures. Combining all information on empirical use patterns and their regional and temporal variation, we calculated indicators of use intensity and environmental risk. Toxicity exposure ratios (TER) were calculated using standard toxicity data (aquatic & soil organisms) from two publicly available databases PPDB (Lewis et al. 2016) and ECOTOX (US EPA 2017) and the mere application rates without consideration of exposure pathways. Only for focal sprayseries, first and higher-tier tier risk indices (including mandatory risk management measures) for different compartments were computed according to the standard approach as conducted by the UBA within the PPP-authorization procedure in Germany. In sum, 29 risk indices were computed (TER-values). Mixture risk indices were calculated based on the concept of concentration addition from single-substance TER and summed up per application date. In general, patterns of pesticide use showed that tank mixtures and spray sequences are predominantly in all crops that were considered in our studies. From the data, we revealed that crops group together in classes of use patterns. The cereals received a wide range of different pesticides classes in medium intensities during the whole spraying sequence; vine and apple orchards were dominated by fungicides and high-intensities. Risk exceedances became relevant if multiple individual TER-values (TERsingle) were already close to the critical TER trigger values. Our results emphasize the relevance of the nowadays largely non-regulated tank mixtures for the risk assessment of non-target organisms. In conclusion, we clearly see the necessity to consider realistic exposure assessments of typical treatment regimens as well as effect estimates from appropriate mixture toxicity models in order to describe the "total risk" of the common chemical plant protection practice.

Advances in Soil Ecotoxicology and Risk Assessment of Terrestrial Ecosystems (I)

556

Time-dependent effects of two fungicides and their mixture on enchytraeid and earthworm communities under field conditions

J. Amossé, S. Bart, INRAAgroParisTech; C. PELOSI, INRA (Institut National de la Recherche Agronomiqu; P. Thiel, INRA; A. Péry, INRAAgroParisTech

According to the current regulation for the registration of plant protection products on the market, the environmental risk assessment of pesticide use is generally performed under laboratory conditions. Very little information is available *in natura*,where multiple stresses occur. In this study, we assessed the effects of two commercial formulations of fungicides, i.e., Cuprafor Micro® (composed of 500 g.kg^{-1} copper oxychloride) and Swing Gold® (composed of 50 g.l^{-1} epoxiconazole and 133 g.l^{-1} dimoxystrobin), and the mixture of both on two groups of terrestrial oligochaetes (*Lumbricidae* and *Enchytraeidae*) after 1, 6 and 12 months (i.e., t1, t6 and t12) of exposure under field conditions. We also assessed the feeding activity of soil organisms using the bait lamina method. Our results showed a lower Shannon index for earthworms in the treatment with the mixture of both pesticides (i.e., 1,5 l. ha^{-1} of Swing Gold® and 4 kg.ha^{-1} of copper) and in the treatment with the Swing Gold® at ten times the recommended dose (i.e., 15 l. ha^{-1}) after one and six months. We also found a lethal effect of Swing Gold® on anecic earthworms at t1, while an effect of copper on annelids at ten times the recommended dose (40 kg.ha^{-1} of copper) was observed later at t12. We showed no overall significant difference in total feeding activity, enchytraeid density and diversity between treatments with or without pesticide at different sampling periods. In the Swing Gold® treatment, earthworm community did not recover twelve months after pesticide application. We suggest thus going beyond the ISO norm 11268-3 (2014) - for the study of the effects of pollutants on earthworms under field conditions - by studying *Oligochaeta* community and other functional endpoints (e.g. organic matter decomposition with the tea bag method) over two years to better assess environmental risks of plant protection product use and their mixture. Keywords: Cuprafor Micro®, Swing Gold®, agroecosystems, feeding activity

557

Toxicity of imidacloprid and thiacloprid towards four Collembolan species

C. Lima, Vrije Universiteit Amsterdam / Animal Ecology; C. van Haren, G. Mainardi, Vrije Universiteit Amsterdam / Department of Ecological Science; C. van Gestel, Vrije Universiteit Amsterdam / Ecological Science

Folsomia candida has been used for assessing the toxicity towards non-target soil invertebrates since the 1960s, but only in the 1990s a standard reproduction test was developed. In 2009, after a ring test, OECD also accepted *Folsomia fimetaria* as a model organism The first species, has been transported all over the world, therefore being considered a tramp species, having a parthenogenetic mode of reproduction. *F. fimetaria* is present in most of natural and agricultural soils worldwide, and has a sexual mode of reproduction. Following a suggestion of the ring test to use different species of springtails to assess the toxicity of contaminants, in this study two additional species, *Heteromurus nitidus* and *Sinella curviseta*, were used together with *F. candida* and *F. fimetaria* to determine the toxicity of imidacloprid and thiacloprid in Lufa 2.2 soil. The tests aimed at answering 2 main questions: (i) Is there a difference in the sensitivity to neonicotinoids between the different species?; (ii) Are these species suitable for assessing the toxicity of neonicotinoids? Imidacloprid was most toxic, with *F. fimetaria* presenting around the same sensitivity as *F. candida* for survival (LC_{50} 0.56 mg/kg dry Lufa 2.2 soil), and a slight difference in the sensitivity for reproduction, with EC_{50}s for *F. fimetaria* of 0.10 mg/kg dry soil and for *F. candida* of 0.26 mg/kg dry soil. *H. nitidus* was slightly less sensitive with an LC_{50} of 1.6 mg/kg dry soil and an EC_{50} of 0.40 mg/kg dry soil. Thiacloprid was tested on *S. curviseta*, *F. candida* and *H. nititdus*, with survival of the first one being least sensitive (LC_{50} 27 mg/kg dry soil), followed by *F. candida* (LC_{50} 5.2 mg/kg dry soil) and *H. nitidus* being the most sensitive with an LC_{50} of 2.3 mg/kg dry soil. Thiacloprid was more toxic to the reproduction of *S. curviseta* (EC_{50} 2.6 mg/kg dry soil) followed by *F. candida* (EC_{50} 1.5 mg/kg dry soil), and *H. nitidus* (EC_{50} 1.3 mg/kg dry soil). The different species tested presented similar sensitivity towards the neonicotinoids for both endpoints, with the exception of *S. curviseta*. The results suggest a specific mode of action of thiacloprid towards reproduction, a trend that has been found in all tests, except for of *H. nitidus* that presented around the same sensitivity to both survival and reproduction. The species tested presented good control performance and consistency in the results, pointing towards a possibility to be used in toxicity tests.

558

Dirty dancing: measuring mite movement responses to pesticide residues

J. Witton, Environment Dept, University of York / Environment; T. Alvarez, EcoRisk Solutions Ltd / Dept of Ecological Sciences; M. Reid, HSE Health and Safety Executive / Chemicals Regulation Division; G. Weyman, ADAMA; M. Hodson, University of York / Environment Department; R. Ashauer, University of York / Environment

For a pesticide to be registered for use, the lethal and sublethal effects on non-target arthropods (NTAs) must be studied. Sublethal effects such as behavioural changes have been reported in NTAs exposed to some pesticides, with avoidance behaviour – where individuals display signs of irritation or repellence when exposed to a pesticide – being of particular interest. More research is necessary to better understand pesticide avoidance behaviour so that population consequences of such behaviour can be estimated. We aimed to develop an efficient method to quantify changes in movement behaviour and identify avoidance behaviour in relation to pesticide exposure in the predatory mite *Typhlodromus pyri*, a model species and natural predator found in fruit orchards across the globe. Using video analysis, we exposed individual adult mites to 3 insecticidal active ingredients (acetamiprid, deltamethrin, dimethoate), each at 3 concentrations, and evaluated mite movement behaviours when exposed to these in comparison to a control arena. We found that distances walked by mites were reduced by up to 87% compared to the control when exposed to 0.1 μg mL^{-1} deltamethrin, and that 54% of individuals exhibited repellence through becoming trapped in the test arena glue boundary at this concentration compared to 0% in the controls. When exposed to 18 μg mL^{-1} acetamiprid mean distance covered fell by 34%; however, when exposed to 0.45 μg mL^{-1} dimethoate the mean distance covered increased by 11%. No individuals exhibited avoidance behaviour when exposed to acetamiprid or dimethoate. We report the variable effects of 3 insecticides on a range of standard movement behaviours in *T. pyri*, including distance walked, time moving/not moving, velocity and meandering behaviour. We also report avoidance behaviour measured as the time taken to become trapped in the test arena glue boundary due to attempts to escape the arena. Our results complement existing knowledge of sublethal pesticide effects in NTAs by quantifying movement behaviour changes in *T. pyri*. We are also adding to the knowledge base relating to avoidance behaviour, which is an area of growing interest. We hope to improve the understanding of population-level consequences from changes in movement behaviours caused by pesticide exposure.

559

Should oral exposure in Hypoaspis aculeifer tests be considered in order to keep them in Tier I test battery for ecological risk assessment of PPPs?

T. Natal-da-Luz, CFE - Centre for Functional Ecology / Department of Life Sciences; T. Gevaert, CFE   Centre for Functional Ecology and Hogeschool Gent, Education, Health and Social Work, University College Ghent; C.S. Pereira, CFE - Centre for Functional Ecology / Department of Life Sciences University of Coimbra; M. Arena, EFSA - European Food Safety Authority / Pesticides; J. Sousa, University of Coimbra / Department of Life Sciences

The recent scientific opinion of EFSA addressing the state of the science on risk assessment of plant protection products (PPPs) for non-target arthropods highlighted the need for the inclusion of other relevant exposure routes, besides contaminated soil, in tests from lower tiers. The reproduction test with the predatory mite *Hypoaspis aculeifer* (OECD 226) is currently being included in the new EU data requirements for the ecological risk assessment (ERA) of PPPs. However, the low sensitivity often shown by this mite towards PPPs, when compared to other invertebrates, makes the test with this species, as it is currently performed, not very useful for tier I test battery. The current test protocol does not take into account the fact that *H. aculeifer* is a predatory species, and only considers exposure to contaminants via contaminated soil, disregarding exposure via contaminated food. Therefore, an adaptation of the test performance, by including exposure via contaminated food, is necessary. The methods described in the standard protocol for mite reproduction test advise feeding the test organisms with fresh preys (e.g. cheese mites *Tyrophagus putrescentiae*) from uncontaminated breeding containers over the test period but, in a real scenario, this exposure is simultaneous for *H. aculeifer* and their preys. Thus, through this protocol, the toxicity of contaminants to *H. aculeifer* might be underestimated. The present study aimed to evaluate the importance of oral exposure to the contaminant as an exposure route to be considered in reproduction tests. Two reproduction tests with *H. aculeifer* were performed (OECD 226) using artificial soil spiked with increasing concentrations of copper (Cu): 0, 450, 675, 1013, 1519, 2278, 3417 and 5126 mg kg^{-1}. Cheese mites were used as food in both tests but, while in one test cheese mites obtained from normal laboratory breeding cultures (clean preys) were added, in the other test, cheese mites previously exposed to Cu (pre-exposed preys) were used. Results showed that *H. aculeifer* fed with pre-exposed preys were more sensitive to Cu than mites fed with clean cheese mites. These data support that the route of exposure represented by feeding on contaminated preys should be taken into account when using reproduction tests with predatory mites for ERA of PPPs. This enhance the need for a revision of the procedures described in the standard protocol for mite reproduction test to avoid underestimation of toxicity of contaminants.

560

Plant protection products in agricultural soils - Do active ingredients show a comparable pattern in worms and in soil?

T. Schmidt, IES Ltd / Ecotoxicology; H. Viric Gasparic, University of Zagreb Faculty of Agriculture / Department for Agricultural Zoology; R. Bazok, University of Zagreb Faculty of Agriculture / Department of Agricultural Zoology; S. Kimmel, Innovative Environmental Services (IES) Ltd. / Ecotoxicology; S. Höger, Innovative Environmental Services (IES) Ltd / Environmental Toxicology

The environmental risk assessment of plant protection products on soil organisms is mainly based on the outcome of laboratory and extended laboratory studies (EFSA 2017). However, the link from the laboratory to realistic field conditions over several seasons is not well established. Currently no validated trigger for bioaccumulation in soil ecosystems is available (EFSA 2017). One possible approach for filling this gap is proposed by combining experimentally determined residue data from earthworms and data from degradation studies in the field. Earthworms were sampled at different seasons from eight fields in Croatia and analysed for 26 active ingredients. The concentrations of 26 analysed active ingredients were 0.000-0.247 mg/kg worm fresh weight with a mean of 0.005 mg/kg. The percentage of samples with values below the limit of detection (LOD), values below the limit of quantification (LOQ = 0.001 mg/kg) and values above LOQ was 29, 42 and 29 %, respectively. Based on publicly available draft assessment reports from EC and EFSA, degradation parameters (DT_{50}, DT_{90}) were used to calculate degradation curves and the current concentration in soil at the date of worm sampling. By comparing analysed residues in worms and calculated concentrations in soil, a substance-specific bioaccumulation factor could be calculated. In the case of imidacloprid, the analysed residue levels in earthworm samples from the fields tended to increase with time whereas the calculated concentrations in soil decreased with time as expected, resulting in a supposed increasing bioaccumulation potential of imidacloprid under field conditions. The procedure proposed here – in the absence of analysed soil data – is a simple estimation which combines field history data with data from publicly available draft assessment reports. This approach may be useful for the assessment of the bioaccumulation potential of an active ingredient from a plant protection product under realistic field conditions.

561

PBT assessment of substances - Proposal of a trigger value for bioaccumulation in terrestrial oligochaetes

M. Simon, Fraunhofer IME / Applied Ecology; C. Schlechtriem, Fraunhofer IME / Department Bioaccumulation and Animal Metabolism; P. Egeler, ECT Oekotoxikologie GmbH; M. Weil, ECT Oekotoxikologie GmbH / Institute for Technology Assessment and System Analysis ITAS; J. Römbke, ECT Oekotoxikologie GmbH; B. Karaoglan, German Environment Agency   UBA; A. Wiemann, UBA Umweltbundesamt; W. Drost, Federal Environment Agency (UBA) / Chemicals

Assessment and regulation of PBT (Persistent, Bioaccumulative and Toxic) substances, are necessary to ensure a high level of protection of human and animal health, and of the environment. In aquatic organisms, trigger values for the identification of bioaccumulative (“B”) and very bioaccumulative (“vB”) substances are bioconcentration factors (BCF) of >2000 and >5000, respectively, obtained from fish flow-through studies according to OECD 305. However, Annex XIII of the REACH regulation does not define similar trigger values for bioaccumulation in terrestrial organisms. The objective of this project is to provide a suitable data base which will help to clarify how to address terrestrial bioaccumulation in the B assessment and to define trigger values for the bioaccumulation factor (BAF) obtained from bioaccumulation studies with terrestrial oligochaetes according to OECD 317 that are comparable to the B/vB criteria in the scope of the PBT guidance revision. For this aim, the study comprised the following three steps: 1) Literature research on available bioaccumulation factors (BAFs) both in open scientific literature and in regulatory data from several OECD 317 studies and performance of correlation analysis between soil-/substance-properties, BCF and BAF values. 2) Performance of bioaccumulation studies according to OECD 317 with the earthworm *Eisenia andrei* using the four model substances endosulfan, methoxychlor, o-terphenyl and PCB153. 3) Proposal of a trigger value for identification of bioaccumulative substances in terrestrial organisms based on the literature research and the performed experimental results. The results clearly showed that organic carbon content of the test matrix used in the tests influences the BAF, whereas no clear correlations between log K_{OW}, log K_{OC} or similar substance properties and the BAF were observed. Additionally, no correlation was observed between substance-specific BCF from fish studies and BAF determined with earthworms. Therefore, lipid- and C_{org}-normalized BSAF should be used for the assessment of terrestrial BAF. Kinetic BSAF from both experimental studies and literature-derived values ranged from 0.21 to 14.8. Based on the data evaluated in the present work, a BSAF trigger value of 1.00 is proposed as a general trigger to indicate bioaccumulation in terrestrial organisms. Other aspects like non-depurated residues at the end of the elimination phase are discussed.

Challenges, methodological developments and practical solutions for Social Life Cycle Assessment in industry and policy

562

Developments and recommendations on the practical use of Social LCA

S. Di Cesare, CIRAD / Department of Economic Studies; A. Zamagni, Ecoinnovazione / LCA and Ecodesign Laboratory; J. Garcia, SCORE LCA; F. Silveri, University of Chieti-Pescara / Department of Economic Studies; A. Lanfranconi, Ecoact; L. Petti, University of Chieti-Pescara / Department of Economic Studies

S-LCA is a multi-criteria, multi-stakeholder and multi-step methodology that provides useful, transparent and science-based information on social and socioeconomic performance of a product throughout its entire life cycle. In this study, a systematic literature review was carried out dealt with within these aspects: the scope of S-LCA, its purpose, the social dimension outside S-LCA (i.e., analytical tools, procedural and managerial tools, currently used for monitoring, assessing, reporting and communicating social aspects, and their main differences with respect to S-LCA); impact assessment methods; users and uses of S-LCA. In addition, an experimentation was conducted through a practical case study based on literature, with the following goals: (1) to test the applicability and practicability of S-LCA; (2) to highlight the capability of the methodology to identify social hotspots along the whole life cycle, and in particular in the remote phases of the life cycle, such as raw material production and end-of-life; (3) to show how those results may complete environmental LCA and other social approaches. The product chosen is a Photovoltaic (PV) Module. The analysis carried out clearly pointed out that S-LCA is an evolving field, and main developments are envisaged, both at the level of methodology and results ‘interpretation and communication. More in detail, the main limits of the S-LCA methodology identified in this analysis are related to: methodological framework for S-LCA, goal&scope definition (in particular regarding the system boundary definition), data access, and use of qualitative data, methodologies and selection of indicators for the impact assessment phase. The strengths of the methodology are related to its capability of making the assessment of the product more complete, adding its social aspects to the environmental and economic ones, in addition to the increased transparency and traceability of product along the value chain. On the basis of the main challenges for S-LCA identified in this study, recommendations were identified, for the further development and implementation of the methodology. The implemented study showed that there is a need for broad improvements of both methodological and communicative issues. To carry out a S-LCA study could be a way to manage social risk thanks to the identification of social hotspots, and to help companies building a targeted strategy for future development of social policies.

563
TBD

564
Social significance analysis of products - considering negative and positive social impacts along the supply chain of leather products
S. Neugebauer, RWTH Aachen University / INaB - Institute for Sustainability in Civil Engineering; M. Traverso, RWTH Aachen
Global textile and especially leather industries facing constant discussions on social compliance mostly in relation to bad working environments and thus cause severe social impacts for different stakeholder groups along the supply chain due to e.g. unhealthy working conditions. In order to determine social impacts occuring during the leather production processes, social life cycle assessment´s (S-LCA) implementation is of major importance. Practical implemetations have to consider indicators and impacts determining social hotspots along the supply chain and should in addition provide information on social challenges and chances by means of negative and also positive social impacts. When assessing products´ life cycles the inclusion of posititve social impacts is crucial, as most of the S-LCA indicators can be both positive or negative. Thus, this study aims at providing a social significance analysis (SSA) determining social hotspots along the leather supply chain including social impacts considering negative as well as positive consequences. Existing social indicators are included, addressing relevant stakeholder groups and impact pathways, e.g. fair wage. New indicators are defined where needed, e.g. to represent the rights for indigenous people affected. The assessments are performed by means of secondary databases, e.g. Social Hotspot Database, and by including primary data gathered at production sites of the European leather producer. The results will provide the challenges and chances of European leather production including the different stakeholder groups affected (e.g. workers) but also positive/negative directions of each social impact category defined (e.g. fair wage as a positive and negative indicator and utility as a purely positive indicator). The SSA is based on the S-LCA of European leather production using a criteria evaluation determining crucial topics along the supply chain, e.g existence of labor laws. Relevant social hotspots are identified. Depending on the indicator direction, social consequences (e.g. benefits in societal health resulting from non-exhausting work hours) can be determined for the stakeholder groups. The inclusion of positive impacts may function as an incentive for improvement and guide the way towards future developments within the European leather industry. The results may also be transferred to further product groups in the global textile and leather industry.

565
Integration of sustainability in industrial research and innovation: perspectives from ArcelorMittal's experience
A. Hettinger, M. Caraty, R. Turconi, ArcelorMittal / Sustainability RD; P. Cortijo, Utopies
The iron and steel industry is both a large contributor to greenhouse gases emissions and a provider of a key material for society's development, being used in a wide range of market sectors such as infrastructure, transport, construction and packaging. Because of its ubiquity and complex supply chain, to promote sustainable development it is essential to ensure efficient production processes, optimizing the use of resources, valorization of byproducts, but also to explore the other stages of lifecycle of products that use steel, i.e. to adopt the holistic approach of Life Cycle Assessment. Lifecycle thinking enables ArcelorMittal - the world's largest steel producer - to promote sustainability not only in its own operations but also in the use of its products by customers. For more than ten years, a research team dedicated to sustainability and lifecycle assessment has been supporting the process and product research within the group. However, with more than 1,000 researchers in 12 research centers globally and hundreds of projects carried out every year, it is impossible for a single team to cover systematically the research performed. To this goal, the "Sustainable Innovation Tool" has been developed to enable the researchers of the group to self-assess their projects sustainability. Using the tool, they evaluate environmental and social aspects of their new processes and products and engage in a learning curve for an improved sustainable performance. The presentation will describe the collaborative development of the tool, the different phases of testing and the current start of the deployment across 6 research programs. We will draw on this experience to provide elements that supported its success, pitfalls to avoid. The company is at the start of this journey and seeks a continuous progress, and possible paths for a better integration between our current assessment of industrial research and generic frameworks such as the sustainable development goals will be discussed.

566
Social footprint of a packaging waste deposit-refund system in Spain
I. Mu�oz, 2.-0 LCA consultants; B. Weidema, Aalborg University; A. Bala, UNESCO Chair in Life Cycle and Climate Change (ESCI-UPF) / UNESCO Chair in Life Cycle and Climate Change. Escola Superior de Comerç Internacional ESCI; P. Fullana, Universitat Pompeu Fabra UPF / UNESCO Chair in Life Cycle and Climate Change Escola Superior de Comerç Internacional ESCI
We present a social footprint assessment of implementing a deposit-refund system (DRS) applied to beverage packaging waste in Spain. In a DRS consumers pay a certain amount in concept of "packaging deposit" added to the price of a product and receive the refund back when they return the used packaging. The social footprint developed by Weidema [2] constitutes a monetary summary measure of income redistribution and the sum of all productivity-reducing externalities related to an activity. It is calculated by a top-down approach using input-output data. This method can be understood as a 'streamlined' social LCA. We applied its two general components: the income redistribution impact (IR): the increase (or loss, if negative) in utility caused by the transfer of money from one societal group to another, andthe productivity impact (loss) from missing governance (PI): the difference between the actual purchasing-power corrected value added and the potential value added when all productivity impacts are internalised. The social footprint of an activity can be defined as SF = IR + PI. We compared two scenarios, namely the current situation for household packaging waste in Spain in 2014 (system A), and a hypothetical scenario (system B) where a DRS is implemented. The functional unit was the total amount of packaging waste to be managed annually.Primary data for the two scenarios were obtained from the environmental and economic studies performed as part of this project. Data to quantify the social footprint were obtained from the database Exiobase v.3.3.10, which was implemented in the software SimaPro. The results show that the social footprint for both systems involves a net social benefit. However system B reduces this benefit by 50% compared to system A. Introducing this DRS system in Spain is expected to lead to a net loss in social benefit compared to the existing system. The social benefit of the increased recycling is more than outweighted by the social costs induced by the activities required to achieve these higher recycling rates (collection manually or automatically of packaging waste in shops and supermarkets and the associated transport). This study is an example of how the concept of social footprint, together with a powerful tool like Exiobase, can pave the way for an operational approach to social LCA, avoiding excessive data requirements and the long lists of impact indicators currently proposed for bottom-up approaches.

567
Poster spotlight: TH226, TH227, TH228

Developments in the use of bioassays for chemical and environmental risk assessment (I)

568
Application of Equilibrium and Toxicokinetic Models to Understand the Behaviour of Organic Chemicals in In Vitro Toxicity Tests
J.M. Armitage, University of Toronto - Scarborough / Physical and Environmental Sciences; J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology
Toxicity testing in the 21st century is expected to rely increasingly on in vitro assays, which now cover a wide range of endpoints including cytotoxicity, receptor binding, protein interactions and DNA binding. In most cases, dose-response relationships from in vitro toxicity tests are reported using nominal concentrations in the test medium despite the known challenges such data introduce for comparing results across different test conditions and between different chemicals and for quantitative in vitro in vivo extrapolation (QIVIVE). Equilibrium partitioning (EQP) and toxicokinetic (TK) models have been proposed in the literature to address some of these issues. The main limitation of equilibrium partitioning models is that instantaneous distribution is assumed and hence uptake kinetics into cells/tissue, cell growth/division and the potential for degradation in the test system cannot be directly included in the calculations. The main objective of this study was to develop a toxicokinetic model (TK) for simulating the behaviour of organic chemicals in in vitro toxicity tests and compare the results to a previously developed EQP model. The toxicokinetic model was applied to a set of hypothetical neutral organic chemicals under different scenarios (e.g., biotransformation half-life) and then the results compared with a previously developed in vitro mass balance modeling tool based solely on equilibrium partitioning. We also applied the

EQP model to a specific ToxCast assay (ACEA_T47D_80hr_negative assay; cytotoxicity) to illustrate the value of the modelling approaches for QIVIVE and hazard/risk assessment. For relatively persistent chemicals (or in cells/tissue with limited metabolic competency), the simulated mass distribution using the toxicokinetic model is similar to the equilibrium partitioning model output for test durations greater than 12 h. In such cases, the EQP modeling approach is deemed sufficient for translating nominal concentrations to more meaningful/reliable toxicity metrics for QIVIVE (e.g., membrane concentrations). Of the 306 chemicals included in the ACEA_T47D_80hr_negative assay simulations, approximately 2/3rds had predicted membrane concentrations in the range expected to result in baseline toxicity (membrane dysfunction/narcosis). Chemicals with predicted membrane concentrations well below the baseline toxicity range may act via a specific mode of action and could therefore be prioritized for further investigation.

569

Experimental exposure assessment in in vitro bioassays for organic acids

L. Henneberger, Helmholtz centre for environmental research - UFZ / Cell Toxicology; M. Mühlenbrink, Helmholtz Centre for Environmental Research GmbH   UFZ / Cell Toxicology; F. Fischer, Helmholtz Centre for Environmental Research GmbH   UFZ; B. Escher, Helmholtz Centre for Environmental Research GmbH - UFZ / Cell Toxicology

Improved assessment of exposure in *in vitro* toxicity assays is essential for the application of *in vitro* data for chemical risk assessment. Equilibrium mass balance models have been developed to convert the reported nominal effect concentrations of the chemicals in the test system to freely dissolved (C_w) or cellular concentrations (C_{cell}), which are considered more meaningful dose metrics than nominal concentrations. *In vitro* exposure assessment might be challenging for pesticides and pharmaceuticals that are organic acids, due to their unusual partitioning behaviour. Hydrophobic acids are typical ligands for serum albumin and are consequently strongly bound to medium proteins in *in vitro* assays, while the affinity to lipids is lower. While reliable models are available to calculate the binding of neutral chemicals to lipid, protein, medium and cells, the binding of organic acids to biological materials cannot be easily predicted. Here we applied a third phase partitioning method to measure binding of organic acids to biological matrices like cell culture media and cell suspensions on the one hand, but also for direct measurement of exposure (i.e., C_w). Because polymers like polydimethylsiloxane that are typically used for solid phase microextraction (SPME) are not suitable for charged chemicals, C18-coated SPME fibres were used in this study, that have been previously reported to have high sorption capacities for charged chemicals. Eight organic acids were chosen for the experiments: diclofenac, 2,4-D, ibuprofen, naproxen, torasemide, warfarin, triclosan, and genistein. For all test chemicals, equilibrium between the SPME fibre and water was established within 4 h and the determined fibre-water distribution ratios were reproducible (SD ≤0.1 log units). Because the sorption of some of the chemicals to the fibres was concentration dependent, it was required to calibrate the fibres for the desired concentration range. The SPME method was applied to measure C_w in cell culture media. At low chemical concentrations the results from the binding experiments agree with the predictions from a mass balance modelling approach. However, saturation of the medium was observed at high chemical concentrations and further experiments will be necessary to investigate for which chemicals and at which concentration levels saturation occurs and if it is required to incorporate non-linear binding into existing exposure models for *in vitro* bioassays.

570

A versatile and low-cost open source pipetting robot for automation of toxicological and ecotoxicological bioassays

L. Nuesser, RWTH Aachen University, Institute for Environmental Research / Department of Ecosystem Analysis; S. Steffens, RWTH Aachen University, Institute for Environmental Research / Institute of for Environmental Research; T. Seiler, RWTH Aachen University / Ecosystem Analysis; E. Salomons, OptiWater; N. Ruchter, Universität Duisburg-Essen / Aquatic Ecology; M. Schumann, University of Duisburg-Essen / Aquatic Ecology; R. Doering, RWTH Aachen University / Institute of Hydraulic Engineering and Water Resources Management; C. Bruell, RWTH Aachen University; H. Schuettrumpf, RWTH Aachen University / Institute of Hydraulic Engineering and Water Resources Management; A. Ostfeld, Technion   Israel Institute of Technology / Civil and Environmental Engineering; H. Hollert, RWTH Aachen University / Institute for Environmental Research; M. Brinkmann, University of Saskatchewan / School of Environment and Sustainability and Toxicology Centre

The contrasting demands of performing bioassays in compliance with regulatory testing strategies, and their potential use in online monitoring of water quality call for automation technology to assist with automated handling and analysis of multiwell plates. Such systems are typically highly sophisticated and thus costly. As a consequence, the availability of pipetting robots, liquid handlers, and stacking units in environmental monitoring is generally scarce. As a potential solution, we developed a simple and low-cost, versatile open-source pipetting robot that has a small footprint. The construction of the pipetting robot was realized mostly using readily available parts, and partly using open-source hardware. We tested its precision in automated 2-fold dilution series and used it for exposure of zebrafish embryos (*Danio rerio*) – a common model species in ecotoxicology - to cadmium (Cd) and permethrin. As expected, concentrations of permethrin rapidly decreased after initiation of static exposures and after each renewal in the semi-static exposure experiments. No such drastic differences were observed for exposures conducted using the pipetting robot.The accuracy of the pipetting steps was generally high. The apparent toxicity was not only greater in zebrafish embryos exposed to permethrin and cadmium using manual semi-static renewal (24 h interval) compared to static exposure, but also greater in embryos exposed using automated semi-static exposure using the pipetting robot (1 h interval). Thus, we were able to confirm that any attempt to keep exposure concentrations as constant as possible will yield more realistic assessments of toxicity. In this respect, exposure using our pipetting robot can be hypothesized to be similar to flow-through exposure, which is, however, typically more labor- and cost-intensive. With minor modifications, the presented system can be used in a variety of different setups and environments. Because its construction and operation are very cost-effective and do not require any specialized personnel, provisioning of instructions to replicate this design has makes automation technology accessible to a much higher number of laboratories around the world.

571

An intestinal fish cell barrier model to assess absorption of poorly soluble organic chemicals in vitro

H. Schug, Eawag - Swiss federal Institute of Aquatic Science and Technology / Environmental Toxicology; F. Begnaud, Firmenich / DRAP; C. Debonneville, Firmenich / Research and Development; F. Berthaud, V. Laubscher, Firmenich SA / DRAS; J. Maner, Eawag / Environmental Toxicology; S. Gimeno, Firmenich / Product Safety and Regulatory Affairs; K. Schirmer, Eawag / Environmental Toxicology

Permeation of organic chemicals from the aquatic environment across cellular barriers is a critical step for accumulation in organisms, such as fish. To better understand the underlying processes, we aim to study the role of the fish intestine as barrier for hydrophobic and volatile chemicals. The function of the intestine in these processes is experimentally impractical to assess on a routine basis. Additionally, hydrophobic and volatile chemicals are difficult to work with, due to their low water solubility and high volatility. Therefore, we here combine a recently developed *in vitro* epithelial barrier model using the rainbow trout (*Oncorhynchus mykiss*) intestinal cell line, RTgutGC, and a newly constructed chamber that enables stable chemical exposure concentrations. In this setup, we measured the permeation of 10 fragrance compounds with a range of volatility (logHLC = -5.8 to -2.2) and hydrophobicity ($logK_{OW}$ = 3.6 to 5.7). The RTgutGC monolayer partly presented a physical barrier for the permeation restricting the fragrance transfer from the apical to the basolateral compartment. The calculated permeation rates across the cell layer combined diffusion controlled permeation and intestinal biotransformation. The involvement of biotransformation within the cell monolayer was further supported by experiments at 4°C and the measurement of cell associated chemical concentration. We determined the chemical distribution in all different compartments of themodel, which correlated with the $logK_{OW}$. The chamber enabled stable exposure concentrations and close to full recovery at the end of the exposure time in the absence of cells. The presence of a chemical sink in the basolateral maintained the concentration gradient and increased the permeation by approx. 1.5 to 3 times, depending on the $logK_{OW}$. At present, there are no comparable intestinal permeation data for fish available, which precludes a direct comparison of the *in vitro* measured rates with *in vivo* observation. However, exactly this unavailability of data highlights the importance of the development of such methodology for uptake studies at the intestinal epithelium. Data derived with this barrier model can help to develop strategies to link *in vitro* permeation with the prediction of bioaccumulation factors for fish. Moreover, the well-defined exposure scenario in our system comprises a prerequisite for studying mechanisms underlying chemical permeation, such as active transport or biotransformation.

572

A new paradigm in water sampling - how can we challenge the needs of effect-based monitoring?

T. Schulze, Helmholtz centre for environmental research - UFZ / Effect-Directed Analysis; L. Beckers, Helmholtz-Zentrum für Umweltforschung GmbH - UFZ / Effect-Directed Analysis; M. Koenig, B. Escher, Helmholtz Centre for Environmental Research GmbH - UFZ / Cell Toxicology; M. Krauss, Helmholtz centre for environmental research - UFZ / Effect-Directed Analysis; m. muz, UFZ - Helmholtz Centre for Environmental Research / Cell Toxicology; P.A. Neale, Griffith University / School of Environment; J. Slobodnik, Environmental Institute; Z. Tousova, Masaryk University / RECETOX; P. Välitalo, Finnish Environment Institute / Laboratory Centre; K. Walz, MAXX Mess- und Probenahmetechnik GmbH; W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis

In vivo and *in vitro* bioassays (effect-based methods, EBM) are increasingly used for the water quality monitoring to complement chemical analysis. In a holistic point of view, sampling is the starting point and an integrated part of the whole analysis workflow. However, sampling for effect analysis is more challenging than for chemical analysis. Thus, the aim of this paper is to discuss (1) the requirements and challenges of sampling for EBM and (2) to present the recently developed large-volume solid phase extraction approach and apparatus (LVSPE) as a promising technology to overcome the disadvantages of traditional sampling techniques with respect to EBM. The first challenge is the demand of water to be enriched which is dependent from the number and extracts consumption of the bioassays used for the assessment. The second challenge is the recovery and carry-over of the potential toxicity from the water sample to the vessels or wells of the bioassay. The third challenge is the representativeness of the sample. The successful implementation of EBM strategies requires the availability of automated sampling devices which allow the sampling of larger water volumes, guarantee the sampling integrity and makes it possible to take representative samples over a longer period or during events such as heavy-rain- and flood-events. A solution to overcome the disadvantages of classical sampling methods and devices is the recently released LVSPE approach and apparatus. It brings the SPE onshore, allows fully automated sample processing and avoids the transport of larger water volumes to laboratory for filtration and extraction. LVSPE was comprehensively assessed with respect to recovery and carry-over of effects. It has been shown that LVSPE is applicable in monitoring and survey programs, to assess surface water and wastewater with effect-based tools and to unravel one of the causes of mutagenicity in the river Rhine using effect-directed analysis. Thus, LVSPE is a promising technology for the implementation of EBM for water quality monitoring in European and worldwide water quality monitoring. LVSPE is commercially available from MAXX GmbH. However, we declare no conflict of interests as all results presented are scientifically justified.

573
Prioritization of non-target screening suspects based on semi-quantitative concentrations and ToxCast in vitro toxicity data
M. Dingemans, A. Brunner, KWR Watercycle Research Institute; K. Baken, KWR Watercycle Research Institute / CWG; A. van Wezel, Copernicus Institute Utrecht University
In addition to target analyses of chemicals in water samples, non-target analyses are increasingly being applied. The aim of this study was to develop an innovative prioritization tool for chemicals of emerging concern for drinking water, by combining HRMS data with high throughput toxicity data from EPA's ToxCast database. To increase the health relevance of the prioritization method, both semi-quantitative concentrations (internal standard equivalents) in the water samples (as a measure of exposure) and toxicity classes based on 5th percentile AC_{50} values (as a measure of hazard) were included as these form the basis for health risk assessment. A procedure to collect chemical-specific toxicity data from the ToxCast database and a scoring methodology for detected suspects were developed and applied to different types of water samples (sewage treatment plant effluent, surface water, ground water and drinking water) to prioritize suspects for identification and further risk assessment. ToxCast data were collected from the EPA's online ToxCast data repository. Assay endpoint AC_{50} values (the concentration at which 50% of the maximum response is achieved) were extracted from the ToxCast database for the tested chemicals (suspects). All ToxCast assays were included in this hypothesis-free analysis. To reduce the impact of very sensitive assay endpoints, the 5th percentile of the range of AC_{50} values of a suspect chemical in ToxCast assays was used as a measure of its toxicity. More than 7000 structures were detected in these water samples by HRMS non-target screening analyses, and these could be linked to >1000 suspects from a curated suspect list of >5000 EU and water relevant chemicals. The ToxCast database contains *in vitro* effect data for 549 of the 1073 suspects present in the water samples. Many suspects were prioritized based on toxicity and semi-quantitative exposure levels that were not prioritized earlier based on exceedance of the Threshold of Toxicological Concern. After confirmation of their identity, the prioritized suspects are candidates for a in-depth risk assessment based on all available toxicity data, for introduction in monitoring programs or for further risk management measures. Standardization of prioritization schemes for suspect screening approaches may be needed for further introduction of these techniques in water quality regulations. Funded by the Joint Research Program of the Dutch water companies (BTO; project 400554-214).

From detection to action: advancements in assessing and managing highly fluorinated compounds

574
Toward the Comprehensive Profiling of Zwitterionic, Cationic, and Anionic Perfluoroalkyl and Polyfluoroalkyl Substances in Firefighting Foam Impacted Soils
G. Munoz, Universite de Montreal / Chemistry; P. Ray, Universite Pierre et Marie Curie; S. Vo Duy, Universite de Montreal / Chemistry; T. Do, Universite de Montreal; S. Mejia, McGill University / Civil Engineering and Applied Mechanics; J. Liu, McGill University / Department of Civil Engineering; S. Sauvé, Universite de Montreal / Chemistry
In recent years, the comprehensive analysis of both aqueous film forming foam (AFFF) formulations and environmental samples aided to reveal the identities of novel classes of perfluoroalkyl and polyfluoroalkyl substances (PFASs). Following the deployment of AFFFs in firefighting training activities or fire emergency response, the soil is typically the first environmental compartment impacted. In such samples, newly identified PFASs could surpass the concentrations of legacy PFASs by orders of magnitude, indicating the need for in-depth characterization of their transport potential and effects in ecosystems. The methods currently available for the analysis of perfluoroalkyl acids (such as PFOS or PFOA) could, however, seriously underperform for certain newly-identified PFASs. Severe discrepancies were noted as regards the extraction efficiency of cationic and zwitterionic PFASs between soils of variable textural class and organic matter (OM) content, which could not be compensated through isotope dilution due to the lack of matching internal standards. If consistent whole-method recovery and accuracy cannot be ensured in a set of environmental samples of variable physicochemical characteristics, any comparison drawn between samples (e.g., inter-site differences) could be questionable. Failure to obtain quantitative recoveries from soils/sediments could also preclude a reliable assessment of environmental fate properties (e.g., soil/water partitioning coefficient, soil/earthworm bioaccumulation factor). Given the aforementioned limitations, the present study set out to propose a suitable preparation procedure for the multi-residue analysis of PFASs in AFFF-impacted soils. A total of 89 PFASs, representing >20 distinct chemical classes previously discovered in AFFF formulations or across AFFF-impacted sites, was therefore evaluated. Multiple extraction methods were assayed to recover PFASs from AFFF-impacted field-weathered soils and soil matrixes amended in-house with AFFFs and aged. The optimized method presented quantitative or near-quantitative PFAS recoveries from diverse soils and limited matrix effects were noted. The method was applied to a limited survey of firefighting training areas in eastern Canada, showing the prevalence of betaine-based (e.g., 6:2 FTAB, 9:1:2 FTB) and amine-based (e.g., PFHxSAm) PFASs at such sites.

575
Investigation of perfluoroalkyl and polyfluoroalkyl substances in products used for building industry as well as industrial textiles and their possible contribution to water contamination
R. Janousek, Hochschule Fresenius, University of Applied Sciences; S. Lebertz, SGS Institut Fresenius GmbH; T.P. Knepper, Hochschule Fresenius, University of Applied Sciences / Chemistry and Biology
Perfluoroalkyl and polyfluoroalkyl substances (PFASs) have been widely studied in environment, outdoor clothes and fire fighting foams. As a consequence of the hazardous environmental properties of some PFASs, such as persistence, bioaccumulation and toxicity, their fate has been widely discussed. Due to their water, dirt and moisture repelling properties, PFASs are suitable for a variety of applications and possess a lot of possible entry pathways that were identified in prior studies. Although, advertisement and material safety data sheets indicate a rather extensive use of PFASs in building materials and industrial textiles, only few studies dealt with investigation of these materials. A total of 23 samples from products used in building industry and 28 industrial textiles have been investigated in the course of this project. Monitoring covered 29 PFASs with a chain length of C4 to C14, including carboxylic acids, sulfonic acids, sulfonamides and fluorotelomer alcohols. PFASs of diverse chain length (C4-C14) were detected in 31 of 51 investigated samples. Concentrations of perfluoroalky acids were up to 430 µg/kg for highly contaminated samples. FTOHs were even detected in concentrations up to several mg/kg. However, FTOHs need to be further investigated since they may be false positives resulting from the low selectivity of utilized transition for FTOH quantification. In addition to performed investigations, rinse of samples of e.g. buildings should be performed to prove suggested entrance pathways. Furthermore, additional method development has to be performed in order to better illustrate entrance pathways.

576
The growing role of seafood consumption for exposures to legacy PFASs Evident in Longitudinal Birth Cohorts from the Faroe Islands
C. Dassuncao, X. Hu, Harvard University; F. Nielsen, University of Southern Denmark; P. Weihe, The Faroese Hospital System / Department of Occupational Medicine and Public Health; P. Grandjean, Harvard University; E.M. Sunderland, Harvard University / School of Engineering and Applied Sciences
Rapid declines in legacy poly- and perfluoroalkyl substances (PFASs) have been reported in human populations globally following changes in production since 2000. However, temporal shifts in exposure sources are not well understood and this is important for mitigating future risks. Here, we report serum concentrations of 19 PFASs (SPFAS) measured in children between 1993 to 2012 from a North Atlantic fishing community (Faroe Islands) where pilot whale is part of the traditional diet. Median SPFAS concentrations in children (ages 5 to 13 years) peaked in 2000 (47.7 ng mL^{-1}) and declined significantly by 14.4% yr^{-1} to 8.7 ng mL^{-1} in 2012. Perfluorocarboxylic acids (PFCAs) with nine or more carbons ($C\geq9$) were strongly associated with mercury in children's hair, a well-established proxy for seafood consumption, especially perfluoroundecanoic acid (PFUnDA, $r = 0.72$). Toxicokinetic modeling revealed PFAS exposures from seafood have become increasingly important (53% of perfluorooctane sulfonate: PFOS in 2012), despite a decline in whale consumption in recent years. A previous study reports PFASs in Faroese drinking water were below detection. We thus infer that even in a major seafood consuming population, declines in SPFAS exposure after 2000 were achieved by the rapid phase out of PFOS and its precursors in consumer products.

577

Membrane-water partition coefficients to aid PFAS risk assessment.
S. Droge, University of Amsterdam/IBED Institute / IBED
Widely varying structures of fluorinated compounds have been detected ubiquitously in humans and the environment. Due to the limited understanding of basic physical-chemical properties of any of these PFASs, risk assessment (RA) models still provide highly uncertain outcomes. Most PFASs are ionogenic, and act as surfactants. As a result, octanol-water partition coefficients (K_{ow}) can not be determined experimentally. Due to the lack of experimental data, QSARs to predict K_{ow} are not properly calibrated for any perfluorinated ionogenic compounds. Furthermore, the dissociation constant (pK_a) of PFASs has proven to be difficult to determine experimentally and is simply unknown for most emerging alternative PFASs. This may lead to high uncertainty on the fraction of ionized and neutral species at a certain environmental pH of emerging PFASs, and the link to the chemical's "hydrophobicity". One of the main applications of a K_{ow} value in RA models is to relate a chemical's "hydrophobicity" to bioaccumulation and toxicity. It is therefore utmost surprising that hardly any data is available on measurements of sorption data to (phospho)lipid, for which assays are readily available and that lack all the concerns about experiments with octanol. Phospholipids are in general the key tissue component to sorbs ionogenic surfactants, and relate directly to baseline toxicity levels and bioaccumulation. The current study evaluates the results of two experimental tools to measure sorption of standard PFAS structures to artificial phospholipids: retention on immobilized phosphoplipid chromatographic column and solid supported lipid membranes. The current study also evaluated to what extent quantum-chemical software COSMOtherm, which does not require experimental calibration data but simply takes 3D-molecular charge densities into account, is able to predict for emerging PFASs both the membrane-water partitioning (K_{mw}) of the ionic perfluor species, and the predictions on pK_a. Whereas COSMOtherm accurately predicts K_{mw} for cationic hydrocarbon surfactants, it strongly misinterprets the membrane affinity of anionic perfluorinated surfactants. It does show promising predictions on pK_a of alternative PFASs (e.g. GenX). The electronegative fluorine atoms exert a great pull on electrons of any ionizable group, thereby e.g. rendering perfluorooctane sulfonamide (PFOSA) to be a strong acid, whereas recent studies and reports suggested PFOSA to be a neutral PFAS.

578

Impacts of ocean circulation on the marine PFOS burden in an era of geographically shifting emissions
C. Wagner, Harvard University / Harvard John A Paulson School of Engineering and Applied Sciences; C. Thackray, Harvard University / School of Engineering and Applied Sciences; X. Zhang, Wisconsin Department of Natural Resources / State Key Laboratory of Pollution Control and Resource Reuse, School of the Environment; E.M. Sunderland, Harvard University / School of Engineering and Applied Sciences
Concerns over the persistence, bioaccumulation potential and toxicity in organisms prompted the inclusion of perfluorooctane sulfonate (PFOS) in the Stockholm Convention in 2009. The ocean is thought to be the terminal sink for most PFOS releases, and high concentrations in marine organisms, as revealed through biomonitoring studies, pose a threat to human and ecological health. The dramatic reduction in PFOS releases around the year 2000 after phase-out of the parent compound to PFOS and its precursors is well documented in Europe and North America. By contrast, some studies have suggested a potential increase in releases from Asian sources, which may drive continued exposure in marine food webs. In our previous work, we developed a PFOS ocean simulation for the North Atlantic. Per capita PFOS emission factors were derived from waste-water treatment plant measurements. This work showed that in 2015, 60 percent of historic inputs from North America and Europe continued to be present in the North Atlantic, whereas 30 percent had been had been transported into the Arctic Ocean and 10 percent to the Tropical Atlantic. Here we extend this work to develop a global PFOS ocean simulation including emissions from China. The global ocean model is forced by historic PFOS releases from 1958-2015 and simulates realistic ocean physics and chemistry. Based on lateral and vertical transport processes and particle associated export we estimate PFOS residence times in the biologically relevant zone of the ocean and present the contribution of different source regions to the oceanic PFOS burden, as well as the importance of precursors. This work will provide insights into future risks associated with shifting source regions and PFOS precursor releases.

579

PFAS pollution at airport sites: point and diffuse sources, fate and transport and remediation
S. Hale, G.D. Breedveld, Norwegian Geotechnical Institute
Using Norwegian airports and fjords as case studies this work will present the identification of point and diffuse source PFAS inputs, the environmental behavior (partitioning) of these compounds in relation to their presence in soil, ground water, surface water and biota as well as suitable remediation methods for different sites. The once highly desirable physicochemical properties of PFAS (resistance to heat, water and oil) has led to a large-scale environmental problem as these properties go hand in hand with a low degradative potential, high persistence, high mobility and toxicity. At airport firefighting training facilities aqueous firefighting foams (AFFF) containing PFAS have been used in order to practice extinguishing fires. This has led to a point source input of pollution of soils, waters and biota in close proximity to these areas. The use of PFAS containing substances by the textile and paper making industries, as well as their presence in waste sent to landfills represent several diffuse source pollution pathways. The case studies presented encompass all of these inputs, and methods used to monitor the behavior of PFAS in soil, ground water, surface water and biota (including the use of passive samplers) will be shown. Understanding the partitioning and leaching behavior of these compounds allows more informed regulatory decisions to be made and given that the regulation of PFAS is currently under the spot light this is of great importance. Perfluorohexane sulfonate were recently included on the list of Substances of Very High Concern in REACH. PFOS is included in the water framework directive and an environmental quality standard is often used in order to set clean up targets. The remediation of PFAS impacted sites presents unique challenges and current remediation of water often relies on pump and treat using activated carbon filters to sorb PFAS. There are fewer suitable remediation methods for soils. Through the case studies presented, different remediation methods that are currently being used in the field and lab (pump and treat and sorbent amendment) will be presented. Sorbent amendment has been shown to be a promising approach with reductions of PFAS leaching up to 99 %.

Improvements in environmental exposure assessment: Development and application of tools across industry sectors, regulatory agencies, and international boundaries (I)

580

Environmental fate and exposure models: Advances and challenges in 21st century chemical risk assessment
M. Scheringer, ETH Zurich / Institute for Chemical and Bioengineering; A. Di Guardo, University of Insubria / Department of Science and High Technology; T. Gouin, TG Environmental Research / Safety and Environmental Assurance Centre; M. MacLeod, Stockholm University / Department of Environmental Science and Analytical Chemistry
Environmental fate and exposure models are a powerful means to integrate information on chemicals, their partitioning and degradation behaviour, the environmental scenario and the emissions in order to compile a picture of chemical distribution and fluxes in the multimedia environment. A 1995 pioneering book, resulting from a series of workshops among model developers and users, reported the main advantages and identified needs for research in the field of multimedia fate models. Considerable efforts were devoted to their improvement in the past 25 years and many aspects were refined: the inclusion of nanomaterials among the modelled substances, the development of models at different spatial and temporal scales, the estimation of chemical properties and emission data, the incorporation of additional environmental media and processes, the integration of sensitivity and uncertainty analysis in the simulations, etc. However, some issues are still challenging and require research efforts and attention: the need of methods to estimate partition coefficients for polar and ionizable chemical in the environment, a better description of bioavailability in different environments as well as the requirement of injecting more ecological realism in exposure predictions to account for the diversity of ecosystem structures and functions in risk assessment. Finally, to transfer new scientific developments into the realm of regulatory risk assessment, we propose the formation of expert groups that compare, discuss and recommend model modifications and updates and help develop practical tools for risk assessment.

581

Development and update of environmental exposure assessment tool EUSES for REACH and BPR Regulations
S. Frattini, ECHA-European Chemicals Agency; R. Cesnaitis, European Chemicals Agency; H. Schimmelpfenning, European Chemicals Agency ECHA; H. Magaud, European Chemical Agency ECHA
Introduction Both REACH Regulation and the Biocidal Products Regulation requires that the chemicals addressed by the respective legislations are used safely. Both regulations require environmental exposure and risk assessment of chemicals. EUSES (the European Union System for the Evaluation of Substances) is a tool developed by authorities in the 90's to support environmental exposure and risk assessment chemicals in line with the methods described in the technical guidance document (TGD 2003) that harmonised the assessments practices for these three policy areas at that time. EUSES last relevant update was in 2004, so that the tool is now partly outdated. An update is needed since the availability of an up-to-date tool is critical for both REACH and the BPR to function efficiently, both for applicants/registrants, MSCAs and ECHA. EUSES has several modules (release estimation, fate and distribution, effect assessment and risk characterisation). Fate and distribution module (including interaction with the release scenarios) as well as release estimation module are in the focus of the update process. **Update needs and developments** The release, fate and distribution modules should be improved based on the development having occurred over the last years, in particular: Implementation of existing and newly developed emission scenario documents (ESD) for biocides in the release module of EUSES. Complementing the release scenarios covered by EUSES. Expanding the applicability domain and exposure

estimation capability of the model. Alignment of the exposure estimation methods to the scientific developments over the last 20 years. Implement the new module of SimpleTreat (4.0). Exploring how to address site specific assessment by EUSES, like for example within authorisation process. A process has been initiated by ECHA with stakeholders to assess the need for update of EUSES. The kick off for this update process is a workshop with stakeholders which will take place in ECHA, in April 2018. The expected outcome of the workshop is the identification and prioritisation of the update needs and inputs on the update process set up (scientifically and IT support/setting). Workshop outcomes planned to be available before May 2018 and will be presented to the wider audience of the SETAC conference.

582

Advances in exposure assessment of fertilizers: development of a fertilizers environmental exposure tool and generic exposure scenarios under REACH

L. Della Pietra, Fertilizers Europe; S. Navis, Arche consulting / Laboratory of Aquatic Ecology, Evolution and Conservation; R. Puska, Yara Suomi; M. Bjørgan, Yara International ASA; K. Oorts, ARCHE

Fertilizers are considered as formulations and therefore no registration or chemical safety assessment is required under REACH. However, use of the individual constituents needs to be covered in the chemical safety reports (CSR) of the respective substances. Currently, exposure and risk assessment of fertilizer uses is mainly described in a qualitative way because of the lack of appropriate environmental release categories (ERCs) and exposure models. Under the umbrella of Fertilizers Europe and the FARM REACH consortium, the fertilizer sector has developed a fertilizer sector uses map. In addition, four sector specific ERCs (SPERCs) were developed, by grouping similar uses, mainly based upon their physical form and application mode. Next, a Fertilizers Environmental Exposure tool (FEE) tool was developed, since in the standard REACH models for environmental exposure assessment (EUSES, ECETOC TRA, CHESAR), no local scenario for direct emissions to soil exists and direct releases from treated agricultural fields to surface water are not taken into account. In addition, important output pathways for fertilizers via crop uptake and harvest are generally not considered in these tools. Quantitative exposure scenarios, resulting in the calculation of realistic worst-case local Predicted Environmental Concentrations (PEC local) for fertilizer constituents in the various environmental compartments (soil, water, sediment) were established. The main focus of the FEE tool is on micronutrients such as manganese, copper and zinc, which are identified as hazardous for the aquatic environment; but the tool allows for assessment of other inorganic and organic substances in fertilizers as well. Conceptually, the tool has been based upon existing REACH exposure modelling, but is adapted for fertilizer uses by adopting relevant information of fate models from other chemical legislations. In order to improve harmonization and communication within the supply chain, generic exposure scenarios have been developed for a number of micronutrient and SPERC combinations. Collectively, the development of SPERCs, the fertilizers environmental exposure tool and generic exposure scenarios, allow for a systematic review of environmental exposure assessments of fertilizer compounds under the REACH legislation. Further information on the project, including downloads of the FEE tool and SPERC factsheets can be found via www.fertilizerseurope.com.

583

Bioaccessibility of grease thickeners and the implications for REACH registration

R.J. Brown, wca consulting; R. Smith, wca; P. Whitehead, wca consulting; J. Dawick, G. Whale, Shell Health / Risk Science Team; A. Dodos, Eldons; T. Halmans, Shell Global Solutions International / Anakytical Department

An intrinsic component of greases are the grease thickeners which include a diverese range of substances including metal soaps, metal-complex soaps and polyureas. These different thickeners impart different technical properties on the final grease. Although individually registered under REACH as isolated substances (i.e. extracted from base oil), grease thickeners are typically manufactured *in situ* in base oil and seldom exist except within a grease base. Under normal environmental conditions, grease thickeners would be expected to remain within the grease base because, during the grease manufacturing process, unique physical interactions (or matrix effects) occur between the grease thickener and the base oil. These interactions are important because, to be effective, the grease thickener matrix has to keep the lubricating base oil entrained. It is proposed that these matrix effects have a significant impact on the bioaccessibility of the grease thickener substances *in situ* in base oil in comparison to their isolated form. These matrix effects are expected to decrease the bioaccessibility of the grease thickener as it is not available to cross an organism's cellular membrane. The European REACH Grease Thickeners Consortium (ERGTC) have characterised the bioaccessibility of their grease thickeners by conducting leaching studies based on a Water Available Fraction" (WAF) approach, but using relevant media i.e. deionised water for the environment or synthetic fed state intestinal fluid (FeSSIF - Biorelevant, Switzerland) to assess exposure route via the gut (human health). Data is presented for different types of thickener substance which shows that most thickeners will not be bioaccessible and therefore, there will be minimal exposure to these substances. As the main form in which grease thickeners are manufactured and used, is entrained in a grease base, it is proposed that a lack of exposure based on low solubilities and/or bioaccessability is taken into consideration when registering the substances under REACH. This is a pragmatic approach for a group of substances that have low hazard potential and avoids conducting unnecessary vertebrate animal testing. The ERGTC strategy for registering grease thickeners under REACH, taking into consideration bioaccessibility, will be presented, including proposed "limits for leaching". This approach could be expanded to include other types of similar substances which occur in situ in an inert carrier such as base oil

584

The durability criteria: a pragmatic and sound approach to the exposure assessment of nano-enabled agrochemicals

M. Kah, University of Vienna / Department of Environmental Geosciences; R.S. Kookana, CSIRO / Land and Water

After many years of research and development, nano-enabled agrochemicals are starting to make their way into the market. Evaluating nano-enabled agrochemicals against conventional analogues is essential to assess the new risks and benefits associated with the technology, and this raises a number of issues for regulators. The ecological risk assessment of nano-enabled agrochemicals is likely to differ from that of conventional products and new parameters are needed to allow an adequate evaluation of the new products. The majority of products currently in development consists in nanocarrier systems loaded with a registered AI. For this type of products, a priority for assessment is to establish the time during which the AI remains associated with the nanocarrier, i.e. the "durability" of the AI−nanocarrier complex [1]. Kookana et al. [2] presented a series of guiding principles for the regulatory evaluation of environmental risk associated with nano-enabled pesticides, including a conceptual strategy relying on the durability parameter. A group drawn from regulatory agencies, academia, research, and the agrochemicals industry recently offered a perspective on relevant considerations pertaining to the problem formulation phase [3]. A case study (pendimentalin nanosized hydrogel) was consider to test and the framework proposed for exposure assessment. The scheme serves as a useful base to guide additional data requirement and help regulators to take informed decisions without having to systematically perform a comprehensive new nano-specific assessment. One of the key conclusions is that an early and reliable measure of the durabilityof the AI-nanocarrier complex under relevant conditions is key to the assessment of nano-enabled pesticides. There is currently no standard protocols to measure the durability of the AI−nanocarrier complex and robust methods for its measurement are urgently needed. References: 1. Kah M, Hofmann T. 2014. *Environ. Int.* 63:224–235. 2. Kookana RS et al. 2014. *J. Agric. Food Chem.* 62:4227–4240. 3. Walker GW et al. 2017. *J. Agric. Food Chem.* doi:10.1021/acs.jafc.7b02373.

585

Can we model emissions, fate and exposure on a global scale? A case study of PCB 153 in human milk

M. McLachlan, Stockholm University / Environmental Science and Analytical Chemistry (ACES); E. Undeman, Stockholm University / Baltic Sea Centre; F. Zhao, Stockholm University / Department of Environmental Science and Analytical Chemistry ACES; M. MacLeod, ITM - Stockholm University / Department of Environmental Science and Analytical Chemistry

One of the grand challenges of environmental chemistry is to be able to predict human exposure to an environmental contaminant based on its emissions. In this paper we explore how, after decades of excellent research by scores of scientists, we measure up to this challenge on a global scale. PCB 153, a widely studied persistent, hydrophobic and semi-volatile pollutant was chosen as the test chemical. We used physico-chemical properties recommended by Schenker et al.[1] and global historical emissions estimates developed by Breivik and co-workers[2] to drive the global multimedia fate and transport model BETR Global.[3] The fugacities of PCB 153 in air, water and soil, modeled at a spatial resolution of 3.75° × 3.75°, were re-gridded to give the historical fugacity records on the basis of individual countries. These were entered into the bioaccumulation and exposure model ACC-HUMAN, which modeled the concentrations of PCB 153 in fish, meat, dairy products and human milk.[4] The human diet in ACC-HUMAN was parameterized for each country based on the WHO Global Environment Modeling System (GEMS) cluster diets.[5] The modeled concentrations of PCB 153 in human milk were compared with the concentrations measured in the WHO/UNEP global monitoring program for POPs.[6] The predicted and observed concentrations were highly correlated, with a correlation coefficient of 0.76. For 49 out of 78 data points, the predictions and observations agreed within a factor of 4. The model over-predicted the concentrations in central Europe and under-predicted the concentrations in much of Africa, in particular West Africa. Potential weaknesses identified in the chain of models include an under-prediction of the rate of decrease in PCB concentrations in air since the 1980s and inadequate treatment of food sourcing.[7,8] We conclude that we have come a long way towards meeting this grand challenge for PCB 153, but there remains room for improvement! References U. Schenker et al. *Environ. Sci Technol.*, 2005, **39**, 8434-8441. K. Breivik et al., *Environ. Sci. Technol.*, 2016, **50**, 798-805. M. MacLeod, et al., *Environ. Pollut.*, 2011, **159**, 1442–1445. G. Czub and M. S. McLachlan, *Environ. Toxicol. Chem.*, 2004, **23**, 2356–2366.
https://undatacatalog.org/dataset/gemsfood-consumption-database M. van den

Berg et al., *Arch. Toxicol.*, 2017, **91**, 83-96. M. MacLeod et al. *J. Ind. Ecol.*, 2004, **8**, 89-102. C. Ng and N. von Goetz, *Environ. Health Perspect.*, 2017, **125**, 1-7.

Risk assessment of Nanomaterials: innovative approaches and application of recent research developments to regulatory science

586

Regulatory tools and activities for environmental risk assessment of nanomaterials in ECHA

A. Kapanen, European Chemicals Agency - ECHA; L. Deydier Stephan, European Chemicals Agency / Evaluation Directorate; V. Rodriguez Unamuno, A. Karjalainen, J. Holmqvist, European Chemicals Agency ECHA

European Chemicals Agency (ECHA) implements the REACH Regulation (EC) No 1907/2006 (Registration, Evaluation, Authorisation and Restriction of Chemicals), the Biocidal Products Regulation (BPR, Regulation (EU) 528/2012), and the Classification, Labelling and Packaging (CLP) Regulation ((EC) No 1272/2008). Industry and authorities need to fulfil their obligations regarding these regulations also for nanoforms as for any other form of a substance. Nanomaterials are implicitly covered by the substance definition of REACH Regulation 1907/2006 although there are no explicit requirement laying down NM specific obligations. ECHA's experience has shown that REACH would benefit from nano-specific provisions. The BPR has partly implemented the Commission recommendation of 18 October 2011 on the definition of nanomaterials article 3(1)(z)). It states that the approval of an active substance does not cover nanomaterials except where explicitly mentioned (Article 4). ECHA currently performs three type of activities to implement REACH, CLP and BPR regulations and to support these processes aiming at ensuring safe use of nanomaterials (NM): REGULATE: formal processes under regulatory frameworks, whereby ECHA uses the legal instruments available under REACH (substance/dossier evaluation, authorisation and restriction), CLP and BPR; SUPPORT: helpdesk, meetings with stakeholders and with Registrants, Nanomaterials Expert Group (NMEG); COMMUNICATE: ECHA Nanomaterials web-site, conferences, workshops, communication throughout the supply chain and in a broader context e.g. EUON and press. This presentation will provide a summary of the multiple actions taken by ECHA to address NM under REACH, CLP and BPR: Dossier and substance evaluation, NMEG, EUON and ECHA's involvement at OECD level. Commission is currently considering modifying some of the technical provisions in the REACH Annexes. This would allow more efficient efforts towards safe use of NM and decreased uncertainties in the regulatory processes. In addition, ECHA highlights the need for good coverage of standard methods applicable to NM to produce adequate information for regulatory risk assessment.

587

Building a Risk Assessment Framework for Nanomaterials in Canada

M. Sauve, Environment Canada; A. Shahsavarani, Environment and Climate Change Canada

Despite the potential benefits associated with the use of nanomaterials, concerns also exist as to potential environmental and/or human health risks as a result of environmental exposure to nanomaterials. Canada regulates chemical substances, including nanomaterials, under various regulatory mechanisms. The *Canadian Environmental Protection Act, 1999* (CEPA) and the Chemical Management Plan (CMP) are key in safeguarding Canadians and the Canadian environment from potentially harmful substances. Under the CMP, the current activities to address nanomaterials include identifying data needs, developing tailored strategies and approaches, work planning and strengthening collaboration with national and international partners and stakeholders. To support these efforts, Canada is developing a science based risk assessment framework (RAF) to guide legislative and regulatory risk assessments of nanomaterials under CEPA. This science based RAF will inevitably touch on many issues faced by other countries and regulators. Our communication strategy includes consultation and collaboration with multidisciplinary experts, the public and stakeholders. Online collaboration tools will be used to encourage participation. A national expert panel will be consulted in the spring 2018, and SETAC Europe would be the first attempt to reach out to the international scientific community for support. We hope to use this venue to generate new partnerships and to strengthen existing ones. Critical questions based on the challenges identified by Canada over more than a decade of regulating new nanomaterials, and by the international scientific community looking at risk assessment challenges for nanomaterials, will be highlighted in this presentation.

588

Inventory of available tools, methods, approaches and best practices on nanomaterials/nanoechnologies

B. Duuren-Stuurman, TNO, Utrechtseweg 48, 3704 HE Zeist, The Netherlands; s. manzo, ENEA / SSPT-PROTER-BES; S. Scalbi, ENEA / UTVALAMB- Technical Unit Model, Methods and Technologies for the Environmental A; P. Reale, ENEA; H. Witters, VITO / Applied Bio & molecular Systems; A. Duschl, PLUS; H. Stockmann-Juvala, FIOH; T. Berezniak, NRCWE; A. Fornara, SP

The EC4SafeNano initiative, founded by Horizon 2020 is an ongoing effort to build a European Centre for Risk Management and Safe Innovation in Nanomaterials and Nanotechnologies. EC4SafeNano aims to bridge the gap between scientific knowledge on hazard and risk, and 'fit-for-purpose' risk management tools and strategies supported by measurement and control methods. The Centre of European organisations will offer services for Risk Management and Safe Innovation for Nanomaterials & Nanotechnologies. One of the first actions of the project was developed an inventory of available resources related to nanosafety issues. Indeed, until now, many tools and studies which aim to improve human and environmental nanosafety have been developed by research organizations and EU funded projects. Therefore this inventory have the aim to give a useful overview on tools, methods, trainings, standards, standard operating procedures (SOPs), guidance documents and best practices in nanosafety. quality criteria are included to give users the possibility to select or search for resources based on, for example, the organization that approved the standard (and thereby indirectly the procedures followed to come to a standard), the level of evaluation and validation of the resources or the acceptance of the resource in view of the REACH legislation. During the project and after the duration of the project this overview will be updated when new information or updated versions of resources become available or when new resources are introduced by a mechanism later to be defined. Moreover this inventory will be published on the EC4SafeNano website. There is a large number of Standards (77) and SOPs (136) that can help the end user to conduct testing on toxicity and eco-toxicity, or measurements in workplaces and environment. One important issue is the scarce number of trainings available only 5. This aspect will be addressed in the EC4SafeNano project to identify the specific training needs and the approaches useful for the end user. Moreover, the analysis emerged that the amount of information related to the environmental assessment of nanomaterials and nanotechnology is very low. This aspect is covered only in 6 out of the 28 included tools, and 4 out of 43 Guidance and Best practices. This shows that, at the moment, aspects such as eco-toxicity and methodologies for the environmental risk assessment and life cycle assessment have most likely not been deeply investigated

589

The Application of Ecotoxicological Tools to Safer-by-Design Strategies for Engineered Nanomaterials

A. Barrick, Association Saint Yves / UCO / UBL, Mer molecules et sante; N. Manier, P. Pandard, INERIS / Expertise and assay in ecotoxicology unit; A. Chatel, Catholic University of the West / UBL, Mer Molécules Santé; C. Mouneyrac, Université Catholique de lOuest / UBL, Mer Molécules Santé

OECD test guidelines using aquatic organisms have been identified as suitable starting points for identifying risk that engineered nanomaterials (ENMs) risk represent for the environment. However, standardized testing (mainly relevant for freshwater species) may not be able to adequately characterize the risk and dynamic behavior of ENMs in the environment, in particular for aquatic regions like coastal areas which are the ultimate sink for all land based contaminants. As a result, the addition of less conventional organisms to regulatory projects can promote the characterization of environmental risk nanomaterials pose. *Mytilus* species have a long history of being used as sentinel organisms to characterize ecosystem health and can be used to promote the understanding of environmental risk that emerging contaminants like ENMs pose. This information can be particularly useful for the development of safer-by-design strategies as implementation in risk characterization can promote the identification of which products an industry creates pose the lowest environmental risk. In this context, under project the Horizon 2020 project NanoReg2, the aim of this study was to demonstrate the implementation of a safer-by-design strategy for ENM development. Three carbon nanofibers (CNFs) were provided by an industrial partner in order to characterize the potential environmental risk their products posed. The aim of the study was to compare the original product (GANF) to a new scaled up production process for the CNF (GATam) as well a graphitized verison of the product (GANFg). The study implemented regulatory testing using *Daphnia magna* and *Pseudokirchneriella subcapitata* as well as hemocytes from the marine mussel *Mytilus edulis* (*M. edulis*) following *in vitro* and *in vivo* testing on subcellular endpoints. The testing strategy was conducted in order to demonstrate the suitability of both regulatory testing (OECD tests) as well an *in vitro* screening strategy on *M. eduilis* hemocytes to characterize the environmental risk posed by ENMs in the context of safer-by-design and its application to industry. In addition, recommendation and discussion on protocols used to test this CNF are provided.

590

Minimising the risk posed by TiO2 nanomaterials used in sunscreen throughout the entire product lifecycle

D. Slomberg, R. Catalano, Aix-Marseille Université; J. Hubaud, Helioscience; V. Bartolomei, S. Motellier, CEA Liten; D. Boutry, CEA - Grenoble; L. Hedouin, CNRS CRIOBE; C. Santaella, CNRS/CEA/Aix-Marseille Université / Bioscience and biotechnology Institute of Aix Marseille; P. Hennebert, INERIS; A. Pinsino, IBIM CNR Palermo; S. Lehmann, University of Grenoble Alps; J. Labille, CNRS

Sunscreens are of emerging concern regarding both human and environmental health. While TiO2 nanoparticles used as UV-blockers may offer a safer alternative to organic filters, their fate and impact and resulting regulation are still under consideration, largely related to the potential risk of nanotechnology-based products. After leaving the skin either through bathing or cleaning, the TiO_2

nanomaterials contained in the sunscreen can be released into rivers, lakes, sea shores, and/or sewage treatment plants. Their fate and impact in these different systems is largely determined by the surface properties, i.e. the coating type and lifetime. This project aims to develop the eco-design of sunscreens through the minimization of risks associated with nanomaterials incorporated into the formulation. All stages of the cream life cycle must be considered in this light, from its manufacture to its end of life, through its use by the consumer and its impact on the exposed environment. By considering each development stage of the sunscreen, from the choice of UV-blocker and its integration into a cosmetic formulation, to the knowledge of the risk involved in this choice all along the product lifecycle, an eco-design approach can be achieved and risk can be minimized. The present work combines industrial companies specialising in cosmetic formulation with academic research experts in the fields of exposure, toxicity and lifecycle assessment. Sunscreen fabrication, risk for the consumer by dermal exposure, risk for the direct aquatic environment and risk related to the end of life of the product are as many key steps of the sunscreen lifecycle that were investigated in this project.

591

Environmental risk assessment of engineered nano-SiO2, nano iron oxides, nano-CeO2, nano-Al2O3, and quantum dots

B. Nowack, EMPA; Y. Wang, Empa Swiss Federal Laboratories for Materials Science and Technology

Many research studies have aimed to investigate the ecotoxicological hazards of engineered nanomaterials (ENMs). However, little is known regarding the actual environmental risks of ENMs, combining both hazard and exposure data. The aim of this study is to quantify the environmental risks for nano-Al_2O_3, nano-SiO_2, nano iron oxides, nano-CeO_2, and quantum dots by comparing the predicted environmental concentrations (PEC) with the predicted no effect concentrations (PNEC). The PEC values of these five ENMs in fresh waters in 2020 for northern Europe and southeastern Europe were taken from a published dynamic probabilistic material flow analysis model. PNEC values were calculated using probabilistic species sensitivity distribution (PSSD). The order of the PNEC values was quantum dots< nano-CeO_2< nano iron oxides< nano-Al_2O_3< nano-SiO_2. The risks posed by these five ENMs were demonstrated to be in the reverse order: nano-Al_2O_3> nano-SiO_2>nano iron oxides>nano-CeO_2>quantum dots. However, all risk characterization values are four to eight orders of magnitude lower than one and no risk was therefore predicted for any of the investigated ENMs at the estimated release level in 2020. Compared to static models, the dynamic material flow model allowed us to use PEC values based on a more complex parameterization, considering a dynamic input over time and time-dependent release of ENMs. The PSSD approach makes it possible to include all available data to estimate hazards of ENMs by considering the whole range of variability between studies and material types. The risk assessment approach is therefore able to handle the uncertainty and variability associated with the collected data. The results of the current study are able to provide a scientific foundation for risk-based regulatory decisions of the investigated ENMs.

Natural toxins and harmful algal blooms (HABs): water and food safety, analysis, toxicity, and risks (I)

592

Occurrence of cyanotoxins in Greek lakes

A. Hiskia, C. Christophoridis, National Center for Scientific Research / Institute of Nanoscience and Nanotechnology; S. Zervou, NCSR Demokritos / Institute of Nanoscience and Nanotechnology; K. Manolidi, NCSR Demokritos; T.M. Triantis, NCSR Demokritos / Institute of Nanoscience and Nanotechnology; T. Kaloudis, EYDAP SA / WATER QUALITY CONTROL

Cyanotoxins (CTs) produced from cyanobacteria in surface water during harmful algal blooms can pose significant threat to human health and the environment. Their analytical determination in both biomass and water is a demanding task as CTs comprise a large variety of compounds with different structural and physicochemical properties, i.e. cyclic peptides (microcystins, MCs and nodularins, NODs) and alkaloids (cylindrospermopsin, CYN, anatoxin-a, ANA-a and saxitoxins, STXs). The most important issues that make the CTs' analysis challenging are mainly the large number of variants of various classes, the limited availability of analytical standards and insufficient validation data. Moreover, different methods of analysis are usually required for each class of CTs in order to achieve acceptable analytical performance. Based to the above there is a need to develop efficient multi-class/varriant method protocols for analysis of as many as possible CTs. Our laboratory has recently developed and validated SPE-LC-MS/MS methods for simultaneous determination of multi-class CTs. As an example, analysis of 12 MCs ([D-Asp3]MC-RR, MC-RR, MC-YR, MC-HtyR, [D-Asp3]MC-LR, MC-LR, MC-HilR, MC-WR, MC-LA, MC-LY, MC-LW and MC-LF), NOD, CYN and ANA-a in one run can be achieved. In addition, validated multi-variant methods for the analysis of STXs based on HILIC-MS/MS have been developed. Those methods has been combined in workflow to analyze multi-class toxins efficiently. The aim of this study was to demostrate the applicability and efficiency of a proposed workflow for multi-class/varriant determination of CTs. Futhermore, to detect and identificy a wide range of CTs in Greek lakes never studied before, using this validated tool. Results of a monitoring survey in Greeks lakes showed that the proposed LC/MS/MS based workflow provided unequivocal and definitive identification of multi-varriant/class toxins, avoiding the drawbacks of bioassay techniques that have been used previously. Using the proposed workflow a wide range of MCs ([D-Asp3]MC-RR, MC-RR, MC-YR, MC-HtyR, [D-Asp3]MC-LR, MC-LR, MC-HilR, MC-WR, MC-LA, MC-LY, MC-LW and MC-LF), CYN, ANA-a, STX and neoSTX were identified in Greek lakes for the first time. *Acknowledgement* - The authors thank CYANOCOST – COST Action ES 1105 www.cyanocost.net

593

Interactions between cyanobacteria and daphnia

G. Bojadzija, UMR CNRS Ecobio; M. Bormans, UMR CNRS Ecobio / UMR Ecobio; C. Edwards, L. Lawton, Robert Gordon University / IDEAS Research Institute; E. Briand, IFREMER - Centre Atlantique / Laboratoire Phycotoxines / Unité DYNECO / Dept. ODE; C. Wiegand, Université de Rennes 1 / UMR CNRS ECOBIO

Thanks to their adaptation cyanobacteria colonized limnic, marine and terrestrial environment. Eutrophication of waters has promoted and will increase cyanobacteria blooms in future, posing hazards to the aquatic ecosystem and human health due to the capability of cyanobacteria to produce bioactive or toxic compounds. One of the groups firstly affected by cyanobacteria is planktivorous zooplankton, such as Daphnia. On the other hand, Daphnia can also suppress cyanobacterial population up to certain density and toxicity. A development of tolerance apparently enables them to withstand cyanobacterial compounds, and is transferable to the next generation. The role of cyanobacterial toxins and other bioactive compounds has not yet fully been elucidated, neither has the question, if the presence of zooplankton grazers could modify their production. This study investigates the mutual two-way interactions, in terms of biochemical and life trait responses of both, cyanobacteria and daphnia. *Microcystis aeruginosa* PCC7806 and its non-microcystin producing mutant *M. aeruginosa* PCC7806 mcy-, and *D. magna* were employed. In order to disentangle mutual interactions between both organisms, a co-culture chamber was designed, where two chambers are physically separated by a 0.2 μm cellulose nitrate membrane filter, preventing the grazing effect but allowing exchange of chemical compounds released into the medium.Exposures lasted one week. First results confirmed the detrimental impact of cyanobacterial metabolites released into their culture medium on *D. magna*. Cyanobacterial culture medium of *M. aeruginosa* PCC7806 obtained after 2 weeks culture, equivalent to 10^4 cells/mL, reduced feeding and survival, moreover altered detoxication and antioxidant response as well as the energetic budget. Exposures to spent media from *M. aeruginosa* PCC7806 mcy- are currently in progress. Vice versa, *M. aeruginosa* PCC7806, reacted to spent medium from *D. magna* cultures of two weeks: During the first days there was an increase of growth rates, followed by a decrease in physiological performance. Moreover, the antioxidant response increased, which, even though not significant itself, caused a significant reduction in the hydrogenperoxide content in the cyanobacteria. First results indicate that cyanobacteria not only harm aquatic organisms, but that vice versa they react to the presence of potential grazers, hence yet unknown substances present in the spent media impair their performance.

594

Teratogenic retinoid-like compounds produced by cyanobacteria into surface waters

K. Hilscherova, Masaryk University, Faculty of Science, RECETOX / Research Centre for Toxic Compounds in the Environment RECETOX; E. Sychrova, Masaryk University, Faculty of Science, RECETOX / Research centre for toxic compounds in the environment; M. Krauss, Helmholtz centre for environmental research - UFZ / Effect-Directed Analysis; A. Jonas, Masaryk University, RECETOX / Faculty of Science; J. Priebojová, Masaryk University, Faculty of Science, RECETOX / Research centre for toxic compounds in the environment RECETOX; J. Vecerkova, Masaryk University, Faculty of Science, RECETOX; L. Sehnal, Masaryk University Faculty of Science RECETOX / RECETOX Research centre for toxic compounds in the environment; T. Prochazkova, Research Centre for Toxic Compounds in the Environment, Faculty of Science, Masaryk University / Research centre for toxic compounds in the environment; S. Scholz, Helmholtz Centre for Environmental Research / Department Bioanalytical Ecotoxicology; M. Smutna, Masaryk University, Faculty of Science, RECETOX / RECETOX Research Centre for Toxic Compounds in the Environment

One of the biggest worldwide problems in aquatic ecosystems is the formation of cyanobacterial water blooms that can have adverse effects on organisms. It has been well recognized that cyanobacteria are able to produce diverse groups of toxins. Recent reports show evidence of new toxic products of cyanobacterial metabolism - retinoid compounds, but there is very limited knowledge regarding to their producers, occurrence or potential associated risks. This research provides environmentally significant information about total retinoid-like activity in the biomass of cyanobacterial water blooms as well as in their surrounding water. It documents production of compounds with this bioactivity into the surface water by various cyanobacterial species. The level of retinoid-like activity reaches values that can cause adverse developmental effects in exposed organisms. Retinoid-like activity in cyanobacterial exudates was in a very good agreement with developmental effects in zebrafish (*Danio rerio*) and frog (*Xenopus laevis*) embryos. Both the phenotypes and effective concentrations of exudates corresponded to all-trans retinoic acid (ATRA) equivalents, supporting the hypothesis that the teratogenic effects of cyanobacterial exudates are likely to be associated with retinoid-like activity. Non-target analyses and comparison of the spectra of compounds present in exudates with different retinoid-like activities pointed to structural features of compounds contributing to the retinoid-like activity. In the approach of a "virtual EDA" we have characterized in detail retinoid-like activity of 53 samples of exudates from 4 algae species and 15 cyanobacteria species/strains. Only several species of cyanobacteria showed detectable activity, while there was no activity in any algal exudates. We have identified a set of compounds that contribute to the total retinoid-like activity in both laboratory and field samples, including ATRA, 9/13cis retinoic acid, but also several novel cyanobacterial metabolites, such as 5,6epoxy-RA or 4keto-ATRA with high retinoid-like potency. In general, retinoid-like activity has been shown to be associated with cyanobacterial water blooms dominated by many different species, which documents that production of retinoids by cyanobacteria in the environment is a common phenomenon. This study was supported by the Czech Science Foundation project No.18-15199S and FP7 SOLUTIONS project No. 603437.

595

(Co-)Production Dynamics of Cyanobacterial Peptides

R. Sanches Natumi, E. Vonwyl, Eawag Swiss federal Institute of Aquatic Science and Technology / Department for Environmental Chemistry; E.M. Janssen, Eawag Swiss federal Institute of Aquatic Science and Technology / Department of Environmental Chemistry

Cyanobacterial algal blooms are expanding temporally and spatially, which is promoted by eutrophication and likely climate change. Cyanobacteria can produce a wide range of bioactive compounds with different modes of action, including a variety of toxic cyanopeptides. Information regarding the occurrence and production dynamics of the majority of novel cyanopeptides is mostly unknown even for common cyanobacterial strains. Such information is crucial to assess the risk of these emerging natural toxins for human health in evaluating their potential to reach drinking water supply plants. While it is known that many cyanopeptides are produced simultaneously from one species, the co-production of these potential toxins has not been explored comprehensively for cyanopeptides other than microcystins. This project focused specifically on the production and co-production dynamics of cyanopeptides from common cyanobacterial species including *Microcystis aeruginosa* and *Anabaena flos-aquae* strains. Batch culturing was done under varied environmental conditions and the cell abundance was followed by optical density, cell counting, and biomass weight. Simultaneously, the cyanopeptide production was followed. Therefore, biomass extracts were purified and measured by liquid chromatography, high resolution mass spectrometry with refined analytical protocols. Comprehensive data analysis was performed to identify cyanopeptides and follow their abundance. These new insights of co-production dynamics are critical to better understand which peptides and peptide mixtures are present during cyanobacterial bloom events.

596

Development of methods for Measuring Total Microcystins in Fish Tissue using the 2-methoxy-3-methyl-4-phenylbutyric acid (MMPB) procedure.

J.M. Lazorchak, U.S. EPA / Office of Research and Development; T. Sanan, United States Environmental Protection Agency / National Risk Management Research LaboratoryWater Supply and Water Research DivisionTreatment Technologies Evaluation Branch; D. Sundaravadivelu, J. Kickish, Pegasus Technical Services, inc.; J. Jones, R. Venkatapathy, Pegasus co US EPA Cincinnati

There are limited methods for the analyses of multiple algal toxins in aquatic food webs, phytoplankton, zooplankton, periphyton, macroinvertebrates, forage fish, bottom feeders and top carnivore fish. Algal toxins in freshwater systems do not necessarily occur as single contaminants; mixtures of toxins may be produced from Cyanobacteria, *Prymnesium parvum (Prymnesins)*, and *Euglena sanguinea*, including microcystins, saxitoxins, cylindrospermopsin, anatoxin-a., prymnesins and euglenophycin. The objective of the first phase of this research was to spike existing fillet and whole fish homogenates with 3 congeners of microcystins (LR, LA and RR) individually and as mixtures, and to develop a method for their recovery and measurement using the MMPB derivatization method. The second phase of the project is to field-test this method on fish collected from water bodies experiencing algal blooms and compare results with individual congener measurements. Extraction methods and analytical methods being developed for this research will be a starting point for developing extraction procedures for plankton, periphyton, and macorinvertebrates. Ten and 100 mg of fish homogenates from fish containing 1, 4 and 14% lipids were spiked with 4 and 40 ng of each of the microcystin congeners, LR, LA and RR. Various extraction techniques and conditions were tested to optimize recovery and simplify the procedure. Overall toxin recoveries were found to range from 30 to 50%. The lipid content was found to not interfere with generation of MMPB; however, it did impact the workup/extraction procedure in ways which were accountable through the use of a surrogate standard. The MMPB technique can be reliably employed for microcystin quantification in fish tissue. Detections in non-spiked samples (10-20 ug/kg) are comparable to literature precedent. For tissue quantification the MMPB method provides considerable improvements over extraction of individual toxin congeners and is consistent even with very polar or hydrophobic MCs.

597

Saponins in the aquatic environment: hydrolysis and toxicity

X. Jiang, University of Copenhagen; H. Hansen, University of Copenhagen / Department of Plant and Environmental Sciences; B.W. Strobel, University of Copenhagen / Plant and Environmental Sciences; N. Cedergreen, University of Copenhagen / Department of Plant and Environmental Sciences

Saponins are a class of bioactive natural compounds. Due to their detergent-like structure, saponins have a lot of applications, e.g. as biopesticides in crop protection. They may leach into the aquatic environment due to their low octanol/water partition coefficient and poor binding to organic matter. They may therefore pose a risk to the aquatic organisms. However, their fate and toxicity in the environment are not fully understood. Hence, we aim to investigate the pH and temperature dependence of saponins hydrolysis together with their dissipation in sampled lake waters and to determine the aquatic toxicity of saponins from quillaja bark, tea seed coat, and quinoa seed coat towards different aquatic organisms. The hydrolysis of saponin (quillaja saponin) showed to be a highly pH dependent base-catalyzed reaction. The half-life was around 330 ±220 days at pH 5.1 and 26 ?, while decreased to 0.06 ±0.01 d at pH 10.0. The hydrolysis was also influenced by temperature with an activation energy of 56.9 ±14.2 kJ/mol at pH 7.2. Lake waters with pH varying between 6.4 and 8.2 showed completely different hydrolysis patterns, with a fast initial dissipation of up to 60% of the initial saponin concentration, followed by an extremely slow to nil reaction, making saponin partially persistent in lake waters. The maximal concentrations protecting 95% of the aquatic species (HC_5) derived from the SSD's of saponins from quillaja bark, tea seed coat, and quinoaseedcoat were 2.91 ±1.00, 0.22 ±0.11 and 22.9 ±5.84 mg/L, respectively. The 100-fold difference in toxicity between the saponin-rich extracts from different plant species, indicate that saponin toxicity depends on the species where it origins from, making "read-across" between saponins a dubious exercise. In addition, the predicted environmental concentrations of different saponins are close to or higher than their water quality standard, which means that the saponins might pose a risk to the aquatic environment if not used cautiously. Therefore, we recommend not using surrogate or expected data/conclusion in the regulation of saponin-rich plant extracts and pay more attention to the potential risk of saponins to the aquatic environment.

Advancing the Adverse Outcome Pathway Framework - An International Horizon Scanning Approach

598

Setting the Stage to Advance the Adverse Outcome Pathway Framework through Horizon Scanning

C. LaLone, U.S. EPA / Mid Continent Ecology Division; G.T. Ankley, U.S. EPA / National Health and Environmental Effects Research Laboratory; S.E. Belanger, The Procter & Gamble Company / Environmental Stewardship and Sustainability Organization; M.R. Embry, ILSI Health and Environmental Sciences Institute (HESI); G. Hodges, Unilever / Safety and Environmental Assurance Centre SEAC; D. Knapen, University of Antwerp / Zebrafishlab Dept Veterinary Sciences; S. Munn, European Commission; E.J. Perkins, US Army Engineer Research and Development Center / Environmental Laboratory; M. Rudd, University of York; D.L. Villeneuve, U.S. EPA / National Health and Environmental Effects Research Laboratory; M. Whelan, University of leicester / Geography; C. Willett, the Humane Society of the United States / Animal Research Issues; X. Zhang, Nanjing University / Environmental Science; M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre

Recognizing the international interest surrounding the adverse outcome pathway framework, which captures existing information describing causal linkages between a molecular initiating event through levels of biological organization to an adverse outcome of regulatory significance, an effort was undertaken to provide the scientific community the opportunity to engage in determining the direction of the AOP framework. Specifically, a horizon scanning effort was used to solicit questions from the international scientific community asking participants to propose questions that consider key outstanding challenges and/or limitations that must be overcome to advance the AOP framework for both research and regulatory decision making. From March-June, 2016, 340 valid questions were collected from 158 global submissions, spanning all continents, to an online horizon scanning survey. Respondents to the survey self-identified as 35% academia, 35% government, 20% industry, and 5% non-government organizations. Following question solicitation, questions were separated into broad topic areas including, AOP networks, quantitative AOPs, collaboration and communication on AOPs, AOP discovery and development, extrapolation, exposure/toxicokinetics considerations, and AOP application. An expert-ranking exercise was then conducted to identify high-priority questions for each category and from this, four key themes emerged that could aid in guiding future AOP research and regulatory initiatives. These themes were used as workgroup topics for a Pellston^{TM} Workshop, including: AOP networks and their applications; quantitative AOPs and

their applications; regulatory use of the AOP framework, and expanding awareness of, involvement in; and acceptance of AOPs to support aspects of predictive toxicology and regulatory decision-making. Charge questions for each workgroup were directly modified from those submitted during horizon scanning. Additionally, from the horizon scanning exercise, frequently asked questions (FAQs) were identified and addressed by experts in the field. Together the horizon scanning approach, expert ranking exercise, and answers to FAQs, were used to set the stage for the SETAC Pellston™ Workshop titled "Advancing the Adverse Outcome Pathway Concept: An International Horizon Scanning Approach," that took place in Cornwall, Canada during April 2017. The contents of this presentation neither constitute nor necessarily reflect US EPA policy.

599

Adverse Outcome Pathway networks: development, analytics and applications

D. Knapen, University of Antwerp / Zebrafishlab Dept Veterinary Sciences; M. Angrish, US EPA, National Center for Environmental Assessment; M.C. Fortin, Alcami / Environmental and Occupational Health Sciences Institute; I. Katsiadaki, Cefas / Environment and Animal Health; M. Leonard, lOREAL SA; L. Margiotta-Casaluci, Brunel University London / Institute of Environment, Health and Societies; S. Munn, European Commission; J. O'Brien, Environment and Climate Change Canada / National Wildlife Research Centre; N. Pollesch, US EPA / ORD NHEERL Mid Continent Ecology Division; L. Smith, University of Florida / Physiological Sciences; X. Zhang, Nanjing University / Environmental Science; D.L. Villeneuve, U.S. EPA / National Health and Environmental Effects Research Laboratory

Adverse outcome pathways (AOPs) are an important framework that can help support more effective use of mechanistic, pathway-based, data in risk assessment and regulatory decision-making. AOPs have rapidly evolved from a conceptual paradigm into a formalized framework for organizing biological and toxicological knowledge according to a set of generally accepted principles and guidelines. In response to the recognized need to continue advancing the framework, SETAC sponsored a global horizon scanning exercise to identify major outstanding topics and challenges related to the AOP framework and its application. The development of guidance related to AOP network development and analysis was identified as a critical need. This not only included questions focusing directly on AOP networks, but also on related topics such as mixture toxicity assessment and the implementation of feedback loops within the AOP framework. This presentation briefly outlines critical concepts concerning the development of AOP networks, how they may be analyzed, and illustrates how information derived from them can be applied. First, derivation of AOP networks is considered in the context of how it differs from development of individual AOP descriptions. Next, the application of filters and layers is discussed, which can be used to refine and enrich derived AOP networks so that they may be tailored to address specific questions of interest. We then introduce a number of analytical and computational approaches that may be used to characterize and analyze the structure of AOP networks to derive information that can guide research and regulatory decision-making. A number of application case studies is used to illustrate concepts underlying development and analysis of AOP networks, and how those concepts tie in with ultimate application. The contents of this presentation represent the personal opinions of the authors and neither constitute, nor necessarily reflect the policies or viewpoints of their employers or institutes.

600

Building and Applying Quantitative Adverse Outcome Pathway Models for Chemical Hazard and Risk Assessment

S. Scholz, Helmholtz Centre for Environmental Research / Department Bioanalytical Ecotoxicology; R. Ashauer, University of York / Environment; R. Conolly, US EPA RTP; B. Landesmann, JRC, European Commission; C. Mackay, Unilever; C. Murphy, Michigan State University / Department of Fisheries and Wildlife; N. Pollesch, US EPA / ORD NHEERL Mid Continent Ecology Division; J. Wheeler, Dow Agrosciences; A. Zupanic, Eawag Swiss Federal Institute of Aquatic Science and Technology / Environmental Toxicology; E.J. Perkins, US Army Engineer Research and Development Center / Environmental Laboratory

Quantitative prediction and assessment of chemical impacts on human and environmental health is important in understanding the potential hazards and risks of using, or being exposed to, chemicals. Here we examine how the Adverse Outcome Pathway (AOP) concept and knowledge base can be used to develop quantitative models (qAOPs) to predict and assess hazards and risks of chemicals. Quantitative models can be developed with a clear problem definition and using AOPs as initial conceptual models. Modeling methods range from semi-quantitative to quantitative modeling approaches or combination of these (e.g. fully mechanistic mathematical /ordinary differential equation based, individual-based models, statistical, or Bayesian network models). We discuss best practices for choosing modeling approaches, model building and the necessity for transparent and comprehensive documentation in order to gain confidence in the use of a model. Finally, we present examples of how qAOP models can support decision making: a screening level assessment of the health hazards of chemicals and chemical mixtures using a qAOP Bayesian network model of steatosis, the use of qAOPs in a prospective risk assessment context (e.g. in vitro to in vivo extrapolation using aromatase inhibition as an example) and for extrapolation between species or life stages.

601

Use of Adverse Outcome Pathways to Inform Decisions on Chemical Innovation, Regulation & Stewardship

T. Hill, US EPA NHEERL/ISTD/CB / ORD NHEERL Integrated System Toxicology Division; P. Browne, OECD / OSCP; K.K. Coady, The Dow Chemical Company / Toxicology Environmental Research and Consulting; M.R. Embry, ILSI Health and Environmental Sciences Institute (HESI); T. Hutchinson, School of Biological Sciences, Plymouth University / School of Biological Sciences; E. Lienala, OECD; L. Maslankiewicz, National Institute for Public Health and the Environment (RIVM); T.M. Steeger, U.S. EPA / Office of Chemical Safety and Pollution Prevention

An invited group of scientists participated in a SETAC Pellston Workshop™, "Advancing the Adverse Outcome Pathway (AOP) Concept – An International Horizon Scanning Approach," in April 2017. The workshop addressed key challenges or limitations of AOP constructs as tools for informing research and regulatory decisions that were identified by responses to a global Horizon Scanning survey. This presentation will summarize the findings of Pellston Workgroup 3, which was tasked with the explication of practical considerations for this use of AOP constructs in regulatory decision making. The use of AOPs and related concepts have increased in scientific and regulatory sectors over the past decade, coinciding with pressures to find innovative solutions to evaluate chemical safety in a more efficient and effective manner that better directs resource utilization. This workgroup focused on how AOPs can be a useful tool for chemical decision makers in the government and private sector. At the various points where chemical decision making is employed across the "life" of a chemical – from research and development within the commercial sector, government registration and regulation, through to post-marketing use/stewardship – AOPs can be used as an organizing principle. Pragmatic evidence is provided for how AOPs can be and are currently being used in chemical decision-making processes. Considerations for evaluating the suitability of AOP for decision makers are discussed, recognizing that the acceptable level of uncertainty varies based upon the nature of the decision and the context in which it is being applied. The presentation provides multiple examples of AOP use and practical considerations for evaluating whether use of AOPs is fit-for-purpose in different circumstances. *This abstract does not necessarily represent the views or policies of the U.S. EPA.*

602

Ensuring Long-Term Utility of the AOP Framework and Knowledge for Multiple Stakeholders

G.T. Ankley, U.S. EPA / National Health and Environmental Effects Research Laboratory; A. Carusi, University of Sheffield; H. Davies, WA State Dept of Health / Dept of Ecology; G. De Grandis, Norwegian University of Science and Technology; B. Escher, Helmholtz Centre for Environmental Research GmbH - UFZ / Cell Toxicology; G. Hodges, Unilever Research / Safety and Environmental Assurance Centre; K. Leung, The University of Hong Kong / The Swire Institute of Marine Science and School of Biological Sciences; M. Whelan, University of leicester / Geography; C. Willett, the Humane Society of the United States / Animal Research Issues

The adverse outcome pathway (AOP) framework serves as a knowledge assembly and communication tool to facilitate translation of mechanistic (e.g., molecular, biochemical, histological) data into adverse apical outcomes meaningful to chemical risk assessment. Although initially designed for ecotoxicology applications, the framework has also received extensive attention relative to chemical safety assessments for human health. Moreover, as the AOP concept and associated knowledgebases have evolved, it has become recognized that the potential stakeholder community is broader than scientists and regulators directly involved in chemical safety assessment. For example, the application of AOP-based thinking for addressing biomedical challenges has become increasingly evident. This presentation will identify various stakeholders who currently, or could potentially, benefit from application of the AOP framework and knowledge to specific needs, and describes challenges and strategies to effectively engaging these stakeholders. We also present a "roadmap" on how to maintain a viable, sustainable network to support AOP stakeholders, including recommendations for governance and coordination of AOP development and knowledge dissemination in a multi-stakeholder consortium. *The contents of this abstract neither constitute, nor necessarily reflect, official USEPA policy.*

603

Adverse Outcome Pathways: Moving from a scientific concept to a globally accepted framework

M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre; C. LaLone, U.S. EPA / Mid Continent Ecology Division

The adverse outcome pathway (AOP) framework has gained significant international traction as a systematic approach for capturing existing knowledge to transparently link mechanistic data to apical toxicity endpoints as a means to inform research and risk assessment. While the framework has evolved significantly since

its introduction in 2010, it was recognized that a survey of the broader scientific community would be useful in identifying shortcomings and in guiding future initiatives. In 2016, we reached out to national and international scientific and regulatory communities to collect questions and provide an opportunity to discuss key outstanding challenges that must be addressed in order to realize the full potential of the AOP framework. Four key themes emerged from this "Horizon Scanning" exercise (see presentation "Advancing the Adverse Outcome Pathway Framework - An International Horizon Scanning Approach" in this session), which were then addressed at a Society of Environmental Toxicology & Chemistry (SETAC) Pellston™ Workshop comprised of international participants representing industry, government, academia, and NGOs was held in Cornwall, Ontario, in April 2017. This presentation will provide an overview of the overall outcomes and common themes that emerged during this Pellston Workshop. In brief, common themes that spanned across these main topics included the need to simplify, translate, and better communicate the AOP framework to the broader international stakeholder community, and a consensus that the AOP framework does not represent a rigid tool but rather a knowledge repository for diverse stakeholders ranging from epidemiologists to mainstream experimental toxicologist to risk assessors and managers. Furthermore, when considering the AOP framework and its applications, the field of environmental toxicology and human health naturally merged into a continuum that is at the nexus of Toxicology in the 21st century. In particular, it was felt that the current momentum the AOP framework has gained across a wide range of professional sectors provides the unique window of opportunity to reach out to and gain acceptance of this framework by society, which will be required for it to become an integral part of the international chemical and environmental risk assessment landscape. *The contents of this presentation neither constitute nor necessarily reflect US EPA policy.*

Environmental specimen banks in research and regulation for a better environmental quality

604

Monitoring of POPs in the Swedish aquatic ecosystem and in human milk

E. Nyberg, A. Bignert, S. Danielsson, Swedish Museum of Natural History; C. Ek, Department of Applied Environmental Science / Department of applied environmental science

In the 1960s, the Baltic Sea was found to be severely polluted by persistent organic pollutants (POPs). These discoveries were the starting point of a continuous Swedish national monitoring program for contaminants in biological matrices, mainly from the marine and freshwater environment, with samples dated as far back in time as 1969. Today's marine and freshwater monitoring programs consist of 32 lakes and 28 marine sites where matrices as perch, pike, arctic char, herring, cod, eelpout, blue mussel and egg from guillemot, oystercatcher and common tern are annually collected. In 1967, examination of human exposure to POPs was initiated by Karolinska Institutet in Stockholm through measurements in human milk from the area, and since 2007 milk has also been collected from Gothenburg in the southwest of Sweden. The milk samples were in 1997 transferred to the Environmental Specimen Bank at the Swedish Museum of Natural History. The main objectives of the monitoring program are to investigate changes over time, to estimate geographical differences and to assess compliances with set target values. Moreover, the program is designed to answer these different questions with a high statistical power. Since the start of the monitoring, concentrations of PCBs, DDTs, HCHs and HCB have decreased in fish and bird eggs from both the Baltic Sea and the Swedish freshwater environment. Several of the classical POPs have also decreased considerably in human milk. However, the non-linear trends differ between the monitoring matrices for several contaminants. In some cases the peak differs, and in others, concentration is levelling out for one matrix but continues to decrease for another. In addition, the concentrations of PCBs, DDTs and HCHs are, despite continuous decreases since the 1970's, still higher in the Baltic Sea compared to, for example, the North Sea.

605

Jumping out of the frying pan and into the fire? Spatial and temporal trends for PBDE, Dechlorane Plus and alternative flame retardants in samples of the German environmental specimen bank

A. Dreyer, Eurofins GfA GmbH / Air Monitoring; F. Neugebauer, Eurofins GfA Lab Service GmbH / R&D; N. Lohmann, Eurofins GfA LabService GmbH; M. Paulus, Trier University; H. Ruedel, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Department of Environmental Monitoring; C. Rauert, Umweltbundesamt / International Chemicals Management; J. Koschorreck, Umweltbundesamt

In the last century, *conventional* brominated flame retardants (FR) such as polybrominated diphenyl ethers (PBDEs) were identified as persistent organic pollutants and subsequently regulated. *Novel* or *alternative* FRs were introduced as their replacements to meet ongoing market demands. Many of these *alternative* FRs are also highly chlorinated or brominated and their fate and effects in the environment may be similar to those of their regulated counterparts. Until now there are only few comprehensive data sets about alternative FRs in the environment, particularly for Germany. In order to provide for a systematic overview about the current state of contamination of the German environment to FRs, a large set of terrestrial, freshwater and marine samples from the German environmental specimen bank were analysed for 45 FRs (PBDEs, Dechlorane Plus and brominated aromates, brominated ethers, cyclic BFRs). The substances will be discussed with respect to their spatial occurrence in the environment (including different matrices as well as land use and ecosystem types), their substance patterns and their bioaccumulation potential. Sample series going back to the 1980s, e.g. from coastal herring gull eggs, freshwater fish, tree leaves and roe deer will be used to illustrate time trends for regulated flame retardants and their substitutes. Recommendations will be given to European and international chemical management.

606

New Uses of Archived Specimens from the U.S.A. NIST Marine Environmental Specimen Bank

R. Pugh, C. Bryan-Sallee, W. Davis, J. Lynch, B.A. Neely, J. Ness, S. Schuur, National Institute of Standards and Technology / Chemical Sciences Division

The National Institute of Standards and Technology (NIST) has been involved in the long-term archival of biological and environmental specimens for over 40 years. Specimens originally intended for monitoring geographic and temporal trends in emerging contaminants as well as changes in transport and accumulation of legacy contaminants have added value today. Tissue and fluid specimens from marine animals, including mammals, seabirds, sea turtles, bivalves, fish, coral and coral ecosystems, collected through various projects are archived at the Marine Environmental Specimen Bank (Marine ESB), Hollings Marine Laboratory, in Charleston, South Carolina, USA, using standardized protocols for collecting, processing, and cryogenic storage. The protocols ensure a high quality sample is provided for downstream analysis that is fit-for-purpose and that homogenous aliquots are uniform, reproducible, and stable over time. New investigations exploring if the standardized protocols, 1) affect the quality and suitability of RNA for genetic expression studies and, 2) are feasible in determining trends in concentrations of perfluorinted alkyl acids (PFAAs) retrospectively, using samples stored and processed in polytetrafluoroethylene (PTFE) based materials, have recently been conducted. In addition, alternative uses of these cryopreserved specimens including, 1) a detailed proteomic profiling of tissues used to evaluate a new high-coverage well-scaffolded genome, and 2) the discovery of using total mercury as an alternate method to genetic species identification, have also been conducted and will be discussed.

607

Monitoring of the indoor environment of ESB laboratories with selected target and non-target screening methods

P. Bohlin Nizzetto, Norwegian Institute for Air Research; M. Schlabach, A. Halse, P. Rostkowski, NILU   Norwegian Institute for Air Research

The environmental specimen banks (ESBs) handles and store a wide range of high-quality samples with a main objective to perform retrospective analyses of newly identified contaminants (chemicals of emerging concern, CECs). It is of highest importance that the samples in the ESBs are representative of its environment and not contaminated during handling and storage. Strict quality assurance (QA) protocols are applied at the individual ESBs, including specially dedicated laboratories, no use of personal-care products, cleaning routines and more. More complicated to control is the indoor air and dust in the laboratories and storage locations. Building materials and indoor objects may contain and further emit or release CECS to the indoor environment. To evaluate this we have taken indoor air and dust samples from two European ESBs. Air samples were taken using passive air samplers while dust samples were taken using wet wipes on horizontal surfaces (not floors). Four room types were included at each ESB; main laboratory, changing room, storage room, and a corridor. The results show that several of the non-regulated CECs are found at high levels in the indoor matrices. These include: chlorinated paraffins, new flame retardants, and siloxanes. In contrast, regulated contaminants (e.g. polychlorinated biphenyls, PCBs, polybrominated diphenyl ethers, PBDEs) are found at low levels. Weather the high levels observed in indoor air and dust affect the ESB samples or not is to be further studied but the findings highlight an important aspect to take into consideration in the QA protocols of ESB.

608

DNA banking and its relevance for biodiversity research

J. Astrin, Zoological Research Museum Alexander Koenig

Within their genomes, the organisms on our planet contain an immense wealth of information about the diversity of life. These genomes conserve the code to identify organisms, comprehend population structures, etc. Fast progress in molecular technologies dramatically speeds up research on genetic biodiversity and increases the demand for professionally preserved and managed genome-quality samples in many disciplines, e.g. in ecology, conservation biology, etc. Biodiversity biobanks cater specifically to these demands, and in a standardized way. Environmental samples collected periodically by ESBs following defined routines constitute a very valuable source of DNA for biodiversity research, as they keep open a window that allows the parallel, correlative analysis of the chemical and of the species community composition of a given environment over time. Through species identification via DNA barcoding and high throughput sequencing, such samples can be most helpful in documenting and interpreting environmental change. Accumulated snippets of free DNA in soil or water samples (freshly collected or from ESBs), so-called environmental or eDNA, enable the comprehensive appraisal of species compositions in a multitude of environments. DNA extracted from ESB samples should be deposited in dedicated DNA banks in order to make this resource available to the scientific community and the fast-evolving DNA analysis methodologies (which may prompt repeated analyses of samples over time, with ever-increasing amounts of genomic data recovered). Thus, by adding DNA banking to their service spectrum, ESBs can considerably increase their visibility and public demand. Furthermore, they can foster knowledge aggregation at the biodiversity level around their ESB samples, making these more valuable.

609
Discussion on environmental specimen banking in research and regulation

Hazard and exposure assessment of chemical mixtures: steps towards increasing the realism of chemical risk assessment (II)

610
Poster spotlight: TH273, TH288, TH285

611
Environmental risk assessment of multiple stressors - chemicals and ionizing radiation
K. Petersen, NIVA - Norwegian Institute for Water Research; J. Brown, Norwegian Radiation Protection Authority; K. Tollefsen, NIVA / Ecotoxicology and Risk Assessment
Increased focus on cumulative effects of pollutants in the environment has led to development of several methods for environmental risk assessment (ERA) of chemical mixtures and for ionizing radiation. Even though no generic impact and risk assessment model exists to accommodate different types of stressors (e.g. multiple stressors such as radiation, metals and organic compounds), larger harmonization and integration of approaches taken can be achieved to improve and combine the existing models for ERA of chemical mixtures and for ionizing radiation from radionuclides. Here we present a potential 2-tired approach for risk assessment of multiple stressors by assuming additivity of chemicals and ionizing radiation as a first approach in order to combine a framework for ERA of chemical mixtures with the ERICA-tool for ERA of radionuclides. The proposed approach was applied to a real case scenario: emission from decommissioning of old oil platforms performed on-shore close to Vatsfjorden (Norway). Several metals, NORMs and organic pollutants are monitored as part of the activity. Effect data for the monitored compounds were compiled from various databases and literature. The Tier 1 identified a cumulative environmental risk of the stressors, and several metals and organics had a risk quotient above 1 (preliminary data). The potential for a cumulative environmental risk was verified in Tier 2 where species group specific risk was investigated. Metals were identified as the main risk drivers for algae, crustaceans and fish, where fish was identified as the most sensitive species group for this exposure scenario. Based on the used exposure scenario, compiled effect data and the suggested approach for ERA of multiple stressors, a potential environmental risk was observed. The main challenges and uncertainties for the proposed approach are linked to exposure data in terms of speciation and bioavailability; time consuming and subjective effect data compilation and assumption of additivity of ionizing radiation and chemical stressors. The approach for estimating environmental risk of multiple stressors requires validation through experimental studies, but could already serve as a suitable tool for prioritization of stressors and organisms of concern, and to identify knowledge-gaps in terms of exposure and effect data. **Acknowledgements:** The project was funded from NRC project 223268 (CERAD) and in-kind from Norwegian Institute for Water Research (NIVA)

612
Assessing health risk associated with micro-pollutant mixtures in drinking water: an innovative combination of in vivo and in vitro assays and analytical screenings
Y. Levi, Univ. Paris Sud / ESE UMR 8079; J. Loret, o. schlosser, CIRSEE Suez Environment; a. guillon, Suez / CIRSEE; m. favier, INSERM Institut Cochin; v. domergue dupont, Université Paris Sud; M. Bimbot, V. Huteau, Univ. Paris-Sud / UMR ESE; m. plewa, b. marinas, University of Illinois at Urbana Champaign
Assessing health risks associated with organic micropollutants in drinking water is a major challenge for public health and improvement of the drinking water production units. Given the low concentrations, the diversity of emerging contaminants and chronic exposure, it is essential to combine chemical analysis with biotests. In vitro bioassays or in vivo tests on aquatic animals allow effect screenings but interpretation for health risk assessment is difficult. We have designed an innovative research program (to our knowledge, the first of its kind applied to drinking water) that aimed to simulate human life-long exposure to micro-pollutants in drinking water for in vivo tests on mammals... Over 4 seasons, we have concentrated by 100 the organic fraction from large volumes of 4 types of water: river water, drinking water produced by a treatment plant with this raw water, reverse osmosis-treated water and bottled mineral water. 2 generations of mice (10 males, 10 females for each type of water) were fed with concentrates during a 1 year period, in order to reproduce in the animals, the exposure as a man drinking the water over 70 years. We carried out complete chemical analysis (quantitative targeted analysis, innovative non-targeted screening by HPLC and GC-2D with mass spectrometry), offering a new vision of the contaminants diversity. In vitro bioassays were used to evaluate endocrine disrupting effects (ER and AR recept.), cell toxicity tests (mammalian and Microtox) and genotoxicity on CHO cell assay. Mice growth, survival rate and behavior in open field and in elevated plus maze were studied. Histological analysis on 19 organs and blood hormonal assays were performed. This complete and innovative protocol did not show significant difference of survival rate and growth between the four mice groups. In contrast, histological and hormonal effects were observed in the mice exposed to the raw river water concentrates as opposed to the mice exposed to the waters of better quality. Several tests suggested a greater sensitivity of the 2nd-generation mice as compared with the 1st-generation. Biotests confirmed the overall good quality of the treated water, whatever the treatment process, despite the presence of organic micropollutants. The presentation will show the detailed protocol and the major conclusions of the study. This work, allowed health risk assessment with regard to long-term exposure to real mixtures of organic pollutants in drinking water.

613
The application of DGT to assess the risk of metal mixtures in polar environments
D. Koppel, University of Wollongong / Chemistry; M.S. Adams, CSIRO; C.K. King, Australian Antarctic Division; D.F. Jolley, University of Wollongong / School of Chemistry
Contaminants predominantly occur in mixtures, posing a challenge to environmental management practices which are usually based on single-contaminant toxicity. Chemical interactions of the contaminants and non-specific biological responses to these mixtures may result in effects that differ from the sum of the toxicity of individual components. These differences can be classed as antagonism (less toxicity than expected from the sum of the individual contaminants in the mixture), non-interaction (toxicity equal to that expected from the sum of individual contaminants), and synergism (more toxicity than expected from the sum of individual contaminants). Diffusive Gradients in Thin-films (DGT) has been established as a robust method for analysing the biologically-available contaminants *in situ* and is well-positioned to assess the toxicity of contaminant mixtures. This study explores the use of DGT (with a Chelex-100 resin) to assess the toxicity of Cd, Cu, Ni, Pb, and Zn in mixtures, to two common Antarctic marine microalgae, *Phaeocystis antarctica* and *Cryothecomonas armigera*.
Non-interactive and synergistic toxicity were observed in the two algal species in response to increasing multiples of an environmental mixture (where the ratios of metals were based on reported concentrations at a historically contaminated Antarctic marine bay). Non-interactive toxicity was observed in response to an equitoxic mixture (five metals at their EC10 concentrations), as determined by Independent Action and Concentration Addition modelling. DGT measured concentrations were able to predict toxicity to the two algae at environmentally realistic contaminant concentrations. The use of DGT in Antarctica was validated by field deployments to sediments and waters of the near-shore coastal environment of a contaminated site around an Australian Antarctic research station. Their use in assessing the risk of historical contaminants to the polar environment is discussed.

614
Ecotoxicity testing of environmentally realistic contaminant mixtures using passive samplers: what can we learn from repeating toxicity tests over an extended period of time?
S. Moeris, Ghent University (UGent) / Laboratory for Environmental Toxicology and Aquatic Ecology; F. Vanryckeghem, Ghent University / Sustainable Organic Chemistry and Technology; S. Huysman, Ghent University; K. Demeestere, Ghent University / Sustainable Organic Chemistry and Technology - Research Group EnVOC; L. Vanhaecke, Ghent University / Veterinary Public Health and Food Safety; H. Van Langenhove, Ghent University / Sustainable Organic Chemistry and Technology; C.R. Janssen, Ghent University / Applied Ecology and Environmental Biology; K. De Schamphelaere, Ghent University (UGent) / Applied Ecology and Environmental Biology
In current environmental risk assessment, researchers assess effects caused by single substances to single species and extrapolate for realistic conditions where organisms are usually exposed to complex contaminant mixtures. The use of passive sampling opens new possibilities to work with such mixtures and to transfer them into biotest systems by either applying passive dosing (for equilibrium based samplers) or extract spiking (for integrative samplers). Our research objective was to investigate whether or not environmentally realistic contaminant mixtures

(ERCMs) have effects on marine phytoplankton and how effects could be explained by measured contaminant concentrations. Further we looked at the repeatability of our test results over an extended time period of 16 months. In the presented research we used extracts of Speedisk™ passive samplers deployed in and outside of the harbour of Zeebrugge (Belgium) to spike several 72 h growth inhibition tests with the marine diatom *Phaeodactylum tricornutum* following ISO 10253. The different growth inhibition tests were performed over a period of 16 months with tests 0, 8 and 16 months after extraction. We observed statistically significant ($p < 0.05$) growth stimulation of up to 6.4 ± 0.5 % and 11 ± 2 % (in the harbour) and 7.0 ± 0.5 % and 14 ± 3 % (outside of the harbour) after an extract storage time of 0 and 8 months, respectively. After 16 months the previously observed effects disappeared completely. In order to explain the differing ecotoxicological responses a targeted analysis was performed for the quantification of more than 100 personal care products, pesticides, pharmaceuticals, (alkyl)phenols, phthalates and steroids. The analysis revealed that testing occurred at contaminant concentrations similar to those measured in water grab samples taken during sampler deployment. Remarkably the observed stimulation effects remained above 5 % when diluting the extracts up to 125 times. These findings suggest that *P. tricornutum* would remain affected by ERCMs even if their environmental concentrations would be reduced considerably. The disappearance of the observed stimulation effects after an extract storage time of 16 months led to the hypothesis that the main contributing contaminants causing stimulation must have degraded over time. In future work it would be of high interest to apply multivariate analysis (i.e. principal component analysis) to identify main contributing contaminants to the observed effects.

615

Marine Diatom Exposure to a Complex Mixtures of Fourteen Chemical Pollutants at Environmental Concentrations. What did we learn?

D. Napierska, European Commission Joint Research Centre / Directorate D Sustainable Resources Water and Marine Resources Unit; R. Carvalho, European Commission Joint Research Centre; A. Lahm, Bioinformatic consultant; S. Tasselli, European Commission Joint Research Centre / Directorate D Sustainable Resources Water and Marine Resources Unit; I. Sanseverino, European Commission Joint Research Centre; S. Balzamo, M. Potalivo, ISPRA Institute for Environmental Protection and Research; R. Loos, European Commission Joint Research Centre; D. Marinov, T. Lettieri, European. Commission - Joint Research Centre / Directorate D Sustainable Resources Water and Marine Resources Unit

Thousands different chemicals are discharged into the environment from agriculture, industry, medical facilities, house-holds. Currently, there is an increasing concern for the environmental impact of mixture of compounds since the additive and eventual synergistic effects are unknown and could produce serious adverse effects. To address this issue, a joint-effort of 16 European and associated research groups participated to an exercise to test a 14-substance synthetic reference mixture at safety environmental concentration under the Water Framework Directive (Environmental Quality Standard, EQS). The mixture, was tested on the own routine bioassays to investigate the chemical mixtures effects (Carvalho et al., 2014). The bioassays covered the entire ecosystem from bacteria to fish as well in vitro assays providing an unique scenario from ecological risk assessment perspective. The results showed that effects were observed at very low concentration on algal-bacteria composition in a marine microcosm, immobilization in crustacean, fish embryo toxicity and frog embryo development. Transcriptomics analysis was performed for the marine diatom *Thalassosiria pseudonana* exposed either to single compound or the mixture to investigate whether the single exposure and multiple exposure would show different gene expression profile pattern. The results show that the mixture induces a pattern similar to the ones induced by the single herbicides Diuron and Isoproturon. Signatures induced by the Cadmium or Nickel partially overlapped with the mixture signature. The exposure to the other compounds did in general not induce relevant signatures, although a weak overall consistent signature is present for some of them. In conclusion the effects of the mixtures could be explained mainly by the exposure to the two herbicides.

Advances in Soil Ecotoxicology and Risk Assessment of Terrestrial Ecosystems (II)

616

How protective is the current risk assessment for soil invertebrates?

P. Kotschik, Umweltbundesamt / Federal Agency of Environment / Risk assessment for plant protection products; J. Roembke, ECT Oekotoxikologie GmbH; J. Sousa, University of Coimbra / Department of Life Sciences; T. Natal da Luz, University of Coimbra / Department of Life Sciences, University of Coimbra; S. Chelinho, CFE Centre for Functional Ecology / Department of Life Sciences of University of Coimbra; N. Capela, CFE Centre for Functional Ecology; S. Pieper, German Federal Environment Agency (UBA) / Plant Protection Products

The present risk assessment evaluating effects from plant protection products (PPP) as well as other chemicals towards soil organisms encloses uncertainties. According to the current guidance on terrestrial ecotoxicology, the risk assessment for soil organisms exposed to PPP is based, in a first step, on results of standardised tests performed in artificial soil. This substrate is a mix of sand, peat, kaolinite clay and calcium carbonate and is not comparable to natural soils. However, limited data is available on the degree to which soil parameters influence the toxicity of chemicals for soil invertebrates in field soils, since toxicity is modulated by chemical sorption and bioavailability. Possibly, soil parameters like pH, organic matter content or texture, interact with each other in affecting the toxicity of different compounds to different soil organism groups. These uncertainties in the accurateness of the extrapolation from lab to field might possibly lead to an underestimation of the toxicity of test chemicals for soil organisms in natural soils. A screening project has been initiated in spring 2016, comprising a literature review aiming to investigate the effects of soil properties on the impact of PPP´s on soil organisms. The practical part of the project included laboratory studies on *Eisenia sp.* and *Folsomia sp.* on 4 PPP in 5 different soils. The results showed deviations on toxicity values obtained for single substances in different soils up to factor 6.4. The highest differences were detected for *Folsomia sp.* exposed to the active substance Pendimethalin in OECD10% and Lufa 2.1, respectively. So, the current risk assessment schemes for soil organisms based on standard laboratory studies performed with the surrogate species *Eisenia sp.* as well as *Folsomia sp.* is not always protective. Uncertainties in the assessment do exist regarding the effects of chemicals in natural soils towards other soil organisms, and other varying soil parameters that have not been investigated systematically so far (e.g. pH, clay content and interaction between them). The conducted literature research as well as the performed laboratory studies should be classified as preparatory work for more comprehensive studies. Focus of the future research should be set on the identification of key parameters influencing toxicity in different groups of species in order to create conceptual models allowing the extrapolation from the lab towards the field situation.

617

Risk assessment of soil organisms in field: dealing with earthworm community

Y. Bayona, F. Brulle, ANSES / U3EIV; A. Boivin, ANSES

For Plant Protection Products (PPP), regulatory risk assessment for soil organisms followed the Tiered Approach procedure. It covers worst-case situations (*i.e* conservative estimates and toxicity laboratory studies) to the most realistic assessment (*i.e.* Field studies). The recent EFSA opinion proposed a framework for risk assessment of soil organisms and definition of protection goals. Still, no statistical tool was proposed when dealing with field studies as higher tier. The aim of this communication is to test tools routinely used for regulatory risk assessment of communities. The tools for the ecological communities were mostly developed for aquatic organisms risk assessment. The multivariate analysis (PRC) is used for the community comparison. Individual populations are compared date-by-date using various statistical analysis commonly including Dunnett test and Williams-t-test. The main weakness pointed out for the higher tier studies is the low number of replicates which could lead to false negative or false positive. The recently improved Minimum Detectable Difference is used to assess the robustness of the data used in these statistical tests. Then, through the analysis of results, we propose some lead and improvements for the soil community risk assessment, from the experimental design to the sampling choice and statistical analysis in the context of higher tier regulatory risk assessment of chemicals for earthworm and soil communities.

618

Metal soil threshold calculator tool: use of available data for derivation of metal soil quality standards for different scenarios and protection goals

K. Oorts, ARCHE; I. Schoeters, Rio Tinto; J. Chowdhury, International Lead Association / Senior Scientist -Environment

During the last 2 decades, intensive research has been performed to improve the risk assessment of metals in soil and numerous chronic metal toxicity data were generated for various terrestrial species and microbial functions in different soil types. Models were developed for correction for differences in bioavailability among soils and for differences between laboratory and field conditions. For many metals, the large amount of chronic toxicity data for different species and functions allow the application of the species sensitivity distribution approach (SSD) to derive soil quality standards. This work was mainly triggered by the European legislation on chemical management (REACH) and the data were therefore primarily used to derive predicted no effect concentrations (PNEC) for prospective risk assessment. To facilitate a more flexible derivation of ecological quality standards for metals in soil for different protection goals (e.g. remediation thresholds), jurisdictions, regions or sites, while still making maximal use of the wealth of data and models already available, a metal soil threshold calculator tool has been developed. This freely available spreadsheet reports almost 1200 reliable chronic toxicity data for the direct effects of the metals Cd, Co, Cu, Pb, Mo, Ni and Zn to soil organisms (plants, invertebrates and microbial processes) and calculates ecotoxicological threshold concentrations expressed as (pseudo-)total (i.e. aqua-regia extractable) metal concentrations in soil (mg/kg dry weight). All metals covered have sufficient chronic toxicity data allowing the derivation of an SSD. The soils used for ecotoxicity testing cover for each metal a wide range of soil properties, making the results representative for most regions in the world. Several options are included to allow calculation of metal soil threshold concentrations for various goals (e.g. risk assessment or setting of remediation thresholds for different land uses): selection of organism groups or species to be considered, selection of effect levels from the original dose-response curves (ECx), selection of protection level (probability level in SSD), bioavailability models to be included etc. The advantages of this approach are the maximal use of available toxicity data and bioavailability corrections and the enhanced transparency in the derivation of ecological quality standards for metals in soil for different goals and different scenarios.

619

Assessment of pesticides on a landscape level- What is basically needed?

A. Toschki, Research Institute gaiac; M. Hammers-Wirtz, Research Institute gaiac / gaiac - Research Institute for Ecosystem Analysis and Assessment; B. Scholz-Starke, M. Ross-Nickoll, RWTH Aachen University, Institute for Environmental Research / Institute for Environmental Research

It is recently discussed and is widely accepted that species diversity and habitat quality dramatically decreased in the last decades due to the increasing intensity of agricultural land use. The decline was proved for various species groups, such as soil arthropods, insects, butterflies, birds etc. With the loss of species also a variety of benefits and ecosystem services that were provided by the species dropped away. The intensive use of pesticides can be quoted as a decisive factor, but the all explaining reason for this inacceptable loss of biodiversity cannot be assigned easily to a single factor and is more due to a multifactorial complex of influences which is responsible as a whole. In current risk assessment procedures, single pesticides were authorized individually independent of the current status of biodiversity in the field and possible effects of the combined or sequential use. Additionally, there is no transparent documentation of the use of pesticides on specific sites available. At the same time there is a lack of knowledge about the development of biodiversity in different agricultural landscapes because monitoring programs have not been established so far. The establishment of a systematic monitoring and a landscape risk assessment is necessary to connect future risk assessment with biodiversity as the protection target. Furthermore good ecological values must be derived and thresholds for the safe operating range must be defined. When the biodiversity level falls below the thresholds measures must be carried out. Geospatial models can help to optimise sustainable agricultural practice and measures for risk mitigation. The presentation will summarize result from different projects.

620

Potential new soil test requirements for the risk assessment of pesticides in the European Union: do we have the right methods?

J. Roembke, ECT Oekotoxikologie GmbH; J. Sousa, University of Coimbra / Department of Life Sciences

In February 2017, the European Food and Safety Authority (EFSA) published a Scientific Opinion entitled "Scientific Opinion addressing the state of the science on risk assessment of plant protection products for in-soil organisms". This document summarizes various scientific issues relevant for the risk assessment of plant protection products (PPPs) for in-soil organisms, i.e. micro-organisms and invertebrates (plants are covered in another EFSA document). The aim of this document is to provide an overview on existing regulatory requirements in soil ecotoxicology, but – more importantly – it also summarizes the scientific background for a new approach. In the future, soil risk assessment should be based on the ecosystem service approach, meaning that important services at agricultural landscapes (e.g. nutrient cycling, soil structure maintenance, just to name a few) have to be protected. In consequence, protection goals for soil organism communities, i.e. their biodiversity and functions relevant for providing these services, have to be defined. This contribution will focus on one important question, assuming that the new risk assessment approach is put in practice: Do we have the appropriate (e.g., scientifically sound, robust, standardized) methods to cover the new data requirements? In this contribution we focus on four aspects of the new methodology: Organism groups, endpoints, soils, regions (i.e. agricultural practices are not covered). In fact, there are various research needs which have to be addressed asap, i.e. before the new requirements will be put in practice. We focus our discussion on those test systems which are already standardized (or which are in the process of being standardized in the foreseeable future). Besides OECD also methods published by ISO or national organisations such as Environment Canada are considered, knowing that the latter often have to be adapted in order to cover the specific needs of PPP environmental risk assessment schemes. In addition, the same criteria as in other comparable approaches will be used and examples of suitable methods will be given. Most important is whether they can be modified in a way that they are useful under the upcoming EFSA regulations.

621

Poster spotlight: TH154, TH155, TH156

Emerging technologies and related raw materials requirements scenarios: the role of life cycle thinking

622

Wood-fibres composite in substitution of a synthetic material to enhance sustainability purposes for automotive sector

S. Maltese, Magneti Marelli SpA / Powertrain Division; a. bonoli, DICAM- Alma Mater Studiorum - University of Bologna / DICAM; L. Zanchi, M. Delogu, University of Florence / Department of Industrial Engineering

Nowaday bio-composite materials have increased automotive market penetration, whose intend is to produce environmental friendly products while maintaining their competitive edge [1]. In particular wood-fibers is considered a more environmental friendly alternative to the synthetic reinforcements as talcum, glass and carbon fibers. The reason behind is due to their less impact on production issues (sharp reduction of toxict and fossil materials as well as for the energy expedenditure, light-weight profile and more energy credit through their incineration [2-4]. This study presents the application of a new industrially engineered wood-based product, called *Woodforce*®, for the production of an automotive PedalBox Support (PBS); in particular, the environmental advantages/disadvantages are presented and discussed when such material is thought to substitute the glass fibres. At this scope, a comparative environmental analysis has been performed between two materials - a standard material (with glass-fibres content) and an innovative (with woodchip content) - which perform the same function. 1. References [1] Partanen A., Carus M. 2016. Wood and natural fiber composites current trend in consumer goods and automotive parts. Reinforced Plastics 60: (3) 170-173. [2] Joshi S.V., Drzal L.T., Mohanty A.K., Arora S. 2003. Are natural fiber composites environmentally superior to glass fiber reinforced composites? Composites: Part A 35 (2004) 371–376. [3] Boland S. Claire, De Kleine R., Keoleian G.A., Lee E.C., Kim H.C. Wallington T.J. 2015. Life Cycle Impacts of Natural Fiber Composites for Automotive Applications. Effects of Renewable Energy Content and Lightweighting. Journal of Industrial Ecology 20: (1) 179-189. [4] Holbery J., Houston D. 2006. Natural-Fiber-Reinforced Polymer Composites in Automotive Applications. Low-Cost Composites in Vehicle Manufacture.

623

Resource depletion of a Lithium ion battery cell technology

M. Cusenza, Università degli Studi di Palermo; S. Bobba, G.A. Blengini, Politecnico di Torino; M. Cellura, M. Mistretta, University of Palermo

Lithium-ion batteries (LIBs) are the dominating storage technology for electric vehicles (EVs). Different types of LIBs, using diverse cathode materials are available in the market, such as $LiMn_2O_4$, $Li(Ni_{1/3}Co_{1/3}Mn_{1/3})O_2$. The cathodes contain a wide range of raw materials (RMs), among which e.g. cobalt is in the 2017 list of CRMs for the Europe Union (EU). CRMs are both of high economic importance for the EU, and vulnerable to supply security. In the last years, the increasing demand of LIBs has triggered a growing interest in the need to ensure the security and the sustainability of the supply of the CRMs used in LIBs and in general in EVs. In this context, lithium rich layered oxides from the class xLi_2MnO_3-$(1-x)LiMO_2$ (M=Ni, Co, Mn), known as LMO – NCM, have drawn attention as cathode material due to their high discharge capacity and lower cobalt content, compared with the Ni-Co-Mn cathodes (NMC). In this context, the authors carried out a Life Cycle Assessment of an 11.4 kWh LMO-NMC battery cells useable in Plug-in EVs with the following main goals: to assess the impact on the mineral, fossil and renewable resources depletion (MFRRD); to estimate the requirement of CRMs; to identify the contribution of each cell component to the MFRRD; to compare the LMO-NMC LIB cell technology with an NMC cell technology available in the literature, with reference to the MFRRD and CRMs requirement. The LMO-NMC battery cell technology is modelled as $0.5LiMnO_2$ – $0.5Li\ (Ni_{1/3}Co_{1/3}Mn_{1/3})O_2$, using both primary and secondary data. The cells of the 11.4 kWh LMO-NMC battery are selected as functional unit. The system boundaries include RMs supply, manufacturing, transports and infrastructures. The results show that the LMO-NMC cells have an impact on MFRRD of 0.34 kg Sb_{eq}. The relevant share of MFRRD (34%) is caused by the cobalt sulphate production used in the cathode. Of the 27 CRMs for the EU, the analysis shows the relevance of only two of them: cobalt and barite. From the comparison with the NMC cell, carried out with reference to 1 kWh of nominal capacity, results that the MFRRD impact and the cobalt requirement of the LMO-NMC technology is lower, respectively, of a percentage equal to -4.4% and -29% than those of the NMC. The results indicate that the LMO-NMC cell could be a suitable technology to meet the demand of the EV market as it involves a lower impact on MFRRD and a lower consume of CRMs compared to the NMC cell.

624

Analysing the environmental impacts of alternative solutions for passenger transportation: LCA of a charging station for e-bicycles

G. Mondello, University of Roma Tre / Department of Business Studies; R. Salomone, L. Giuttari, G. Saija, G. Ioppolo, University of Messina / Department of Economics; M.C. Lucchetti, University of Roma Tre / Department of Business Studies

The transport sector causes higher environmental impacts that are mainly connected to the passenger car activities. In this context, strategies for reducing the environmental impacts related to the transport sector are required by moving from cars to alternative transport vehicles, such as electric bicycles. Although many studies have been focused on the application of the Life Cycle Assessment (LCA) method for assessing the environmental impacts of electric vehicles (EVs), there are few information regarding the environmental impacts connected to the life cycle of the charging infrastructures to operate the EVs, and, in particular, regarding the environmental performance of charging station for e-bicycles This study aims to evaluate, through the application of the LCA method, the environmental burdens of a stand-alone charging station for electric bicycles manufactured in Italy, assuming its installation and utilisation in Italy with a lifetime of 15 years. The investigated stand-alone charging station is composed of eight designated positions for charging the e-bicycles' battery and of two wind-solar modular systems that are respectively equipped with one photovoltaic (PV) module and three low profile vertical axis wind turbines. The structure is also geared with a system connected to the conventional electricity grid which allows to provide energy when the wind and solar conditions are insufficient. The analysis also includes the whole life cycle of eight e-bicycles, as well as the potential avoided production of conventional electricity which may be replaced by the electricity surplus produced by the wind-solar system. The functional unit (FU) adopted to perform the analysis is one charging station installed in Italy and used for a lifetime of 15 years. System boundaries follows a cradle-to-grave approach and include four main phases: 1) production phase, 2) installation phase, 3) use and maintenance phase, and 4) end of life phase. The results undersore that the Climate Change impact related to the whole life cycle of the investigated charging station is 13,816.5 kg CO2 eq per FU. The main environmental impacts are due to the production phase and, in particular, are connected to the production of the e-bicycles, while higher environmental benefits are connect to the replacement of conventional electricity with the energy surplus produced by the wind-solar modular system installed on the charging station.

625
Raw materials requirements scenarios for the electric mobility penetration in the Italian urban vehicle fleet: a life cycle thinking approach combined with raw materials assessment
L. Cutaia, C. Chiavetta, P. Porta, ENEA; M. La Monica, C. Scagliarino, CINIGeo
The automotive fleet is gradually changing from internal combustion to hybrid and electric engines. This is fostered by the European policies concerning the need of reduction in pollutant emissions and in the improving of urban air quality. Currently most of the analyses carried out focus on the comparison of the emissions related to the different typologies of cars and on the CO_2 emission cut generated by the consumption of electricity produced from renewable sources in the use phase of the car. The proposed approach wants to investigate the need of raw materials in future scenarios of electric technology penetration in the urban vehicle fleet. Focusing on the Italian urban vehicle situation and considering the current technology used for the electric batteries and the recharging systems, the study account for the raw materials needed for different penetration scenarios of the electric mobility, adopting a Life Cycle Thinking perspective. The authors have been beyond a mere quantification of the primary materials required for the progressive electrification of the urban fleet, evaluating the raw materials availability from a market perspective and defining the limiting factor among three basic material of the current electric technologies (lithium, graphite and manganese). Therefore this work, starting from an evaluation of the material needed for different electric mobility penetration scenarios in the Italian urban fleet, wants to highlight the market dynamics especially for 3 primary materials widely required in the electric mobility focusing also on the possibility offered by a transition to a circular economy, investigating the green mining potential available for these materials both for their recovering in the same technology and in others fields.

626
Coupling dynamic carbon accounting and partial-equilibrium economic model for energy policy assessment
A. Albers, P. Collet, D. Lorne, IFPEN / Economics & Technology Intelligence; A. Benoist, CIRAD / UPR BioWooEB ELSA research group; A. Hélias, Montpellier SupAgro / LBE ELSA
Energy production and consumption is the main driver for anthropogenic GHG emissions, and in the French context, the transportation sector is the principal emitter accounting for almost one third of these emissions. The growing need to reduce GHG emissions and mitigate climate change demands tapping alternative energy resources, as currently enforced by energy policies (e.g. the French Transition for Green Growth Act). LCA scholars increasingly assess the environmental performance of the advance biofuels, but mainly from a static perspective. Results are therefore limited to linear simplifications, whereby long-term impacts might be neglected or underestimated. New dynamic LCA approaches have been suggested, however no consensus is available on how to treat Cbio sequestration dynamics over different timeframes. This study further addresses the temporal shortcomings of bioenergy systems while considering future outlooks and consequences on the market dynamics. The approach consists of a hybrid-approach combining the MIRET energy systems model with dynamic Cbio accounting models towards dynamic LCA. The former —a prospective techno-economic partial-equilibrium model covering the French energy-transport sector— represents scenario-dependent outputs over a long timeframes (2007 to 2050), exploring optimisation options under no-policy and policy-driven constraints. The latter assesses biomass growth and allometric relations representing the Cbio fixation of a vegetation species per hectare on an annual basis, and thus the time-dynamic Cbio flows between the atmosphere and the technosphere. The assessed Cbio flows primarily originate from lignocellulosic biomass and their co-products generated from MIRET outputs under business as usual and normative scenarios. The transformed Cbio inventories are then combined with both dynamic and static LCA characterisation factors, towards a comparison of both approaches. The results show that the time factor is an essential component to properly assess long-term Cbio sequestration potentials and climate benefits of lignocellulosic biofuels. The consideration of technological innovation and market dynamics in a transitioning energy system expands the assessment boundaries providing insights into least cost (economic optimisation) and low carbon (Cbio sequestration) options influenced by policy and decision constraints. Future refinements addressing other bioenergy paths are envisaged.

627
Poster spotlight: TH304, TH309, TH314

Developments in the use of bioassays for chemical and environmental risk assessment (II)

628
SIMONI: Smart integrated monitoring of the water quality
R. van der Oost, Waternet / Onderzoek en Advies; G. Sileno, Waternet / Research and Development; M. Thao Nguyen, Waterproef; L. Moria, Waternet / Water Systems
At present, regular water quality assessment is almost exclusively performed by target chemical analysis of substances. Scientific research over the last decades, however, demonstrated that water quality assessment with only chemical analyses is not reliable. Since over 100,000 harmful substances may be present in the aquatic environment, a paradigm shift from 'substances' to 'effects' has to be encouraged in order to implement a more holistic approach in regular monitoring. Therefore, an alternative Smart Integrated Monitoring (SIMONI) strategy has been developed by Dutch water research institutes. The purpose of this 2-tiered strategy is to obtain more reliable information on the chemical water cycle quality without increasing the monitoring costs. Key factors for generating this model were the selection of the most relevant bioassays and the design of effect-based trigger values (EBT). Tier 1 of the strategy combines micropollutant concentration by passive sampling with testing of 15 bioanalytical endpoints. This hazard identification makes the distinction between low, acceptable and increased ecological risks. Only at sites where tier 1 indicates increased ecological risks, a customized tier 2 research is performed to identify the chemicals that cause the bioanalytical effects and to evaluate if observed *in vitro* effects pose a serious *in vivo* threat to the ecosystem. The present paper is focused on demonstrating the field feasibility of the SIMONI strategy. The strategy has been applied and validated in more than 100 Dutch field studies. Results of several of these monitoring studies will be presented in order to demonstrate the field feasibility of the SIMONI strategy for identifying hot-spots of chemical pollution. It appears that highest ecological risks generally occurred at agricultural sites. In addition, increased ecological risks were also observed at waters receiving wwtp effluents, sewage overflows and landfill runoff. A tier 2 approach is the assessment of the mixture toxic pressure on the ecosystem by using chemical analytical results for msPAF determination (potentially affected fraction of water organisms due to multiple substances). At most sites with increased ecological risks due to micropollutants, similar classifications were obtained with bioanalytical and extensive chemical analyses. The tier 2 research at agricultural greenhouse areas identified eight pesticides contributing most to the increased environmental risks.

629
Bioassay battery responses to POCIS and Speedisk passive sampler extracts
M. de Baat, University of Amsterdam / IBED-FAME; M. Thao Nguyen, Waterproef; R. van der Oost, Waternet / Onderzoek en Advies; W. van den Berg, Waterproef Laboratory, Research and Validation; P. de Voogt, University of Amsterdam / IBED; M. Kraak, University of Amsterdam / IBED-FAME
A large portion of the toxic effects observed in surface waters cannot be explained by compounds that water authorities regularly monitor. Since chemical analysis of the myriad of substances present in the aquatic environment is practically and economically not a viable option, there is a growing interest in effect-based monitoring. Additionally, concentrations of compounds typically vary over time and therefore traditional grab sampling only provides a snapshot of the chemical make-up of a water body. Passive sampling can overcome this limitation by exposing a sorbent to the target environment, accumulating compounds from the water over time. Hence, there is an urgent need for a time-integrated effect-based monitoring strategy that employs a combination of passive sampling and bioassays, thus detecting bioanalytical responses caused by mixtures of all bioavailable compounds. Many pollutants of emerging concern are polar compounds, underlining the need to standardize the employment of polar passive samplers in such monitoring strategies. The aim of the present study was, therefore, to determine bioassay battery responses to extracts of two types of polar passive

samplers, the often used polar organic chemical integrative sampler (POCIS) and the recently introduced Speedisk. POCIS and Speedisk passive samplers were simultaneously deployed at sites likely to be contaminated with polar compounds, including agricultural greenhouse sites and wastewater treatment plant (WWTP) impacted locations. The extracts of the passive samplers were subjected to a battery of bioassays, specifically responsive to polar compounds, including ERα, anti-AR and GR chemical activated luciferase gene expression (CALUX) bioassays, as well as the RIKILT WaterSCAN for antibiotics activity. In addition, the Microtox test for non-specific toxicity to bacteria was run. The two investigated passive sampling devices generated a different toxicity profile in the applied bioassay battery, with several unique responses per passive sampler. Nonetheless, POCIS caused bioassay responses more frequently and more intensely, leading to more frequent trigger value exceedances and thereby to the detection of ecotoxicological risks. Hence, POCIS outperformed Speedisk in most bioassays at the majority of the investigated locations. These results thus suggest that POCIS is best fit for application as passive sampling device targeting polar compounds in effect-based water quality monitoring strategies.

630

Endocrine modulation and toxic effects of sunscreen chemicals, Octocrylene and Benzophenone, on zebrafish

Q. MENG, The Chinese University of Hong Kong; K. Chan, The Chinese University of Hong Kong / Life Sciences

Sunscreen chemicals are used to prevent the skin and hair of human from the harm of ultraviolet light, and they are widely applied in a variety of personal care products. These materials are discharged to the environment continuously as daily care products. At present, many kinds of UV filters are found in surface water and polluted water, and their concentrations in the urine and blood of human are much higher than that in the environment, indicating that the UV filters have the possibility of bioaccumulation to cause toxic effects in the accumulated organisms. Benzophenone-1 (BP1), benzophenone-3 (BP3), benzophenone-8 (BP8) and octocrylene (OC) are four such chemicals that have been detected in environmental samples and linked to alterations in estrogen receptor signalling pathways and oxidative stress. In this study, zebrafish larvae and a liver cell model of zebrafish liver cells, the ZFL cell line, will be used to investigate the potential risks of BPs and OC and the molecular mechanism underlying the toxic effects. Docking analyses, 24 h and 96 h chemical exposures will be conducted on the ZFL cells to determine the potential binding affinities to the estrogen receptor (ER) and the half-lethal concentration (LC50) for the UV filter chemicals. The gene expression profiles on the ER pathway and the aryl-hydrocarbon receptor (AhR) pathway will also be measured by quantitative real-time PCR in ZFL and larvae exposed to the sunscreen chemicals. A dual-luciferase reporter gene system with AhR and ER clones transfected in ZFL cells will be used to confirm the biological activities of these sunscreen chemicals in ZFL cells.

631

Current status of in vitro bioassay approach in environmental risk assessment of abiotic environmental mixtures and individual organic contaminants

M. Machala, Veterinary Research Institute / Chemistry and Toxicology; K. Pencíková, S. Strapácová, Veterinary Research Institute, Brno / Chemistry and Toxicology; L. Svrzková, Veterinary Reseach Institute, Brno / Chemistry and Toxicology; J. Neca, M. Ciganek, Veterinary Research Institute, Brno / Chemistry and Toxicology; I. Bartonková, Z. Dvorák, Faculty of Science, Palacky University, Olomouc; J. Topinka, Institute of Experimental Medicine, CAS, Prague; J. Vondrácek, Institute of Biophysics, CAS, Brno

Since 2001, our laboratory has continuously employed an ever growing set of in vitro bioassays combined with a detailed chromatographic analysis, and in some cases the effect-directed analysis, in order to identify principle modes of action of contaminants bound to river sediment, airborne or diesel exhaust particles. Toxicity profiling of selected individual contaminants was used as a complementary approach, which aimed to identify major toxic modes of action and principle contributors to specific toxicity effects. The AhR-mediated activity has been recognized to play a key role in toxicity of organic extracts of abiotic environmental mixtures. Using the DR-CALUX assay, we established relative effective potencies (REPs) for a large series (>100) of polycyclic aromatic compounds, including PAHs, methyl-, nitro- and oxy-PAHs, thiophenes, dinaphthofurans, benzacridines and carbazoles, which complemented the available data on polychlorinated biphenyls, dibenzo-p-dioxins and dibenzofurans. Recently, we also developed REP values in human AhR-dependent reporter gene assay, in order to compare the potencies in rodent and human models. Using a set of various CALUX assays, receptor-specific gene expression analysis (using qRT-PCR methodology), as well as non-receptor-based toxicity screening methods including estimation of stable DNA adduct production, oxidative DNA and lipid damage, and inhibition of cell-to-cell communication, we further studied toxic potencies of both mixtures and selected individual compounds to induce (anti)estrogenic, (anti)androgenic, TRalpha-, CAR-, PXR- or other receptor-mediated activities of both environmental mixtures and selected individual classes of organic contaminants. The general outline of those studies will be illustrated using an example of a detailed in vitro toxicological evaluation of standard reference material of diesel exhaust particles (SRM 2975), with an aim to document both the complexity of the observed effects and the difficulties faced when applying these data in risk assessment of complex mixture. [The study was supported by the Czech Science Foundation, grant no. P503-12-G147.]

632

Hormone-like activities in waste water characterized by CALUX bioassays, chemical analysis and Effect-directed Analysis

Y. van Oorschot, R. ten Broek, The Water Laboratory; M. Lamoree, VU University, Department Environment & Health / Department Environment & Health; N. Zwart, VU University Department Environment & Health; C.J. Houtman, The Water Laboratory

Emission of compounds with biological activities from waste water treatment plant (WWTP) effluents into surface waters is a topic of concern for ecology and drinking water companies. We investigated the occurrence of hormone-like activities in WWTP samples and pursued to identify compounds responsible for them. To this aim, CALUX bioassays and a UPLC-tQ-MS target analysis method for hormones used in high volumes in pharmacy were applied. In addition, a novel high throughput Effect-directed Analysis (HT-EDA) platform was used to separate compounds in the extracts with high resolution LC-fractionation creating 288 4.79 sec.-fractions that were tested in the bioassays. In parallel, QToF high resolution MS data were recorded to correlate compound identity to peaks from the 'bioassay chromatograms' reconstructed from the bioassayed fractions. All five types of activities tested were observed in the WWTP samples. Androgenic and estrogenic activities were almost completely removed during WW treatment, anti-androgenic activity was only found in treated WW. Glucocorticoid and progestagenic activities were present in similar concentrations in untreated as in treated WW. The glucocorticoid activity in influent was fully explained by prednicarbate, triamcinolone acetonide, dexamethasone and amicinonide. In effluent however, detected hormones could only explain 15% of the activity, indicating the presence of unknown (metabolites of?) glucocorticoids in effluent. The androgenic activity in both influent and effluent could predominantly be attributed to the presence of androstenedione and testosterone. Application of the HT-EDA-platform delivered bioassay chromatograms of the WWTP effluent in which active compounds were separated into sharp peaks. The glucocorticoid activity appeared to be caused by at least four different compounds (peaks), not being dexamethasone. The androgenic activity was fractionated into a small peak probably attributable to co-elution of testosterone and androstenedione. The QToF MS data are currently interpreted to elucidate the identity of the unknown glucocorticoids. Also bioassaychromatograms of other endpoints will be discussed. This study demonstrates the value of toxicity profiling with bioassays as first tier in the monitoring of water quality. In case observed activities exceed trigger values, additional risk assessment is needed and the HT-EDA platform can help to characterize and ultimately identify the responsible compounds.

633

Non-target screening and identification of emerging pharmaceuticals and their transformation products in wastewaters

c.G. Parège, Université de Bordeaux / EPOC / LPTC UMR 5805 CNRS; M. Dévier, University of Bordeaux / EPOC / LPTC UMR 5805 CNRS; E. Maillot-Maréchal, INERIS / UMRI SEBIO ECOT; E. Geneste, University of Bordeaux / EPOC / LPTC UMR 5805 CNRS; S. Ait-Aissa, INERIS / UMRI SEBIO ECOT; H. Budzinski, University of Bordeaux

Wastewaters represent a major pathway of introduction of EDCs into the aquatic environment. Considering the large diversity of compounds and their possible transformation products, many EDCs are currently unknown. Therefore, characterization of the presence and identification of EDCs in wastewater are major issues in order to assess their occurrence in natural waters and the associated risks for wildlife. To date, *in vitro* assays based on luciferase reporter gene expression, are available to assess the biological activities of samples in a quantitative, sensitive, specific and fast way. The comparison of concentrations derived from bioassays and from chemical analyzes allows assessing the contribution of micropollutants to the overall biological or toxic activity of a sample. In this study, a systematic approach combining effect-directed analysis (EDA) and high resolution spectrometry was applied to several urban WWTPs to establish an overall contamination diagnostic and to identify major contaminant that could be released in environment. For this purpose, crude extract from influent and effluent of an urban WWTP were analyzed by LC-QTOF and tested on estrogens, androgens and glucocorticoids receptors. The non-target screening allowed detecting more than 7000 and 4000 signals in influent and effluent extracts of WWTP, respectively. We observed that 70% of compounds detected in effluent were produced by the treatment process suggesting the formation of transformation products. Concerning the strategy implemented to identify compounds of interest, it was decided to use effect-directed analyses methodology to have a tool to target active compounds in relation with selected biological activity. To this end, the crude extracts were fractionated by HPLC and biologically active fractions were isolated for further chemical identification. Biological fractionation profile of samples indicated the presence of estrogenic and glucocorticoid activities at all studied sites with very similar fractionation patterns between sites, highlighting major and recurrent individual fractions. The identification of active compounds was performed using LC-QTOF and several drugs and their transformation products (*e.g.* O-desmethyl venlafaxine, O-desmethyl tramadol) were recurrent found. This approach combined to LC-QTOF has allowed establishing a list of systematic detected non-target compounds in several wastewaters.

Indigeneity and Science: A collaborative work in progress

634

The Nechako White Sturgeon Recovery Initiative: A discussion of species at risk conservation, scientific outreach, community and First Nations support

T. Lane, University of Saskatchewan; C. Williamson, Freshwater Fisheries Society of British Columbia; J. Alcaraz, K. Shekh, University of Saskatchewan / Toxicology Centre; M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre

Nechako white sturgeon (*Acipenser transmontanus*) are a genetically unique population of fish which have inhabited the Nechako watershed for roughly 10,000 years. Within the last 50 years this population has suffered significantly because of anthropogenic activities. In 2003, the Committee on the Status of Endangered Wildlife in Canada identified the Nechako white sturgeon as a Nationally Significant Population. In 2006, this population was further listed as *endangered* under the Species at Risk Act. Prior to both designations in 2000, the Nechako White Sturgeon Recovery Initiative (NWSRI) was established in Vanderhoof, British Columbia. The goal of the initiative is the conservation and recovery of this one-of-a-kind population of white sturgeon which hold intangible cultural value within communities surrounding the Nechako watershed. A 5-million-dollar aquaculture facility, the Nechako White Sturgeon Conservation Centre was designed specifically for the NWSRI, built, and opened in 2014 to provide the resources to further support this conservation effort. This facility was a product of over a decade of work by a Technical Working Group (TWG) and a Community Working Group (CWG). The TWG includes biologists, industry and First Nations members who have a vast knowledge of white sturgeon. The goal of the TWG is to use the best available science, local and traditional knowledge to determine why the Nechako white sturgeon population is declining and to develop a plan to rehabilitate this population of fishes. The CWG is composed of First Nations members, local and regional governments, industry, and public volunteers. The CWG plays a vital role in communication, public outreach, and promoting community involvement. Activities of TWG and CWG support the mandate of the NWSRI through direct involvement of First Nation communities, volunteers and students. The Emergency Sturgeon Live Release Boat Kit program is an example of multiple First Nation communities working in union with the NWSRI to promote conservation and stewardship of Nechako white sturgeon. The annual Juvenile Sturgeon Release event involves students, volunteers and First Nations who release thousands of juvenile sturgeons, which were reared from eggs by the TWG, back into the Nechako watershed. The NWSRI is a unique conservation effort that promotes and utilizes the support of scientists, First Nations, volunteers and students to engage the community in the conservation of an endangered species.

635

The NSERC CREATE H2O Program on First Nations Water and Sanitation Security: Case Studies on Drinking Water Quality

A. Farenhorst, Universsity of Manitoba / Soil Science; W. Ross, University of Manitoba / Centre for Human Rights Research; R. Mi, University of Manitoba / Department of Soil Science; R. Patidar, University of Manitoba / Department of Microbiology; G. Amarawansha, K. Anderson, University of Manitoba / Department of Soil Science; E. Khafipour, University of Manitoba / Department of Animal Science; A. Kumar, University of Manitoba / Department of Microbiology

The NSERC CREATE H2O program is the first science-engineering research training program in Canada that combines technical water and wastewater management training with Indigenous theory, law and methodological skills training. Since its inaugural year in 2013, the program has trained 86 students who have collectively worked with 30 First Nations communities in the provinces of Manitoba, Ontario and Saskatchewan, Canada. 33% of the university students and postdocs enrolled in the program self-identify as Indigenous. This presentation provides an overview of the approaches the program is utilizing for: engaging communities and students in research training activities, Indigenizing science and engineering curricula, and designing advocacy strategies to support clean drinking water as a human right in First Nations communities in Canada. Case studies are presented to demonstrate the community-based monitoring programs implemented to examine drinking water quality in First Nations homes. First Nations communities participating in the research had various types of water distribution systems. Overall, water samples were collected from: lakes and groundwater (source water); water treatment plants, water trucks and community standpipes; homes relying on piped water, wells, above-ground cisterns and underground cisterns; and buckets/drums in homes without running water. Water analysis included standard measures of chemical and bacterial parameters, DNA and RNA techniques for microbial profiling, and the quantification of antibiotic-resistance genes in water samples. The main findings are that despite effective water treatment plants in communities, the tap water in many First Nations homes contained fecal bacteria at alarmingly high levels and antibiotic-resistance genes were also detected in a range of drinking water samples. The issue of poor drinking water quality in First Nations communities in Canada remains unsolved and there is an urgent need for improved monitoring and upgrading of infrastructure, especially in communities relying heavily on cisterns and community standpipes for drinking water supplies. Most importantly, investments to connect homes directly to water treatment plants via improved pipelines is key to reducing human exposure to waterborne illnesses, while enhancing options for families to participate in economic development, food security and spiritual and cultural wellbeing in their communities.

636

Rare earth elements (REEs) in the Canadian Subarctic: scientific perspectives and community engagement with environmental monitoring in Nunavik, Northern Quebec

G.A. MacMillan, Centre détudes nordiques, Université de Montréal / Department of Biological Sciences; J. Gérin-Lajoie, Université du Québec à Trois-Rivières / Centre détudes nordiques, Département des sciences de lenvironnement; J. Chetelat, Environment and Climate Change Canada / Ecotoxicology and Wildlife Health; E. Hébert-Houle, Université du Québec à Trois-Rivières / Département des sciences de lenvironnement; J. Rowell, University of Montreal / Department of Chemistry; J. Heath, The Arctic Eider Society; H. Snowball, The Northern Village of Kangiqsualujjuaq; R. Mickpegak, Sakkuq Landholding Corporation Kuujjuaraapik; M. Amyot, Universite de Montreal / Département de sciences biologiques

Many communities in Canada's North are increasingly concerned about the impacts of large-scale socio-environmental changes, such as climate change and industrialization, on their ways of life. One main concern for northern communities is the environmental impact of mining, including the large number of rare earth element mining projects that are currently being developed in northern Canada. Rare earth elements (REEs) are a chemically-similar group of contaminants of emerging concern, which includes the 15 trivalent lanthanide metals. Not particularly rare, REEs are increasingly exploited for essential uses in high-tech industries, including electronics, clean energy, and agriculture. Although knowledge of the fate of REEs and their impact on natural ecosystems is critically needed as emissions increase, few ecotoxicological studies exist for REEs particularly field-based studies on their bioaccumulation and food web dynamics. To investigate how REEs behave in remote northern food webs, samples were collected from community-driven collaborative research projects with the Inuit communities of Kuujjuarapik-Whapmagoostui (K-W) and Kangiqsualujjuaq in Nunavik (Northern Québec). The combined objectives of these projects were a) to study the behaviour of REEs in northern ecosystems before the start of mining activities and b) to engage community members in the research process through the co-development of objectives, sample collection and the sharing of research results. We asked questions from the scientific perspective and on community engagement: Can we detect REEs in freshwater, marine and terrestrial plants and animals? Do REEs bioaccumulate and bioamplify in northern food webs? How to best establish a sustainable community-based environmental monitoring program? Can we engage the youth in environmental monitoring and science education? Wildlife harvesting and tissue sampling was partly conducted by local hunters, including a range of key species important for both biomonitoring and country food. Indigenous knowledge was used in the study design, to coordinate local sampling, and to decide when, where and what species to collect. Our study presents novel data on the behaviour of REEs in northern ecosystems and recommendations for establishing sustainable and effective community-based environmental monitoring projects with indigenous communities for emerging contaminants.

637

Te Ohu Mō Papatūānuku: A Collective Response to Healing

T. Godfery, H. Hireme, Te Whare Wananga O Awanuiarangi / School of Undergraduate Studies

The use of pentachlorophenol (PCP) as an anti-sapstain in timber treatment, with subsequent disposal of chemically treated wood waste in the Whakatane District of New Zealand has resulted in a legacy of contamination. There are 36 identified wood waste sites located on private and public lands, as well as the customary lands and waters of the indigenous Ngati Awa people. The pervasive effects of contaminants upon both human and environmental health has led to the formation of the collaborative group Te Ohu Mo Papatuanuku. The collaboration is steered by indigenous members of Ngati Awa, whilst being strongly supported by scientists, local government agencies, and industry. As a consequence of ongoing research, the use of a rather unique approach utilising combined myco- and phyto-remediation to remediate dioxin-contaminated land has been adopted. Whilst implementation of the approach is underpinned by science, the use of "nature to heal nature" is an approach that resonates with the indigenous community. Contemporary environmental problems resulting from anthropogenic activities often require the use of scientific based solutions. Hence, even when indigenous participation is encouraged by the scientific community as part of the problem solving process, the contribution of indigenous knowledge may be considered of less value than scientific knowledge. Of vital importance to ongoing environmental care however, is the role of indigenous knowing – indigenous relationships informed by binding and enduring familial links with lands, waters, and people. This presentation builds upon previous presentations detailing the journey of the Te Ohu Mo Papatuanuku research collaboration – using a synchronistic approach –

whereby science is an integral remedial component and provides a vehicle for remediation to occur, but cultural and soul connections of the indigenous people are the drivers of reciprocal remediation, and subsequent healing. Scientific knowledge applies science and indigenous knowing revitalises relationships, informing and infusing behaviour with an ethos of respect, empathy, and reciprocity.

638
Discussing the Unfamiliar but Contentious: Hydraulic Fracturing Consultation with Remote, Indigenous Communities in the Northern Territory, Australia
R. Smith, Hydrobiology Pty Ltd; R. Pepper, Land and Environment Court of New South Wales; D. Ritchie, Ninti ONe Foundation
On 14 September 2016 the Northern Territory (NT) Government announced a scientific inquiry into hydraulic fracturing of onshore unconventional reservoirs in the Northern Territory (the **Inquiry**) under the *Inquiries Act (NT)*. The Inquiry was required to assess the available scientific evidence to determine the likely nature and extent of the environmental impacts and risks, including cumulative impacts and risks, associated with hydraulic fracturing of unconventional reservoirs and the Associated Activities in the NT. The Inquiry was specifically instructed to assesswhether or not there would be unacceptable impacts or risks to economic, cultural and social conditions, by developing and implementing a stakeholder engagement program. Indigenous people make up most of the resident populations in the areas of the shale-gas basins in the Northern Territory. Indigenous people are linked with their land (including waterbodies) by their ancient traditions and contemporary use of their land in accordance with those traditions. As a community, they must be able to maintain their cultural traditions relating to that land in order that their ownership rights continue to be recognised, from one generation to the next. Indigenous communities are therefore particularly vulnerable to degradation of the landscape and the natural systems it supports. Therefore, the Inquiry undertook focused stakeholder consultation with remote indigenous communities across the NT in three rounds of visits for community forums and hearings. These could not be conducted in the same manner as larger community consultation, and featured language and background knowledge barriers that are not typical of even remote non-indigenous community consultation in Australia. A number of these issues, and how they were addressed are discussed. Although the timeframe allotted to the Inquiry was particularly challenging for indigenous community consultation, and hence the extent of engagement in the process varied greatly between communities, nonetheless the Inquiry did achieve substantial engagement with most. The community feedback gained thereby was a vital input into the Inquiry's assessment of the potential cultural impacts of shale gas development in the NT.

639
Incorporating cultural values and perspectives of First Peoples' (Aboriginal People) into water planning, science and environmental water management
B.J. Moggridge, Institute for Applied Ecology, University of Canberra / Institute of Applied Ecology
Australia is the driest inhabited continent on Earth, yet is has been the traditional lands of its original inhabitants Australia's First Peoples for thousands of generations. Protecting water landscapes (surface and ground water) has always been a high priority for survival in a dry landscape, and protecting water remains a cultural obligation. The challenge for First Peoples is to ensure their value and relationship with water is not diminished or excluded by modern day water planning and science or from environmental flow management. First Peoples acquire the right wisdom and traditional science and knowledge and many indicate that their worldview is seeing water as inseparable from the land and the sky, bound by traditional lore and customs for its protection. For Australia's First Peoples, occupying an ever drying landscape, traditional knowledge of finding, re-finding and protecting water sites has been integral to their survival for so long. This paper will explore relationships between First Peoples and water planning and environmental water management in three ways. Firstly, history, challenges and institutional responses in integrating First Peoples cultural values into water planning, science and management. Secondly, propose a series of on-the-ground applications of cultural water and environmental water empirically. Finally, integrating First People's science into water management will be assessed through comparisons between the Australian situation through case studies looking at models and methodologies.

Improvements in environmental exposure assessment: Development and application of tools across industry sectors, regulatory agencies, and international boundaries (II)

640
Tap water intake of poly- and perfluoroalkyl substances (PFASs) in relation to serum concentrations in a nationwide prospective cohort of U.S. women
X. Hu, F. Laden, Q. Sun, P. Grandjean, Harvard University; L.W. Yeung, University of Örebro / Department of Chemistry; E.M. Sunderland, Harvard University / School of Engineering and Applied Sciences
Drinking water advisory levels have been adopted by many regulatory agencies to reduce chronic exposure to persistent fluorochemicals. However, most U.S. advisory levels are based on the assumption of approximately 20% of overall PFAS intake comes from drinking water. Better characterizing the relative importance of drinking water to overall human exposures is important for developing health protective guidelines. Most previous investigations that have associated drinking water PFAS exposures and total body burdens of these compounds have focused on highly contaminated sites. Exposures to PFASs for the general population of individuals from geographically diverse areas are thus less understood. Here we investigate the relative importance of drinking water for total PFAS exposure among women in the Nurses' Health Study (NHS), a large U.S. based cohort study. Our analysis included U.S. women who provided a residential drinking water sample in 1989-1990. We measured concentrations of 11 PFASs in a subset (n=111) of matched archived drinking water samples and serum samples. We evaluated the relative importance of home tap water for measured levels of PFASs in human using both statistical and toxicokinetics (TK) models. Results suggest that home tap water is a significant exposure source for general American women. In 1989-1990, the median contribution of drinking water to serum PFASs in women in the NHS cohort was 8.8% to 30% for the five PFASs modelled. This ratio varies across individuals and compounds by up to a factor of 2-3. We will next investigate how this ratio varies geospatially and whether it is associated with distance to well-known point sources. The spatial analysis results will also be discussed in the presentation. By comparing PFAS concentrations in archived tap water sample with recent tap water samples matched on the township, we found the fraction of quantifiable PFASs (i.e. known PFASs) has decreased in most towns and unknown extractable organic fluorine (EOF) has increased. Our analysis suggests tap water may be a significant exposure source for five PFASs among a group of U.S. women from diverse geographical areas. Increases in unquantified EOF in recent tap water suggest additional quantification would be worthwhile. Other exposure sources such as consumer products are suspected to dominant overall exposure of individuals in the NHS cohort prior to the restrictions and regulations of legacy PFASs in the U.S.

641
Consideration of the bioavailability of metals and metal compounds in freshwaters in regulatory frameworks
H. Ruedel, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Department of Environmental Monitoring; C. Díaz Muñiz, Cantabrian Basin Authority; H. Garelick, Middlesex University; N. Kandile, Ain Shams University / Department of Chemistry, Faculty of Women; B.W. Miller, US Environmental Protection Agency / National Enforcement Investigations Center; L. Pantoja, Middlesex University / Natural sciences; W. Peijnenburg, RIVM / Center for Safety of Substances and Products; D. Purchase, Middlesex University / Department of Natural Sciences, Faulty of Science and Technology; Y. Shevah, Consulting Engineers and Planners Ltd; P. Van Sprang, ARCHE; M.G. Vijver, CML Leiden University / Conservation Biology; J. Vink, DELTARES / Dept Soil and Groundwater systems
Recent research has demonstrated that both total and dissolved metal concentrations are not the most appropriate parameters for the risk assessment of metals in aquatic systems. There is an increasing awareness by leading regulatory bodies in the USA, Europe and other countries that the bioavailable fractions of metals could be better descriptors of their risks. The principal concept of metal bioavailability is the Biotic Ligand Model (BLM) which allows site-specific assessments of metals' risks by considering the environmental factors which determine the bioavailability of dissolved metals in the aquatic environment. For many metals bioavailability in freshwaters is modulated by dissolved organic carbon (DOC) concentrations, water hardness, the pH of the water and other factors such as temperature, concentrations of further ions and suspended solids as well as metal speciation. Metal-specific BLMs were proposed for different biological species and both, acute and chronic exposures. The BLM approach has been described extensively in the scientific literature, and BLMs have been applied for the risk assessment of metals and metal compounds (e.g., for copper and zinc in the EU). In the past, the broader use of the BLM approach for the site-specific evaluation of surface water monitoring data was hampered by the huge data requirements of the original BLMs (several site-specific water parameters). But the recent development of user-friendly BLM-based bioavailability tools (e.g., www.Bio-met.net, www.PNEC-pro.com) now allows the consideration of bioavailability for the evaluation of freshwater monitoring data of relevant metals. Such tools, which only need a basic set of easily available water parameters as input (mostly pH, Ca concentration, DOC, and dissolved metal concentration), are currently available for metals such as lead, nickel, copper and zinc. The new EU WFD environmental quality standards for lead and nickel according to Directive 2013/39/EU now consider the bioavailable fractions of these metals. In this contribution, the advantages and possible drawbacks of BLM-based bioavailability tools are presented highlighting feasibility, ranges of validity, and comparability between tools. Finally, recommendations for the regulatory implementation are given. This contribution is based on the outcome of the IUPAC-supported project "Consideration of bioavailability of metals/metal compounds in the aquatic environment" (#2011-060-1-600).

642

Age-Based and Time Trends of Exposure Chemical Biomarkers in the US Population 1999-2014

V. Nguyen, University of Michigan / Department of Computational Medicine and Bioinformatics; J. Colacino, University of Michigan / Department of Environmental Health Sciences; J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology; J. Kvasnicka, University of Michigan / Department of Environmental Health Sciences; O. Jolliet, University of Michigan

When interpreting biomonitoring data, we are limited by the cross-sectional nature of biomonitoring data and incomplete longitudinal data. It is important to differentiate between the potential influential of temporal determinants on legacy exposure, versus current exposure that may be due to relevant consumer product usage. In addition, an overarching systematic approach to studying exposure for a broad set of chemicals is missing. We developed a method to evaluate and compare age-specific exposure trends for 229 chemicals in the US population. Chemical biomarker measurements and demographic traits were obtained from the National Health and Nutrition Examination Survey (NHANES) datasets for the years 1999-2014 (n = 74,942). We extracted the persistency of chemical biomarkers from 16 different classes of chemicals from databases, literature review, and Quantitative Structure Activity Relationships (QSAR) when needed. To compare differences in chemical biomarker concentrations across the age groups, we partitioned the distribution of each chemical biomarker by 12 different age groups, which were defined based on age-specific behaviors. To evaluate the influence of age on the chemical exposure biomarker concentrations, we performed a series of chemical-specific linear regressions with adjustments for time, sex, ethnicity, poverty income ratio, and levels of cotinine. Effect estimates for age were converted to percent changes in the chemical biomarker. We have identified that chemicals with half-lives shorter than a year to either show no variations in ages or high concentration in younger individuals. In addition, for chemical with half-lives longer than a year, the age percent difference increases with higher persistence. We determined that chemicals indicative of legacy exposure have a human half-life longer than one year and a long time lapse between the ban/phase-out and sample collection, while chemicals reflective of current exposure are primarily due to the usage. We observed that different age group trends are explained by the product types in which the chemical is used and are not driven by belonging to a chemical family. The current study has defined criteria necessary to differentiate legacy exposure chemicals from currently exposed chemicals. Integrating generalized linear models with comparison of chemical distributions by the age groups led to identifying an age pattern of concern for children.

643

Biomarkers for the assessment of water quality in tropical estuarine environments in northeast Brazil

M. Jorge, Universidade Federal Maranhão - UFMA / Oceanografia e Limnologia; A. Bianchini, Universidade Federal do Rio Grande - FURG / Instituto de Ciências Biológicas

In attempt to define and measure the effects of pollutants in the aquatic ecosystem, biomarkers have attracted a great deal of interest. The principle of the biomarker approach is to analyze the organism's responses to pollutant exposure. Therefore, the aims of the present study were to verify the suitability of biochemical responses of estuarine fish (*Micropogonias furnieri*), copepod (*Acartia tonsa*) and crabs (*Callinectes spp*) as bioindicators to evaluate the environmental quality assessment in tropical estuaries of Maranhão State, Northeast Brazil. Thus, were evaluated biomarker of metal exposition as metallothionein-like proteins (MTLP), biomarker of organic exposition as 7-ethoxyresorufin-O-deethylase (EROD), and biomarkers of general effects like lipid peroxidation (LPO) and acetylcholinesterase (AChE). MTLP and LPO concentration were analyzed in copepod whole-body, crabs and fish hepatopancreas, while EROD activity were analyzed just in fish hepatopancreas and AChE inhibitionin in copepod whole-body, crabs hepatopancreasm and fish muscle and brain. Samples were collected in three different sites along two estuaries (São Marcos Bay and São José Bay), in two seasons (dry season/August and rainy season/January) in three different years (August/2012; January/2013; August/2013 and January/2014). In both estuaries, a high degree of heterogeneity were observed in biomarkers response over the two years of study, with significant spatial and temporal changes. However, analyzing all biomarkers studied, regardless organism and tissue, it is possible to observe at least two biomarkers alteration in both estuaries and season, reaching up to seven different biological responses in rainy seasons. The responses confirmed the initial expectation that both São José Bay and São Marcos Bay are subjected to the impact of the adjacent river basin drainage. In this context, biomarkers response were able to distinguish the types of contamination (organic and/or inorganic) which are affecting the estuaries along different seasons and year as well as the effects on local species. Regards test organisms, estuarine fish (*Micropogonias furnieri*) showed to be more sensitive to environment alterations, revealing most of the results obtained. Therefore, these results show that biomarkers is a promising tool for the assessment and monitoring of macro-tidal estuaries from tropical aquatic ecosystems impacted by anthropogenic activity.

644

Monitoring of priority substances in German freshwater fish of different age, size and trophic level

G. Radermacher, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Environmental Specimen Bank and Elemental Analysis; H. Ruedel, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Department of Environmental Monitoring; N. Lohmann, Eurofins GfA Lab Service GmbH; P. Lepom, A. Duffek, German Environment Agency

Beside monitoring of chemicals in the water phase, the European Water Framework Directive (WFD) also requires the quantification of certain priority substances (PS) in fish tissue. With the WFD daughter directive 2013/39/EU additional compounds were categorized as PS and for several of these environmental quality standards (EQSs) for biota have been introduced. This project was initiated to support the implementation of the WFD biota monitoring by comparative investigations and to propose an appropriate fish monitoring strategy which integrates all WFD monitoring requirements (e.g., compliance testing for human health- and secondary poisoning of wildlife-based EQS, comparability of monitoring data between sites and trend monitoring). To this end a dedicated sampling campaign was designed which covered six different freshwater sites. Fish were caught at sampling sites in the rivers Weser, Havel, Elbe and Moselle, in Lake Starnberg and a lagoon at the Baltic Sea. At each site three of the fish species were sampled that are listed in a German guidance document (RAKON IV.3): bream, chub, perch, roach and whitefish. During each sampling campaign it was tried to obtain twenty fish per species from two different age classes allowing multiple comparisons. Fish were dissected into fillets and carcasses, which were processed separately. Biometric data (e.g. fish size, weight, sex) were documented. Age was determined by examination of scales and trophic position by 15N/13C stable isotope measurements. Total mercury concentrations were determined for both individual fish fillets and carcasses as well as for age-grouped pools of both. Organic PS and fat contents were determined only in the fish fillet and carcass pools. Data are evaluated to derive recommendations for an optimal WFD biota monitoring and reporting. An important aspect is how biota burdens of PS are influenced by sample choice: Which fish species are most appropriate? Which age/size class is appropriate? Which tissue should be chosen? Fish of which trophic level should be caught? In this contribution, influences of these factors on fish levels of PS (e.g., mercury, PFOS, HCB, HBCDD) will be evaluated. It will also be assessed whether the data allow deriving factors for the conversion of whole body to fillet monitoring data and *vice versa*. Another aspect is to test whether the normalization of biota monitoring data (e.g., lipid normalization, trophic level normalization) enhances comparability.

645

Using Paleoecotoxicolgy to Assess the Toxicity of Lake Sediments Impacted by Legacy Gold Mining in Yellowknife, NT, Canada

C. Cheney, University of Ottawa / Biology; M.P. Pothier, J.R. Thienpont, University of Ottawa / Department of Biology; J.B. Korosi, York University / Department of Geography; L.E. Kimpe, University of Ottawa / Department of Biology; J.M. Blais, University of Ottawa / Biology

Natural resource extraction has supported the development of Canada's far north for many decades, but the legacy of these extraction processes remains apparent in northern Canadian landscapes today. From 1948-2004 Giant Mine operated 5 km north of the City of Yellowknife, and contributed to the economic growth of one of Canada's largest northern cities. Giant Mine roasted arsenopyrite ore at high temperatures to liberate gold, however a by-product of this roasting process was over 20000 tonnes of particulate arsenic trioxide, which was deposited to the surrounding landscape. The full extent of historic contamination in the area has yet to be characterized, although lakes within 17km of the roaster stack presently have elevated surface water arsenic concentrations. Here we use novel methods in paleoecotoxicology to characterize the full extent of contamination by analyzing ^{210}Pb dated lake sediment cores collected from 18 lakes within a 30km radius of the roaster stack. Metal profile concentrations of arsenic, antimony, and lead track a peak in concentration during the height of mining operations, which decreases with distance from the roaster stack. Principal Component Analysis (PCA) of sedimentary metals show clustering of chemically similar lakes prior to mining, with divergence during operations, and little change occurring since after the mine closure. Microbial bioreporters were applied to assess the bioavailability of arsenic in porewater. This novel application of a developing methodology indicates that arsenic in porewater samples is 70% bioavailable. Classic *Daphnia* toxicity tests indicate that lake surface water collected 1 km from the mine decreases *Daphnia* survivorship, despite the mine being inactive for over 10 years. Novel applications of *Daphnia* toxicity tests with historic sediment indicates that although *Daphnia* can survive in more recent sediments, mortality increases when *Daphnia* are exposed to sediments corresponding to the time of mine operation. These results suggest that lakes in the region continue to show lingering contamination from past mining activities. Sedimentary analysis indicates a much larger radius of contamination than previously documented at the site. Future work examining multi-trophic level responses to contaminant exposure in dated sediments will improve our understanding of the impact of mining operations to the aquatic environments near Yellowknife, and will help predict the timing of ecosystem recovery.

Ecotoxicology of micro and nanoplastics: Mechanistic

646
Wastewater-based microplastics: Presence in wastewater effluent and effects on freshwater organisms
S. Ziajahromi, Griffith University - Smart Water Research Centre / Griffith School of Environment; P.A. Neale, Griffith University / School of Environment; A. Kumar, CSIRO / Center for Environmental Contaminants Research; L. Rintoul, Queensland University of Technology; F.D. Leusch, Griffith University / Australian Rivers Institute
Microplastic pollution has become a serious environmental concern. Microplastics associated with wastewater treatment plant (WWTP) effluent have been found globally in marine and freshwater environments. Nevertheless, the concentration of microplastics discharged via wastewater after different treatment processes remains less understood. In this study, we identified microplastics in the wastewater effluent from three major WWTPs utilizing primary, secondary and tertiary treatment processes in Sydney, Australia. A novel validated sampler was designed for *in situ* collection of microplastics from wastewater effluent. The sampling method was combined with an efficient sample processing procedure to enhance the accurate detection of microplastics. The results indicated that primary effluent contained an average 1.5 microplastics/L. The amount of microplastics reduced to 0.6 microplastics/L after secondary treatment and 0.2 microplastics/L after tertiary treatment. Polyester fibres and polyethylene beads were predominantly detected in wastewater effluent, which possibly originate from synthetic clothing and cosmetic products, respectively. This suggests that WWTPs can act as a significant pathway to release microplastics to the aquatic environment, given the large volume of treated wastewater being discharged on daily basis. The effects associated with wastewater-based microplastics (e.g. fibres and beads) were thus investigated by exposing two freshwater organisms, a waterflea (*Ceriodaphnia dubia*) and a sediment-dwelling midge larvae (*Chironomus tepperi*), to microplastics in water and sediment, respectively, at concentrations within the range of environmentally realistic concentrations. A dose-dependenteffect was observed after acute and chronic exposure of *C. dubia* to microplastics, with fibres showing more significant effects, though chronic effects were only observed at concentrations six times higher than reported environmental levels. Further, exposure to an environmentally relevant concentration of microplastics adversely affected the survival, growth and emergence of *C. tepperi*. Size-dependent effects were observed with microplastics, with beads in the size range of 10-27 µm showing more pronounced effects. Our study demonstrates that microplastics are released into the environment by WWTPs and can have effects on freshwater organisms at concentrations within an order of magnitude of environmentally relevant levels.

647
What is in our plastic? In vitro toxicity of extracts from plastic products
L. Zimmermann, Goethe University Frankfurt am Main; C. Völker, Institute for Social-Ecological Research; M. Wagner, Norwegian University of Science and Technology / Department of Biology
The ubiquitous abundance of plastic litter in aquatic ecosystems causes concern over their potential ecological and health impacts. So far, most toxicity studies focus on physical effects of plastic particles and chemical effects of sorbed environmental pollutants. The effects of chemicals intrinsically present in plastics, i.e. plastic additives and side products, attract less attention in this context. Nevertheless, it is well established that these chemicals migrate from consumer products, thus representing a source of exposure to wildlife and humans. The aim of the current study is to investigate the *in vitro* toxicity of chemicals leaching from various plastic products and to characterize them using non-target chemical analysis. Different plastic types shall be ranked according to the toxicity of their leachates. Thirty-four plastic consumer products made of high/low-density polyethylene (HD/LDPE), polystyrene (PS), polypropylene (PP), polyethylene terephthalate (PET), polyvinyl chloride (PVC), polyurethane (PUR) or polylactic acid (PLA) were cut and extracted in a sonication-assisted, solvent-based procedure mimicking worst-case leaching. The extracts were tested for unspecific toxicity in the Microtox assay, for estrogenic and anti-androgenic activity in yeast-based reporter gene assays as well as for their potential to induce an oxidative stress response in the AREc32 assay. Two-thirds of the products leached chemicals triggering unspecific toxicity and one third of the samples induced an oxidative stress response. Nine plastic products released chemicals that were antiandrogenic and four slightly estrogenic. Overall, PVC and PUR extracts induced the highest toxicity in terms of potency and number of affected endpoints as almost all extracts triggered a high unspecific toxicity, oxidative stress and antiandrogenic activity. In contrast, PET extracts were less toxic with only one sample inducing oxidative stress. Interestingly, all PLA extracts produced a very high unspecific toxicity. Effects of HDPE, LDPE, PP and PS extracts strongly depended on the product. Our findings indicate that extracts of plastic products induce a range of toxicological endpoints, including unspecific toxicity, oxidative stress and endocrine activity. Some polymers, e.g. PVC, PUR, consistently trigger a higher and broader toxicological response. This demonstrates that a substantial part of plastic products are a potential source of exposure to toxic chemicals.

648
Microplastic size-dependent toxicity, oxidative stress induction, and multixenobiotic resistance (MXR) inhibition in the monogonont rotifer (Brachionus koreanus)
C. Jeong, J. Lee, Sungkyunkwan University
Plastic pollution in marine environment is a great concern in these days due to their world-wide distribution, persistence, and increasing amount of small-sized plastic particles due to degradation of larger plastic debris. However, little is known about their impacts on marine organisms, particularly at the molecular level. Here the dependence of microplastic toxicity to the monogonont rotifer (*Brachionus koreanus*) on particle size was investigated by studying the ingestion and egestion of fluorescently labeled 0.05, 0.5, and 6 µm polystyrene microbeads. Exposure to polystyrene microbeads led to significant size-dependent negative effects on growth rate, reproduction, and lifespan. In consistent, transmission electron microscope (TEM) analysis have revealed cellular damages in the rotifer *B. koreanus* exposed to 0.05 µm microbeads, indicating nano-sized microbeads would cause more serious impacts on aquatic organisms. To further explore the defense mechanism in response to different sizes of microbeads, the activities of several antioxidant-related enzymes and phosphorylation statuses of mitogen-activated protein kinases (MAPKs) were determined. Particularly, 0.05 µm microbeads have inhibited multixenobiotic resistance (MXR), resulting increase of sensitivity of rotifers to environmental pollutants. Our study provides a better understanding of molecular responses in the rotifer *B. koreanus* in response to microplastics and their potential impacts on the aquatic ecosystem.

649
Sorption of model pollutants on microplastics and toxicity assessment using early life stage of zebrafish (Danio rerio)
B. Cormier, EPOC University of Bordeaux; M. Larsson, Orebro University / Man-Technology-Environment research centre (MTM); L.W. Yeung, University of Örebro / Department of Chemistry; C. Clérendeau, EPOC University of Bordeaux; A. Karrman, Orebro University / MTM Research centre; B. Morin, University of Bordeaux / EPOC; M. Engwall, Orebro University / Man-Technology-Environment research centre (MTM); M. Bégout, X. Cousin, IFREMER / Laboratoire de Ressources Halieutiques de La Rochelle; J. Cachot, University of Bordeaux / EPOC; S. Keiter, Orebro University / MTM Research centre
The growing production of plastics increased the amount of plastic debris in aquatic ecosystems. Their degradation lead to the emission of microplastics (MPs) when their size is between 1-5000 µm. MPs can result from runoff and degradation (biodegradation or weathering breakdown) of macroplastics or from microplastics in cosmetics, industries and domestic sources. Virgin plastic polymers are, in principle, biologically inert and thus non-toxic. However, plastic production includes additives such as plasticizers, colorants or fire retardants that can be toxic. In addition to the potential toxicity caused by additives, MPs offer surfaces where hydrophobic pollutants can be adsorbed, including persistent organic pollutants (POPs). In marine environments, such chemicals are found at high concentrations in the surface layer, where low-density microplastics are most abundant. Indeed, these small particles can act as vectors and carriers for a wide range of pollutants and be ingested directly by organisms, causing chronic physical and/or toxicological effects. While ccumulation of MPs in aquatic ecosystems is a growing concern in society, the toxicity of MPs for wildlife and the processes of sorption of organic pollutants onto MPs are very complex and poorly understood. Therefore, objectives of the present study were to investigate the sorption kinetics of two models pollutants to LDPE (Low Density PolyEthylene) microparticles for 3 months, and the toxicity linked to pollutants sorbed on microplastics, using zebrafish embryos and larvae. Results of the sorption experiment showed that a longer exposure time did not affect the sorption rate of PFOS, but affects BaP sorption to the particles. The sorption of BaP was increasing during the first 7 days, while for the remaining 3 months of the experiment, the concentration of BaP absorbed by the MPs was constantly deacreased. On the toxicity aspects, no acute toxic effects were observed, using FET test with microplastics particles (virgin or spiked). No differences were found regarding sub-lethal endpoints (genotoxicity, photomotor response (PMR) and EROD activity) between the control group and fish exposed to virgin MPs, spiked MPs or compound alone. Results showed that MPs can be vectors of pollutants which sorbed at the surface over time. The toxicity of MPs as carriers of POPs cannot be demonstrated with 96h of exposure to LDPE microparticles. Experiments are currently being done to test the toxicity of MPs.

650
Comparative role of microalgae and microplastics in the effects of chlorpyrifos on molecular biomarkers in marine mussels
L. vidal-linan, Universidad de Vigo; B. Fernández, IEO; M. Albentosa, Instituto Español de Oceanografia / Marine Environment and Environmental Protection Area. Fisiology and Ecotoxicology of Bivalve Molluscs Department; J.A. Campillo, Instituto Español de Oceanografia / Centro Oceanográfico de Murcia; J. Bellas, IEO
Plastic particles within the microns range (microplastics, MP) are increasingly present in marine ecosystems. One of the most concerning aspects of MP in marine

habitats is that they might act as vectors of pollutants to marine organisms, since hydrophobic organic contaminants with low water solubility tend to concentrate on the surface of these particles. In this study we have compared the role of polyethylene MPs and microalgae (MA) of similar size, as vectors of the organophosphorus insecticide chlorpyrifos (CPF) to *Mytilus galloprovincialis* marine mussels. With that aim, CPF pre-exposed MP and MA were offered to the mussels and a battery of biochemical biomarkers of exposure and effects was measured. AChE activity in digestive gland and gills was significantly inhibited at all CPF treatments, disregarding exposure time. Levels of GST activity in the digestive gland in the three CPF treatments (CPF, MA+CPF and MP+CPF) after 7 days exposure were significantly higher than levels in treatments without CPF. However, after 21 days exposure, GST activity in the controls significantly increased, and differences with controls disappeared. For GST in gills, a significant increase in activity was observed in the MP, CPF and MA-CPF treatments after 7 days, compared to the MA control. When the nine biomarkers recorded are combined using the Integrated Biomarker Response (IBR) index a similar response in the three CPF treatments is initially observed (7 d), but after 21 d an enhanced response in observed in the MA+CPF and MP+CPF treatments only. In conclusion, AChE inhibition was similar in all CPF treatments disregarding the presence of particles. However, both MP and MA particles in CPF-exposed mussels produced in the long term an increase in biomarker response compared to waterborne exposure. Therefore MP seem to play a similar role than natural organic particles as vectors of organics to marine organisms.

651
Poster spotlight: TH001, TH002, TH003

Natural toxins and harmful algal blooms (HABs): water and food safety, analysis, toxicity, and risks (II)

652
Dissipation of the carcinogen ptaquiloside in water resources
L. Rasmussen, Metropolitan University College; J.S. Wu, F. Clauson-Kaas, University of Copenhagen; J. Sandersen, D. Lindqvist, Metropolitan University College; B.W. Strobel, University of Copenhagen / Plant and Environmental Sciences; H. Hansen, University of Copenhagen / Department of Plant and Environmental Sciences
Ptaquiloside (PTA) is a natural carcinogen found in a number of ferns Worldwide. The distribution and occurrence of PTA is best known from the Bracken ferns (genus *Pteridium*) which are classified in Group 2B *Possibly carcinogenic to humans* by WHO/IARC. The content of PTA in Bracken is highly variable (up to 5 w-%). PTA is readily leached from Bracken stands from where it can enter the soil, waterways or potentially contaminate groundwater. Several records of PTA contamination of surface water and upper groundwater exist from Denmark and Great Britain. The fate of PTA in surface and ground water has not been studied. Under sterile conditions, dissipation (= hydrolysis) of PTA in aqueous solution follows classical first order kinetics: $k_{obs} = k_{acid}[H^+] + k_{neutral} + k_{alkaline} \cdot k_{water}[H^+]$. The rate constants are: $k_{acid} = 25.7 \pm 1.0$ M^{-1} h^{-1}; $k_{neutral} = 9.5 \pm 6.0 \cdot 10^{-4}$ h^{-1} and $k_{alkaline}$ $4.8 \pm 0.0 \cdot 10^4$ M^{-1} h^{-1}. The activation energy for PTA hydrolysis at pH 4.6 is approx. 75 kJ mol^{-1}. Hence, hydrolysis is a function of both pH and temperature. The purpose of this investigation was to study the degradation of PTA under near-natural conditions using 10 different surface and groundwaters from Denmark and to compare the degradation kinetics with the existing model for hydrolysis. Degradation was fast in natural non-sterilised lake waters with half-lives from 5 to 25 h. All PTA were degraded within 200 h. Sterile controls had no degradation. Winter samples exhibited slower degradation (half-lives up to 100 h). Sterile samples followed the existing model for hydrolysis closely, i.e. no degradation at neutral pH. PTA persist considerably longer in groundwater. Half-lives in groundwater ranged from 7 to approx. 50 days with fastest degradation in alkaline waters. The existing model for hydrolysis could generally predict the rate of degradation in groundwater. However, the model did not perform well at low or high pH. Pterosin B was formed as the end product by hydrolysis in the tested groundwaters at a molar ratio of approx. 1:1 for PTA:Pterosin B. However, under weak acid condtions, only approx. 10% of the potential pterosin B were formed. The risk of leaching from Bracken stands and contamination of surface waters is highest during the winter season. PTA has the potential of contaminating mainly shallow aquifers as the compound can persist for up to several months in contact with groundwater. The risk is expected to be highest at near-neutral conditions in aquifers.

653
On-line detection of algal toxins in sea water
S.F. Bodini, SYSTEA; F. Pasquazzi, Systea SpA; A. Porchetta, L. Micheli, G. Volpe, L. Fabiani, University of Tor Vergata; L. Sanfilippo, P. Moscetta, Systea SpA; G. Palleschi, University of Tor Vergata
Natural toxins produced by plants, algae and microorganisms represent a serious concern for public health. Current detection methods need expensive equipment, trained personnel and complicate protocols. In particular, the determination of marine biotoxins, released as a consequence of toxic algal blooms, is performed offline on fish / shellfish homogenates rather than on-line, resulting not appropriate for monitoring programs that require real-time warnings like in specific situations relevant for public health, such as bathing sites and acquaculture plants. Thus, there is a special need for on-line, continuous, rapid and sensitive field tests. To fill this gap, it was developed and tested a direct Enzyme–Linked Immuno–Magnetic Colorimetry microplate assay for the detection of Domoic Acid, Saxitoxin and Okadaic Acid in seawater. The assay is based on the fact that, in the presence of the target toxin, competition occurs and consequently the color production decreases proportionally to the toxin concentration. This analytical approach allows to combine antibody selectivity, convenience of a separation step through the use of magnetic beads and simplicity of colorimetry. Next, the manual assay was integrated within a fully automated PC-controlled on-line analyzer based on the micro Loop Flow Reactor technology able to host three immunosensor sub-modules. Specific volumes of reagents were injected to a flow cell, equipped with a heater, a magnet and an optical group. Because most of the reagents needed to be kept at 4 °C, a Peltier refrigerated compartment was designed and incorporated in the instrument. Laboratory measurements were executed to validate the prototype efficiency to detect sub-ppb concentrations of the algal toxins. The obtained calibration lines were consistent with the strict requirements limiting the presence of the toxins in environmental waters. On-line suitability was demonstrated by a field installation on a floating platform in the port of La Spezia, Italy, for daily monitoring of real marine water samples, in which the instrument was integrated with a communication module for real-time data transfer to a control center. In conclusion, results obtained showed that the automated measurements are repeatable and sensitive. Further work must go into developing additional specific antibodies to extend the application on other natural pollutants released by plants, algae and microorganisms, with a particular eye on freshwater cyanotoxins.

654
A decade of chemical studies on Ostreopsis. What's left?
C. Dell'Aversano, University of Napoli Federico II, Department of Pharmacy / Pharmacy; L. Tartaglione, University of Napoli Federico II / Department of Pharmacy; M. Forino, University of Napoli Federico II Department of Pharmacy / Department of Pharmacy; A. Penna, S. Casabianca, S. Capellacci, University of Urbino / Department of Biomolecular Sciences; C. Totti, S. Accoroni, Università Politecnica delle Marche / Department of Life Science and Environment; R. Pistocchi, F. Guerrini, L. Pezzolesi, Alma Mater Studiorum University of Bologna / Department of Biological, Geological and Environmental Sciences; G. Honsell, University of Udine / Department of Agricultural and Environmental Sciences; M. Pelin, S. Sosa, University of Trieste; A. Tubaro, University of Trieste / Department of Life Sciences
Over the last decade massive blooms of the benthic dinoflagellate *Ostreopsis* cf. *ovata*, once confined to tropical and subtropical areas, have occurred in more temperate regions worldwide including the Mediterranean Sea. Concurrently, negative impacts on human health mainly due to inhalation of toxic aerosols and/or skin contact were observed together with death of marine invertebrates. When first toxic outbreaks related to *O.* cf. *ovata* occurred in 2005 along the Ligurian coasts (Italy), little was known on several aspects of the phenomenon. Although some *Ostreopsis* spp. were known to produce congeners of palytoxin (PLTX), *O.* cf. *ovata* was not known as a toxic species and its metabolic profile had never been investigated. Secondly, although PLTX itself was reported as one of the most potent non-protein marine toxins known and tentatively suggested as the causative agent of fatal food poisonings in the tropics, it had never been suspected to exert toxicity through inhalation. Last but not least, the role of the environmental conditions on *O.* cf. *ovata* proliferation and toxin production had been poorly studied. Therefore, the need for increase knowledge on potential risks for humans and ecosystem stimulated research in the field. An Italian interdisciplinary network was created, including scientists from the Academia and operators of the regional environmental protection agencies and food safety laboratories. This joint effort led to clarify many of the aspects related to the *Ostreopsis* phenomenon that still represents one of the major threats to humans in the Mediterranean area. This presentation is meant to summarize the results of our studies on *O.* cf. *ovata*, highlighting inter- and intra-specific variability of the toxin profiles, structural variability of the detected toxins and, in some cases, linking such differences to the risk that PLTX congeners pose to humans following inhalatory, dermal and oral exposure. Phylogenetic relationships among many isolates of Mediterranean *O.* cf. *ovata* were investigated as well as innovative molecular qPCR based assay was developed for monitoring activities. A general overview of the environmental conditions that favour *O.* cf. *ovata* proliferation and toxin production will be also provided based on laboratory data and in field observations. The methodological approach, besides addressing many of the palytoxin-related issues, may serve as template for facing in due time any emerging toxin-related threat to humans.

655
Untangling the geosmin appearance in a Mediterranean river: relationship of geosmin concentration and physicochemical parameters over a year
C. Espinosa, L. Llenas, Universitat de Vic Universitat Central de Catalunya / BETA Technological Centre; M. Ordeix, Universitat de Vic Universitat Central

de Catalunya / Centre de Estudis dels Rius Mediterranis; M. Abril, Universitat de Vic Universitat Central de Catalunya / BETA Technological Centre; M. Ricart, Universitat de Vic Universitat Central de Catalunya / BETA Technological; N. Sellarès, Universitat de Vic Universitat Central de Catalunya / Centre dEstudis dels Rius Mediterranis; J. Colon, S. Ponsá, Universitat de Vic Universitat Central de Catalunya / BETA Technological Centre
The Mediterranean region is one of the most densely populated and industrially developed area in Spain. As a consequence, most of the rivers in this region are impacted by multiple anthropogenic stressors. One of these rivers is the Ter River (NE Spain), where human pressures are diverse and have increased in the last decades because farming, urban development and industry practices depends on its water. In addition, fluctuations in water discharge due to the Mediterranean climate create a high variability of conditions along the Ter River. One of the major problems detected in Ter River in the recent years is the appearance of geosmin. This is a metabolite generated mainly by cyanobacteria and actinomycetes that, when die, is released into the water, giving it a bad smell and taste. Although some studies have described that the production of this metabolite depends on environmental conditions, the factors associated with its production are still not clear. This supposes an economic problem for the water supply companies, since they cannot predict its appearance and have to act when customers complains arrive. The aim of this study is to evaluate the co-relation between physicochemical parameters and geosmin appearance along the Ter River during one year, and to study seasonal variability of geosmin concentration. The study has been performed in four sampling sites across the upper-middle part of the Ter basin. The sampling frequency varied throughout the year. During the potential geosmin formation period (February to June), sampling was performed weekly while from June to December, sampling was performed monthly. The parameters analysed have been nutrient concentration, suspended solids, organic material, turbidity and geosmin concentration in water. Biofilm samples were taken in order to analyse the chlorophyll *a* content. The results obtained clearly reflected the seasonal variation in the appearance of geosmin, being its concentration higher in winter (32 ng/L). They also evidenced the N/P ratio as one of the key factors involved in the geosmin formation. However, a more in-depth analysis of the N/P ratio in water is still necessary in order to explain the mechanisms that generate the geosmin formation within the organism. For this reason, a mesocosms experiment that tests the influence of the N/P ratio on the geosmin formation within the biofilm could be the next step to follow.

656

Italian guidelines to assess and manage the risk associated to cyanobacteria blooms in water during bathing and recreational activities

M. Manganelli, Istituto Superiore di Sanità / Dip. di Ambiente e Salute; E. Funari, F. Buratti, Istituto Superiore di Sanità / Dept. of Environment and Health; E. Testai, Istituto Superiore di Sanità / Dip. di Ambiente e Salute
Many species of cyanobacteria thrive in different aquatic environments, where they can produce cyanotoxins with different toxicological profile. The still growing anthropic pressure and climate changes are causing the expansion in terms of time and space of their blooms, increasing the concerns for human health in several exposure scenarios. The Italian guidelines for the management of cyanobacterial blooms in bathing water, firstly drowed up for the implementation of European bathing water directive (Directive 2006/7/EC), have been recently updated. A risk-based approach has been developed after a thorough revision of the current scientific knowledge on cyanobacteria distribution in the Italian Lakes and on chemical, toxicological and epidemiological aspects of different cyanotoxins. The possible exposure scenarios have been considered: oral, dermal and inhalation exposure to cyanotoxins, during recreational activities, have been individually examined, to develop a framework of thresholds and actions aimed at preventing harmful effects for bathers. Three phases of attention relative to monitoring plans have been consequently defined: routine, alert and emergency, suggesting the actions to take at any moment. Parallel to environmental monitoring, a multi-step health-surveillance system has been proposed, aimed at collecting important epidemiological information and at limiting unnecessary accesses to the hospital through a screening action by local workers (lifeguards, local health units, pharmacists, etc.). All the technical/practical information on strategies, methods and protocols to carry out all the foreseen activities, from the sampling of different matrixes, to the analysis and the reporting to health authorities are ptovided. IN summary, guidelines, also by comparing international guidance values and/or guidelines, provide criteria to plan environmental monitoring activities, health surveillance and public communication systems. Finally the still important scientific gaps and research needs are highlighted.

657

Identification and prioritization of emerging risks for food safety: climate change as a driver

A. Maggiore, European Food Safety Authority (EFSA) / Risk Assessment and Scientific Assistance Department; A. Afonso, EFSA European Food Safety Authority / Risk Assessment and Scientific Assistance Department; A. Rortais, European Food Safety Authority; G. De Sanctis, D. Verloo, C. Gardi, S. Dhollander, Y. Van der Stede, F. Boelaert, M. Binaglia, J. Tarazona, EFSA European Food Safety Authority
According to the General Food Law, the European Food Safety Authority (EFSA) is required to identify emerging risks in the fields within its mission. EFSA has developed a methodological framework for identification of emerging risk, starting from a preliminary identification of priority emerging issues through knowledge networking activities. The long term anticipation of emerging risks includes the identification of drivers. Drivers are the underlying natural or human-induced factors that directly or indirectly cause emerging risks. Climate change is recognised as a critical driver and its impact on the occurrence and toxicity of toxin producing phytoplankton, bacteria and pathogenic viruses and on other food safety domains was demonstrated. With the aim of further exploring tools to identify and prioritise emerging risks, EFSA initiated a project focusing on climate change as a driver of emerging risks for food and feed safety, including plant and animal health. A knowledge discussion group involving the major institutions involved with climate change has been created. The group will define criteria to identify relevant subdrivers (eg. rising and more fluctuating temperatures, changing precipitation patterns, increase in natural disasters etc), the issues relevant to different food safety domains including plant health and animal health, and to develop a harmonised and transparent scoring system applicable to the identified emerging issues in order to prioritise future research and risk assessment activities.

Advances in evaluating and regulating endocrine disruptors

658

Hazard identification of endocrine disrupting properties of pesticides on non-target organisms: state of the art and future perspectives

s. barmaz, EFSA - European Food Safety Authority / Pesticides Unit; M. Arena, EFSA - European Food Safety Authority / Pesticides; D. Auteri, EFSA - European Food Safety Authority / Pesticides Unit
According to the Regulation (EU) no 283/2013, setting out the data requirements for active substances, in accordance with Regulation (EC) No 1107/2009 concerning the placing of plant protection products on the market, the endocrine disrupting properties of pesticides should always be assessed, as substance identified as an endocrine disruptor should not be approved. Most of the current knowledge about endocrine disruption is related to EATS (Estrogen, Androgen, Thyroid and Steroidogenesis) modalities in vertebrate organisms i.e. there is a good mechanistic understanding on how those modalities can lead to an adverse effects. The OECD conceptual framework and the OECD 150 provide an overview of the existing assays for the endocrine disruptors (ED) hazard identification and guidance on how to interpret the results of those tests. A suitable testing strategy which allows the identification of ED properties of pesticides through EATS modalities is available for some taxa of non-target organisms (i.e. fish and amphibians). The analysis confirmed that the available test methods and knowledge on birds' endocrinology do not allow a full ED assessment although they can provide supportive information. In the case of reptiles, appropriate standard test methods are completely missing. In some circumstances, extrapolation between taxa could be scientifically supported. However, consideration should be given to taxon-specific differences. Extrapolation between mammals and amphibians regarding the ED hazard identification through the thyroidal modality has been investigated in the past. A similar analyses has not been done for EAS modalities, however, in some cases extrapolation among oviparous vertebrates is scientifically justified e.g. in the case of the steroidogenesis pathway leading to reproductive dysfunction (more specifically egg production). The main scope of this work is to present a critical overview of the available standardised test methodology for the ED assessment of non-target organisms, including consideration on the extrapolation between taxa. In addition, possible future perspective and research needs are discussed.

659

Contaminants of emerging concern in the North American Great Lakes: Evidence of reproductive disruption from field and laboratory studies

H.L. Schoenfuss, St. Cloud State University / Aquatic Toxicology Laboratory; L. Thomas, St. Cloud State University / Biological Sciences; L. Wang, St. Cloud State University; N. Cipoletti, St. Cloud State University / Aquatic Toxicity Laboratory; Z. Jorgenson, St Cloud State University / Environmental Contaminants; S. Elliott, U.S. Geological Survey / Minnesota Water Science Center; M.E. Brigham, U.S. Geological Survey
Analysis of nearly 500 surface water samples collected as part of the Great Lakes Restoration Initiative at 54 sites in tributaries of the North American Great Lakes Watershed confirmed the ubiquitous presence of Contaminants of Emerging Concerns (CECs) in anthropogenically impacted aquatic environments. Cluster analyses of commonly detected CECs in this data matrix suggests that the co-occurrence of approximately half of the CECs can be attributed to dichotomous urban or agricultural upstream land use. Mixtures found in watersheds with urban influences commonly contained steroidal estrogens, BPA, alkyphenols, pharmaceuticals and personal care products. Agriculturally influenced sampling sites contained herbicides and pesticides in addition to BPA and alkylphenols, but mostly lacked pharmaceuticals and personal care products. Almost 3,000 resident and caged sunfish (*Lepomis ssp*.) were collected from 27 of the 54 sampling sites and analyzed for indicators of stress associated with CEC exposure. In the presence of high aqueous CEC concentrations, glucose concentrations spiked in sunfish

plasma and liver cells exhibited toxic stress response. Canonical correspondence analyses revealed that concurrent with indicators of toxic stress, biomarkers of reproductive potential declined. To further examine the population level consequences, fathead minnows (*Pimephales promelas*) were exposed in the laboratory for three generations to the empirically derived urban CEC mixture at three environmentally relevant concentrations. Mixtures at environmentally measured concentrations enhanced fecundity in the F2 and 3 generations, while higher mixture concentrations resulted in declining fecundity. Taken together, this integrated series of studies indicates that CECs in Great Lakes tributaries may impact fish population health and sustainability.

660

AOP-informed assessment of Endocrine Disruption in freshwater crustaceans

K. Tollefsen, NIVA / Ecotoxicology and Risk Assessment; M. Cronin, Liverpool John Moores University; . Evensen, Norwegian School of Veterinary Science; L. Evenseth, The Arctic University of Norway; F. Falciani, University of Liverpool; T. Iguchi, YOkohama City University / Molecular Environmental Endocrinology; C. LaLone, U.S. EPA / Mid Continent Ecology Division; Y. Li, NIVA Norwegian Institute for Water Research; C. Mellor, Liverpool John Moores University / School of Pharmacy and Biomolecular Sciences; E. Perkins, US Army Corps of Engineers ERDC; T. Rundberget, Norwegian Institute for Water Research; B. Salbu, Norwegian University of Life Sciences; I. Sylte, The Arctic University of Norway / Department of Medical Biology; D.L. Villeneuve, U.S. EPA / National Health and Environmental Effects Research Laboratory; N. Vinas, Mississippi State University / Engineer Research and Development Center; Y. Song, NIVA Norwegian Institute for Water Research / Ecotoxicology and Risk Management

A number of exogenous compounds have the potential to interfere with the endocrine system of animals and may perturb vital endocrine processes to a degree causing an adverse effect (outcome) on ecologically relevant endpoints such as growth, development and reproduction. These endocrine disrupting (ED) effects have been well characterised in aquatic vertebrates and mammals due to a well-defined endocrine system and substantial research effort in the last decades, but knowledge on ED effects in a larger range of species are still poorly characterized. Lack of knowledge on ED effects in invertebrates is currently a major limiting factor to properly perform risk assessment of endocrine disrupting chemicals (EDCs) across taxa. The present project have focussed on developing Adverse Outcome Pathways (AOPs) for EDs in aquatic crustaceans, and applying these to assess the hazard and risk of ecologically relevant complex mixtures of pollutants. Although several ED mechanisms have been proposed to be of relevance for crustaceans, perturbations of endocrine processes related to chemical interactions with the ecdysone receptor (EcR) and the Juvenile Hormone (methyl farnesoid) receptor (MfR) have been identified to be of particular concern. The present paper focus on the application of AOPs to 1) develop linkage between endocrine mechanisms and adverse outcomes, 2) identify knowledge gaps and inform testing strategies, 3) identify sensitive species/taxa, 4) identify likely define toxicity endpoints suitable for Integrated Approaches for Testing and Assessment, IATA, 5) identify potential EDCs and 6) practical implementation of AOP information into cumulative hazard and risk assessment of ecologically relevant exposure scenarios. *Acknowledgement* - Funding from RCN-221455 â_Adverse Outcome Pathways for Endocrine Disruption in Daphnia magna, a conceptual approach for mechanistically-based Risk assessment (www.niva.no/edrisk)", RCN-268294 "Cumulative hazard and risk assessment of complex mixtures and multiple stressors (www.niva.no/mixrisk)" and EU-FP7 project SOLUTIONS (http://www.solutions-project.eu/project/).

661

Assessing impacts of place-based mixtures of emerging contaminants on endocrine activity and adverse outcome pathways: comparisons of different life stages

R. Klaper, University of Wisconsin-Milwaukee / School of Freshwater Sciences; J. Crago, Texas Tech University/TIEHH / Environmental Toxicology The Institute of Environmental and Human Health TIEHH

Emerging contaminants often appear as mixtures of differing concentrations across a landscape. Traditional toxicological assays as well as receptor binding assays that examine the impacts of individual chemicals to endocrine disruption pathways do not adequately detect all EDC compounds and do not describe the collective impact of mixtures as there can be cross-talk among molecular pathways. Using place-based mixture concentrations of emerging contaminants in combination with multiple molecular initiating events from adverse outcome pathways can help to identify potential hotspots of potential environmental impact that cross multiple mechanisms of action. This talk will discuss the use of transcriptomics to modify the OECD fish embryo acute test (FET) and chronic exposures to juevenile and adults fish are being used to examine EDC pathway related disruption. Examples discussed will include several experiments using exposure mixtures representing those measured in several locations in Lake Michigan.

662

Interference of hepatotoxicity with endocrine activity in zebrafish (Danio rerio)

L.A. Baumann, University of Heidelberg / Aquatic Ecology and Toxicology; H. Holbech, University of Southern Denmark / Biology; L. Weltje, BASF SE / Crop Protection Ecotoxicology; H. Schmidt-Posthaus, University of Bern / Institute of Animal Pathology; H. Segner, University of Bern / Centre for Fish and Wildlife Health; T. Braunbeck, University of Heidelberg / Centre for Organismal Studies

Vitellogenin (VTG), a well-established biomarker for the diagnosis of endocrine activity in fish, is used in the OECD test guidelines 229, 230 and 234. A reduction of VTG production (mainly in females) is usually associated with androgenic or anti-estrogenic activity, whereas an increase of VTG (mainly in males) is associated with estrogenic activity. However, the synthesis of VTG may not only be modified by typical endocrine-related pathways, but also through non-endocrine-mediated processes. In particular, hepatotoxicity, i.e. toxicant-induced impairment of liver structure and function, can influence VTG as a biomarker, since it is synthesized in the liver. Changes in VTG caused by non-endocrine hepatotoxicity in a screening assay would unnecessarily trigger very labor-, animal- and cost- intensive higher tier testing (e.g. a fish life cycle test). Therefore, an intimate understanding of the interplay between primary endocrine-related and non-endocrine-related pathways influencing VTG production is crucial for the avoidance of false diagnoses. The present study is driven by the hypothesis that hepatotoxicity may interfere with VTG synthesis in the liver of exposed fish. Thus, we investigated the effects of two well-known hepatotoxicants, acetaminophen (APAP) and isoniazid (INZ), on zebrafish (*Danio rerio*) in a 21-day flow-through exposure test according to OECD guideline 230. Various hepatotoxicity- and endocrine system-related endpoints were recorded: - mRNA expression of different endocrine-related (vtg1, vtg3 and esr1) and hepatotoxicity-related marker genes (fabp10a, apoa1, cyp2k19 and cyp3a65) in the liver; - protein levels of VTG in head/tail homogenates; - hyaluronic acid (a biomarker for liver toxicity) levels in head/tail homogenates; - liver histology and ultrastructure; Both APAP and INZ had different effects on exposed fish. While APAP did not cause any histopathological alterations in the liver, INZ significantly induced hepatocyte degeneration. VTG levels in APAP-exposed females were elevated, while no effect was observed in INZ-exposed fish. Likewise, transcriptional responses in the liver differed between both compounds and indicate that both did interact with different endocrine- and hepatotoxicity-related pathways. The results from both studies will further be evaluated with respect to their potential for the development of an adverse outcome pathway (AOP) for interference of hepatotoxicity with the VTG response in fish.

663

Single Pulse Exposure of Different Life Stages of Zebrafish to the Selective Estrogen Receptor Modulator Tamoxifen Citrate

S. Kroesen, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology; C. Schaefers, Fraunhofer-Institut / Ecotoxicology; E. Bruns, Bayer AG, Division Bayer CropScience / Ecotoxicology; E. Eilebrecht, M. Teigeler, Fraunhofer IME / Ecotoxicology

The discussion about the regulation of endocrine disruptors (ED) is on-going between groups of scientists, authorities and stake-holders. Especially dose-dependency, low dose effects and effect thresholds are still under debate. To address parts of these questions, a static life-cycle test with zebrafish (*D. rerio*) has been performed to examine if a pulse exposure of an ED might lead to distinguishable effects and the establishment of a dose-response relationship is possible. The results willl be used for a comparison with available data originating from a flow-through study with TC in zebrafish. A water-sediment system has been set up to expose different life-stages (group A: 40 eggs, group B: 20 juveniles, group C: 16 adults) to a known ED, Tamoxifen citrate (TC). Observed endpoints included early-life stage survival, juvenile growth, reproduction, as well as adult growth, sex ratio, vitellogenin levels and F_1-generation early life-stage. Four concentrations of TC were applied as a pulse in three replicates each, ranging from 125 µg/L to 1000 µg/L (spacing factor 2). Four controls replicates were included. Mortalities occurred in all developmental stages (groups A to C), especially in high concentrations (500 µg/L, 1000 µg/L). In sexually mature fish (group C) mortality was higher in males. A decline in fertility could be observed for group C, possibly related to the higher male mortality. Total egg numbers appeared unaffected. The results were mirrored for fish introduced as juveniles (group B): While fertility rates were not influenced negatively, fecundity was lower in remaining concentrations (125 µg/L, 250 µg/L). Changes in egg morphology were noticed shortly after exposure (group C). Consequently, F_1-fish originating from group C showed a dose-dependent decrease in survival rates and growth. Although reproduction data are difficult to be attributed to endocrine activity, an influence on the endocrine system of the test animals seems apparent. Particularly sex specific effects in F_0-animals as well as an impaired early life-stage in F_1-animals are of highest interest. Further data on vitellogenin and reproduction will help clarifying pending questions. Additionally, several other accessible datasets from zebrafish studies featuring paired pulsed and flow-through exposures of EDs with diverse dissipation times will be integrated in the concluding assessment. The final objective is to deduce possible effect thresholds based on internal concentrations.

BiER is good for you: How biotransformation and elimination rate information can improve chemical assessments

664

A Tiered Approach for Screening Chemicals for Biomagnification Potential in Humans

A. Sangion, University of Insubria / Department of Theoretical and Applied Sciences (DiSTA); J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology; P. Gramatica, E. Papa, University of Insubria / Department of Theoretical and Applied Sciences (DiSTA)

Bioaccumulation is a process in which the chemical concentration in an organism exceeds the concentration in the respiratory medium, the diet or both and is an integral aspect of hazard and risk assessment. Strong correlations between partitioning properties such as the octanol-water partition coefficient (Kow) and the octanol-air partition coefficient (Koa) and bioaccumulation metrics in water-respiring and air-breathing organisms have led to Kow and Koa being the primary screening criteria for bioaccumulation assessment. However, primary biotransformation rate constants (k_B) and half-lives (HL_B) are also critical determinants of bioaccumulation. Here we present a tiered approach for screening the bioaccumulation potential of organic chemicals in air-breathing organisms. The tiered approach progresses from screening-level conservative assumptions based on Kow and Koa only to more realistic assumptions for, internal distribution, chemical properties and biotransformation (Tiers from 1 to 4 respectively). Biomagnification Factor (BMF) derived from a typical human diet as calculated by the Risk Assessment IDentification And Ranking (RAIDAR) mode is the metric for assessing bioaccumulation potential in air-breathing organisms of approximately 13,000 chemicals including industrial chemicals, pharmaceuticals, personal care products and chemicals used in consumer goods. Tiers that do not consider biotransformation (1, 2 and 3) estimate a high percentage of chemicals with BMF greater than 1 (i.e. about 93%, 95% and 93%). In particular, in Tier 2 and in Tier 3, the introduction of the biotic partition coefficient kStorLipW, kMembLipW and kProtW and about the ionic state at pH 7.4 reduce the BMF estimate for some chemicals, but in general the effects are limited. In Tier 4 the introduction of the HL_B has a high impact on the screening results, strongly reducing the BMF estimate to < 1 for most of the compounds (i.e. about 90%). This shows how models based only on partition coefficients are not sufficient to describe and address the bioaccumulation and biomagnification processes, and can lead to overly conservative estimates ("false positives"). Moreover the study highlights the key role of biotransformation in bioaccumulation assessment for air-breathing organisms and highlights the need for reliable data on biotransformation to effectively categorize chemicals for hazard.

665

Critical Evaluation of a Human In Vitro Biotransformation Rate Database: Case Study of Seven Chemicals

K.L. Foster, ARC Arnot Research and Consulting Inc. / Adjunct Professor, Trent University, Applications of Modelling & Quantitative Methods (AMOD); A. Looky, ARC Arnot Research and Consulting Inc.; J.M. Armitage, University of Toronto - Scarborough / Physical and Environmental Sciences; M. Embry, ILSI; J.W. Nichols, U.S. EPA / ORD NHEERL Mid Continent Ecology Division; B. Wetmore, U.S. EPA / National Exposure Research Laboratory; J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology

Despite the fundamental value of biotransformation rate information, relatively few measured *in vivo* data are available for humans compared to the thousands of chemicals requiring evaluation. Reliable models, laboratory measured *in vitro* biotransformation rate data, and *in vitro-in vivo* extrapolation (IVIVE) methods can be applied to address *in vivo* biotransformation rate data gaps and, coupled with data confidence assessment methods, uncertainty and data utility. We have developed a new database of >11,000 human *in vitro* biotransformation rate estimates (half-lives, clearance rates and rate constants) derived from liver microsomal, S9 homogenate, and hepatocyte-based assays for >8,500 organic chemicals from the literature and publicly available databases (i.e., ChEMBL). The database is comprised primarily of pharmaceuticals and pharmaceutical candidates from various experimental sources. The organic chemicals in the database represent a broad range of physical-chemical properties (Log K_{OW}=-4 to 13, Log K_{OA}=0.01 to 47, Log K_{AW}=-42 to 3) and the *in vivo* intrinsic clearance values ($CL_{Int,In\ Vivo}$ $mL.h^{-1}.kg^{-1}$) span about 8 orders of magnitude. We developed and applied novel data quality assessment methods based on proposed standardized testing guidance to address variability and uncertainty in the database. The data quality assessment methods included compiling physical-chemical property data (e.g., K_{OW}, pKa, water solubility) for all of the chemicals and applying a mass balance *in vitro* model. The ensuing data quality scores (e.g., high or low confidence) may help identify datasets that are most appropriate for QSAR development and for other potential applications (e.g., bioaccumulation screening, prioritization). The score results are further examined in a case study of seven chemicals and the utility of high and low confidence biotransformation rate data, its merits and limitations for various use contexts, are discussed and overall key findings of the critical review of existing human *in vitro* biotransformation rate data are summarized.

666

Sediment-associated cyclic volatile methylsiloxanes: Biotransformation in a freshwater oligochaete and an estuarine polychaete

H. Selck, Roskilde University / Dept Science and Environment; R. Windfeld, Roskilde University / ENSPAC

Chemical regulatory legislation of organic contaminants is generally based on an assessment of the chemical potential to persist (P) in the environment, bioaccumulate (B) in biota, and possess potential toxicity. Applying standardized exposure setups (i.e., water-only exposure) as historically has been employed in environmental risk assessment, may underestimate bioaccumulation of hydrophobic organic contaminants (HOCs) in sediment-dwelling organisms because: 1) HOCs often accumulate in sediments to concentrations greatly exceeding the concentration in the overlying water; and 2) a number of papers illustrate that sediment-associated HOCs are available for uptake in benthic organisms. Alternatively, benthic invertebrates may be able to metabolize organic contaminants (i.e., biotransform), thus reducing their body burden. However, available information on the biotransformation capacity of benthic organisms is very limited. We conducted a number of experiments examining uptake and biotransformation of sediment-associated cyclic volatile methylsiloxanes (i.e., D4 and D5) in two deposit-feeding worms, namely, the estuarine polychaete, *Capitella teleta* and the freshwater oligochaete, *Tubifex tubifex*. This presentation will provide examples of how biotransformation capacity varies among the two benthic deposit feeders, and how biotransformation may reduce body burden and facilitate the removal of sediment-associated siloxanes. Including these factors in a hazard or risk assessment are likely to impact PBT assignment and categorization, and exclusion of benthic organism behaviour may add compound uncertainty to predictions of bioaccumulation and trophic transfer.

667

Toxicokinetics and biotransformation products of diuron and 3,4-DCA in the developing zebrafish embryo (Danio rerio)

E. Jarosz, UFZ Leipzig / Bioanalytical Ecotoxicology; M. Krauss, Helmholtz centre for environmental research - UFZ / Effect-Directed Analysis; S. Aulhorn, Helmholtz Centre for Environmental Research UFZ / Department Bioanalytical Ecotoxicology; E. Küster, Helmholtz Centre for Environmental Research, Dept.Bioanalytical Ecotoxicology / Department Bioanalytical Ecotoxicology; T. Luckenbach, Helmholtz Centre for Environmental Research UFZ / Department Bioanalytical Ecotoxicology

Diuron is a commonly used phenylamide herbicide which acts by interrupting the photosynthetic electron transport chain. Like other phenylamide herbicides diuron is bio-transformed to 3,4-dichloroaniline (3,4-DCA) which occurs in plants, the liver of vertebrates and in soil. Fish embryos do not possess the same metabolic potential as adult fish. It was not determined so far if different embryo stages differ regarding toxicokinetics of diuron and 3,4-DCA and whether toxicities come from parent compounds or metabolites. We addressed the following questions: What are rates of uptake and elimination of diuron and 3,4-DCA in different zebrafish embryo stages? Is diuron biotransformed by the embryo *via* which metabolic pathway? Does the embryo`s chorion form a barrier for diuron and 3,4-DCA mitigating the compounds` toxicities? Information on these aspects is valuable for the understanding of the toxicity of phenylamide herbicides to fish embryos. For determining tissue concentrations after different times of exposure, freshly fertilized zebrafish eggs were exposed to the $EC20_{(48h)}$, i.e., for diuron 2,86 mg/L and for 3,4-DCA 1.41 mg/L, pools of 7 embryos were shock-frozen at 13 time points from 1.5 to 120 hours post fertilization (hpf). The test compounds were extracted from the embryo tissue with $MeOH/H_2O$ and quantified using liquid chromatography coupled to high resolution mass spectrometry (LC-HRMS). Depuration of test chemicals from the embryo tissue was examined in five different developmental stages of embryos that upon exposure to chemicals were transferred to clean medium and then sampled after 0.5, 1.5, 3, 6 and 24 hrs. The tissue concentrations for diuron reached T_{max} around 48 hpf, T_{max} for 3,4-DCA was between 8 and 24 hpf. Based on the data for internal concentrations upon different exposure times uptake and elimination rate constants (k_1, k_2) were determined. Both elimination rates and residue of initial concentration after 24 hrs. of depuration differed between embryo stages. The search for possible metabolites showed that 3,4-DCA was transformed into 3,4-dichloroacetanilide in the embryo and two products of N-demethylation of diuron were found. This confirms that both phase I and phase II metabolic enzymes are active from the first hours of embryo development and pinpoints to the biotransformation capability of the zebrafish embryo at this early stage.

668

Application of a generic fish PBTK model for binary mixtures of chemicals

C. Tebby, INERIS / METO; R. Beaudouin, A. Grech, C. Brochot, INERIS / Models for Ecotoxicology and Toxicology METO

The integration of mechanistic approaches in Environmental risk assessment requires the integration of processes to move towards estimating internal dose from exposure or environmental concentrations (external dose) to predict toxicity in each taxa or the whole ecosystem. In this context, the overall objective of this work is to develop models to integrate TK data for environmental risk assessment of single and multiple chemicals. Three steps were defined to fulfil this objective: (i) Data collection of biological, physiological, and toxicological variables to calibrate and develop PBTK models, (ii) Development of PBTK models for environmental risk

assessment of single chemicals, (iii) Development of PBTK models for environmental risk assessment of multiple chemicals. Generic PBTK models for single compounds in rainbow trout (*Oncorrynchus mykiss*), fathead minnow (*Pimephales promelas*), stickleback (*Gastrosteus aculeatus*) and zebrafish (*Danio rerio*) have been developed. Physiological description and parameters proposed by Nichols et al. [1] were updated by an extensive litterature search. New mathematical functions were proposed to integrate the main factors influencing the toxicokinetics (water temperature, growth dilution, reproduction cycle,…). Default values for compound-specific parameters were estimated by QSAR models based on hydrobycity [2, 3]. An optional interaction terms was added to the mixture PBTK models for metabolic interactions such as competitive inhibition. Two case studies were selected based on availability of toxicokinetic (TK) and toxicdynamic (TD) data for single compounds and for mixtures. In the first case study, the interaction between melamine and cyanuric acid was studied and in a second case study, the interaction between chlorpyrifos and permethrin was modelled. The models developped enable to model interactions that are observed between exposure concentrations and final effects. The QSAR estimations of certain compound-specific parameters can compensate for the lack of data in fish. Extrapolation from one species to another with the various models developed can also help bridge gaps. [1] Nichols et al. 1990. Toxicol Appl Pharmacol 106:433-447. [2] Bertelsen et al. 1998.. Environ Toxicol Chem 17:1447-1455. [3] Nichols et al. 2006. Aquat Toxicol 78:74-90.

669

Application of Aqueous and Dietary In-Vivo Bioaccumulation Tests to Determine Biotransformation Rates, Elimination Rates and other Bioaccumulation Metrics

F. Gobas, Simon Fraser University / Resource & Environmental Management; M. Dimauro, K. Compton, Simon Fraser University; Y. Lee, Simon Fraser Univeristy / Resource and Environmental Management; V. Otton, Simon Fraser University / Resource and Env Management; J.C. Lo, Simon Fraser University / Biological Sciences; G. Allard, Simon Fraser University / Faculty of Environment

Bioaccumulation assessment is quickly evolving to respond to the need to conduct bioaccumulation assessment faster, better, cheaper while reducing animal use. In this presentation, we present novel methods for conducting both aqueous and dietary bioaccumulation tests that provide more information on bioaccumulation than currently used methods while also reducing animal use, effort and costs. Stream-lined aqueous bioconcentration test designs and dietary bioaccumulation studies following OECD 305 test guidelines are shown and discussed. A key element of the novel methods is the use of reference chemicals. In addition to experimental methods, we also present computational methods for deriving biotransformation rate constants, elimination rate constants and other bioaccumulation metrics with their associated error from the results of bioaccumulation tests. This involves an Excel worksheet, referred to as ADME calculator, that is specifically developed to interpret the results from aqueous and dietary bioaccumulation tests in terms of Absorption, Distribution, Metabolism and Excretion (ADME) rates including somatic and intestinal biotransformation rate constants, elimination rate constants, BCF and other bioaccumulation metrics. We further demonstrate that the application of reference chemicals can help to develop a full mass balance of the internal distribution of test chemicals in fish in the test and in the field under environment-specific exposure conditions. The application of the test results for determining exposure pathways of the test chemical under field conditions is illustrated. We conclude that bioaccumulation tests can become more effective in developing bioaccumulation profiles of chemicals when including reference chemicals.

Poster Abstracts

Advances in environmental risk assessment of oil spills and offshore oil & gas operations (P)

MO001

An in-situ amphibian metamorphosis assay to evaluate oil spill-related toxicity in receiving freshwater systems

R. Krohn, University of Calgary / Dept. of Ecosystem & Public Health, Faculty of Veterinary Medicine; J. Muscatello, Lorax Environmental Services Ltd; J. Smits, University of Calgary / Ecosystem & Public Health Faculty of Veterinary Medicine

Diluted bitumen (dilbit) transported from the oil sands in northern Alberta, consists of a mixture of chemicals, such as aromatic hydrocarbons, metals and other compounds, which may pose risks to wildlife and human health, if spilled into the environment. There is a major knowledge gap regarding remediation of oil spills into freshwater environments. The relative efficacy of different remediation strategies for these spill emergencies are untested. We have established an *in situ* amphibian assay to serve as an indicator of health and recovery in freshwater ecosystems, which can be applied to assess risk and remediation efficacy. In spring 2017, Wood frog tadpoles were placed in 5, partially submerged cages (50 animals/cage), which were tethered to the peat-organic shoreline of Lake 260 of the International Institute for Sustainable Development-Experimental Lakes Area (IISD-ELA), Ontario, Canada. Tadpoles were fed and monitored every other day and were euthanized when >50% reached their metamorphic climax (the day of forelimb emergence), to perform gross anatomical examinations, sample collection and relevant biochemical analyses. Major outcomes: 1. Time to metamorphosis (an established, sensitive biomarker) 2. Mortality rate 3. Morphometrics (total body mass, length and hepatic mass) Analyses: 1. Hepatic detoxification effort (ethoxyresorufin-O-deethylase (EROD) enzyme activity); 2. Thyroid hormones levels (sensitive biomarkers of endocrine disruptors); 3. Triglyceride levels (reflecting body condition & energy stores). 4. Tissue contaminant levels (metals, PAHs) Baseline data for Wood frog development in Lake #260 were acquired in 2017, and potential pitfalls and solutions for the metamorphosis assay were identified. This assay will be used in the 2018 field season with the experimental shoreline dilbit spills and remediation strategies planned at Lake #260 at the IISD-ELA. \n Keywords: oil spill, endocrine disruptors, metamorphosis, Wood frogs

MO002

APPLICABILITY OF RISK BASED, TIERED ASSESSMENT OF PRODUCED WATER DISCHARGE IN NIGERIAN SHALLOW OFFSHORE ENVIRONMENT

M.G. Smit, Shell International; O. Anako, SPDC Nigeria Ltd

The pursuit of excellence in managing risks associated with produced water discharges has led to continuous innovation of internationally acceptable risks assessment tools for determining the risks associated with produced water discharges. This risk based assessment have provided a structured approach for the assessment of potential risks from water discharges. At the centre of this structure is a comparative analysis of predicted environmental concentrations (PECs) of chemicals and effluents to predicted no effect concentrations (PNECs) of environmental receptors. However, the determination of the likelihood and severity of effects is complicated and based on an integrated evaluation of several Lines of Evidence (LoEs). This study utilized risk-based assessment tools from Shell's tiered assessment framework for discharges. This framework was developed based on international good practice and includes screening tools that allow for a rapid assessment of discharge properties and associated risks like SPME-GC and Microtox. In addition higher tier tools were applied like two way GCxGC, PETROTOX modelling and several levels of plume dispersion assessment. The objective is to determine the applicability of risk-based practice to the specific shallow offshore discharge, with possible replication to other shallow offshore or near shore discharges in Nigeria. It also assessed if discharges would be acceptable from an international perspective and whether indeed the risks are tolerable and as low as reasonably practicable (ALARP). Results of the assessment indicate that at oil in water levels at or below 25mg/l there is low concern related to the environmental risk of the hydrocarbons in the PW discharge. Phenols and BTEX came up as the highest risk contributors but Tier-2 modelling indicated that these substances quickly dissipate after discharge. PETROTOX modelling showed that the hydrocarbon fraction in the PW could not fully explain the observed PW toxicity. This led to a recommendation for qualification and registration of offshore chemicals and identified a need for the alignment considerations of future Biological Monitoring programs to international protocols such as OECD and ISO. Application of smart screening tools (Tier-1) for frequent PBT monitoring to address variability and for tuning and focusing the larger Tier-2 and 3 assessments.

MO003

Assessment of the biological impact of using chemical dispersants to remediate oil spills in different environmental conditions using zebrafish embryos

N. Gonzalez-Soto, A. Esteban-Sánchez, University of the Basque Country UPV/EHU / Dept. Zoology and Animal Cell Biology and Research Centre for Experimental Marine Biology and Biotechnology PIE; A. Orbea, University of the Basque Country / Dept. Zoology and Animal Cell Biology and Research Centre for Experimental Marine Biology and Biotechnology PIE

Oil spills are a global concern due to their capacity to affect wide areas of the ocean and the difficulty of the subsequent restoration of the ecosystem. Early life stages of fish are especially sensitive to oil spills as they are unable to flee the area and lack mature detoxification systems to withstand the chemicals. They also represent a direct link to population consequences and resilience. The addition of chemical dispersants can facilitate the dissolution of some chemicals present in the oil and make them more bioavailable. In addition, some chemical dispersants have been proved to be toxic themselves. The impact of crude oil on a specific ecosystem and its recovery potential are determined by the biotic and abiotic elements of the ecosystem such as species composition, temperature, oxygen level and salinity. At low temperatures the persistence of hydrocarbons in the environment increases. Based on the standard OECD test with zebrafish embryos, we have tested the toxicity of the chemical dispersant FINASOL OSR52 and of the water accommodated fraction of a naphtnenic North Sea crude oil produced with dispersant (WAF_{OIL+D}) or without dispersant (WAF_{OIL}) at different conditions of temperature and salinity. For WAF produced in marine water, polydimethylsiloxane (PDMS) sheets were incubated in the WAF_{OIL}/ WAF_{OIL+D} and then used as passive dossers. Exposure to the dispersant caused 100% of mortality at concentrations ≥500 mg/L. Increased prevalence of malformations were observed at concentrations as low as 0.01 mg/L. Direct exposure to WAF_{OIL} and WAF_{OIL+D} resulted in a greater embryo mortality than the exposure through PDMS sheets. Significant differences were observed in hatching rate and in the prevalence of malformations of embryos exposed to WAF_{OIL} and WAF_{OIL+D} produced in different conditions. Although no clear differences were observed in relation with the temperature production of WAF_{OIL+D} / WAF_{OIL} , in general greater sublethal effects were observed in the case of embryos exposed to WAF_{OIL+D} than to WAF_{OIL}. Zebrafish embryos appeared as a good model to study the toxicity of WAF depending on the temperature and on the addition of chemical dispersants. Funded by the EU H2020-BG-2005-2 project GRACE (grant agreement #679266), Spanish MINECO (NACE project CTM2016-81130-R) and MECD (FPU grant to A.E.), the Basque Government (consolidated research group IT810-13) and the University of the Basque Country (UFI 11/37).

MO004

Behaviour and effects of a marine diesel oil in a semi-static exposure experiment using mussels (Mytilus spp.) from the Baltic Sea

R. Turja, Finnish Environment Institute, SYKE / Marine Research Centre; A. Ahvo, Finnisk Environment Institute / Marine Research Centre; H. Kankaanpää, A. Reunamo, K.K. Lehtonen, K.S. Jørgensen, Finnish Environment Institute / Marine Research Centre

Marine diesel oil is produced and transported in large volumes in the Gulf of Finland and also used extensively as fuel in marine traffic in the Baltic Sea area. The heavily intensifying marine traffic in the area increases the occurrence of smaller spills and leads to higher risk of major oil spills, which would certainly have drastic consequences to the local ecosystem. Chemical composition, mainly the polycyclic aromatic hydrocarbons (PAHs), can be more variable between the different diesel fuels, affecting the toxicity of the diesel to exposed marine organisms. The aim of this study was to determine the changes in the concentration of PAHs in water, accumulation of PAHs, and a battery of biomarkers in Baltic Sea mussels (*Mytilus* spp.) exposed to a common type of low-sulphur marine diesel oil produced by Neste Oil's Porvoo refinery in Finland. The diesel oil was applied to mussel aquaria as a water accommodated fraction (WAF). The exposure set-up consisted two replicate aquaria in each treatment; control, WAF-high and WAF-low, each with 200 mussels in 20 liters of artificial seawater (10°C). Water and WAF treatments were renewed every two days. Changes in PAH concentrations in water were constantly quantified using a TriOS Enviroflu HC-500 fluorometer sensor. Another sensor was used to collect auxiliary data on temperature, turbidity and chl a concentration (mussels fed with algae). Biomarkers of oxidative stress, biotransformation, neurotoxicity and bioenergetics were measured from mussels after 1 and 7 days of exposure and after a one week recovery period in clean water. Water and mussel tissue samples were also taken to chemical analysis of PAHs. Based on the sensor fluorescence data the initial PAH concentrations were ca. 30µg/L in WAF-high and 15µg/L in WAF-low treatments. In a semi-static system with mussels the concentrations decreased during 24h after which the level remained stable until the next water exchange. During the recovery period PAHs occurred in water after every water exchange, suggesting significant release of PAHs from mussels (both from shell surfaces and internal parts). Differences between the treatments were observed in various biomarkers measured. Combined fluorescence, chemical and biomarker data give important insights to the fate and toxic effects of marine diesel oil in the northern Baltic Sea environment.

MO005

Biliary PAHs and enzymatic biomarkers in the teleost Eugerres brasilianus along four tropical estuaries in the Brazilian Northeast

J.S. Silva, R.N. Alves, UFPE Universidade Federal de Pernambuco / Zoology;

P.S. Carvalho, UFPE - Universidade Federal de Pernambuco / Zoología
Polycyclic aromatic hydrocarbons (PAHs) are oil derived compounds known for their toxicity to aquatic organisms. Estuarine regions are frequently contaminated with PAHs as a result of urbanization processes and industrial activities, including the oil productive chain. This study aimed to evaluate PAH biliary bioconcentration and biochemical effects in the fish *Eugerres brasilianus* sampled along four estuaries in the state of Pernambuco, northeastern Brazilian coast. Fish were sampled in Aquirindá river, Formoso River Estuarine System (AR-FRES), Massangana river, inside Suape Estuarine Complex (MA-SEC), Barra de Jangada Estuarine System (BJES) and Bacia do Pina Estuarine Complex (BPEC). Fish bile samples were analyzed using fixed wavelength fluorescence to estimate equivalent concentrations of the PAHs naphthalene, phenanthrene and chrysene. Liver samples were analyzed for activities of biotransformation enzymes *Ethoxyresorufin-O-deethylase* (EROD), and glutathione S-transferase (GST), antioxidant defense enzymes catalase (CAT) and glutathione reductase (GR), and acetylcholinesterase (AChE). Bile PAHs and biochemical biomarkers in fish sampled during an annual cycle in AR-RFES and MA-SEC indicated similar bile PAH concentrations and enzymatic activity levels between these estuaries, despite the different anthropogenic activity patterns. Suape Estuarine Complex includes a developing industrial port complex, while Rio Formoso Estuarine System is within a low population density area focused mostly on tourism. Fish sampled in the two other estuarine systems near Recife metropolitan area, BJES and BPEC, showed chrysene equivalent bile concentrations between 13 x and 19 x higher than AR-FRES, respectively. EROD, GST and CAT activities were also increased in BJES and BPEC, reaching 30 x for EROD, and approximately 2 x for GST and CAT when compared to AR-FRES. Higher PAH bioconcentration and enzymatic induction in *E. brasilianus* fish from BJES and BPEC indicate that these fish are spending energy to biotransform and excrete these contaminants, which may have consequences to their growth and survival in such regions. The results indicate that BJES and BPEC receive a greater input of PAHs, associated with the higher population density and anthropogenic activities in these regions. The parameters used will be useful for monitoring of these estuarine systems, especially Suape Estuarine Complex, which is under a rapid urbanization and industrialization process.

MO006

Bioaccumulation of Sulfur and Nitrogen Containing Hydrocarbons in Petroleum Substances

T. Parkerton, ExxonMobil Biomedical Sciences Inc. / Toxicology & Environmental Science; A. Bleich, ExxonMobil Biomedical Sciences Inc; J. Butler, ExxonMobil Biomedical Sciences, Inc / Environmental Toxicology and Chemistry Laboratory; C.A. Sutherland, ExxonMobil Biomedical Sciences, Inc. / Toxicology and Environmental Science; A.D. Redman, Exxon Mobil Biomedical Sciences / Toxicology and Environment Science Division; M. Lampi, ExxonMobil Biomedical Sciences, Inc.
A recent analytical effort to characterize the composition of petroleum substances from different categories indicated that a variety of sulfur and nitrogen heterocyclic compounds were detected in the range of 0.01-3.24% and < 0.01-0.08%, respectively. Given the limited information available on the bioaccumulation potential of these substance classes, a dietary bioaccumulation study with rainbow trout was performed. Representative compounds with log K_{OW} values > 4.2 from five classes (sulfides, thiols, thiophenes, carbazoles and acridines) were investigated along with a positive control (hexachlorobenzene). Test compounds were administered simultaneously in the diet at a 2% body weight ration per day to minimize growth dilution and limit vertebrate use. Chemical and lipid analysis were performed on diet and fish tissues at different sampling times. Results were used to calculate substance-specific lipid-normalized biomagnification factors, dietary assimilation efficiencies and growth-corrected eliminated rates. Comparison of experimental results to model predictions for non-metabolizable chemicals was used to infer the role of tissue and gut biotransformation in mitigating observed bioaccumulation. This study provides new data to inform bioaccumulation assessments of heterocyclic compounds and to support development of quantitative structure-property relationships for improved bioaccumulation prediction of sulfur and nitrogen containing compounds.

MO007

Biochemical biomarkers and histopathology in juvenile Solea senegalensis for early warning assessment of marine ecosystem health

T. Briaudeau, University of the Basque country UPVEHU; A. Alves Dos Santos, University of the Basque country UPVEHU / CBET Research Group Dept Zoology and Animal Cell Biology Faculty of Science and Technology and Research Centre for Experimental Marine Biology and Biotechnology PIE; G. Guerrero Limón, University of the Basque country UPVEHU; I. Marigomez, Euskal Herriko Unibertsitatea / Zoology & Animal Cell Biology (Sci & Tech Fac); I. Zorita, FUNDACION AZTI; M. Soto, University of the Basque Country / Zoology and Animal Cell Biology, Research Centre for Experimental Marine Biology and Biotechnology PIEUPVEHU; U. Izagirre, University of the Basque Country UPV/EHU / CBET Research Group Dept Zoology and Animal Cell Biology Faculty of Science and Technology and Research Centre for Experimental Marine Biology and Biotechnology PIE
Human originated contaminants can appear diluted in estuarine and marine waters or accumulate in sediments. Chemical analysis provides key data on toxicant levels but gives limited inputs on their potential biological effects. The combination of biological responses with chemical data is essential to improve the assessment of environmental pollution. In this context, the use of benthic species for the assessment of biological effects of marine pollution is crucial for marine environment monitoring. In the Bay of Biscay, the common flatfish *Solea* sp., is candidate to be recognized as sentinel species in pollution monitoring programmes. The present study uses juvenile *Solea* sp. (23.24± 1.22cm standard length) exposed to contamination conditions to better understand toxicity processes involved based on biochemical biomarkers and histopathology. *Solea senegalensis* was exposed to three different experimental set ups: (a) contaminated sediments; waterborne metal (Cd) and (c) waterborne organic pollutant (Benzo(α)pyrene). A battery of biochemical biomarkers was analysed in samples of liver and brain: Catalase, glutathione S-transferase, acetylcholinesterase and superoxide dismutase. Exposure to contaminated sediments led to reduction of catalase, glutathione S-transferase and superoxide dismutase activities and induction of acetylcholinesterase activities. Exposure to waterborne toxicants provoked a reduction of catalase and glutathione S-transferase activities. Biochemical biomarkers in sole were sensitive enough to differentiate degree of response after three days of exposure. Histopathological responses were detected after long-term exposures showing higher prevalence of liver alterations such as hyperaemia, melanomacrophage centres and necrosis. The present laboratory experiments helped characterizing the impact of pollution in sole at different levels of biological organization and different time scales. Work funded by Spanish MINECO (CTM2012-40203-C02-01 and PhD fellowship to T.B.), University of the Basque Country– UPV/EHU (UFI 11/37) and Basque Government through Consolidated Research Groups fellowship (IT810-B).

MO008

BIOMARKER AND GENE TRANSCRIPTION VARIABILITY IN PERCH IN REFERENCE SITES USED FOR BIOMONITORING STUDIES

L. Förlin, N. Asker, University of Gothenburg / Department of Biological and Environmental Sciences; M. Töpel, University of Gothenburg / Department of Marine Sciences; T. Österlund, Chalmers University of Technology / Mathematical Sciences; J. Parkkonen, J. Sturve, University of Gothenburg / Department of Biological and Environmental Sciences
Perch (Perca fluviatilis) has been used in biological effect monitoring of point sources in Sweden for many years, for example in studies of effects of industrial effluents. Since 1988, perch has also annually been included in a program for integrated coastal fish monitoring in three reference sites along the Swedish east coast, sites characterized by no or minor local anthropogenic influences. Long term studies of reference sites offer the possibility to follow the natural variability of physiological and biochemical endpoint (i.e. biomarkers) as this defines the changes of relevance in polluted sites. Using a set of physiological and biochemical endpoints (i.e. biomarkers) clear time trends for "early warning" signs of impaired health are noted in the perch from these three reference sites possibly as a result of increased baseline pollution. The data sets also show relatively large variations between years. To further investigate these time trends and to identify additional temporal variation in biological parameters, global gene transcription studies using RNA sequencing was performed. Perch collected in 2010 and 2014 were selected as they showed variation in several biomarkers such as the activity of the detoxification enzyme CYP1A (EROD), plasma levels of vitellogenin, markers for oxidative stress, white blood cells count and gonad sizes. The RNA sequencing study identified approximately 3800 genes that were differentially expressed in the five sexually mature female perch collected in 2010 compared to the five individuals from 2014. Also principal component analysis (PCA) using all sequenced transcripts identified large differences in gene transcription as individual perch collected during the separate time points were clearly divided into two groups. Gene Ontology enrichment analysis showed that the differentially expressed genes were involved in biological processes such as *innate immune responses, response to toxic substance, response to hypoxia* and *cholesterol biosynthetic process*. In conclusion, differences in immune system parameters and responses to exposure of toxic substances have now been verified on two different biological levels (mRNA and protein) in perch collected in 2010 as well as 2014. Additional biological processes having temporal variation have been identified compared to the previous measurements of biomarkers.

MO009

Cellular and tissue-level biomarkers in mussels (Mytilus edulis) sampled in two different study areas in the Northern Atlantic

D.B. Fernández, ESTACIÓN MARINA DE PLENTZIA. UPV/EHU / Departamento de Zoología y Biología Celular Animal; U. Izagirre, University of the Basque country UPVEHU / CBET Research Group Dept Zoology and Animal Cell Biology Faculty of Science and Technology and Research Centre for Experimental Marine Biology and Biotechnology PIE; X. Lekube, PiEUniversity of the Basque Country; I. Marigomez, Euskal Herriko Unibertsitatea / Zoology & Animal Cell Biology (Sci & Tech Fac); B. Zaldibar, University of the Basque Country UPV/EHU / Department of Zoology and Animal Cell Biology. Centre of Experimental Marine Biology & Biotechnology; M. Soto, University of the Basque Country / Zoology and Animal Cell Biology, Research Centre for Experimental

Marine Biology and Biotechnology PIEUPVEHU
Biomarker approach has been widely used in mussel monitoring programs for several years. However, up to know it has not been commonly used in high latitude study areas. In order to establish reference values of cellular and tissue-level biomarkers in the Northern Atlantic Ocean, mussels of two sizes (small, 2-3 cm; large, 3.5-4.5 cm) from selected polluted (commercial harbor & ports, WWTP dumping area) and reference sites in Tromsø (69º 40´N) and Trondheim (63º 26´N) were sampled in early autumn of 2016 and late summer 2017. Different tissue-level biomarkers including cell type composition (VvBAS) in digestive gland epithelium, structural changes of digestive alveoli (MLR/MET), relative proportion of digestive and connective tissue (CTD) and histopathological alterations in the digestive gland were measured. In addition, lipofuscin and neutral lipid accumulation, lysosomal membrane stability (LMS) and structural changes in the endo-lysosomal system (LSC) of digestive cells were also determined. Higher VvBAS values were recorded in polluted sites than in mussels from reference sites in both study areas. Moreover, mussels from impacted sites exhibited enhanced atrophy of the digestive alveoli (high MLR/MET values) and retraction of digestive diverticula resulting in apparently higher relative extent of interstitial connective tissue (high CTD ratio). Regarding inflammatory responses, parasitic burden and atresia, higher weighed prevalence values than in the reference site were recorded in the two polluted sites from Trondheim. Differences between the two mussel sizes were recorded in parasitic burden, large mussels exhibiting a higher level of parasitization than small mussels. Lipofuscin accumulation was higher and neutral lipid accumulation lower in the polluted sites than in reference sites in both study areas. Lysosomal biomarkers were different between the two sizes. Overall, all biomarkers respond similarly in both study areas indicating the suitability of the selected biomarkers in order to be applied in the Northern Atlantic Ocean.Acknowledgements: Work funded by, EU GRACE Project (Grant Agreement Number 679266), Basque Government (IT810-13) and UPV/EHU (UFI 11/37).

MO010

Cytotoxicity of the WAF of naphthenic North Sea crude oil with and without dispersant in hemocytes of the marine mussel Mytilus galloprovincialis (L.)

G. Nicolussi, University of the Basque Country / CBET Research Group, Dept. Zoology and Animal Cell Biology, Science and Technology Faculty and Plentzia Marine Station, University of the Basque Country (UPV/EHU). Basque Country, Spain; A. Katsumiti, University of the Basque Country / CBET Research Group, Dept. Zoology and Animal Cell Biology Faculty of Science and Technology and Research Centre for Experimental Marine Biology and Biotechnology PIE; D. Bilbao, University of the Basque country (UPV/EHU) / IBeA Research Group, Dept. Analytical Chemistry; N. Etxebarria, University of the Basque Country UPV/EHU / Plentzia Marine Station (PiE-UPV/EHU) & Dep Analytical Chemistry; M.P. Cajaraville, University of the Basque Country / CBET Research Group, Dept. Zoology and Animal Cell Biology Faculty of Science and Technology and Research Centre for Experimental Marine Biology and Biotechnology PIE

Oil pollution coming from accidental oil spills and from activities related to oil production and transport pose a risk to marine ecosystems. Impact of oil spills may vary depending on environmental parameters such as temperature. There is a need to develop efficient tools to assess the risks of oil pollution and of oil spill response strategies, such as the use of dispersants. The aim of this work was to apply an *in vitro* approach using hemocytes of the marine mussel *Mytilus galloprovincialis* as a model to evaluate the toxicity of the water accommodated fraction (WAF) of naphthenic North Sea crude oil produced at different temperatures (10, 15 and 20°C) with and without the dispersant Finasol OSR 52. In order to evaluate the contribution of the dispersant on the toxicity of the WAF plus dispersant, the cytotoxicity of the dispersant alone was also tested. Primary cultures of hemocytes were exposed in glass covered microplates to different dilutions of WAF (0.25, 2.5, 25, 50 and 100%) with and without the dispersant and to the dispersant alone at the same concentrations present in the WAF dilutions of oil plus dispersant (1.25, 12.5, 125, 250 and 500 mg/L). After 24 h exposure, cytotoxicity (MTT test) and ROS production were measured. WAF was moderately cytotoxic to mussel hemocytes. WAF produced at different temperatures showed similar cytotoxicity to hemocytes. A slight but significant decrease in cell viability occurred at 25, 50 and 100% WAF (produced at 10 and 15°C) and at 50 and 100% WAF (produced at 20°C). These results suggest that relevance of temperature of WAF production on its cytotoxicity is limited. WAF caused a significant induction of ROS production in hemocytes, indicating occurrence of oxidative stress. When tested alone, the dispersant caused a slight but significant decrease in cell viability at the two highest concentrations. However, WAF produced with dispersant at the three different temperatures was not toxic to hemocytes. These results appear to indicate that the dispersant efficiently reduced the toxicity of the crude oil WAF in the selected cell model. Overall, the *in vitro* toxicity testing approach in mussel hemocytes could be used as a rapid screening tool for environmental risk assessment of oil spills and oil response strategies in the marine environment. *Funded by EU H2020 GRACE project (679266), Spanish MINECO NACE project (CTM2016-81130-R), Basque Government (consolidated research group IT810-13) and UPV/EHU (UFI 11/37).

MO011

Determination of inorganic cations and amines in wastewater, surface water, and neutralizing amine solutions by IC coupled with a single quadrupole MS

T. Christison, Thermo Fisher Scientific / Strategic Ion Chromatography Applications; G. Ellison, ThermoFisher Scientific / Chromatography and mass spectrometry division; T. Cross, Thermo Fisher Scientific; J. Rohrer, Thermo Fisher Scientific / Chromatography and mass spectrometry division

Inorganic cation and amine determinations are important to assess salt build-up in amine neutralizing solutions, or to meet regulatory discharge compliance from petroleum and municipal treatment plants wastewater. Additionally, municipal water plants require cation determinations to monitor secondary water characteristics. In the petroleum industry, alkanolamines (monoethanolamine, diethanolamine, and methyldiethanolamine) are used routinely to prevent corrosion during transportation to the refinery or to remove sour gases during the refining process. Processing plants require accurate analytical methods to characterize and determine the next refining steps needed for oil and gas products received from various oil, gas, and fracking wells and to meet wastewater discharge requirements. Ion chromatography coupled to a single quadrupole mass spectrometer (IC-MS) is an ideal and economical way to determine and confirm cations and amines. Here we demonstrate cation, alkylamine, and alkanolamine determinations in amine neutralizing solutions, amine wastewater, municipal wastewater samples, drinking, and surface water samples by cation-exchange separation followed by suppressed conductivity and mass spectrometry detections in a serial configuration. Cations, alkylamines, and alkanolamines were determined in full scan from m/z 18 to 250 and individual SIMs as bare ions and when further sensitivity is needed, as their hydrated adducts. Unlike earlier IC-MS methods for cation determinations, the new single quadrupole MS used in these experimentsrequired no organic solvent for desolvation. Limit of Detections were single digit or double digit µg/L for most analytes. The experiments showed that typically sodium, ammonium, and primary amines were the primary contaminants in the scrubbing amines.

MO012

Distribution and ecological risk assessment of palm stearin in coastal marine environments of Hong Kong after an accidental pollution in Pearl River Estuary, South China

G. Zhou, R.W. Lai, R.C. Sham, C. Lam, The University of Hong Kong / The Swire Institute of Marine Science and School of Biological Sciences; K. Yeung, J.C. Astudillo, The University of Hong Kong; K.K. Ho, The University of Hong Kong / The Swire Institute of Marine Science and School of Biological Sciences; M.M. Yung, J.K. Yau, The University of Hong Kong; K.M. Leung, The University of Hong Kong / The Swire Institute of Marine Science

On 3 August 2017, two container vessels collided in the Pearl River Estuary, southwest of Hong Kong, leading to release over 1,000 tonnes of palm stearin into adjacent waters. About 200 tonnes of the stearin reached south coasts of Hong Kong after two days. However, toxicity of palm stearin to marine organisms is unknown. This study, therefore, aimed to examine its contamination levels in seawater, sediment and animal samples collected from seven locations along the south coasts of Hong Kong; determine its toxicities to selected marine organisms including microalgae (*Isochrysis galbana* and *Chaetoceros gracilis*), the copepod (*Tigriopus japonicus*), the artemia (*Artemia franciscanas*), and the fish (*Oryzias melastigma*), and derive interim water quality guidelines (WQG) of the palm stearin and thereby assess its ecological risks to local marine ecosystems. Samples of the palm stearin, surface seawater, sediment and three intertidal gastropods were collected twice (within seven days and four months after the incident) in six locations along the south coast of Hong Kong. Fatty acids in these samples were detected using gas chromatography-mass spectrometry. Standard toxicity tests were conducted with the selected species. The results showed that all seawater samples collected from the six sites were heavily contaminated by the palm stearin after one week of the accidental pollution. We also found that although the palm stearin had little effects on marine animals, it was toxic to microalgae species as reflected by growth inhibition. Its toxic mechanisms on the microalgae may be associated with its adsorption onto microalgal cells, and reudction of light prenetration to the cells due to obstruction from the stearin and palm oil. At present, we are generating additional toxicity data on other microalgal species (*Thalassiosira weissflogii* and *Tetraselmis suecica*). Using all toxicity data generated from this study, we will ultimately determine an interim WQG for the palm stearin, and use this WQG to assess its ecological risks to local marine ecosystems. This study represents the first comprehensive investigation on the ecological risk of the palm stearin in the world, and the results will facilitate informed decision-making by the environmental authority.

MO013

Ecological impacts of larvicidal oil on the marine ecosystem: implications on its management

K. Yeung, The University of Hong Kong; K.M. Leung, The University of Hong Kong / The Swire Institute of Marine Science

Mosquitoes are some of the most influential insects to mankind in the world due to their ability to transmit diseases to humans, resulting in millions of deaths every year. Prevention of mosquito-borne diseases and elimination of mosquitoes are important to protect human health. Among different methods of eliminating mosquitoes, the use of larvicidal oil is the most common way being employed in Hong Kong. However, larvicidal oil will be eventually released into the marine environment due to rainfall and surface runoff, and thus it may affect marine organisms. However, its toxicity and ecological risk to marine organisms remain largely unknown. Therefore, this study aims at investigating the environmental fate of larvicidal oil in the marine environment and its toxicities towards marine organisms at different trophic levels along the food chain. The composition of larvicidal oil was characterized by gas chromatography-mass spectrometry. It was found to consist mainly of aliphatic petroleum hydrocarbons (*n*-alkanes) ranging from *n*-octane to *n*-pentacosane. The concentrations of larvicidal oil were determined with the range from 6.92 mg/L to 53.89 mg/L., by analyzing water samples collected along coastal areas in Hong Kong. Standard acute toxicity tests were conducted to investigate their toxic effects to the marine microalgae *Isochrysis galbana* and *Chaetoceros gracilis* (primary producers), the intertidal copepod *Tigriopus japonicas* (a primary consumer), the brine shrimp *Artemia franciscana* and fish embryos of the marine medaka *Oryzias melastigma*. Our results showed that although all test marine species were not very sensitive to larvicidal oil with the ranking of their acute median lethal concentrations (LC50) that were all above the estimated hazardous concentration for 5% of species (HC_5), the results of a probabilistic risk assessment showed that the local marine ecosystem had 65.7% of chance to be at risk (i.e., hazardous quotients > 1) from exposure to larvicidal oil using Monte Carlo simulation, indicating that the current risk was unacceptably high. Hence, monitoring and control on the use of larvicidal oil as mosquito control pesticide would be urgently needed to mitigate its ecological risks.

MO014

Effects of a coastal oil spill on marine invertebrates and their potential to recover

M.F. Lemos, S. Silva, Instituto Politécnico de Leiria / MARE IPLeiria

There has been an increasing public concern and focus on marine contamination issues mainly due to the arising of emergent pollutants, posing a major threat to human and environmental health. Still, the contamination by polycyclic aromatic hydrocarbons (PAHs) remains one of the most ubiquitous sources of pollution in the marine environment, being reported to elicit toxic, carcinogenic and mutagenic effects on marine biota. Moreover, the assessment of these impacts in costal invertebrates after a spill, the extent of these effects and energetic trade-offs, potential recovery, and even which species to use is still deemed for an effective environmental contamination assessment. After an accidental industrial oil spill at the rocky shore of Peniche, Portugal in the summer of 2018, the water was analyzed during the low-tide for PAHs one week later and regularly throughout six months in the spilled beach and in 7 other rocky beaches in the vicinity. Also, at all locations, *Patella depressa* and *Gibbula umbilicalis* organisms were collected, and several biomarkers addressed. For both species, the neurotransmission enzyme acetylcholinesterase, oxidative stress enzyme catalase and superoxide dismutase, oxidative damage DNA damage and lipid peroxidation, energy metabolism lactate and isocitrate dehydrogenase, and electron transfer system, and carbohydrates, lipids and proteins energy reserves were assessed. The impacts of this oil spill over the two coastal invertebrate species' biomarkers was compared over the differentially PAH contaminated sites and their sensitivity evaluated. Also, organism's ability to recover over time was also addressed and these tools and species potential for costal monitoring pollution scenarios discussed.

MO015

Effects of oil exposure on visual function in early life stage fishes

J.T. Magnuson, University of North Texas / Biology; A.J. Khursigara, The University of Texas at Austin / Marine Science Institute; E. Allmon, The University of Texas at Austin; A. Esbaugh, University of Texas Marine Science Institute / Department of Marine Science; R.M. Heuer, University of Miami / Marine Biology and Ecology; J.D. Stieglitz, M. Grosell, RSMAS University of Miami / Marine Biology and Ecology; A.P. Roberts, University of North Texas / Advanced Environmental Research Institute

The *Deepwater Horizon* oil spill released millions of barrels of oil into the Gulf of Mexico, coinciding with peak spawning periods of ecologically important fish species, such as the mahi-mahi (*Coryphaena hippurus*), red drum (*Sciaenops ocellatus*), and sheepshead minnow (*Cyprinodon variegatus*). Downregulation of genes important in eye development and function, as well as morphological abnormalities have resulted from polycyclic aromatic hydrocarbons (PAHs) present in the oil at concentrations less than 10 µg/L, impacting fish vision. Mahi-mahi, red drum, and sheepshead minnow embryos were exposed to weathered crude oil and assessed for visual function using the flicker-fusion principle to monitor an optomotor response, with subsequent histological analysis taken of each larvae's retina. Oil-exposed larvae exhibited a reduced PAH-dependent optomotor response with a reduction in retinal layers and neuronal connections that play an important role in visual function and image processing. The present study provides evidence that weathered crude oil affects the visual system in developing larval fish, and relates oil-induced histological effects to behavioral endpoints. This research was made possible by a grant from The Gulf of Mexico Research Initiative. Grant No: SA-1520; Name: Relationship of Effects of Cardiac Outcomes in fish for Validation of Ecological Risk (RECOVER).

MO016

Effects of oil spill on coastal seaweed in the Arctic

S. Wegeberg, Aarhus University / Department of Bioscience; J. Fritt-Rasmussen, K. Gustavson, Aarhus University / Department of Bioscience - Arctic Environment

In case of an acute oil spill response operation, decision making regarding the operational response strategy and prioritizing biology at risk must be resolute. For that a Net Environmental Benefit Analysis, NEBA, is often performed to achieve the optimal environmental benefit with respect to choice of oil spill combat methodology and biology at risk. To provide data for assessing beaching oil spill impacts in the Arctic areas, the effects of oil smothering of the macroalgae *Fucus distichus*, which is a dominant species in the intertidal zone of the coasts in the Arctic, as well as its self-cleaning potential by wash in sea, were studied. Effects of four different oil types were tested, including crude oil types, bunker oil and marine diesel. Different oil types have varying properties depending on the origin of crude oil and refinery process, and hence may have different effects due to their physical and chemical characterizations. Photosynthetic activity was measured as proxy for effect on growth and the self-cleaning potential was tested by wash in sea for oil smothered tips of *F. distichus* over a period of 2 weeks. The removal of the oils from the seaweed surface was considered as relatively fast ($T_{1/2}$ ~ 3-4 days). Depending of oil type, the oil inhibited or stimulated photosynthetic activity. Marine diesel inhibited photosynthetic activity, whereas the three other oil types stimulated the activity. Thus, in general, the results indicated 1) that oil smothering was relatively fast washed off in the sea water; 2) that, depending on the oil type, photosynthetic activity were stimulated or inhibited; and 3) that the photosynthetic activity was still affected (stimulated or inhibited) even after 14 days, although oil on the tip surface was completely or almost completely washed off. The studies were funded by the European Commission Horizon 2020 programme and the Government of Greenland.

MO017

Effects of water accommodated fractions of crude oil on the Baltic Sea blue mussel Mytilus trossulus at different salinities

A. Ahvo, Finnisk Environment Institute / Marine Research Centre; R. Turja, Finnish Environment Institute, SYKE / Marine Research Centre; A. Reunamo, Finnish Environment Institute / Marine Research Centre; J. Nuutinen, Finnish Environment Institute / Laboratory Centre; K.K. Lehtonen, K.S. Jørgensen, Finnish Environment Institute / Marine Research Centre

In the Baltic Sea accidental oil spills are mainly combatted using mechanical collection. However, this method is insufficient in harsh weather conditions such as high waves or in the presence of ice. The use of dispersants is an alternative counteractive method but in the Baltic Sea their use is restricted by HELCOM recommendations since the chemically dispersed oil may cause severe toxic effects on marine biota. In addition, the behaviour of dispersants with oil under brackish water conditions is not well studied. In the present study, impacts of a crude oil and the dispersant Finasol 51 on marine biota were investigated under cold conditions (5ºC) at two salinities corresponding to the German (15.0) and southern Finnish coastal areas (5.6). Baltic Sea blue mussels (*Mytilus trossulus)* were exposed to the water accommodated fraction (WAF) and dispersed WAF (WAF-D) of naphthenic North Sea crude oil in a semi-static aquarium experiment. Concentration of WAF or WAF-D in the aquaria was 5%. The mussels were sampled after 0, 1, 7 and 21 days of exposure, and analyzed for accumulation of polycyclic aromatic hydrocarbons (PAHs), and biological effects including acetylcholinesterase, glutathione-*S*-transferase, catalase and glutathione reductase activities, lipid peroxidation, and protein carbonylation. In addition, changes in *Mytilus*-associated bacterial communities extracted from the gills and digestive glands of the mussels were investigated by sequencing of 16S rRNA genes and quantitative PCR targeted to bacterial PAH-degradation genes. Water samples from the exposure aquaria were taken for oil and PAH analysis. Dispersant augmented the amount oil in the exposure water with 0.13 mg/l oil in 5.6 WAF compared to 44 mg/l oil in 5.6 WAF-D (GC-FID, petroleum hydrocarbons C_{10}-C_{40}). A significantly higher oil concentration was observed at the lower salinity WAF-D water with 44 mg/l oil at 5.6 and 1.82 mg/l oil at 15. The higher salinity and WAF-D elicited more oxidative stress and neurotoxic effects already after one day of exposure. *Mytilus*-associated bacterial communities also varied depending on salinity and the use of dispersant. The results indicate that during the application of dispersants salinity plays a key role both in regard to oil concentrations in water as well as biological effects observed in the exposed biota. This should be taken into careful consideration when designing oil spill mitigation procedures in the Baltic Sea.

MO018

Multiple biomarkers on the estuarine guppy Poecilia vivipara to monitor two brazilian tropical estuaries

A.G. Torreiro-Melo, UFPE Universidade Federal de Pernambuco / Department of Zoology; J.S. Silva, UFPE Universidade Federal de Pernambuco / Zoology; E. Zanardi-Lamardo, Universidade Federal de Pernambuco / Department of Oceanography; P.S. Carvalho, UFPE - Universidade Federal de Pernambuco / Zoología

Integrated approaches using biological responses in multiple organization levels are essential for environmental monitoring of tropical estuaries with ecologically relevant tools. The guppy *Poecilia vivipara*, native species with a broad tropical distribution, was utilized in such an approach, using *in situ* field exposures in cages

(IS) and resident individuals (RES) collected close to the mouth of two tropical estuaries, Bacia do Pina Estuarine System (BPES), and Barra de Jangada Estuarine System (BJES), in the Brazilian northeastern coast. This study is based on the analysis of water concentrations and internal accumulation of bile metabolites of polycyclic aromatic hydrocarbons (PAHs) by fixed fluorescence (FF), as well as biochemical responses related to the biotransformation of contaminants ethoxyresorufin-O-deethylase (EROD) and glutathione S-transferase (GST), and to neurotransmission acetylcholinesterase (AChE). Behavioral activities related to swimming speed and resistance were also evaluated. Individuals grown in the laboratory were used for *in situ* exposure and also as controls (CON) for IS and RES. Significant contamination by PAHs was evidenced from both estuarine systems, with higher phenanthrene and chrysene concentrations in the bile of resident fish at BPES, which in turn partially justified the significant induction of EROD and GST in these individuals. Resident fish at BJES showed high EROD and GST induction that cannot be explained by PAHs contamination, and suggests the presence of other contaminants with mechanisms of action similar to dioxins, possibly from a paper industry. Elevation of GST activity was detected in three of the four sites assessed on both estuaries, and loss of swimming resistance was verified on individuals exposed at the same sites, indicating a correlation between GST and this behavioral effect relevant to survival of the species. Indications of acetylcholinesterase inhibitors were not detected, except at the BPES inner region. This study shows the potential and feasibility of using the guppy *P. vivipara* on the evaluation and monitoring of pollution in estuaries along the Brazilian coast.

MO019

NEW METHODOLOGY TO DETERMINE BTEX IN SOIL SAMPLES BY HPLC-DAD

L. Silva, Universidade Federal do ABC / PROGRAD - CLD; C. da Silva, E.C. Lima, UFABC / CCNH; D. Rosa, UFABC / CECS

Benzene, toluene, ethylbenzene and xylene, commonly referred as BTEX, are constituents of fossil fuels that cause serious negative impacts on the environment and human health. At fuel stations whose storage tanks are leaking, these substances may in contact with the soil and even reach the groundwater. In order to detect the concentration of these compounds in contaminated soils, gas chromatography (GC) is the most commonly used technique. In the present work it is proposed the use of high performance liquid chromatography coupled to diode array detection (HPLC-DAD) for the determination of BTEX in soil samples. A methodology was developed using as mobile phase methanol and H_2O acidified with 250µL of H_3PO_4 (70:30, v / v), Eclipse XDB C18 column (5µm x 4,6 x 250mm), flow of 1.5 mL min^{-1}, λ = 205nm and T = 50 ° C. The analysis was carried out using the Agilent 1220 HPLC system equipped with an automatic injector, a column oven, and a diode array detector. Data were acquired using the OpenLAB A.01.05 software. A calibration curve for BTEX standards was constructed in 7 concentration levels: 1 to 68 ppm for benzene, 1 to 80 ppm for toluene, 1 to 80 ethylbenzene and 1 to 85 for xylene. The curves were submitted to inter- and intra-assay repeatability analyzes. Standard curves with adjustments above 0.991 relative standard deviations (% RSD) of less than 1.9% were obtained. Reproducibility tests were performed with two solutions obtained from the standard solution. In the samples containing analytes from the soil contaminated with gasoline, % RSD was obtained below 6.5% and recovery rate was 68% for benzene and 75% for toluene, 78% for ethylbenzene and 78% for xylene. The method of soil analysis via HPLC is therefore efficient and as an alternative to be highlighted for analyzes of soils contaminated with gasoline. It is intended to validate the methodology using appropriate protocols and apply it in contaminated areas for the verification of BTEX levels in a next step.

MO020

Petroleum pollution of alluvial sediments near Sava river, Serbia

M. Ilic, IChTM / Department of Chemistry; S. Bulatovic, Faculty of Chemistry, University of Belgrade; T. Solevic Knudsen, IChTM / Department for Chemistry; J. Milic, Institute of Chemistry, Technology & Metallurgy / Department of Chemistry; S. Miletic, J. Avdalovic, Institute of Chemistry, Technology & Metallurgy, University of Belgrade / Department of Chemistry; G. Devic, Institute of Chemistry, Technology and Metallurgy, University of Belgrade

Heating plant "New Belgrade" is located on the left coast of the Sava River, about 1km from its estuary in the Danube, and represents a potential source of petroleum pollutants for the alluvial area of the river, ground water as well as Sava river. The aim of our research was to determine the presence of petroleum pollutants and their vertical migration in the alluvial area of Sava river. The investigation was started in the summer of 2015. The soil was sampled in three different microlocations (Z1, Z3 and Z7) up to depth of 15m. The sampled material was organized in the layers, and for all microlocations was made a lithological profile. Most of the samples have had a clayey-sand structure with low content of organic matter. Extraction of petroleum pollutants from soil samples were done using the Soxhlet apparatus with dichloromethane. After extraction, the dichloromethane extracts were then fractioned by column chromatography into fractions of: saturated hydrocarbons (Fraction I), aromatic hydrocarbons (Fraction II), and polar compounds (alcohols and keto compounds (Fraction III) [1]. For monitoring changes in the vertical migration of petroleum pollutants, and the relationship of this migration with the soil characteristics, the group composition was determined for each borehole of all microlocations, taking into account their lithological profiles. Results of our research showed that in all samples the most represented were polar compounds (Fraction III), while the saturated hydrocarbon were least represented (Fraction I). This trend is almost unchanged in samples from different microlocation at different depths. It can be concluded that composition of petroleum pollutants can be unchanged through the alluvial sediments up to 15m depth and they can reach the underground waters, Sava river and consequently disturb the quality of the environment. References: Miletic S., Ilic M., Avdalovic J., Šolevic Knudsen T., Beškoski V.P., Branimir Jovancicevic B., Vrvic M.M. (2015) Oil pollution in the vicinity of a heating plant in New Belgrade (Serbia) – influence on the quality of the surrounding soil and sediments. 16th European Meeting on Environmental Chemistry, EMEC16, Book of Abstracts. November 30 – December 03. 2015, Torino, Italy. Acknowledgements This work was supported in part by Ministry of Education, Science and Technological Development of the Republic of Serbia, Project No: III 43004.

MO021

Prey capture to male aggression: the role of ecologically relevant behaviours in the assessment of complex petroleum based contaminants.

D. Philibert, University of Alberta / Biological Sciences; D. Lyons, C. Philibert, University of Alberta; K.B. Tierney, University of Alberta / Biological Sciences

Crude oil and its associated by-products are ubiquitous in the aquatic environment due to both natural and anthropogenic sources (i.e. oil seeps and rivers flowing over surface bitumen, and pipeline ruptures, grounded ships, storage tank leaks and tailing pond seepage, respectively). This diversity in sources gives rise to a large family of complex contaminant mixtures, including weathered and unweathered oil, unconventional oil, such as diluted bitumen (dilbit), and crude oil extraction-based mixtures, such as oil sands process water (OSPW). Historically, studies focused on lethality and cardiotoxicity; complex behaviours have been, for the most part, overlooked despite the merits of including these endpoints in toxicological studies. In this study, we compared various ecologically relevant behaviours (prey capture, male aggression, reaction to alarm odourant) of developmentally exposed fish (*Danio rerio* and *Cyprinodon variegatus variegatus*) across various contaminants. Exposure to oil-based contaminants did not impair outright function, but instead altered the variation in behavioral phenotypes present in the population of exposed fishes. Previous studies suggest cortisol can be associated with behavioural phenotypes, and that developmental cortisol levels may pre-determine the behavioural phenotypes found in a population of exposed fishes. Complex behaviours are sensitive sublethal endpoints that could be used in the risk assessment of contaminant mixtures. The inclusion of complex behaviours in toxicological studies brings ecological relevance to a biomarker dominated field.

MO022

Risk-Based Approach: Assessment of Offshore Discharge Waters

K. Wadhia, National Oilwell Varco (NOV) / Environmental; O. Pelz, BP / Gulf Coast Restoration Organization; S. Cousins, BP

In 2012, OSPAR (Oslo and Paris Conventions) adopted the recommendation 2012/5 for a ‘Risk Based Approach (RBA) to the management of Produced Water (PW) discharges from offshore installations’. The application of the RBA recommendations (2012/5) is implemented by the UK’s regulator, the Department for Business, Energy and Industrial Strategy (BEIS). The objective of the RBA is to assess the environmental risk of a PW discharge in the OSPAR maritime area. This is achieved by analysing the effluent and added substances to obtain a measure of the risk of the discharge. If the result is out-with the recommended criteria, a Produced Water Management Plan (PWMP) must be adopted to comply with the RBA regulatory requirements. Processing the information generated by the RBA, each PWMP would be specific to the discharged effluent, platform and area, aiming to minimise environmental risk of each PW discharge. The RBA method is compiled of a six- step process. The steps are based on a standard method where a Predicted Environmental Concentration (PEC) and a Predicted No Effect Concentration (PNEC) of the PW or individual products are determined, and a PEC:PNEC ratio is calculated. The PEC:PNEC ratio and Environmental Impact Factor (EIF) which describes a PEC:PNEC ratio in a specified volume of water characterises the potential risk imposed to the receiving environment. With use of a dispersion modelling tool, the fate of the PW and thus the relative environmental risk can be mapped specifically to the installation area providing an overall risk profile. The PW is additionally characterised at a substance level, highlighting components which contribute to the overall environmental risk, and will feed directly into the PWMP. Notably in the UK RBA methodology is the absence of PW WEA concerning sensitivity to fish, and we therefore studied the comparative inference of use of different trophic level species, including fish (*Cyprinodon variegatus*). This study provided unique and important empirical data and information to evaluate significant considerations for implementation of regulatory PW management methodology. In addition to the potential environmental impact and comparative contribution from prodution chemicals & naturally occuring substances, and validity of the step-wise tiered screening approach, the invesitgations provided valuable assessment into adequacy and sensitivity of ecologically relevant species and the implications for regulatory monitoring regime.

MO023

Risk-based assessment of produced water discharges - need for alignment

M.G. Smit, Shell International

Produced formation water is the main waste stream from upstream oil and gas activities. For offshore installations, next to produced water reinjection (PWRI), discharge of treated produced water is a commonly used disposal route applied in line with ALARP principles (As Low As Reasonably Practicable). In order to properly manage produced water discharges, a variety of principles have been adopted in national and international regulatory frameworks focusing on e.g. the oil in water content, toxicity of produced water, PBT characteristics of applied offshore chemicals, environmental monitoring, etc. The onus is on the industry to comply with regulations in the country of operation, to properly manage the risk of produced water discharges and to relieve any concerns over the potential environmental effects in the receiving marine environment. For this purpose the industry is applying a diversity of tools and methods within the framework of risk-based assessment (RBA). Tools and methods range from simple (tier 1) screening tools to comprehensive (tier 3) field verification programs and include among others; chemical analysis, determination of PBT characteristics through whole effluent assessment studies and modelling, dilution screening and 3D dispersion modelling, etc. Within those practices a wide range of risk endpoints are being applied, each with their own level of conservatism. Without harmonization of endpoints it is difficult to interpret when risks can considered to be adequately controlled. For example, different approaches base the assessment on either chronic or acute toxicity data. And where one method considers an acceptable mixing zone of 500m (OSPAR) another might apply 100m (US-EPA). This hampers straightforward comparison of results of risk-based assessments across industry and regulatory frameworks. Different objectives of the assessment undertaken might, however, be a reason for existing differences in methods. It is, therefore, crucial that for each assessment method a corresponding assessment objective is formulated and communicated. This presentation will provide an overview of RBA practices commonly applied by the industry to offshore produced water disposal, showing main assumptions, input requirements, risk endpoints applied and corresponding assessment objectives. Examples will be used to highlight the need for further harmonization of approaches. Development of industry guidance including a common tiered framework for RBA is suggested as a first step to achieve this.

MO024

Spatial and temporal analysis of the risks posed by total petroleum hydrocarbon and trace element contaminants in coastal waters of Kuwait

E.E. Nicolaus, Cefas Lowestoft Laboratory / Environment and Ecosystems

Nine trace elements including As, Cd, Cu, Fe, Hg, Ni, Pb, V and Zn, and total petroleum hydrocarbons were analysed from water samples collected from 23 stations since 1984 from Kuwaiti coastal waters. Here it was investigated whether concentrations of these determinants are at levels above Kuwaiti and internationally established assessment criteria (AC). The results indicate that Cu and Cd had the most Kuwaiti AC breaches over time. Comparing the data of the last sampled year to the least stringent international AC, then Cu and Cd showed breaches at all stations. The trends for trace metals are significantly downwards, especially for Cd and Hg. No determinant measured showed a significant upward trend, indicating that water pollution for these contaminants is not a worsening situation. However, further sampling should be carried out to confirm these findings, especially at shoreline locations, where routine monitoring ceased in 2011 to investigate any recent changes.

MO025

Temperature-dependant toxicity of Napthenic North Sea crude oil WAF, dispersant and their mixture: sea urchin bioassays

L.d. Miguel, University of the Basque country (UPV/EHU) / Department of Zoology and Animal Cell Biology; U. Izagirre, University of the Basque Country UPV/EHU / CBET Research Group Dept Zoology and Animal Cell Biology Faculty of Science and Technology and Research Centre for Experimental Marine Biology and Biotechnology PIE; I. Marigomez, Euskal Herriko Unibertsitatea / Zoology & Animal Cell Biology (Sci & Tech Fac)

Maritime traffic and oil platforms in the North and Baltic Sea have been growing during the last years and thus, the risk of oil spills is gaining concern. Seasonally driven changes in water temperature could modify the potential toxicity of spill products including chemical dispersants. The impact of crude oil water accommodated fraction (WAF) and dispersants have been widely studied but their potential toxic effects at given different range of temperature have not been deeply explored yet, to our knowledge. Thus, as a part of a European project called GRACE, the aim of the present work was to assess the potential toxicity of WAF produced from: Napthenic North Sea crude oil (NNS), Finasol OSR52 dispersant (Total Fluids) and their mixture (NSS+Finasol OSR52) in a wide range of temperatures (5, 10, 15, 20 and 25°C). In order to identify temperature-dependant toxicity, acute toxicity bioassays using larvae and embryos of the sea urchin *Paracentrotus lividus* (Lamark) were performed. After the exposure period, EC_{50} values were calculated and length of larvae was measured to assess the inhibition of larvae growth. Additionally, larvae abnormalities were determined to calculate a general index of toxicity (IT). In the present study, at 25°C NSS WAF provoked a lower inhibition of larvae length than the other studied temperatures. Accordingly, oil toxicity seems to be influenced by temperature. Indeed, temperature is known to affect hydrocarbon solubility and evaporation, and dispersant effectiveness. The same trend was found for Finasol OSR52 WAF. However, high temperature seems not to follow the same pattern in the case of the mixture. EC_{50} and IT values were in accordance showing that dispersion increases WAF toxicity. Overall, results showed that temperature affects oil, oil:dispersant and chemical dispersant toxicity. In this study, larvae abnormalities and reduced larvae growth indicate that toxicity was lower in crude oil WAF than in dispersant WAF, with their mixture in between, for all the temperatures tested. *Acknowledgement* – This work has been funded by the EU H2020-BG-2005-2 project GRACE (grant agreement number 679266), Spanish Ministry of Education, Culture and Sport (PhD fellowship L.dM FPU15/05517 grant) and the Basque Government (Consolidated Research Group GIC IT810-13).

MO026

Temporal variability of acute toxicity of Produced Formation Water discharged from offshore platforms: the responses of sea bass (Dicentrarchus labrax L., 1758) larvae

L. Mariani, CNR-IRSA / IRSA; E. Magaletti, B. Di Lorenzo, F. Onorati, C. Virno Lamberti, ISPRA Institute for Environmental Protection and Research

The Higher Institute for Environmental Protection and Research (ISPRA) is responsible for the evaluation of the potential environmental impact on marine ecosystem caused by the Produced Formation Water (PFW) discharged from Italian gas offshore platforms. A multidisciplinary approach has been applied through the monitoring of chemical-physical characteristics of water and sediment, matched with biological investigations, such as ecotoxicological bioassays on bacteria, algae, rotifers, crustaceans, echinoderms and fishes. The PFW is an effluent containing complex mixtures of contaminants, the composition of which may change with time. It is therefore necessary to analyse a large number of samples taken over a long period of time in order to adequately assess the toxicity of this effluent. The present paper reports on a specific topic within the whole study: the variability of the acute toxicity responses of fish to PFW collected on two off-shore gas platforms in three years (2003-2005). More sensitive life stages (post larvae of 25-45 days old) of European sea bass (*Dicentrarchus labrax* L., 1758) were used. Tests were performed over 24^h and 96^h and the dilutions 6.25-12.50-25.0-50.0-100.0 % PFW were used. The $LC50^{24h}$ on post larvae ranged from 17.67 % to 37.42 % PFW. The $LC50^{96h}$ on post larvae ranged from 6.68 % to 16.51 % PFW. The PFW acute toxicity responses showed a temporal variability of PFW as it is highlighted by standard deviation values of LC50 data: exposure 24h (25.61 ± 7.02% PFW); 96h (10.84 ± 3.37 % PFW).In accordance with GESAMP recommendations (2007), the work stresses the importance of accurate estimates and measures of oil inputs into the sea, by increasing the number and frequency of samples needed to estimate the environmental hazard of PFW.

MO027

Tentative identification of halogenated polycyclic aromatic hydrocarbons in biota

Z. Xia, University of Manitoba; P. Thomas, C. Marvin, Environment and Climate Change Canada; W. Johnson, University of Manitoba / Chemistry; O. Francisco, I. Idowu, University of Manitoba; J. Stetefeld, University of Manitoba / Chemistry; G. Tomy, Department of Fisheries & Oceans / Department of Chemistry

Polycyclic aromatic compounds (PACs) are a complex class of compounds that are present in fossil material such as petroleum oils. The most common PACs are the polycyclic aromatic hydrocarbons (PAHs) of which 16 have been identified as priority compounds by the United States Environmental Protection Agency. However, there are other important PACs that to date have received less attention. These include halogenated PAHs, non-halogenated alkylated PAHs and heterocyclic aromatic compounds that contain S-, O- and N-atoms. Halogenated PACs especially those containing chlorine atoms are likely to be more environmentally persistent than their non-halogenated analagoues because of the presence of the halogen atoms. In addition, the toxicity of some halogenated PAHs have been found to be similar to dibenzo-p-dioxins and dibenzofurans. Because Cl and Br ions are present in the marine environment, we hypothesize that halogenated PACs can be formed and will be bioaccumalate in biota samples. Here we present a method based on high resolution gas chromatography coupled to mass spectrometry using specific multiple reaction monitoring (MRM) ion transitions in the elctron ionization mode to detect and quantify halogenated PACs in biological samples. The method was used on a NIST Standard Reference Material (SRM) of mussel (*Mytilus edulis*) tissue (SRM-2974a) collected from a marine environment. Preliminary results show that 1-chloropyrene is present in this sample. In addition, we observed multiple peaks in the MRM ion tranisition corresponding to chloro-anthracene/phenanthrene but we do not have authentic analytical standards to match retention times. Work is ongoing to identify other halogenated compounds present in biological samples from Canada.

MO028

The experience with the use of biomarkers as Risk Indicators in Environmental Risk Assessment of oil based discharges offshore

S. Sanni, International Research Institute of Stavanger / Environment; E. Lyng, D.M. Pampanin, International Research Institute of Stavanger / Environment

Department

An approach to integrate biomarkers into probabilistic risk assessment has recently been developed and published regarding oil based discharges offshore. The main purpose has been to enable the of use monitored biomarker responses offshore as Risk Indicators in the procedures for Environmental Risk Assessment of produced water (PW) discharges. The principles of the approach and experiences obtained in applying it to existing oil field monitoring data will be presented. The approach was tested on biomarker data from the latest surveys in the biomarker based Water Column Monitoring (WCM) program on the Norwegian Continental Shelf for assessment of PW effects. Cases including both a typical PW discharge and an alternative discharge make the data set interesting for testing the interpretation capability of the approach. At the site with no discharge of PW at the time of the survey, sediment contaminated by drill cuttings were the sources of contamination. The experiences gained are discussed in relation to contaminant sources, use of the approach to provide assessment criteria for biomarkers, and for the performance of the biomarkers as risk indicators in relation to assessed environmental risk.

MO029

Tissue-level biomarkers and histopathological alterations in mussels (Mytilus trossulus) from the Baltic Sea exposed to water accommodated fractions of crude oil

J. Bir, Khulna University / FMRT department; E. Gil-Uriarte, University of the Basque country (UPV/EHU) / Zoology and Cell Biology; A. Ahvo, Finnish Environment Institute / Marine Research Centre; R. Turja, Finnish Environment Institute, SYKE / Marine Research Centre; K.K. Lehtonen, Finnish Environment Institute / Marine Research Centre; U. Izagirre, University of the Basque country UPVEHU / CBET Research Group Dept Zoology and Animal Cell Biology Faculty of Science and Technology and Research Centre for Experimental Marine Biology and Biotechnology PIE; I. Marigomez, Euskal Herriko Unibertsitatea / Zoology & Animal Cell Biology (Sci & Tech Fac); M. Soto, University of the Basque Country / Zoology and Animal Cell Biology, Research Centre for Experimental Marine Biology and Biotechnology PIEUPVEHU

The brackish Baltic Sea is a fragile ecosystem potentially sensitive to oil spills. Chemical dispersants are an effective method to mitigate coastal impacts of oil spills; however, oil treated with dispersants may have unknown toxic effects on biota. The Baltic Sea blue mussel (*Mytilus trossulus*) is a particular variety of marine mussels adapted to low salinity. Early winter mussels were collected scuba diving in Tvärminne (Finland) in November 2016, taken to laboratory facilities and acclimatized at the experimental temperature of 5ºC to two different salinity regimes, the local 5.6 and the artificially increased 15.0 representing the southern Baltic Sea). Mussels were exposed to water accommodated fractions (WAF) and chemically dispersed WAF (dispersant Finasol OSR 51) mixtures (WAF-D) and sampled at 0, 1, 7 and 21 d. Tissue level biomarkers were investigated to determine the following biological responses: cell type composition (volume density of basophilic cells, Vv_{BAS}) of the digestive gland epithelium, structural changes of digestive alveoli (mean luminal radius/mean epithelial thickness, MLR/MET), mean epithelial thickness/mean diverticular radius (MET/MDR), connective/ diverticula ratio (CTD), gonadal development and other histopathological alterations in digestive gland, gonad and gills. V_{VBAS} increased significantly after 1 d in mussels exposed to WAF and WAF-D at the salinity of 15.0, and decreased afterwards. MLR/MET changed markedly with exposure time at 15.0 whereas MET/MDR showed no response. High CTD values in mussels observed at the salinity of 5.6 indicate a poorer condition of the digestive gland at low salinities than in mussels maintained at 15 psu. Pathological responses (atrophy, necrosis, vacuolization, haemocytic infiltration, granulocytomas) were assessed, being more evident in mussels exposed to WAF and WAF+D (21d). Salinity is a major factor controlling the biology of mussels in the Baltic Sea. The results obtained here indicate that during the early winter the health of native mussels in the very low salinity central-northern part of the sea is more easily impaired than in those inhabiting the more saline southern regions. The current study is among the first ones applying tissue level biomarkers in *Mytilus trossulus* in the Baltic Sea and provides preliminary reference values for future biomonitoring programmes in the area. *Acknowledgement*s: Funded GRACE project (EU H2020 grant Nº679266) and a Basque Gov. fellowship to EGU

MO030

Toxicity of diluted bitumen to freshwater fish and invertebrates

P.Y. Robidoux, V. Bérubé, AGAT Laboratories Ltd / Specialty services Division; J. Leblanc, Fisheries and Oceans Canada / Biologist, Contaminated Sites; M. Desrosiers, Public Services and Procurement Canada

This study investigates the acute and chronic toxicity of two blends of diluted bitumens ("dilbit") and weathered dilbit on freshwater fish and invertebrates after exposure to vdifferent concentrations of physically-dispersed (water accommodated fraction; WAF)vand chemically-dispersed (chemically-enhanced WAF; CEWAF). Toxicity of weathered, unweathered and dispersed Access Western Blend (AWB) dilbit was evaluated on fathead minnow (Pimephales promelas). Toxicity of weathered and unweathered Cold Lake Blend (CLB) dilbit was assessed on Rainbow trout (Oncorhynchus mykiss), and two invertebrate species, daphnia (Daphnia magna) and ceriodaphnia (Ceriodaphnia dubia). For fathead minnow, unweathered AWB demonstrated a significantly higher toxicity (LC50-96 h = 0.628 g/L) compared to the weathered AWB (LC50-96 h = 2.06 g/L). Chronic toxicity tests showed that fathead minnow lethality was also higher for AWB (LC50-7 d = 0.593 g/L) compared to the weathered AWB (LC50-7 d = 1.31 g/L) whereas larval growth toxicity was lower for AWB (IC25-7 d=0.312 g/L) compared to the weathered dilbit (IC25-7 d = 0.096 g/L). Rainbow trout exposed to unweathered CLB demonstrated a significantly higher toxicity (LC50-96h = 5.66 g/L) compared to the weathered CLB (LC50 > 18 g/L). A lethal toxicity (LC50 = 6.43 g/L) was also observed on ceriodaphnia exposed to the CLB WAF while no mortality was observed with the weathered CLB. The reproductive effects on ceriodaphnia were greater with the CLB (IC25 < 1.0) than with the weathered CLB (IC25 = 3.99 g/L). Volatile organic compounds (VOC), polycyclic aromatic hydrocarbons (PAH) and total petroleum hydrocarbons (TPH) increased as the dilbit WAF increased.

MO031

Toxicity of produced water from offshore oil production in Norway and corresponding polar and apolar fractions

T. Størseth, A. Booth, SINTEF Ocean / Environmental Technology; D. Altin, BioTrix; A. Brunsvik, SINTEF Materials and Chemistry / Mass Spectrometry; M.U. Rønsberg, L. Sørensen, B. Hansen, SINTEF Ocean / Environmental Technology; W. Robson, University of Plymouth; P. McCormack, Universitym of Plymouth; S.J. Rowland, University of Plymouth / School of Geography Earth and Environmental; L. Faksness, SINTEF Ocean / Environmental Technology

Regulation of produced water (PW) discharges on the Norwegian continental shelf is based on a maximum oil-in-water limit of 30 mg L^{-1}. However, the use of conventional oil quantification methods based on traditional GC is limited when it comes to polar compounds that originate from produced crude oils. Furthermore, the use of treatment or production chemicals might also contribute to the overall PW toxicity. As a result, there may be discrepancies between measured concentrations of organic compounds and the total PW components that contribute to toxicity. In the current study, PW was collected at the "point of release" from four oil platforms on the Norwegian continental shelf. PWs were selected from oil fields of different operational ages, which produce oils exhibiting different physical and chemical properties. Samples were subjected to extraction with dichloromethane, followed by fractionation into apolar and polar fractions using solid phase extraction, recovering 80 % of the total GC amenable material in these fractions. The total extracts and fractions were thoroughly characterized using GC--MS, GCxGC--MS, LC-Orbitrap-MS, and by direct infusion FT-ICR-MS. The total PW extract, as well as the apolar and polar fractions were subject to acute toxicity tests using nauplii of the marine copepod *Acartia tonsa*. LC_{50} values for the total PW extracts ranged between 0.05--0.98 mg L^{-1} (based on total GC amenable fraction analysis). For three of the PWs, the toxicity was mainly attributed to the polar fractions, with LC_{50} values ranging between 0.17--0.57 mg L^{-1}. Interestingly, toxicity was mainly attributed to the apolar fraction of the fourth PW, with an LC_{50} of 0.05 mg L^{-1}. For the PWs where toxicity mostly related to the polar fraction, this fraction spanned from 16--55% of the total PW (GC amenable fraction analysis). For the PW where toxicity mostly related to the apolar fraction this was 35%. This study demonstrates that PW toxicity may be associated with compounds that are currently poorly characterized. Polar fractions may contain compounds not amenable to GC, or that contribute to the GC--based quantification of oil in water. This suggests that PW toxicity is not directly correlated with the GC quantifiable compounds that are used for regulating discharges today. Further studies should be pursued with a wider array of PWs from a range of sources to determine if alternative methods of characterization are needed for regulation of PW discharges.

MO032

Toxicokinetics of oil components in Arctic copepods

I. Øverjordet, SINTEF Materials and Chemistry / Environmental Technology; R. Nepstad, SINTEF Ocean / Monitoring and Modelling; B. Hansen, SINTEF Ocean / Environmental Technology; T. Jager, DEBtox Research / Dept of Theoretical Biology; J. Farkas, SINTEF Ocean / Environmental Technology; D. Altin, BioTrix; T. Nordtug, SINTEF Ocean / Environmental Technology

To comprehend the implications of large oil spills in the Arctic marine environment, we need a better understanding of the toxicokinetics of oil in true Arctic species. The central position of *Calanus hyperboreus* in the Arctic food chain, as well as its life history strategies and Arctic adaptation, makes it a relevant and valuable test species to provide empirical data on oil component kinetics. *C. hyperboreus* of developmental stage copepodite three (CIII) and five (CV) were exposed to the water soluble fraction (WSF) of crude oil (Troll B) in continuous renewal system (4 or 8 d) followed by a recovery period (20 or 35 d). Water concentrations and body residues, as well as biometry of lipid volume fractions, were measured at intervals during the exposure and recovery period. One compartment toxicokinetic models were fitted to the experimental data to estimate bioconcentration factors (BCFs) and elimination rates (Ke). The BCFs were consistently higher for the lipid-rich CVs compared to the CIIIs, indicating a higher bioaccumulation potential in the lipid-rich stage. The higher lipid volume fractions may explain the higher BCFs, although other factors like body size and activity levels may have contributed as well. The BCFs are well predicted by the octanol-water partitioning coefficient (log K_{OW}). The slope of the relationship, however, differed between the lipid-poor CIIIs and the lipid-rich CVs. For the

CIIIs, the slope was close to unity, indicating a similarity between structural lipids and octanol. The lower slope for CV signifies that storage lipids are less well represented by octanol. Elimination rates were consistently higher in the CIIIs than the CVs, resulting in a substantially longer half-time of elimination and high retention of oil components in the CVs. We discuss the role that various biological factors that may contribute to this difference.

MO033

Two Dimensional Gas Chromatography for the analysis of polycyclic aromatic compounds and their alkylated homologues in environmental samples

I. Idowu, University of Manitoba; W. Johnson, University of Manitoba / Chemistry; O. Francisco, University of Manitoba; P. Thomas, C. Marvin, Environment and Climate Change Canada; J. Stetefeld, University of Manitoba / Chemistry; C. Sandau, Chemistry Matters; T. Obal, Maxxam Analytics International Corporation / Scientific Services; G. Tomy, Department of Fisheries & Oceans / Department of Chemistry

Polycyclic aromatic compounds (PACs) and their alkylated homologies are ubiquitious and known environmental contaminants. Due to their structural diversity and complexity of alkyl-substituted PACs, the resolution of individual alkyl congeners, especially those that are of environmental significance, is difficult if not impossible with conventioanl one-dimensional gas chromatography (GC). The peak capacity of the two dimensional-GC has immensely improved analysis of these complex compounds in environmental matrices. In this study, the separation and identification of individual isomers of C1-C4 naphthalenes, phenanthrenes/anthracenes, dibenzothiphenes, fluorenes, pyrenes/fluoranthenes, chrysenes and benzo(a)pyrenes were examined on a GCxGC high resolution time of flight mass spectrometry (HRTOF/MS). This was achieved by varying the primary oven column length (30m to 60m) using the same column stationary phase (5Sil MS (low polarity)) and a 2m 17Sil MS (mid-polarity). To address a wider scope of organic matrices, samples analyzed include extracts of biota, used lubricating oil and coal samples. Resolution of individual isomers of interest were observed on the 30m primary column and much more evident on the 60m column. Undoubtedly, the peak capacity and vast database of information provided by 2DGC-HRTOF/MS for different sample matrices is an asset in the field of environmental research. This will aid isomer-specific measurements of known toxic alkyl homologues in environmental samples as well as proper identification of prominent ones. Also, PAHs and alkyl-PAHs profiling patterns have proven to be useful in source apportionment.

MO034

Using the hagfish (Myxine glutinosa) to study biological effects of a wreck filled with chemical munitions

A. Ahvo, Finnisk Environment Institute / Marine Research Centre; H. Niemikoski, Finnish Institute for Verification of the Chemical Weapons Convention / Department of Chemistry, University of Helsinki; K. Straumer, Thünen Institute of Fisheries Ecology; J.A. Tørnes, Norwegian Defense Research Establishment; P. Vanninen, Finnish Institute for Verification of the Chemical Weapons Convention / Department of Chemistry, University of Helsinki; T. Lang, Thünen Institute of Fisheries Ecology; A. Lastumäki, K.K. Lehtonen, Finnish Environment Institute / Marine Research Centre

The sea bottom of the Skagerrak Strait (North Sea) contains ca. 45,000 tonnes of chemical warfare agents (CWA) dumped after the Second World War. Entire ships loaded with CWAs were intentionally sunk and are still laying on the deep bottoms (ca. 600 m) of the area in different states of deterioration by corrosion. The current status of the CWAs in the wrecks is unknown; if released into the environment they may have significant deleterious effects on local marine biota. Within the research programme of the EU Baltic Sea Region Interreg project DAIMON (Decision Aid for Marine Munitions, www.daimonproject.com), one of these wrecks was selected to study the leakage of CWAs and their possible biological effects. From the few fish species that inhabit the studied depth range in the region, the hagfish (*Myxine glutinosa*), a sediment-dwelling chordate, was selected as target organism for chemical analyses of CWAs in tissues and biological effect studies. Samples were taken using bait traps near the wreck and from a reference area known to contain no wrecks. Liver tissue was analysed for oxidative stress biomarkers (including lipid peroxidation, protein carbonylation, glutathione-S-transferase, glutathione reductase and catalase activity) and for histopathological biomarkers, and muscle tissue was analyzed for acetylcholinesterase activity. Chemical analyses were performed from muscle samples and separate whole fish samples, and the results indicated the presence of oxidized forms of CWA-related phenylarsenic compounds in most of the muscle samples. Established biomarker methods used widely in various fish species were shown here for the first time to be applicable also in hagfish. However, only minor differences in the measured biomarker responses between individuals collected from the wreck and the reference area could be observed. Based on this study, the hagfish is regarded as a suitable candidate for ecotoxicological studies of deep marine areas. More information on the biology of hagfish and the natural variability of their biomarkers is needed to distinguish true effects of hazardous substances.

Wildlife ecotoxicology: laboratory dosing studies to field population assessments (P)

MO035

Seabird-derived contaminants and genotoxicity in Collembola from the Arctic

S. Kristiansen, University of Oso / Department of Biosciences; K. Hylland, Department of Biosciences, University of Oslo, Oslo, Norway / Department of Biosciences; H. Leinaas, University of Oslo / Department of Biosciences; G.W. Gabrielsen, Norwegian Polar Institute; D. Herzke, NILU Norwegian Institute for Air Research; K. Borga, Department of Bioscences, University of Oslo / Department of Biosciences

Seabirds occupy high trophic positions and due to biomagnification their bodies contain high concentrations of many anthropogenic contaminants. In the Arctic they have been shown to function as important biovectors of contaminants from ocean to land. The tundra near bird cliffs is heavily influenced by nutritious and contaminant-enriched guano and is generally characterised by rich and diverse vegetation. In these areas, springtails (Collembola) contribute to a high proportion of the soil fauna biomass. They play a vital role in soil ecosystem processes such as decomposition and mineralization. The aim of this study was to determine the exposure, accumulation and effects of seabird-derived contaminants on Collembola. Two Collembola species and their habitat (soil/moss) were sampled at 7 sites with high, medium and low seabird influence in West Spitsbergen, Svalbard, and analysed for a wide range of organic contaminants, mercury (Hg), and stable isotope ratios of carbon and nitrogen ($\delta^{13}C$ and $\delta^{15}N$, respectively). In addition, Collembola were analysed for genotoxic responses, i.e the amount of DNA strand breaks and micronucleus frequency. Seabird influence (indicated by $\delta^{15}N$) and contaminant concentrations were indicated to be higher in soil/moss sampled closer to the bird cliffs (0–150 m) compared to further away (250–400 m) within the same site. When comparing among sites, however, no association between $\delta^{15}N$ and contaminant load was found. The total contaminant loads in habitat samples were dominated by Hg, while no trend was found for organic contaminants. Contaminant concentrations in Collembola were dominated by Hg, followed by hexachlorobenzene (HCB), polychlorinated biphenyls (PCBs) or polybrominated diphenyl ethers (PBDEs), and chlordanes (CHLs). No association was observed between contaminant concentrations in Collembola and habitat. DNA fragmentation was higher in Collembola from sites with high seabird influence, compared to sites with medium and low. No differences in micronucleus frequency (MN) was found between sites or species. This is the first study on MN in Collembola and should be further explored. DNA fragmentation, sensitivity to induced DNA fragmentation and micronucleus frequency were associated with both $\delta^{15}N$ and contaminant levels, increasing with concentrations of lower chlorinated PCBs and CHLs. The sensitivity to induced oxidative stress was negatively correlated with higher chlorinated PCBs (6-7 chlorine substituents).

MO036

Higher contaminants and poorer condition in an Antarctic avian top predator from 2001 to 2013

H.K. Midthaug, Department of Biosciences, University of Oslo / Department of Bioscences; J.O. Bustnes, Norwegian Institute for Nature Research / Fram Centre; A. Polder, Norwegian University of Life Sciences / Department of Food Safety and Infection Biology; K. Borga, Department of Biosciences, University of Oslo / Department of Biosciences

Risks associated with chemical exposure to the Antarctic ecosystem has been considered low. However, recent investigations have shown that south polar skua (*Catharacta maccormicki*) has the highest levels of biomagnifying contaminants among Antarctic seabirds. The present study quantifies OHC levels in south polar skua blood, and evaluate associations between contaminant occurrence, diet, trophic position, biological variables and day of sampling. Furthermore, the study investigates temporal change of organochlorine contaminant (OCs) by comparing with previous data from the same colony, collected during the season of 2001/2002. South polar skuas were sampled during the breeding season of 2013/2014 in Svarthamaren, Dronning Maud Land, Antarctica. Whole blood was analyzed for 87 OHCs of which 56 were detected. Stable isotope ratio of carbon ($d^{13}C$) and nitrogen ($d^{15}N$) in blood, were used to determine carbon source and relative trophic position, respectively. In 2013/2014, predominant contaminants were Mirex (8484 ng/g lw) and Hexachlorobenzene (HCB) (3561 ng/g lw). These levels were higher than those reported from other south polar colonies and Antarctic seabirds at similar ecological niches. Multivariate analysis indicated that skuas sampled late in the breeding season had higher concentration of perfluoralkyl substances (PFASs) and lower relative contribution of polychlorinated biphenyls (PCBs) and polybrominated diphenyl ethers (PBDEs). Due to low intraspecific variance in $d^{13}C$ and $d^{15}N$, no significant associations were found between OHCs and isotopes. However, lack of associations could also be due to influence of migration, wintering habitat and different turnover rates in OHCs and isotopes. Skuas from 2013/2014 had significantly higher concentrations of most OCs and a lower body condition than skuas from 2001/2002. ∑PCB, Mirex and HCB increased with 105%, 40% and 60%, respectively, between 2001/2002 and 2013/2014. Ratios of Mirex/∑PCB and Mirex/HCB decreased between the two seasons, suggesting stabilizing Mirex levels and possibly declining levels in the future. Further studies should elucidate the effects of wintering grounds and diet, as well as the level and contaminant occurrence in Antarctic Petrel (*Thalassoica Antarctica*), the main prey

of south polar skua during the breeding season. Keywords: Antarctic, south polar skua, stable isotopes, temporal variation, OHC

MO037

Evaluation of malformations induced by a hospital effluent of Toluca (Estado de México) in Lithobates catesbeianus

H. Islas-Flores, Universidad Autonoma del Estado de Mexico / Toxicología Ambiental; I. Pérez-Alvarez, Universidad Autonoma del Estado de Mexico / Environmental Toxicology; L. Gómez-Oliván, Autonomous University of the State of Mexico / Chemistry; M. Galar-Martinez, Instituto Politécnico Nacional, Escuela Nacional de Ciencias Biológicas; N. SanJuan-Reyes, Autonomous University of the State of Mexico / Chemistry; O. Dublan-García, University Autonoma del Estado De Mexico / Chemistry; M. Hernández-Navarro, Universidad Autonoma del Estado de Mexico / Toxicology

Hospital effluents are important from the ecotoxicological point of view and are an important source of pollutants emission, among which are pharmaceutical products, chemical residues, radioelements, disinfectants and heavy metals, among others, which are waste of daily activities and has reported that they can reach concentrations between 4 and 150 times higher than those detected in municipal effluents. In addition to this, they usually do not have adequate pre-treatment before their emission, so they can be mixed with other effluents from homes , industries and municipal wastewater, which can subsequently generate interactions, enhance effects and create synergies, which lead to induce adverse effects on the environment. so it is important to study. The Estado de Mexico is located in the center of the country and is considered the entity with the largest population and according to data reported in 2015, it has 1835 medical units of different levels of care. On the other hand, *Lithobates catesbeianus* is a species considered native of the state and has been proposed by the government as an alternative to food supplement (due to its protein content). Due to the aforementioned, the objective of this work was to evaluate the malformations generated by a hospital effluent of Toluca, Estado de México in this species and compare with *Xenopus laevis*, a species that is used as a preferred bioindicator, using the frog embryo teratogenesis assay: *Xenopus* (FETAX). For this purpose oocytes in mid-blastula transition were exposed for 96 h to six different concentrations of the effluent (0.1, 0.3, 0.5, 0.7, 0.9 and 1%), subsequently, the mean lethal concentration (LC50) effective concentration inducing 50% malformation (EC50), and the teratogenic index (TI) was obtained. Results indicates that lower concentrations of the hospital effluent induced slightly higher malformations and lethal effects in *X. laevis* (EC50=0.132%, LC50=0.508%, IT=3.8) and in *L. catesbeianus* (EC50=0.351%, LC50=1.431%, TI=4.0), the main alterations being microcephaly, cardiac and facial edema, malformations in the eye, notochord, tail, fin and intestine. However, the highest TI of *L. catesbeianus* indicates a higher probability that embryos exposed to this hospital effluent will be malformed in the absence of mortality compared to *X. laevis*. and therefore, can be considered as a sensitive and useful species to evaluate toxic effects of contaminants with the FETAX assay.

MO038

Monitoring fish health in a densely populated catchment in Central Germany

M. Schweizer, University of Tuebingen / Animal Physiological Ecology; A. Dieterich, S. Betz, Eberhard Karls Universität Tübingen / Animal Physiological Ecology; N. Corral Morillas, Eberhard Karls Universität Tübingen; C. Dewald, Eberhard Karls Universität Tübingen / Animal Physiological Ecology; D. Leim, Eberhard Karls Universität Tübingen; L. Miksch, S. Nelson, V. Prozmann, J. Rüschhoff, Eberhard Karls Universität Tübingen / Animal Physiological Ecology; R. Triebskorn, University of Tuebingen / Animal Physiological Ecology; H. Köhler, University of Tubingen / Animal Physiological Ecology

In the frame of the joint project *NiddaMan* coordinated by the Goethe University Frankfurt/Main we investigated health parameters of fish from the River Nidda and its tributaries Horloff and Usa in Central Germany. The Nidda river system is regarded as a typical river system for central Europe as it passes a very densely populated area and as it is heavily influenced by anthropogenic factors including agriculture, communal waste water and industrial discharges. To get a broad overview of the situation fish face in this river system and the resulting effects, we investigated biological parameters on different levels: (I) Evaluation of water and sediment samples from the field with the *Danio rerio* embryo test (Dar-T), including the endpoints mortality, hatching success, heart rate, developmental delays and malformations. (II) Investigation of fish health by histopathology of actively (caged rainbow trout) and passively monitored (caught feral) fish focussing on the metabolically most important organ, the liver. (III) Additional inclusion of biomarker data like EROD activity (CYP1A1, indicating pollution with dioxin-like compounds) and number of micronuclei (genotoxicity) within erythrocytes. Results show that the river system – from a biological point of view – is not in a good (as demanded by the EU Water Framework Directive) but rather in a moderate to unsatisfactory condition throughout most of its stretch, whereas upstream areas mainly perform worse than sampling sites downstream. This is noticeable in results obtained by the Dar-T, in particular. However, histopathology of the liver from monitored fish upstream and downstream in general showed vacuolisations, inflammations, haemorrhages in the tissue, and even some necrosis. Our results revealed that, in the case of the Nidda and its tributaries, there is an urgent demand for action to strongly improve the biological integrity of this system.

MO039

Multigenrational toxicity of Fipronil to Folsomia candida

D.d. Oliveira, C.M. Reganhan Coneglian, SCHOOL OF TECHNOLOGY UNICAMP; V.B. Menezes-Oliveira, Universidade de Sao Paulo / Department of Hydraulic and Sanitation

Fipronil is a pesticide widely used on agricultural pest control, especially in sugarcane crops. This compound acts as an inhibitor of nerve signals in insects, and pose as a risk to non-target terrestrial organisms (i.e. the collembolan *Folsomia candida*), which plays important roles in the maintenance of soil quality. The main objective of this study was to evaluate the ecotoxicological effects on the reproduction of three generations of the *Folsomia candida* species when exposed to the pesticide over time, under a natural tropical soil. Test procedures were adapted from the ISO 11267 guideline. The chosen concentrations of fipronil were based on the recommended doses (RD) for the control of the pest *Migdolus fryamus* in sugarcane crops (RD = 1.3 mg of the commercial product / kg^{-1} of dw soil), what means 1.04 mg of fipronil / kg dw soil. Concentrations tested were 0.06; 0.13; 0.26 and 0.52 mg fipronil kg-1 of dry weight soil. The EC_{50} values were 0.21; 0.18 and 0.09 (a.i.) kg^{-1} of soil, for the first, second and third generation, respectively. According to the results, fipronil showed significant toxicity at low concentrations up to the third generation, causing effects on the reproduction and survival of *Folsomia candida*, and could be considered a highly dangerous pesticide for terrestrial arthropod organisms.

MO040

Fipronil effects on freshwater benthic algal communities

J. Val, D. Ballestero, San Jorge University; E. Navarro, CSIC - Spanish National Research Council / Dept. Recursos marins renovables; J. López-Martínez, A.M. Mainar, Universidad de Zaragoza; M. Pino, San Jorge University / Facultad ciencias de la salud

Fipronil is a widely used broad-spectrum phenylpyrazole insecticide, effective against insects resistant to other agents as pyrethroids, organophosphates or carbamates. Accordingly, it has gained popularity worldwide as a pesticide in both agricultural and urban environments. Its wide use, resulted in the presence of fipronil in soil, surface and ground waters involving a risk to the environment and humans. In addition, humans and animals are also exposed to fipronil, by ingestion of products containing residues. Previous studies showed the toxicity of fipronil to aquatic freshwater organisms using model species, but there is scarce information about its impact on wild organisms. This study assesses, for the first time, the impact of fipronil on the photosynthesis of natural freshwater algal benthic community. This community –periphyton- is a key element of aquatic trophic chains, and is routinely used as indicators of water quality. Results show LC_{50} values of 0,74 mg/l (0,63-0,89) (p> 0,001), exposing periphyton to fipronil under standard medium. However, toxicity was almost inexistent when assays were done using natural river water. In this last case, the bioavailability of the fipronil was hypothetized to be reduced by natural substances present in the river water (solid suspended solids, organic matter, etc..). These results would contribute to a more realistic assessment of the environmental impacts of the use of this kind of pesticides.

MO041

Use of organophosphorus insecticides in agriculture lands, in a simple test birds says please no!

A.N. BUJDUD LEÓN, Universidad Juarez del Estado de Durango / Facultad de Medicina Veterinaria y Zootecnia; M. Pereda Solis, Universidad Juarez del Estado de Durango / FMVZ; J.H. Martinez-Guerrero, Universidad Juarez del Estado de Durango / Facultad de Medicina Veterinaria y Zootecnia; M. GUERRERO CERVANTES, Universidad Juarez del Estado de Durango / FMVZ

Due to the human population increase and the consequent high demand for food, each day a larger area of the planet is dedicated to the practice of agriculture. Crops favor the reproduction of various organisms (invertebrates and vertebrates) that are combated with large amounts of pesticides, it are chemical compounds used extensively, and so all organisms are exposed from different sources such as food, water and soil, therefore the toxicity of agrochemicals, as well as the patterns of use and misuse that is made of these compounds generates risks for the survival of the biodiversity of agroecosystems. The main pesticides used in agriculture are organophosphates (OP), they are highly toxic. In birds, the main route of intoxication to OP is through the consumption of contaminated food, although it can also occur by inhalation when flying over or inhabiting the crop fields and its surroundings. The OP affects the nervous system by inhibiting the function of cholinesterase, which is responsible for transforming the neurotransmitter acetylcholine into choline and acetate. A decreased level of cholinesterase (ChE) activity in the animal tissue is a strongly indicative sign that some type of exposure to an inhibitory agent of this enzyme has occurred. Our objectives were to document ChE levels in house sparrows in response to their exposure to an OP product used in agriculture and determine the extent of their negative effects on birds. In the University Campus (UJED, Durango, Mexico), we collected 19 house sparrows and serum ChE activity was determined by spectrophotometry before and after the consumption of food treated with malathion. The results show a ChE inhibition (11.58 %) after treatment (p = 0.03), also we observed nonsignificant

relationships (Pearson, R2=0.11) between the ChE and the weight or sex of the birds. The weight of the birds decreased on average 1.54 g after the exposure, possibly due to the stress of capture and the lack of adaptation to captivity. The house sparrow showed to be a species sensitive to the presence of OP compounds in the environment. It is possible that the house sparrow and other birds that co-inhabit the agricultural areas where malathion is applied, as well as other OP insecticides, are prone to develop different levels of intoxication and that in some cases their health condition is compromised.

MO042

Implementation of a worst-case landscape scenario for population modelling of a fungicide applied in cereals

M. Wang, WSC Scientific GmbH / Dept Efate Modelling; T. Preuss, Bayer Ag / Environmental Safety; M. Ebeling, Bayer AG Crop Science Division / Ecotoxicology - Terrestrial Vertebrates Expert Team

In many cases EU member states prefer the use of country specific scenarios for the exposure or risk assessment of pesticides. This is sometimes requested due to specific geographical features or agricultural practice. In the present example, we demonstrate how the specific agricultural situation of the Netherlands was taken into account for the selection of landscape scenarios for use in population modelling. It is first shown how a country specific landscape scenario is developed. Then, the dose response obtained in a rat reproduction study with an azole fungicide is employed in a population-level risk assessment on small herbivorous mammals (Common vole, *Microtus arvalis*). The margins of safety obtained in that assessment indicate that no adverse population-level effects would be expected under realistic worst-case field conditions.

MO043

Biomonitoring and validation of non-invasive samples for the analysis of metals in freshwater turtles from mining areas

J. Pareja Carrera, IREC-UCLM / IREC-UCLM; A. Rodríguez-Perez, UCLM-IREC; M. Martinez-Haro, IREC-Instituto de Investigación en Recursos Cinegéticos / Department of Life Sciences; R. Mateo, IREC-CSIC- UCLM / Grupo de Toxicología de Fauna Silvestre; M. Ortiz Santaliestra, Institute for Game and Wildlife Research (IREC) UCLM-CSIC-JCCM

The mobilization of metals present in the earth's crust due to anthropogenic activities, such as mining, is one of the main contamination routes for wildlife. With the objective of biomonitoring and validating non-invasive methods for metal analysis in reptiles, levels of lead (Pb) and mercury (Hg) in blood, faeces and carapace scales of freshwater turtles (*Mauremys leprosa*, n=86) from historical mining areas (i.e. Valle de Alcudia-Sierra Madrona district for Pb, Almadén district for Hg) in Central Spain were analysed. The highest levels of blood Pb were found in the animals collected from the area of Valle de Alcudia-Sierra Madrona, more specifically from Solana del Pino, with an average (±SD) of 5.59±3.66 µg/g dry weight (dw). Individuals from this location showed the clearest evidences of oxidative stress, as estimated from the highest values of malondialdehyde (biomarker of lipid peroxidation) and superoxide dismutase activity. However, no correlations were found between these biomarkers and blood Pb levels. Similarly, the highest levels of Hg were detected in the animals collected from the area of Almadén, specifically from Almadenejos, with an average (±SD) of 8.83±8.84 µg/g dw. These individuals showed no evidences of oxidative stress, but presented increased activity of glutathione peroxidase and reduced glutathione levels relative to the rest of populations, which would indicate that antioxidant system is preventing from Hg-derived oxidative damage to occur. Blood levels of these two elements were above those reported as susceptible to cause sub-lethal effects in reptiles for the vast majority of terrapins from the most contaminated sites (100% of terrapins from Solana del Pino with blood Pb levels > 15 µg/dl; 70.3% from Almadenejos with blood Hg levels > 2.76 µg/g dw). Faeces and carapace scales obtained non-invasively correlated significantly with blood levels for the case of Pb ($R \geq 0.705$, $P < 0.001$), but not for Hg ($R \leq 0.362$, $P \geq 0.127$). Thus, these samples could be used as non-invasive methods for the analysis of Pb bioavailability in *M. leprosa*, and by extension in reptiles, which will contribute to the development of ecotoxicology in reptiles, a group very little studied in this regard.

MO044

An analysis of important life stages, exposure routes and test endpoints in amphibians and coverage by existing risk assessment regulatory requirements for plant protection products, part 1

A. Aldrich, Agroscope / Ecotoxicology; C. Berg, Uppsala universitet, Dept. of Environmental Toxicology / Dept of Environmental Toxicology; M. Ortiz Santaliestra, Institute for Game and Wildlife Research (IREC) UCLM-CSIC-JCCM; S. Pieper, German Federal Environment Agency (UBA) / Plant Protection Products; S.M. Weir, Queens University of Charlotte / Department of Biology

Concerns have been raised that the current risk assessment of plant protection products (PPP) may not sufficiently cover the risk to amphibians and reptiles. To address these concerns, the European Food Safety Authority (EFSA) has published a Scientific Opinion addressing the state of the science regarding the risk to amphibians and reptiles exposed to pesticides (EFSA 2017). A central task of the EFSA working group was to understand how well important life stages, exposure routes and endpoints are currently covered (or not) by the current risk assessment paradigm. We had four steps in this analysis: 1) Identify relevant life-stages, important exposure routes and possible endpoints with relevance to the specific protection goals (SPGs) defined for amphibians; 2) Evaluate the coverage by existing standard test guidelines with regard to these important stages/routes/effects; 3) Explore whether tests with surrogate species could provide suitable information allowing for extrapolation to assess the hazard/risk for amphibians; 4) Identify proposed non-standardized test protocols suitable to address the gaps for specific protection goals defined for amphibians. Care was taken to address all relevant routes of exposure, life-stages, and short and long term effects. For amphibians, greater coverage or surrogacy exists for the aquatic larval stage and short term effects, with less coverage of the adult terrestrial stage, reproductive toxicity, and specifically for the potential effects via dermal exposure routes. To cover important life stages, exposure routes and effects, tests addressing dermal overspray and reproductive toxicity in amphibians may be needed. The analysis for reptiles will be presented in a separate poster. The concerns that the current risk assessment of pesticides may not sufficiently cover the risk to amphibians were supported after the analysis of the currently available data. The exercise provides a useful base for further research necessary to advance the ecotoxicological risk assessment of amphibians within the remit of the PPP authorization.

MO045

European common frog (Rana temporaria) larvae show subcellular responses under field-relevant Bacillus thuringiensis var. israelensis (Bti) exposure levels used in mosquito control

S. Allgeier, B. Frombold, University Koblenz-Landau; V. Mingo, Trier University / Biogeography; C. Bruehl, University of Koblenz-Landau / Institute for Environmental Sciences

Bacillus thuringiensis var. *israelensis* (Bti) is presumed to be an environmental friendly agent for use in either health-related mosquito control or the reduction of nuisance associated with mosquitoes coming from temporary flooded wetlands. Amphibians co-occurring with mosquito larvae in these wetlands may be exposed to Bti products several times during their breeding season. Up until now, information regarding effects on the non-targeted group of amphibians has to be considered rather inconsistent. On this account, we evaluated how repeated exposures to frequently used Bti formulations (VectoBac®12AS, VectoBac®WG) in field-relevant rates affect European common frog (*Rana temporaria*) larvae. In a laboratory approach, tadpoles experienced exposure conditions similar to realistic mosquito control in the Upper Rhine Valley (Germany). We assessed potential effects with regard to enzymatic biomarkers (glutathione-S-transferase, glutathione reductase, acetylcholine esterase), development, body condition and survival until the end of metamorphosis. Regardless of the formulation, delivery form or application rate, tadpole survival rates and time to metamorphosis were slightly reduced after repeated Bti exposures, while body condition was similar throughout the treatments. Furthermore, Bti induced significant increases of all enzymatic activities irrespectively of the applied field rate and formulation, indicating oxidative stress as well as unspecific neurotoxic effects. Repeatedly executed Bti applications, especially acting on early developmental stages, seem to increase the risk for adverse effects. The examination of several biochemical markers is needed to evaluate the ecotoxicological risk of Bti for amphibian populations, especially in the context of worldwide amphibian declines. Following the precautionary principle, the implementation of certain thresholds for application numbers and intervals should be considered in order to ensure environmentally friendly mosquito control programs, especially in areas originally designated for nature conservation.

MO046

Influence of salinity and temperature on tadpoles of Xenopus laevis

C. Monteiro, R. Alves, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro; A.M. Soares, University of Aveiro / department of Biology & CESAM; I. Lopes, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro

Worldwide coastal low-lying ecosystems are experiencing an increased salinity due to seawater intrusion. Considering the projections of the International Panel for Climate Changes regarding the increase of mean temperatures, until 2100, and consequent sea level rise, it is foreseen an increase in the number of costal ecosystems suffering from such salinization. Among coastal ecosystems that will be impacted with seawater intrusion are wetlands, which constitute hotspots of biodiversity and represent relevant ecosystems for amphibians. This class of vertebrates holds the highest proportion of endangered species and is considered very vulnerable to salinity changes. In this context, the present study aimed at evaluating the influence of temperature on the adverse effects that increase of salinity may cause to tadpoles of the amphibian species *Xenopus laevis*. To address this objective, *X. laevis* tadpoles (Gosner 25) were exposed to a range of 5 NaCl concentrations under three temperatures: 20, 23 and 26ºC. The following parameters were evaluated at the end of the test: feeding rate, body weight and growth rates. The differences reported for size between control and NaCl concentrations were mainly due to the tail length. For all concentrations, the feeding rate decreased with increasing NaCl concentration. As well, body weight decreased

with increasing salinity. Although significant interactions between temperature and salt concentration were registered, a pattern of influence in the temperature was not observed. Adverse effects were observed at the lowest tested salinity levels, which suggest that these species are highly vulnerable to small salinity increases and would be at high risk under seawater intrusion scenarios. Keywords: Salinity; toxicity; amphibia

MO047

EFFECTS OF THE EXPOSURE OF LARVAS OF Dendropsophus columbianus (ANURA: HYLIDAE) TO WATERS CONTAMINATED BY ANTHROPOGENIC ACTIVITIES IN A RIVER BASIN OF THE COLOMBIAN ANDES

V. Ramírez Castaño, Universidad de Caldas; B. Toro, Universidad de Caldas / Biological Sciences

The pollution generated by agriculture, livestock and mining have impacted the watersheds in the Colombian Andes. Amphibians have been used to evaluate this contamination due their biphasic lifecycle, which has made them ideal models in aquatic ecotoxicology. The objectives of this work were: 1) to determine if tadpoles of *D. columbianus* exposed to contaminants of agricultural, livestock and mining (with mercury: Hg, and with mercury and cyanide: Hg/CN) varied in the snout-vent length (SVL), tail length (TL), head width (HW), and body weight, and 2) to evaluate the effect of exposure on metamorphosis and behavior of the larvae. The AMPHITOX protocol was followed using ten larvae in each of the treatments and in the control, which were exposed from the moment of hatching to complete metamorphosis. Significant differences were found in the LRC between the larvae of the control and the Hg/CN mining treatment (Z= -28.92, p= 0.000) and between Hg/CN mining and agriculture treatments (Z= 25.325, p= 0.001) after 50 days of exposure. Differences in LC were found between the larvae of the control and the Hg/CN mining treatment (Z= -25.57, p= 0.001), and between Hg/CN mining and Hg mining treatments (Z= 21.525, p= 0.009) in the same time. The weight did not show differences. The majority of larvae of the control and the agriculture treatment showed similar development rates, reaching stage 46 between days 60 and 75 of exposure. While tadpoles exposed to Hg/CN mining did not complete the metamorphosis and reached stage 42 between days 50 and 55. Surface flotation was the activity that was most stable at the time of exposure, presenting percentages of individuals greater than 60% in the control (81%), and in the agriculture (70%) and Hg mining treatments (65%), between the last days of exposure evaluated: 22 to 28. It is shown that 1) the effects on growth of anuran larvae due to agricultural, livestock and mining contamination are similar. However, samples contaminated by mining produce smaller tadpoles than agriculture and livestock, and 2) tadpoles exposed to agricultural and livestock contamination, unlike other studies which report long times of metamorphosis, show a time of this process that approximates to the time that the species experimented in the control (134 days) and probably, to the time of this *in situ*.

MO048

Risks for amphibians and reptiles by dermal exposure to pesticides

F. Streissl, EFSA / Pesticides Unit; P.I. Adriaanse, Alterra Wageningen University and Research Centre; R. Sharp, EFSA - European Food Safety Authority / Pesticides Unit; K. Machera, Benaki Phytopathological Institute / Department of Pesticides Control & Phytopharmacy, Athens, Greece

Amphibian and reptilian species are found in agricultural landscapes. Some inhabit water bodies either permanently or during some time of their life cycle. Others use predominantly the terrestrial habitat. Some of those species migrate long distances through fields and some reside in fields and field margins where they can be exposed to pesticide residues in food items, water, soil and plant surfaces. A number of studies indicate that pesticide exposure can lead to severe impacts with up to 100% mortality from overspray at field rates for some pesticides. Dermal exposure was identified as an important exposure route in the EFSA opinion. The aim of the current study was to investigate different options to address the risk from dermal exposure by overspray and contact to soil and plant surfaces. Existing exposure models were reviewed with regard to their suitability for amphibian and reptilian risk assessment. This included comparison of parameters used in model calculations, comparison of sensitivity to dermal exposure and worst case exposure calculations to identify groups of amphibians and reptiles with greatest dermal exposure from overspray and to compare it with exposure from contact to soil and plant surfaces. In addition a novel approach was developed to estimate the uptake from soil and plant surfaces. Mammalian dermal toxicity related to local effects and dermal adsorption data may be used as surrogates for reptiles but not for amphibians. The development of a test method investigating local effects on amphibian skin, as well as the estimation or measurement of body burden following exposure via the dermal route is needed. Comparison of body burden by overspray to body burden by exposure to treated soil or foliage showed that the maximal body burden by overspray was lower than the maximal body burden by passive or active dermal uptake from soil or by contact to foliage. An approach was suggested which can combine oral and all dermal exposure routes in one overall body burden which could form the basis for a realistic risk assessment.

MO049

Evaluating the Role of Fish as Surrogates for Amphibians in Ecological Risk Assessment

S. Glaberman, University of South Alabama / Biology; J. Kiwiet, University of South Alabama; C. Aubee, US Environmental Protection Agency / Risk Assessment Division Office of Pollution Prevention and Toxics

Ecological risk of chemical exposure to aquatic-phase amphibians is historically evaluated using surrogate toxicity data from standard fish species. Recently published meta-analyses of fish and amphibian ecotoxicity data concluded that both groups are similarly sensitive to a range of chemicals. However, these analyses are limited because the amphibian data reported in the peer-reviewed literature are variable both with respect to experimental design and test species. In 2010, the U.S. Environmental Protection Agency began receiving ecotoxicity data for a standard amphibian test species (*Xenopus laevis*) as part of the Endocrine Disruptor Screening Program. Although these studies are primarily designed to inform a determination of potential thyroid interaction within the context of other endocrine screening studies, they also contain valuable data on survival and growth that can be compared to existing fish data for a given chemical. We used this dataset to compare no observed adverse effect concentration (NOAEC) values for survival, body weight, and length data between fish and amphibians for 45 different pesticide active ingredients. Overall, the results indicate that fish are a reasonably good predictor of amphibian toxicity as there were no statistically significant differences in NOAEC values between the two groups for the endpoints examined. However, toxicity endpoints were lower in amphibians as compared to fish approximately half the time across chemicals, challenging the notion that fish are consistently more sensitive than amphibians. Disclaimer: *Disclaimer: The views expressed in this presentation do not necessarily represent the views of the U.S. EPA or the United States.*

MO050

Long-term survival of mancozeb exposed common vole populations from one to the following reproductive season

F. Von Blanckenhagen, J. Ludwigs, Rifcon GmbH

Rodent field effect studies relevant for pesticide risk assessment typically take place during crop development from spring to autumn and thus within a single reproductive season. However, animal survival covering multiple reproductive seasons has only rarely been considered. This includes also microtine rodents such as the common vole (*Microtus arvalis*), as they are considered as rather short-lived vertebrates. However, overwintering individuals are important for the survival of local populations, and potential pesticide exposure during maturation might impact survival during seasonal bottlenecks in winter from one to the next reproductive season. Furthermore, from a regulatory perspective and due to the one-reproductive-season-duration of most field effect studies in wildlife, there is an increasing concern on potential long-term effects from exposure occurring delayed or that the reproduction in the following season might be affected by exposure during a previous application season. Against this background we monitored individually marked common vole populations from a long term effect study on spray applications of Dithane M-45 (Mancozeb 80% WP) during one reproductive season further on into the following reproductive season. The test item Dithane M-45 was applied four times in June according to Good Agricultural Practice at an application rate of 2 kg a.s./ha. Trapping and marking of voles in the same investigation plots was conducted until September, followed by further trapping until spring of the following year and the onset of the new reproductive cycle. Reproductive parameters recorded as indicators of potential long-term effects resulted in very similar patterns in treatment and control plots, and the data show no indication that common voles were negatively affected by multiple applications of the test item also in the following year. Furthermore and up to now not documented for wild living common voles, the data gives evidence that free-living common vole individuals can reproduce for more than one reproductive season.

MO051

An analysis of important life stages, exposure routes and test endpoints in reptiles with regard to coverage by existing risk assessment regulatory requirements for pesticides

A. Aldrich, Agroscope / Ecotoxicology; C. Berg, Uppsala universitet, Dept. of Environmental Toxicology / Dept of Environmental Toxicology; M. Ortiz Santaliestra, Institute for Game and Wildlife Research (IREC) UCLM-CSIC-JCCM; S. Pieper, German Federal Environment Agency (UBA) / Plant Protection Products; S.M. Weir, Queens University of Charlotte / Biology

Concerns have been raised that the current risk assessment of pesticides may not sufficiently cover the risk to amphibians and reptiles. To address these concerns, the European Food Safety Authority (EFSA) has published a Scientific Opinion addressing the state of the science regarding the risk to amphibians and reptiles exposed to pesticides (EFSA 2017). A central task of the EFSA working group was to understand how well important life stages, exposure routes and endpoints are currently covered (or not) by the current risk assessment paradigm. We had four steps in this analysis: 1) Identify relevant life-stages, important exposure routes and possible endpoints with relevance to the specific protection goals (SPGs) defined for these groups; 2) Evaluate the coverage by existing standard test guidelines with regard to these important stages/routes/effects; 3) Explore whether tests with surrogate species could provide suitable information allowing for extrapolation to assess the hazard/risk for amphibians and reptiles; 4) Identify proposed non-standardized test protocols suitable to address the gaps for specific protection goals defined for amphibians and reptiles. Care was taken to address all relevant routes of exposure, life-stages, and short and long term effects. Reptiles have very little data with which to assess the coverage of important life stages and exposure routes. Current knowledge suggests that surrogacy based on bird or mammal data may not be appropriate for the juvenile and adult stages of reptiles, though data are very limited. Reptile eggs are not covered by any aspect of the current risk assessment paradigm, but it is unknown to what extent reptiles lay eggs in crop fields, and how likely exposures are to occur to eggs under realistic scenarios. Therefore, more data are needed to determine if this is a vulnerable life stage that needs specific consideration. The concerns that the current risk assessment of pesticides may not sufficiently cover the risk to reptiles were supported after the analysis of the currently available data. The exercise provides a useful base for further research necessary to advance the ecotoxicological risk assessment of reptiles within the remit of the pesticide authorization.

MO052

AmphiMove: Moving patterns and microhabitat selection of European anurans in agricultural landscapes

J. Sadowski, Julius Kuehn Institute / Institute for Plant Protection in Horticulture and Forests, Vertebrate Research; A. Esther, Julius Kuehn-Institute - Federal Research Centre for Cultivated Plants / Institute for Plant Protection in Horticulture and Forests, Vertebrate Research

The current decline of amphibian populations on global and local scales is discussed by scientists around the world. Studies suggest that enhanced application of plant protection products (PPP) is one of the main reasons that today amphibians represent the most endangered vertebrate group in Europe. The first version of a scientific opinion currently released by EFSA (European Food Safety Authority) highlights the need for including amphibians in the risk assessment of plant protection products. Specifics in terms of physiology (e.g., permeable skin) and ecology (aquatic and terrestrial life-stages in combination to complex moving patterns and habitat preferences) make this taxon in particular vulnerable to pesticide applications in agricultural landscapes. It is emphasized that detailed ecological data of especially terrestrial amphibians is still under-represented but required for a comprehensive risk evaluation of all amphibian life-stages. The aim of the project AmphiMove is to fill the data gap on terrestrial life-phases of European anurans with focus on moving patterns and microhabitat selection in agricultural landscapes. At two study sites individuals of common toads (*Bufo bufo*) and common frogs (*Rana temporaria*) were caught at and around their breeding ponds, fit with a transmitter and afterwards tracked via radio-telemetry. Locations, biotic and abiotic parameters of the selected microhabitats were recorded daily. We show preliminary results of the first period of data collection for common toads from March to October 2017.

MO053

A quantitative AOP for activation of the aryl hydrocarbon receptor leading to early life stage mortality in amphibians and reptiles

A. Schroeder, University of Minnesota-Crookston / Math, Science and Technology; J.A. Doering, US EPA / Mid Continent Ecology Division; M. Brinkmann, University of Saskatchewan / School of Environment and Sustainability and Toxicology Centre; M. Lucio, S. Stoeck, University of Minnesota Crookston / Math, Science, and Technology Department; Z. Currie, University of Saskatchewan Toxicology Centre / Toxicology Centre; M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre

Differences in sensitivities to chemicals among species and taxa is a major challenge for accurate ecological risk assessments. Most toxicity information is collected for a few model species and little is known about the relationship between the sensitivity of the model species compare to non-model species. Quantitative adverse outcome pathways (qAOPs) are quantitative, biologically-based models which describe key event relationships that link a molecular initiating event to an adverse outcome. qAOPs can serve as a useful tool to determine the relationship between the sensitivity of chemicals with a molecular initiating event and an adverse outcome among species. Previously, a qAOP had been described for the indirect relationship between activation of the aryl hydrocarbon receptor (AHR) by dioxin-like compounds (DLCs) and embryo-mortality in birds and fishes. It was hypothesized that this qAOP was also applicable to amphibians and reptiles. However, little is known about whether the sensitivity to activation of AHR is predictive of sensitivity to DLCs of embryos of any amphibians or reptiles. Therefore, in order to test the hypothesis of applicability to amphibians and reptiles, this study investigated sensitivities to activation of AHRs in an in vitro transactivation assay to in vivo embryo sensitivities for an amphibian, the African clawed frog (*Xenopus laevis)*, and a reptile, the common snapping turtle (*Chelydra serpentina*). Embryo-mortality was assessed in African clawed frog embryos exposed to serial concentrations of one of two DLCs: 2,3,7,8-tetrachlorodibenzofuran (TCDF) or 2,3,4,7,8-pentachlorodibenzofuran (PeCDF). Embryo-mortality was assessed in common snapping turtle embryos exposed to serial concentrations of one of four DLCs: TCDF, PeCDF, 3,3',4,4',5-pentachlorobiphenyl (PCB 126), or 2,3,7,8-tetracholordibenzo-p-dioxin (TCDD). Further, in vitro AHR transactivation assays were used to determine sensitivity to activation of the AHR1 isoform of African clawed frog and common snapping turtle to these selected DLCs. It is anticipated that this research will result in a single qAOP linking in vitro activation of the AHR to embryo-mortality with taxonomic applicability across phylogenetically diverse oviparous vertebrates, including birds, reptiles, amphibians, and fishes. This qAOP could guide more objective ecological risk assessments of DLCs to diverse taxa which are not easily studied, such as native species of reptiles and amphibians.

MO054

Do historically metal-exposed amphibian populations acquire resistance to lethal levels?

E. Fasola, University of Coimbra; M. Ortiz Santaliestra, Institute for Game and Wildlife Research (IREC) UCLM-CSIC-JCCM; J. Pareja Carrera, IREC-UCLM / IREC-UCLM; M. Martinez-Haro, Institute of Game and Wildlife Research (IREC) UCLM-CSIC-JCCM; R. Ribeiro, Universidade de Coimbra / Life Sciences; R. Mateo, IREC-CSIC- UCLM / Grupo de Toxicología de Fauna Silvestre; I. Lopes, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro

The aims of this work were to 1) determine oxidative stress and metal burden in anuran tadpoles from historically metal exposed populations (Hg/Pb), and 2) assess if tadpoles from impacted sites have increased tolerance to metals relative to tadpoles from reference sites. Metal body burden, oxidative stress biomarkers and metallothioneins (MT) were measured in *Pelophylax perezi* tadpoles from reference and metal contaminated sites. Additional tadpoles (20 per site) were collected and exposed in lab conditions during 24h to Hg or Pb levels above the median lethal concentrations reported for amphibians (1.5 and 10.5 mg/l, respectively). The parameters mentioned above plus mortality were monitored at the end of the assay. Field-collected tadpoles from Pb and Hg polluted sites had higher metal body burden than those from reference sites (median per site as d.w. 540.4-708.1 vs 2.6-9.5 ng Pb/g, 768.2-31035.3 vs 0.01 ng Hg/g; all $p < 0.01$). Levels of MT (median, µg/g tissue) were significantly higher in tadpoles from Hg polluted sites than in the rest of locations (248.5-307.7 vs. 63.9-138.6; $p < 0.01$), suggesting that MT levels can be induced, in natural populations, by the sum of environmental factors. Exposure to Hg caused mortality of all individuals, while Pb did not result lethal to tadpoles. Laboratory exposure revealed that experimental treatment rather than pollution at the origin site determined Pb body burden (controls: from reference site 96.7-120.4 ng/g, from Pb site 118-491.6 ng/g; Pb-exposed: from reference site 36979.9-54760.4 ng/g, from Pb site: 9043.5-78452.4 ng/g), showing that Pb was readily bioavailable for exposed tadpoles. Lab exposure to Pb increased MT levels in tadpoles from reference sites (exposed vs. non-exposed: 116.13 vs. 41.70 µg/g; $p < 0.01$), but not in those from Pb-polluted areas. Oxidative stress biomarkers did not differ either between origin sites or because of experimental exposure to Pb. MT levels in tadpoles from reference populations that were taken to the laboratory and kept as controls decreased during their permanency in the laboratory (105.99-138.66 vs 29.72-41.70 µg/g; $p < 0.05$). This could be consequence of a reduction in the laboratory of stress sources other than metals that can also induce MT synthesis (e.g. thermal stress). The fact that this decrease was not observed in tadpoles from Pb-polluted sites (105.61-109.41 vs 193.50-130.23 µg/g; $p < 0.05$) would suggest that these animals may have high constitutive MT levels.

MO055

Assessment of metal contamination levels and stress responses of endangered sea turtles of São Tomé and Principe

I.F. Morão, S.C. Novais, Polytechnic Institute of Leiria / MARE IPLeiria; I. Vieira, ATM - Associação para o Estudo, Proteção e Conservação das Tartarugas Marinhas nos Países de Língua Portuguesa; C. Barata, CSIC / Environmental Chemistry; B. Pina, IDAEA-CSIC / Department of Environmental Chemistry; M.F. Lemos, Instituto Politécnico de Leiria / MARE IPLeiria

São Tomé Island harbors important sea turtle nesting and feeding sites. However, insufficient enforcement of environmental laws to avoid illegal take of nesting females and eggs, associated with a great lack of knowledge about how these species interact with their environment and how human activities impact their survival in the region, constitute significant challenges for sea turtle conservation. Through current local conservation projects, some information on genetics and nutrition of sea turtle populations is being unveiled but very little is known about how ocean pollution is impacting these turtles. The main objective of this study was to assess the metal concentrations accumulated by two species of S. Tomé sea turtles (*Eretmochelys imbricata* and *Chelonia mydas*) and infer about possible impacts of such contamination on their general stress responses and health status. More specifically, the final goal was to find correlations between metals in their tissues and the expression of key genes involved in detoxification/sequestration and metal transport, antioxidant responses and oxidative stress, immunological responses, mitochondrial respiratory and energy production, among others, which could be indicative of these organisms health and future viability. To achieve these goals, nesting female turtles were sampled for blood and skin tissues, immediately after egg laying in their well-documented spawning sites in S. Tomé. Skin samples were collected from the right front flipper of the turtles and stored at -20ºC until analysis of metal concentrations. Blood samples were withdrawn from the external jugular vein and stored in RNAlater at -20°C until RNA extraction and gene expression analysis using quantitative real-time PCR (qPCR). Additionally, body mass and carapace length were also recorded, along with all information regarding egg spawning and its success. Results showed significant correlations between expression of some genes and metal contaminant levels, pinpointing some candidate genes to be used as biomarkers of interest for biomonitoring campaigns, which worrying function highlights the need for a close follow-up of these organisms. This study represents the first attempt to address pollutant levels and the biological impairments of such stressors in these turtle species nesting in S. Tomé which, given their classification as endangered species (IUCN red list), is of paramount importance to contribute for conservation measures and management.

MO056

Ecotoxicology of Africa's three largest reptiles: POPs, metals, eggs, and eggshells

H. Bouwman, North-West University / Unit for Environmental Science and Management; R. Nel, Nelson Mandela Metropolitan University / Department of Zoology; H. Kylin, Linköping University / Department of Thematic Studies Environmental Change; D. Govender, SANParks; M. Du Preez, North West University / Zoology

The Nile Crocodile (*Crocodylus niloticus*), Loggerhead Turtle (*Caretta caretta*) and Leatherback Turtle (*Dermochelys coriacea*) are the largest reptiles in Africa. The bioaccumulation and effects of metals and metalloids on large-bodied reptiles are less well known compared with birds and mammals, especially those from Sub-Saharan Africa. Globally, reptiles are experiencing declines, and pollution is one of the hypothesized reasons for the decline. The Nile Crocodile and Loggerhead Turtle are at relatively high trophic levels, with the Nile Crocodile also being the largest predator in Africa. We sampled eggs from these three species (27 crocodile, and 10 each from the two turtle species) and analysed the shells and contents seperately for metallic elments using ICP-MS. Trophic level, body size, and migratory patterns influenced the concentrations in shells and egg contents, but crocodiles generally seem to have lower concentrations than the sea turtles. Compared with data from elsewhere, sea turtle eggs had lower concentrations, but crocodile eggs had relatively higher copper and mercury concentrations. Comparisons between eggshells and egg content elemental compositions for each species clearly showed that eggshells can not be used as proxy for egg contents. Sampling therefore, requires the collection and analyses of unhatched eggs. Relative elemental composition patterns indicated overlaps for the respective egg contents and eggshells of the sea turtles, but not for the crocodiles. We found thicker eggshells significantly associated with higher iron concentrations in the crocodiles. The implications may be that hatchlings may spend more energy to break through the leathery shells, and may therefore affect reproduction. Copper had concentrations that raised concern in all three species. The strontium concentration in the eggshells of the Leatherback Turtle was high. Mercury, copper, strontium, and selenium should be monitored in all species. Eggs should also be analysed for other co-occuring pollutants, such as POPs and endocrine disrupting compounds, since sub-lethal effects, especially when the eggs are covered, is difficult to discern. Based on the work presented here and those of others, it is obvious that more studies are needed to obtain a better picture of the chemical and biological interactions involved with Africa’s three largest reptiles \n.

MO057

Improving knowledge flow: from consumer to environmental risk assessment

L. Villamar Bouza, s. barmaz, R. Sharp, EFSA - European Food Safety Authority / Pesticides Unit; M. Arena, EFSA - European Food Safety Authority / Pesticides; D. Auteri, EFSA - European Food Safety Authority / Pesticides Unit

The assessment of pesticide residues levels in environmental matrices is part of the risk assessment for non-target organisms under Regulation (EC) no 1107/2009. In the case of risk assessments for birds and mammals, according to EFSA (2009), the Tier 1 risk assessment uses default values for residues levels (in terms of residue per unit dose, RUD) and residue decline (in terms of a time weighted average factor, TWA). When the tier 1 risk assessment indicates a high risk a higher tier assessment is required. One option is to refine the estimate for the level of residues that wildlife consume through their diet by calculating specific RUDs or by deriving substance/crop specific DT_{50} values which are then used to recalculate the TWA factor used in the risk assessment. Often the residues studies submitted in the context of the consumer risk assessment are used to derive such DT_{50} values. These are then further evaluated with specific kinetic tools (FOCUS kinetics). It should be noted that the refinement of the RUD values is done only in rare cases since the dataset at the basis of the default values is relatively large. These refinements allows for a more realistic assessment accounting for the differences in residues decline due to the crop type, growth stage, climatic conditions across EU zones and to specific characteristics of the substance under assessment. Other parts of the data used for the consumer risk assessment for pesticides can also provide information for the environmental risk assessment. In particular, metabolism studies in plants are used for the identification of the pertinent metabolites to be further considered in the risk assessment of birds and mammals. The metabolism data for hen and rat/goat can also be used for addressing such metabolites. The main scope of this work is to further analyse the standard dataset available and the specific guidance in use for the consumer risk assessment in order to better define how the data and knowledge developed in the context of the consumer risk assessments (internationally agreed methodologies, existing guidance documents) could be integrated in the environmental risk assessment. Particular consideration is given to the possibility of extrapolation between crops, use patterns (e.g. growth stages, application number) and European zones.

MO058

Increasing salinisation effects on Pelophylax perezi populations - Could historical exposure drive effects?

S. Costa, Universidade de Aveiro / Department of Biology & CESAM, 3810-193 Aveiro; I. Lopes, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro

Coastal wetlands are a priority for conservation because they are biodiversity hotspots and have high ecological importance in ecosystem services. However, they are increasingly exposed to strong anthropogenic pressure and climatic extremes. Additionally, Intergovernmental Panel on Climate Change (IPCC) reported alarming projections for sea levels rise in the AR5 at 2013. Within this scenario, it is foreseen the salinisation of low-lying coastal freshwater ecosystems due to the intrusion of seawater, which will adversely affect many populations of amphibians. *Pelophylax perezi* is distributed along all coastal territory in Portugal, where there are some populations historically exposed to increasing levels of seawater. Though this species has been reported as tolerant to high salinity levels, there is a lack of knowledge regarding long term effects of salinization to natural populations of this amphibian species. The present work aimed at characterizing the lethal and sublethal sensitivity to salinization of early life stages of *P. perezi* originated from reference and salinized natural populations. Embryos (Gosner state 8-10) were exposed for 96h, and tadpoles for 72h to 168h (test dependent) to several dilutions of seawater and concentrations of NaCl (used as a surrogate of seawater to increase salinity). The following endpoints were monitored: time until hatching, growth and feeding. Comparing the results for time to hatch between salinization-impacted and non-impacted populations, differences were not found. For this endpoint, NaCl revealed to be more toxic than seawater (EC_{50} of 14.04 and EC_{50}=11.89 $mScm^{-1}$ for seawater and NaCl, respectively). As well, for the other monitored endpoints (tadpole growth, weight and feeding) NaCl caused significant adverse effects. Highly diluted seawater increase growth on tadpoles from salinization-impacted population, but for non-impacted populations, tadpoles’ growth decrease with the decrease of seawater dilutions.

MO059

Wildfires effects on aquatic invertebrates organisms with in situ bioassays

N. Abrantes, University of Aveiro / CESAM/DAO; A. Ré, University of Aveiro / Department of Biology and CESAM; I. Campos, University of Aveiro / Department of Environment and CESAM; J. Pereira, University of Aveiro / Department of Biology Centre for Environmental and Marine Studies CESAM; J.J. Keizer, University of Aveiro / Department of Environment and Planning CESAM; F. Gonçalves, University of Aveiro / Department of Biology and CESAM

In the last decades, the increased frequency and extent of wildfires have become a societal and environmental problem in Portugal. Among the distinct environmental impacts, the role of wildfire on the water quality has increasingly received research attention, particularly in what concerns to the input of polycyclic aromatic hydrocarbons (PAHs) and metals associated to ashes. However, their toxic effects on the aquatic life have been largely ignored. The main goal of this study was to assess the off-site effects of wildfire on freshwater organisms through the use of *in situ* bioassays. These bioassays were conducted in a recently burned eucalyptus area located in Préstimo (Águeda, central Portugal) and occurred after the first post-fire rainfall events at four sites - two in the main stream (Alfusqueiro) and two in tributary streams. In the river, two sites were chosen, one located upstream of the burned area (RUS-upstream river) and another located downstream, within the burned area (RDS-River downstream). The same was done for the tributaries (SUS-stream upstream and SDS-stream downstream in the burned area). Distinct freshwater organisms, including the shrimp *Atyaephyra desmaresti* (water column organism), the amphipod *Echinogammarus meridionalis* (water-sediment interface organism) and the benthic insect larvae of *Chironomus riparius* were exposed in all four sites, using dedicated test chambers. After two days of field exposition, the mortality and post-exposure feeding inhibition were evaluated. The lethality was not sensitive to discern impacts among the assessed sites because the results showed negligible mortality for all the species and sites. Conversely, the sub-lethal post-exposure feeding inhibition, revealed a decrease in the feeding rate of organisms from the sites impacted by wildfire (RDS and SDS). Unlike, the sites outside the burnt area (RUS and SUS), showed no adverse effects in this endpoint. These results highlight that *in situ* bioassays are a suitable tool to assess the risks of wildfire to aquatic species and that the post-fire runoff, rich in substances such as PAHs and metals, can sub-lethally impair the aquatic organisms in water bodies located downstream the burnt area.

MO060

Estrogenic effects of an Organophosphorous Flame Retardant (TCPP) on Edible Sea Urchin "Paracentrotus lividus"

P.C. López, University of Vigo / Ecology and Animal Biology; E. Pereira-Pinto, University of Basque Country; L. Mantilla-Aldana, University of Vigo / Ecology and Animal Biology; r. beiras, University of Vigo / Toralla marine sciences station (ecimat)

Abstracts New synthetic chemical compounds, like Organophosphorous Flame Retardants (OPFRs), are widely used by the industry as plastic additives in common life objects, or overlaying woods, fabrics, etc. They pose environmental risk due to their efects as androgenic or estrogenic endocrine disrupters, imitating male or female hormones respectively, interfering in vital functions of the organisms. Increasing presence of those chemicals in the water due to the incomplete elimination in the wastewater treatment plants, is emerging as a new problem in water contamination. OPFRs, like Tris (2-Chloro-1-Methylethyl) Phosphate (TCPP), are a typical flame retardant in plastics, being the most detected chemical in the aquatic system. Possible toxic effect of this chemical has not been deeply evaluated yet. This study aims to explore the possible effect of TCPP as an endocrine disruptor on the edible sea urchin *Paracentrotus lividus*. 392 individuals have been distributed in groups: Normal Control, Solvent Control (Acetone) and TCPP exposed (1 and 10 mg/L), they were maintaned in controlled conditions and analyzed at 7 and 28 days. TCPP exposure did not cause histological damages in the gonads, and the bioaccumulation in the tissues was moderate (mean BCF=28 L/Kg WW). However, the results of the GI in this study, support the idea of an endocrine disruption action of TCPP in females exposed to the compound, thus the compound could be catalogued as estrogenic for this marine biological model. **Keywords:** Edible Sea Urchin, Organophosphorous flame retardant, endocrine disruptor.

MO061

Short-term effects of fluoxetine exposure on biomarker and behavioural responses of an estuarine fish

I.A. Duarte, M.P. Pais, P. Reis-Santos, MARE - Marine and Environmental Sciences Centre; H.N. Cabral, Faculdade de Ciências da Universidade de Lisboa / MARE-FCUL; V.F. Fonseca, MARE   Marine and Environmental Sciences Centre

Pharmaceutical compounds are routinely discharged into the aquatic environment. There is growing concern whether they elicit deleterious effects on aquatic organisms, following point source acute exposure as well as chronic exposure. Antidepressants, such as fluoxetine, are frequently detected in both freshwater and coastal systems and have deleterious biological effects at very low concentrations. Nonetheless, contradicting evidence has been reported with lack of consistency in responses across studies. In this context, short-term effects of fluoxetine exposure were analysed in common goby *Pomatoschistus microps*, an estuarine resident species. Two experiments were conducted: where 1) fish were exposed to environmental concentrations of fluoxetine for 96h (0.1 - 100 µg/L) and 2) fish were exposed to high fluoxetine concentrations for 1h (1, 5 and 10 mg/L). Acute toxicity was assessed via multiple biomarker responses, namely antioxidant enzymes activity, detoxification enzymes activity, neurotoxicity and biomarkers of deleterious effects (e.g. catalase, glutathione S-transferase, acetylcholinesterase, lipid peroxidation, DNA damage). Behavioural responses were also examined, concerning activity and feeding behaviour. Overall, results provided insights into the effects of fluoxetine at the sub-individual and individual-level responses in this species and are discussed concerning realistic exposure scenarios as well as their potential implications to estuarine populations.

MO062

Assessment of PCDD/Fs, dioxin-like PCBs and PBDEs in Mediterranean striped dolphins

F. Capanni, University of Trieste / Department of Life Sciences; J. Muñoz-Arnanz, IQOG-CSIC / Department of Instrumental Analysis and Environmental Chemistry; L. Marsili, M. Fossi, University of Siena / Department of Physical Sciences, Earth and Environment; B. Jimenez, IQOG-CSIC / Department of Instrumental Analysis and Environmental Chemistry

Marine mammals are exposed to a variety of persistent organic pollutants (POPs) that bioaccumulate in marine ecosystems. In the present study, blubber samples from ten stranded Mediterranean striped dolphins (*Stenella coeruleoalba*) were used to investigate levels of polychlorinated dibenzo-*p*-dioxins and furans (PCDD/Fs), dioxin-like polychlorinated biphenyls (DL PCBs) and polybrominated diphenyl ethers (PBDEs) using gas chromatograph coupled to a high-resolution mass spectrometer and by using the isotopic dilution technique. The WHO Toxicity Equivalence (TEQ) approach was applied. Median DL PCB concentration was 1820 ng/g lipid weight (l.w.) (range: 474-3840 ng/g l.w.), with males showing statistically higher levels than females (ANOVA, $p< 0.0357$). DL PCBs accounted for over 93.3% of total TEQs. Mono-*ortho* PCB congeners represented on average 99.9% of total DL PCBs, but their TEQ contribution was similar to non-*ortho* PCBs. PCB118 was the most abundant compound (236-1970 ng/g l.w.) and gived the highest contribution to the total TEQs (30.5%), followed by PCB126 (29.4%). The predominant non-*ortho* PCB congener was PCB77, suggesting a higher exposure than elimination rate. Median level of PCDD/Fs was 23.9 pg/g l.w. (14.0-35.0 pg/g l.w.). In contrast to what has been previously reported, the most abundant congeners were 123478-HxCDF (0.85-10.6 pg/g l.w.) and 2378-TCDF (1.69-4.95 pg/g l.w.), while 12378-PeCDD and 23478-PeCDF provided the highest contribution to TEQs. Total TEQ values ranged from 54.0 to 94.6 pg/g l.w. in females and from 99.0 to 250 pg/g l.w. in males, exceeding the TEQ threshold value associated with immunological dysfunctions in harbor seals. Regarding PBDEs, the median value was 456 ng/g l.w. (219-1660 ng/g l.w.), with statistically significant differences between males and females (Mann–Whitney U test $p< 0.0472$). These levels rival and even surpass those reported in other marine mammals in the same area. TetraBDEs were the most abundant congeners, with BDE47 ranging from 39 to 560 ng/g l.w. These results confirmed how the Mediterranean subpopulation of striped dolphin is currently subject to a major threat of environmental pollution. To date, few studies have focused on the evaluation of TEQ values and PBDE levels on this subpopulation. Toxicological and risk assessment studies on this species, recognized as an ocean health sentinel at a sub-basin scale, may provide an early indication of potential adverse health effects.

MO063

Assessment of POPs in stranded sperm whales   (Physeter macrocephalus) from the Mediterranean Sea

A. Bartalini, University of Siena / Department of Environmental, Earth and Physical Sciences; J. Muñoz-Arnanz, IQOG-CSIC / Department of Instrumental Analysis and Environmental Chemistry; L. Marsili, University of Siena / Department of Physical Sciences, Earth and Environment; S. Mazzariol, University of Padova / Department of Public Health, Comparative Pathology and Veterinary Hygiene; M. Fossi, University of Siena / Department of Physical Sciences, Earth and Environment; B. Jimenez, IQOG-CSIC / Department of Instrumental Analysis and Environmental Chemistry

The sperm whale (*Physeter macrocephalus*) is a large toothed whale inhabiting the Mediterranean Sea. This endangered species is subject to a number of threats such as exposure to high levels of contaminants. Several studies show high levels of persistent organic pollutants (POPs) in dolphin tissues from the Mediterranean Sea, but data on sperm whales from the same area are much more scarce. In this study, we analyzed POPs in blubber of nine sperm whales stranded along the Italian coast from 2009 to 2016. Fresh samples were spiked with a suit of PCDD/Fs, PCBs, and PBDEs ^{13}C-labeled standards prior to Soxhlet extraction, and then cleaned-up by using the automated sample preparation system. Samples were analyzed for seventeen PCDD/Fs, twelve DL-PCBs and twenty-seven PBDEs. Quantification was carried out by the isotopic dilution technique by GC-HRMS on a Trace GC Ultra gas chromatograph coupled to a high-resolution mass spectrometer. Samples' lipid content was determined gravimetrically. The relative abundance of the study contaminants followed the order DL-PCBs>PBDEs>>PCDD/Fs. The mean concentration values obtained were 6420 ng g^{-1} (2100-20800 ng g^{-1}) for DL-PCBs, 612 ng g^{-1} (312-1390 ng g^{-1}) for PBDEs and 57.8 pg g^{-1} (45.4-83.5 pg g^{-1}) for PCDD/Fs. Our results were in the same order of magnitude that those reported for the same species in the same area by a recent study from other authors save for PCDD/Fs which were found in an order of magnitude lower.Yet, they were generally much higher that those reported for sperm whales from the Sea of Cortez and from Australia. Regarding PBDEs levels, our results were lower that those reported for sperm whales from North-Atlantic. The PCDF congener profile (penta>hexa>tetra>hepta>octa) was relatively similar to those reported for sperm whales from Australia and to those reported in blubber of striped dolphin (*Stenella coeruleoalba*) from the Mediterranean Sea. In contrast, the PCDD congener profile (hexa>penta>tetra>hepta>octa) was quite different from those with a lower contribution of higher chlorineted congeners and a higher contribution of lower chlorinated congeners.Total calculated TEQs ranged from 275 to 987 pg g^{-1} l.w. and surpassed the threshold of 210 pg WHO-TEQ g-1 l.w. in blubber proposed as starting point of immunosuppression in harbour seals. This high level of contamination is not considered to be the cause of death of these animals, but may have contributed to lowering the defense of their immune system.

MO064

Biochemical and molecular responses to organic contaminants in bottlenose dolphins (Tursiops truncatus gephyreus) from southern Brazil.

B. Righetti, Universidade Federal de Santa Catarina / NEPAQ-CCA; J.J. Mattos, Universidade Federal de Santa Catarina / NEPAQ, Departamento de Aquicultura; M.N. Siebert, Universidade Federal de Santa Catarina / LABCAI Bioquimica; D.d. Lima, Universidade Federal de Santa Catarina / Bioquimica; F.L. Zacchi, Universidade Federal de Santa Catarina / Departamento de Bioquímica; P. Fruet, FURG   Universidade Federal do Rio Grande / Museu Oceanográfico; F. Daura-Jorge, P.C. Simões-Lopes, Universidade Federal de Santa Catarina / ECZ; A. Bainy, Universidade Federal de Santa Catarina / Bioquimica; K. Luchmann, Santa Catarina State University / Engenharia de Pesca

Various adverse health effects have been associated with exposure to persistent organic pollutants (POPs) in cetaceans. Such effects threaten the maintenance of odontocete populations, emphasizing the need for biomarkers that indicate early-on biological responses to POPs. The present work evaluated biomarker response to organic contaminants in bottlenose dolphins subspecies *gephyreus* from two estuarine systems of southern Brazil impacted by agricultural and industrial runoff: Laguna Estuarine System (LES) (n=7) and Patos Lagoon Estuary (PLE) (n=10). Antioxidant enzymes and mRNA transcript levels of genes related to xenobiotic detoxification (*AhR, ARNT, CYP1A, GST, MT2*), antioxidant defense (*GST-π, GPx 4, GR*) and immune response (*IL-1, MHC-II*) were analyzed in integument samples obtained through remote biopsy. POPs were measured in the blubber of the same animals. Generalized linear models (GLMs) were used to analyze the response of each biomarker to ΣPCBs, ΣDDTs, Mirex, Chlordanes (CHL), Hexachlorbenzene (HCB), sampling season (winter or summer) and location (LES and PLE). The best model to describe each biomarker response, with the lowest Akaike Information Criterion (AIC), was chosen using backward selection. GLMs results indicate that the transcript levels of all studied genes were higher in winter when compared to summer, potentially due to enhanced metabolism over colder months. mRNA transcript levels of *AHR*, *GR*, *IL1* and *MT2* genes correlated positively with increasing levels of blubber ΣPCBs, supporting the occurrence of biological response to this class of contaminants. GLMs for *MT2* indicated that the transcript levels of this gene are higher in dolphins from LES, possibly due to greater metal inputs in this area. GR activity was higher in dolphins with higher Mirex levels. Overall, results indicate that biomarker response in the skin of bottlenose dolphins is altered due to exposure to ΣPCBs and ΣPBDEs, which co-varied with ΣPCBs, and Mirex. Absence of influence of other contaminant classes over biomarker response might be due to low contaminant levels in sampled dolphins. This influence might also have been overshadowed by the effects sampling season. Nonetheless, results indicate a sufficiently high exposure to PCBs, PBDEs and Mirex to trigger a biological response in dolphins from these small resident coastal populations, particularly susceptible to the negative effects associated to contaminants.

MO065

Monitoring Eleonora's falcon conservation status both at its breeding and non-breeding grounds, using biological (stress indices) and environmental data

V. Tsarpali, University of Patras / Department of Biology; C. Barboutis, Hellenic Ornithological Society / Antikythira Bird Observatory; C. Kassara, University of Patras / Department of Biology; M. Papadimitraki, S. Giokas, University of Patras / Biology; S. Dailianis, University of Patras / Department of Biology

The present study investigated a battery of stress indices in blood and liver of Eleonora’s falcon (*Falco eleonorae* Géné, 1839), a colonial breeding raptor of the Mediterranean that overwinters in SE Africa, mainly in Madagascar. In particular, cholinesterase (ChE), acetylcholinesterase (AChE) and butylcholinesterase (BChE) activity, as well as cellular abnormalities (MN assay) were measured in blood samples collected from wild individuals, captured on the island of Antikythira (Greece), in May (N=13) and September 2017 (N=19). The results derived from the samples that were collected in May are indicative of the habitat quality at the species’ wintering and/or staging areas, while the ones derived from the samples collected in September are indicative of the habitat quality at the species’ breeding grounds. Moreover, in order to investigate the water quality in the breeding area of *F. eleonorae*, natural water pond samples were collected in September 2017 and further analyzed for the presence of heavy metals. Additionally, heavy metals were measured in liver of an individual found dead near the water ponds. According to the results, total plasma ChE activity ranged between 3.370±0.433 - 11.343±0.829 nmol ml^{-1} min^{-1} in May and 1.444±0.079- 9.314±1.618 nmol ml^{-1} min^{-1} in September. AChE activity remained almost constant between the two sampling periods, while BChE in May was significantly higher than September. Total nuclear abnormalities showed almost similar frequencies in both periods, while cytoplasmic abnormalities observed in September were significantly higher than in May. Cr and Cu levels in water samples were found to be within natural levels, while Cd and Pb concentrations were lower than the MAC-EQS values. All heavy metals levels measured in liver samples were found to be relatively low. The results of the present study showed for the first time that the assessment of a battery of stress indices in tissues of *F. eleonorae*, together with chemical analysis of data derived from their natural habitats, could serve as a valuable tool for elucidating the quality of its foraging grounds and, hence the impact of land use on the species’ conservation status.

MO066

Optimising design and analysis of acute effect field studies

R. Dittrich, Tier3 Solutions GmbH / Wildlife Ecology; I.S. Hotopp, Tier3 Solutions; M.M. Benito, C. Wolf, Tier3 Solutions GmbH

Vertebrate risks assessments of a plant protection product (PPP) may indicate an acute risk to wild birds and mammals or predict effects on population development. This might be driven by (too) conservative assumptions on the exposure side of the equation for the risk evaluation, due to the lack of better data. It is therefore worthy to study deeply the presence of effects in the field in order to obtain a proper risk assessment. We highlight three complementary ways to improve the quality of such field studies. An optimal study design combines the ‘extensive’ approach, by using a great area or number of agricultural fields in different study sites, with the ‘intensive’ approach, by using radio-tracking techniques in a control/treatment design. This double approach covers the natural variation in parameter estimates and enables the identification of possible treatment effects. The radio-tracking technique is sensible enough to monitor the fate of individuals in a treated population over a long time period, and to find them in case of mortality. However, in most cases the critical point is the disappearance of individuals; an increase in their number can indicate a greater vulnerability to other stressors. In the context of a good study design, we also propose an improved statistical evaluation to increase the detectability of effects in comparison to earlier studies. The Kaplan-Meier survival curve and the Cox proportional hazards model are recommended as methods for the analysis of survival information. The Cox model is a well-known statistical technique used in medical tests. It provides an estimate of the treatment effect on survival adjusted for other explanatory variables. Moreover, as it isolates the effects of treatment from the effects of other covariates, the assessment of results of such field studies is facilitated. Additionally, an essential part of every statistical evaluation is to know the minimum number of individuals needed in order to perceive actual treatment effects in the statistical output. Using data from generic radio telemetry studies on real untreated populations of wood mouse and several bird species, we run simulations of acute effects for different scenarios. The results show that minimum sample size is highly dependent on, first, the species, and second, the action mode and persistence of residues of each specific PPP.

MO067

Assessing impacts of legacy pollutants on wildlife of the Trinity River (Texas, USA) using Neotropic Cormorants as indicator species

M.A. Mora, Texas A&M University / Wildlife and Fisheries Sciences; C. Sandoval, Texas A&M University / WFSC

The Trinity River (Texas, USA) has been historically known as a polluted river because of its proximity to the Dallas-Fort Worth area and also because of known discharges of sewage and agricultural irrigation waters to the river. Surprisingly, there are no studies regarding the presence of legacy contaminants in the river and their impacts to wildlife. The objectives of this study were to determine accumulation and potential impacts of persistent organic pollutants such as organochlorine pesticides, PCBS, and PBDEs on nesting aquatic birds of the Trinity River, using Neotropic cormorants (Phalacrocorax brasilianus) as indicator species. Adult and first year cormorants were collected from two sites on the Trinity River Watershed during 2014 and 2015. Tissue sections from liver, spleen, kidneys, and gonads, were used for histopathology analysis, and a portion of the liver was analyzed for OC pesticides, PCBs, and PBDEs. Breast feathers were analyzed for Hg. Surprisingly, all the contaminants were present at low concentrations and below those that could be associated with adverse effects; however, altered structure, composition and function, were detected in the livers and kidneys of most samples. A novel coccidian Eimeria sp. was also detected in the kidneys of several cormorants. Our results suggest that aquatic birds using the Trinity River watershed are not at risk for adverse effects due to OC pesticides, PCBs, PBDEs, and Hg. These results should be useful to wildlife managers regarding concerns over contaminant impacts of the Trinity River on wildlife.

MO068

Tracking the effects of a neonicotinoid insecticide on songbird migration

M.L. Eng, University of Saskatchewan / Toxicology; B. Stutchbury, York University; C.A. Morrissey, University of Saskatchewan / Biology

Recent decades have seen a dramatic increase in the application of neonicotinoid insecticides, which are now the most widely used class of insecticides in the world. Migratory seed-eating birds that use agricultural landscapes for refueling may be particularly susceptible to the neurotoxic effects of neonicotinoids. Effects on refueling or orientation behavior could have significant fitness consequences; however, the influence of neonicotinoids on migratory ability is poorly understood. We used white-crowned sparrows (*Zonotrichia leucophrys*) during spring migration to assess the direct effects of imidacloprid on the migratory behavior of seed-eating passerines. In a previous captive study, we found birds exposed to environmentally relevant concentrations of imidacloprid experienced significant mass loss and stopped orienting correctly in behavioural trials, whereas control birds maintained body mass and a seasonally appropriate northward orientation. To corroborate results from captive trials on free living birds, we conducted a study on radiotagged white-crowned sparrows following a single oral dose of imidacloprid. Birds were caught in Ontario, Canada during spring migratory stopover and exposed via gavage to a single dose of a vehicle control or imidacloprid at either 1.2 mg/kg bw or 3.9 mg/kg bw (n = 12 birds/treatment). Birds were held for approximately 6 hours, and their body mass and food intake were monitored. Individuals were then tagged with uniquely coded transmitters and released into into a MOTUS array of automated telemetry towers in Southern Ontario, Canada to track their movements on a landscape scale. We found that high dose birds significantly reduced food consumption, and imidacloprid exposed groups lost a significant amount of body mass relative to controls within 6 hours of dosing. Using automated telemetry data, we found that birds with the longest stopover durations were in the imidacloprid treated groups. Further analysis of the telemetry data is being used to determine effects on speed of travel and direction of migratory movements across a large water barrier.

MO069

A synthesis of the interactions between anticoagulant rodenticides and wildlife

R. Shore, Centre for Ecology & Hydrology (NERC); N. van den Brink, Wageningen University / Dept of Toxicology; J.E. Elliott, Environment Canada / Science Technology Branch; B.A. Rattner, USGS-Patuxent Wildlife Research Ctr / Patuxent Wildlife Research Center

Anticoagulant rodenticides (ARs) are the mainstay of rodent control throughout the world. Regulatory risk assessments indicate ARs pose a significant risk to non-target wildlife but AR use remains widely authorised because the benefits (particularly to human health) are deemed to outweigh the environmental risks. Recently, an authoritative reference text, prepared by 24 international scientists, reviewed the main issues related to ARs and wildlife, specifically: AR use, regulation, exposure pathways, toxicity, mechanism of action, pathology, pharmacokinetics, genetic resistance, non-target risk and its mitigation, alternatives

to ARs and integrated pest management (IPM). Broad concepts that emerged were: there is high conservation of the blood clotting process and so ARs can affect a wide range of non-target species; development of genetic resistance in target species led to global use of the more acutely toxic and persistent second-generation ARs (SGARs); vitamin K1 can be an effective antidote (unlike for many rodenticides); variation in non-target sensitivity may be due to pharmacokinetic, ecological and behavioural processes; >50% of predatory species contain AR residues; AR residues indicate exposure but a definitive diagnosis of AR-induced mortality requires additional evidence (e.g., clinical signs, haemorrhagic lesions); probability of death in relation to AR residues may help assess extent of mortality in populations; tissue residues are informative of exposure but dietary AR concentrations are more suited to assess risk; and primary AR exposure associated with urban, agricultural and island conservation use may cause localized non-target species population declines, but there is no clear evidence for secondary AR exposure causing population declines. Alternatives to ARs (e.g., bromethalin, cholecalciferol, zinc phosphide) also pose a significant risk to non-target wildlife, livestock, companion animals and people. There are a number of key information needs to better understand the environmental risk from ARs. These are: improved understanding of AR-induced effects and risk to wildlife populations; knowledge of exposure and effects in invertebrates and lower vertebrates; enhancement of resistance management in target species; development of safe alternative chemical and non-chemical methods. It is anticipated that addressing these research priorities would reduce the potential AR-related conflict between protection of human health and wildlife.

MO070

Anticoagulant rodenticides in red kites (Milvus milvus) in Britain

R. Shore, Centre for Ecology & Hydrology (NERC); L. Walker, Centre for Ecology & Hydrology; J. Jaffe, Zoological Society of London / Institute of Zoology; L. Barnett, Fera Science Ltd; J.S. Chaplow, Centre for Ecology & Hydrology; S. Charman, Fera Science Ltd; A. Giela, Science and Advice for Scottish Agriculture; A. Jones, Fera Science Ltd; M.G. Pereira, Centre for Ecology & Hydrology / Lancaster; E.D. Potter, NERC Centre for Ecology & Hydrology / Lancaster; A. Sainsbury, Zoological Society of London / Institute of Zoology; D. Sleep, N.J. Thompson, C. Senior, NERC Centre for Ecology & Hydrology; E. Sharp, SASA Scottish Government / Pesticides and Wildlife

Second generation anticoagulant rodenticides (SGARs) can be toxic to all mammals and birds. Studies have shown that, in Britain, there is widespread exposure to SGARs in a diverse range of predatory mammals and birds, including red kites (*Milvus milvus*). This species may be particularly at risk as it scavenges dead rats, a target species for rodent control. Investigation of SGAR exposure in red kites in Britain is conducted by independent laboratories but recently the WILDCOMS network (www.wildcoms.org.uk/) has facilitated collation of these data sources and a resultant national-scale annual assessment of exposure in red kites. We report the findings of the first such national scale exposure which was conducted for red kites found dead in 2015. Carcasses were typically found by members of the public, sent to an investigating laboratory, necropsied and analysed for liver SGARs by Liquid Chromatogrpahy Mass Spectrometry. Of 26 red kites from England & Wales that were analysed, all had detectable residues of difenacoum and brodifacoum; most also contained bromadiolone. Difethialone was less frequently detected and flocoumafen was not detected in any birds. Sum liver SGAR concentrations ranged from 50 to 1266 ng/g wet wt. (arithmetic mean: 372 ng/g). Post-mortems indicated that 9 (35%) of the kites had internal hemorrhaging that was not associated with detectable trauma; most had elevated sum SGAR liver concentrations. On the basis of these two factors, it is considered probable that SGARs were a contributory cause of death in these birds. Residue data were also available for 6 red kites from Scotland. Three (50%) had liver residues of at least two SGARs (bromadiolone and difenacoum); brodifacoum was also detected in one of these kites. SGARs were assessed to be a contributory cause of death in the bird that had residues of three SGARs. The data for Scotland, although limited, suggest that exposure of red kites in 2015 may have been less marked than in England & Wales, as has been found for other species collected in other years. Overall, these results suggest all red kites in England & Wales at least, are exposed to SGARs and that poisonings are not uncommon. Despite this, the red kite population has greatly expanded in Britain as birds recolonise former haunts. The entent to which exposure to SGARs may affect future population growth merits further investigation.

MO071

Environmental determinants of the exposure to anticoagulant rodenticides in non-target species

J.J. López-Perea, Instituto de Investigacion en Recursos Cinegeticos / Unidad de Ecología y Ciencia Animal; I.S. Sanchez-Barbudo, UCLM-CSIC / Grupo de Toxicología de Fauna Silvestre; P.R. Camarero, Instituto de Investigación en Recursos Cinegéticos, CSIC-UCLM-JCCM / Grupo de Toxicología de Fauna Silvestre; R. Mateo, IREC-CSIC- UCLM / Grupo de Toxicología de Fauna Silvestre

Anticoagulant rodenticides have some similarities with other bioaccumulative persistent organic compounds, because of their frequent presence in many predatory species. In addition, the fact of being highly toxic substances makes this biaccumulation particularly harmful for these predators. Considering that the use of rodenticides occurs mainly in areas with high density of rodents that are in turn prey to multiple predators, we can also expect an ecological trap scenario. We studied the levels of second generation anticoagulant rodenticides (SGARs) and the environmental factors that influence such exposure in non-target species. The analysis included liver samples of wild animals (n=244) found dead between 2007 and 2016 in the region of Aragon (NE Spain). This sampling included 49 species (1 reptile, 16 mammals and 32 birds). Liver samples were analysed by LC-MS and the presence of SGARs was statistically analysed with generalized linear models with a binary logistic response to study the effect of environmental or habitat characteristics including human population and livestock density and types and surface of crops. SGARs residues were detected in 83 (34%) of the analysed animals, corresponding to 25 (51%) species. Ten species (53 individuals), corresponding to four mammals and six birds, had residues >200 ng/g, which is the threshold associated with adverse effects: these included common raven (67%), red fox (50%), red kite (38%), eagle owl (25%), stone marten (23%), common buzzard (17%), Western marsh-harrier (17%) and Eurasian badger (14%). The spatial analysis at the municipality level has allowed to identify the percentage of continuous urban land area, the density of human population and the density of cattle as descriptors of the presence of SGARs in animals. We have also discarded the relationship between the exposure to SGARs and the area occupied by cropland or intensive orchards. The presence of SGARs in predators was therefore more associated with the use of these products as biocides in urban areas or cattle farms rather than as plant protection products in cropland. This information could be used to reduce the exposure to SGARs in predators or scavengers that find an attractive ecosystem in the urban areas or around farms, which can actually constitute an ecological trap for them.

MO072

Four years of NewRaptor: results from in ovo exposure in model species and field sampling in raptors

N. Briels, Norwegian University of Science and Technology / Biology; T.M. Ciesielski, Norwegian University of Science and Technology; M.E. Løseth, The Norwegian University of Science and Technology / Biology; B.M. Jenssen, Norwegian University of Science and Technology / Biology; I. Eulaers, University of Antwerp / Biology; C. Sonne, Aarhus University / Department of Biosciences, Arctic Research Centre; T. Nygård, T.V. Johnsen, Norwegian Institute for Nature Research NINA; P. Gómez-Ramírez, University of Murcia / Department of Toxicology; A. Garcia-Fernandez, University of Murcia / Sociosanitary Sciences; J. Martínez, University of Murcia / Ecology and Hydrology; J.O. Bustnes, Norwegian Institute for Nature Research NINA; G. Poma, G. Malarvannan, University of Antwerp / Toxicological Center; A. Covaci, University of Antwerp, Toxicological Center / Toxicological Centre Dep of Pharmaceutical Sciences; D. Herzke, NILU Norwegian Institute for Air Research; B. Styrishave, University of Copenhagen / Section of Analytical Biosciences Department of Pharmacy; V. Jaspers, Norwegian University of Science & Technology / Biology

The international research project NewRaptor (ID 230465/F20, funded by the Norwegian Research Council and the Norwegian University of Science and Technology) aims to investigate the exposure and effects of emerging chemicals in birds of prey. The raptors under investigation include the terrestrial Northern goshawk (NG - *Accipiter gentilis*) and the marine White-tailed eagle (WTE - *Haliaeetus albicilla*) from Norway and NG from Spain. During the breeding seasons of 2015 and 2016, blood and body feathers were obtained from the chicks (in total n = 160 for NG and n = 70 for WTE) in the nest when they were circa 4-9 weeks old. The samples were analysed for novel brominated flame retardants (nBFRs), organophosphate flame retardants (PFRs) and per- and polyfluoroalkyl substances (PFASs), along with trace elements and legacy persistent organic pollutants (POPs). Significant differences were found between the two species (with WTE generally showing higher levels of pollutants), but also within species, depending on the location. PFASs were generally found at the highest concentrations, with perfluorooctane sulfonate (PFOS) being the most important compound. nBFRs and PFRs were found at very low or non-detectable levels in blood plasma. Further, pollutant effects on different biochemical, immunological and endocrine parameters were assessed. We have performed controlled *in ovo* exposure studies in Japanese quail (*Coturnix japonica*) and chicken (*Gallus gallus domesticus*) as model species, both with single compounds and in a mixture scenario. The compounds for the quail exposure study included Dechlorane Plus (DP), tris(1,3-dichloro-2-propyl) phosphate (TDCIPP) and their 1:1 mixture, while PFOS, F-53B (PFOS replacement product) and their 1:1 mixture were used in chickens. Effects on gene expression and activity of anti-oxidative enzymes (catalase, superoxide dismutase, glutathione-S-transferase and glutathione peroxidase), lipid - and protein oxidative damage and biotransformation (cytochrome P4501A) were investigated. Further, hormonal analysis of corticosterone and progesterone was performed using HPLC-MS/MS. Gene expression and enzyme assays on similar endpoints will be performed on NG samples in January 2018 and will be presented alongside the results from the *in ovo* exposure studies at SETAC. This will enable discussing the potential usefulness and pitfalls when extrapolating from laboratory dosing studies using model species to field assessments in raptors.

MO073

The potential of feathers as a biomonitoring tool for fluoxetine in wild birds

S.E. Whitlock, Environment Department, University of York / Environment; K. Arnold, University of York / Environment; J. Lane, Animal and Plant Health Agency; M.G. Pereira, Centre for Ecology & Hydrology / Lancaster; R. Shore, Centre for Ecology & Hydrology (NERC)

The antidepressant fluoxetine has been identified as a contaminant which may pose a risk to wild birds. However there is little empirical evidence regarding which bird species are most at risk of exposure to fluoxetine, in terms of concentration levels in wild bird samples. A significant barrier to sampling wild birds is that fluoxetine is cleared very quickly from systemic circulation and plasma concentrations might be expected to fall to levels below the limit of detection in less than one hour post-exposure. Faecal sampling presents a similar problem. However, fluoxetine is detectable in the hair of humans and other mammals. We hypothesised that fluoxetine might likewise be detectable in feathers. Moult occurs over a period ranging from weeks to months and during this time, growing feathers have a blood supply. If a wild bird was exposed to fluoxetine during this period, the compound could be laid down in the feathers and subsequently be detectable. We conducted an aviary study with wild caught Eurasian starling (*Sturnus vulgaris*) to determine whether fluoxetine administered during a period of feather regrowth is deposited in the feathers in detectable concentrations. We removed two rectrices (tail feathers) from each bird. We then administered a dose of fluoxetine at an environmentally relevant concentration (3.8 µg d^{-1}) each weekday throughout the regrowth period. We plucked the new feathers once they were fully regrown and analysed them by LC-MS/MS for fluoxetine and its major active metabolite norfluoxetine. Our preliminary results indicate that fluoxetine is detectable in the feathers and we will present information on the concentrations present and whether they are correlated with levels in other tissues such as liver and brain. We discuss the extent to which feathers have potential as tools for monitoring the occurrence and severity of exposure to fluoxetine in wild birds.

MO074

Field-effect studies as a suitable method to assess effects of plant protection products on free-living common voles (Microtus arvalis): A case study with the fungicide iprodione

O. Fuelling, C. Miersch, Tier3 Solutions; S. Steiger, BASF SE, Agrarzentrum Limburgerhof

After foliar spray application of a plant protection product on crops, food sources of small mammals may be potentially contaminated with this product. Ingestion of treated food could possibly lead to effects on the population level (e.g. reproductive impairment etc.). In the presented study, it was examined if there were any long-term effects from repeated foliar spray applications of the fungicide iprodione on populations of the common vole, *Microtus arvalis*. The field-effect study was conducted in Germany during the main reproductive period of the common vole on 14 commercially used grassland fields. Regular life-trapping sessions which followed a capture-mark-recapture design were conducted from June to November 2014 on treated and untreated (control) grassland fields, as well as in adjacent habitats. Population size and development, body weight, reproductive performance, and survival rates were determined and analyzed using linear mixed models. Results of the study will be presented in detail in the poster. The results support the fact that field-effect studies, which investigate effects under real use conditions and are included as higher tier refinement option in the EFSA Bird and Mammal Guidance Document (EFSA, 2009), are a useful tool for the higher tier risk assessment, even though no internationally agreed standard protocol for mammal field-effect studies is existing yet.

MO075

Monitoring NSAIDs in carrion and avian scavengers form Spain: preliminary results after diclofenac registration for veterinary use

R. Mateo, IREC-CSIC- UCLM / Grupo de Toxicología de Fauna Silvestre; M. Herrero, Instituto de Investigación en Recursos Cinegéticos (IREC); P.R. Camarero, Instituto de Investigación en Recursos Cinegéticos, CSIC-UCLM-JCCM / Grupo de Toxicología de Fauna Silvestre; I.S. Sanchez-Barbudo, UCLM-CSIC / Grupo de Toxicología de Fauna Silvestre; R. Velarde, I. Marco, UAB

The serious impact of diclofenac on Asian vultures raised the alarm of the deficient environmental risk assessment of some veterinary drugs. In the case of diclofenac, there was an evident gap in the knowledge of the high toxicity that this non-steroidal anti-inflammatory drug (NSAID) has in Old-World vultures that can feed on treated livestock (LD_{50} in *Gyps bengalensis* of 98-225 µg/kg body mass). Despite the experience acquired after the collapse of the vulture populations in Asia, in 2013 this drug was authorized for use in veterinary medicine in Spain and other countries in the European Union with the consequent risk of repeating the situation generated in Asia. In this work, we have studied the presence of NSAIDs in carrion animals (kidney, liver and muscle of pig, n=125) supplied in “muladares” to feed vultures. We have also studied the presence of NSAIDs residues in tissues of avian scavengers (vultures and kites, n=27) found dead with suspicion of being intoxicated. NSAIDs were detected in tissues of four pigs (3.2%). Low levels of flunixin were detected in liver (4.1 ng/g) and kidney (7.9 ng/g) of two pigs; meloxicam was detected in the liver of one pig (23.8 ng/g); and diclofenac was detected in the muscle of another pig (170.5 ng/g). This level of diclofenac was relatively high, but kidney and liver of the same animal were negative for diclofenac presence. An examination of this muscle sample showed a pale area adjacent to a congestive portion that may correspond to the injection point of the diclofenac in the pig. Flunixin was the only NSAID detected in the studied avian scavengers. Two out of 22 Eurasian griffons (*Gyps fulvus*) analysed had 330 and 23 ng/g of flunixin in liver. Another cinereous vulture (*Aegypius monachus*) showed 2.83 µg/g of flunixin in liver, but it was diagnosed as an iatrogenic poisoning at the wildlife rehabilitation center. Lesions in the kidney and visceral gout have not been observed macroscopically or microscopically in 15 Eurasian griffons analysed. Residue levels in carrion and scavengers indicate a limited risk of poisoning, possibly because treated livestock died several hours after NSAID injection. However, veterinary use of NSAIDs can still be a threat for scavengers if veterinarians and farmers ignore the associated risk when treated livestock is used to feed vultures.

MO076

Different approches comparison for evaluation of hypopharyngeal glands (HPG) in Honeybees (Apis mellifera L.)

A. Drzewiecka, M. Napora-Rutkowska, J. Faron, Institute of Industrial Organic Chemistry Branch Pszczyna / Department of Toxicological Studies; P. Parma, E. Kulec-Ploszczyca, A. Glanas, Institute of Industrial Organic Chemistry Branch Pszczyna / Department of Ecotoxicological Studies; N. Lemańska, Institute of Industrial Organic Chemistry, Branch Pszczyna; M. Frontczak-Baniewicz, R. Strzalkowski, Mossakowska Medical Research Centre, Polish Academy of Sciences / Electron Microscopy Platform; D. Krakowian, Institute of Industrial Organic Chemistry Branch Pszczyna / Department of Toxicological Studies; A. Daniel-Wójcik, Institute of Industrial Organic Chemistry Branch Pszczyna / Deparment of Toxicological Studies

Honeybees (*Apis mellifera* L.) are beneficial arthropods that play important roles in nature but also in the food and pharmaceutical industries. One of the conditions for maintaining healthy colonies is the proper development of the honeybee workers hypopharyngeal glands (HPG) which produce proteinic substance to feed larvae and queen. The aim of this study was to validate the different algorithms (including obtaining the material) to conduct the hypopharyngeal glands development evaluation, in order to select the method that combines the highest reliability (the smallest technical error), the optimal cost, the least effort and time-consumption. The study was conducted on Honeybees subjected to chronic toxicity studies performed according to the EFSA guidelines (EFSA Journal 2013;11(7):3295). Insects were treated with four different chemicals in 3 to 5 concentrations. The HPG were obtained from 5 bees per test item (in the highest concentration, which did not cause mortality below 50%) and the negative control. Six different approaches for HPG evaluation were tested: - histopathology (HP) of isolated glands (linear and quantitative measurements, imaging); - histopathology (HP) of whole heads (linear and quantitative measurements, imaging); - whole mount (WM) method on isolated gland (linear and quantitative measurements, imaging); - protein absorbance (PA) from isolated glands (quantitative measurements); - protein absorbance (PA) from whole heads (quantitative measurements); - scanning electron microscope (SEM) (linear and quantitative measurements, imaging). The linear measurements (small and big axis of symmetry) were taken from ten acini from left and right HPG, however for SEM only left HPG were included. The quantitative measurements (number of acini per 1 mm^2) were taken during HP, WM and SEM testing. The quantitative measurements of protein absorbance from isolated glands were taken from left and right HPG. The analysis of the results showed decreases and increases of acini and protein absorbance depending on the test item. However, observed deviations did not show any statistical significance. According to the performed studies for evaluation of hypopharyngeal glands development the linear measurement combined with imaging should be used.

MO077

Bird and mammal focal species for pesticide risk assessment in rice

M. Vallon, C. Dietzen, S. Laucht, F. Sotti, J. Ludwigs, Rifcon GmbH

Ecotoxicological risk assessment for birds and mammals is required for the registration of pesticides in Europe to assess potential risks to wildlife through uptake of contaminated diet items. To ensure a realistic and reliable risk assessment, bird and mammal focal species should be used that are representative for the species actually occurring in the crop of concern. However, in the relevant guidance document on bird and mammal risk assessment by the European Food Safety Authority (EFSA), rice is to date pooled with other cereals such as wheat and barley, despite the obvious peculiarity of rice cultivation. The generic focal species suggested for rice are thus those known from the dry environments of cereal fields. To address this issue, we conducted a comprehensive literature review on bird and mammal species regularly occurring in rice paddies at the relevant time periods of potential pesticide exposure to identify appropriate focal species candidates for risk assessment. Our results show that the relevant species occurring in the wet environments of rice paddies indeed clearly differ from the focal species suggested for risk assessment for wildlife in cereals and thus provide a baseline for more realistic and rice-specific risk assessments for birds and mammals.

MO078

Non-invasive assessment by feathers of lead exposure and its relationship with stress hormones in bearded vultures from the Alps

R. Mateo, IREC-CSIC- UCLM / Grupo de Toxicología de Fauna Silvestre; L. Giménez-Lozano, IREC Instituto de Investigación en Recursos Cinegéticos; L. Monclus, UAB; I. Champly, ASTERS; M. Lopez-Bejar, UAB

A reintroduction project of bearded vulture or lammergeier (*Gypaetus barbatus*) has been carried out in the Alps since 1987. There are several factors that can affect the success of this reintroduction, and one of them is exposure to lead from the ammunition used for hunting. An exposure to lead, even at sub-lethal levels, can be a stressful situation that implies an increase in circulating corticosterone levels. In birds that are rare and difficult to capture, blood sampling for both lead and corticosterone analysis can be a difficult task and with some risk for birds. For this reason, the development of non-invasive methodologies to study exposure to pollutants and their respective biomarkers is being directed towards the analysis of easy-to-collect samples, such as feathers moulted by birds. The objectives of this study are to quantify the degree of exposure to lead that the bearded vultures have in the Alps by analysing moulted feathers found in the field, relating them to corticosterone levels in the same feather and defining the factors that affect the fluctuations of these two components along transverse segments of feathers. The analysis of the segments of 20 different feathers from different bearded vultures revealed that the abnormal exposure to lead (probably due to the ingestion of ammunition) had a prevalence of 15% (three feathers with levels above 2 µg/g of Pb in rachis), and that the annual incidence of such exposure was 30% (a feather with an abnormal exposure during its development of approximately two months). The concentration of lead in the segments of rachis feathers was negatively related to the concentration of corticosterone, and was significantly influenced by the collection area and the individual (feather). In the present study we observed that exposure to elevated levels of lead can occur in a high percentage of individuals in a population throughout the year, which in the case of the lammergeier of the Alps can entail a risk to the sustainability of the population if this exposure reach lethal levels.

MO079

Post mortem stability of phase I and II biotransformation enzymes in the liver of kelp gull Larus dominicanus

J.J. Mattos, Universidade Federal de Santa Catarina / NEPAQ, Departamento de Aquicultura; D.d. Lima, Universidade Federal de Santa Catarina / Bioquimica; B. Righetti, Universidade Federal de Santa Catarina / NEPAQ-CCA; L.O. Villas Boas, V.H. Dias, Universidade Federal de Santa Catarina / CCB; S. Costa-Silva, C. Kolesnikovas, M. Antonelli, Associação R3 Animal; K. Luchmann, Santa Catarina State University / Engenharia de Pesca; A. Bainy, Universidade Federal de Santa Catarina / Bioquimica

The measurement of biomarker responses to chemical contaminants in wild organisms represents a powerful tool in environmental monitoring programs. However, getting biological samples suitable for biomarker analysis may be challenging, since some programs rely on samples collected from carcasses. Shorter periods from death to analysis of biochemical biomarkers provides more accurate results, but how these parameters change at longer *post mortem* intervals remains unclear. This study evaluated the *post mortem* stabilityof key biotransformation enzymes (glutathione *S*-transferase, GST and 7-etoxyresorufin-*O*-deetilase, EROD) in the liver of kelp gull *Larus dominicanus* for periods up to 24 hours. Liver tissue of two euthanized animals was sectioned into several 1-cm^3 cubes and stored in individual closed tubes at 25 ºC for 0, 1, 2, 3, 6, 12, 18 and 24 h *post mortem* before liquid nitrogen freezing. Cytosolic and microsomal fractions were obtained from 150 mg of each sample individually and used for GST and EROD measurements, respectively. GST activity proved to be stable after 24 h (85-90% of initial activity). EROD activity decreased abruptly after the first hour *post mortem* for both animals. After 3 hours EROD activity presented 65 to 71 % of initial activity and 28 to 50% of its initial activity after 6h, showing an exponential decrease along *post mortem* period. The estimated half-life for EROD varied from 2.8 to 5.2 hours *post mortem*. Our results indicate that time elapsed since death until sample collection plays an essential role for biotransformation enzymes, especially concerning EROD activity. GST seemed to be more resistant to degradation over time, and it thus appears possible to make valid GST activity measurement in selected post mortem liver tissue of kelp gull. Overall, our findings demonstrate that caution is warranted in monitoring programs when comparing biological samples with different intervals between collection and analysis procedures.

MO080

Investigating thyroid disrupting effects of organohalogenated contaminants in White-tailed eagle nestlings

M.E. Løseth, The Norwegian University of Science and Technology / Biology; G.S. Eggen, N. Briels, Norwegian University of Science and Technology / Biology; T. Nygård, T.V. Johnsen, J.O. Bustnes, Norwegian Institute for Nature Research NINA; D. Herzke, NILU - Norwegian Institute for Air Research / FRAM Centre Tromso; G. Poma, G. Malarvannan, University of Antwerp Toxicological Center; A. Covaci, University of Antwerp, Toxicological Center / Toxicological Centre Dep of Pharmaceutical Sciences; B.M. Jenssen, Norwegian University of Science and Technology / Biology; V. Jaspers, Norwegian University of Science & Technology / Biology

White-tailed eagles (WTE; *Haliaeetus albicilla*) can accumulate a wide range of organohalogenated contaminants (OHCs), due to their apex trophic position. Their diet consists mainly of fish and seabirds, thus long food chains and a high potential for biomagnification of OHCs. The nestlings can therefore be exposed to high levels of certain OHCs through maternal transfer to the eggs, and later through diet. Concentrations of per- and polyfluoroalkyl substances (PFASs) have recently been shown to exceed those of other legacy OHCs in this species and accordingly required closer attention. Several of these POPs have shown to interfere with endocrine systems in birds, and especially the thyroid system. The thyroid system is important for birds' thermoregulation, metabolism, growth and development. Assuring appropriate concentrations and actions of the two major thyroid hormones thyroxine (T_4) and triiodothyronine (T_3) in the circulatory system is therefore especially important in nestlings. The aim of the present study was, for the first time, to investigate the effect of POPs and PFASs accumulation in plasma on thyroid hormones (TH) of nestling white-tailed eagles. We also included the body mass and age to assess influence of biological variables on the TH. Blood plasma samples were obtained from 70 nestlings of white-tailed eagles from two archipelagos in Norway, Smøla (n = 35) and Steigen (n = 35), in the summer of 2015 and 2016. In total, 14 polychlorinated biphenyls (PCBs), 7 organochlorinated pesticides (OCPs), 5 polybrominated diphenyl ethers (PBDEs) and 8 PFASs were quantified in over 50 % of the plasma samples at each location and each year. Our results show higher OHC concentrations in Steigen [median and range; Σ_{14}PCBs: 5.1 ng/ml (1.5 – 59.1 ng/ml), Σ_7OCPs: 4.2 ng/ml (1.3 – 52.2 ng/ml), Σ_5PBDEs: 0.3 (< 0.1 – 2.6 ng/ml) and Σ_8PFASs: 20.8 ng (7.2 – 52.9 ng/ml)], than Smøla [median and range; Σ_{14}PCBs: 3.9 ng/ml (0.8-34.7 ng/ml), Σ_7OCPs: 2.4 ng/ml (0.9 – 15.3 ng/ml), Σ_5PBDEs:0.2 (< 0.1 – 1.5 ng/ml) and Σ_8PFASs: 10.9 ng (4.6 – 46.7 ng/ml)]. The analyses of thyroid hormones have been carried out and the results will be presented at the conference, along with biological parameters and OHCs.

MO081

Assessment of exposure and effects of Hg levels in feathers of White-tailed eagles (Haliaeetus albicilla) and Northern goshawks (Accipiter gentilis) nestlings from Norway

P. Gómez-Ramírez, University of Murcia / Department of Toxicology; J.O. Bustnes, Norwegian Institute for Nature Research / Fram Centre; G.S. Eggen, Norwegian University of Science and Technology / Biology; I. Eulaers, University of Antwerp / Biology; G. Lepoint, University of Liege / Department of Oceanology; J.M. Pérez-García, University of Lleida / Department of Animal Sciences; T.V. Johnsen, Norwegian Institute for Nature Research NINA; A. Garcia-Fernandez, University of Murcia / Sociosanitary Sciences; V. Jaspers, Norwegian University of Science & Technology / Biology

Anthropogenic activities have led to a global increase of Mercury (Hg) in the environment. Due to its toxicity, legislative measures have been taken to reduce the levels. To assess the effectiveness of such restrictions and the current levels of these chemicals in the environment, biomonitoring using birds is very useful. Hg has caused detrimental effects in birds such as haematotoxicity, immunotoxicity and endocrine disruption e.g. suppression of baseline corticosterone. The aim of this study was to assess the exposure to Hg and its effects at the biochemical/physiological level in White-tailed eagles (WTE) and Northern goshawks (NG) from Norway. Samples were obtained in 2014 from nestling WTE (n=14) and NG (n=11) in northern Norway (Nordland- N 68.30 – 68.47?, E 24.54 – 25.27?- and Troms- N 68.77 – 67.39?, E 20.39 – 23.34?- counties, respectively). Total Hg in feathers, total and free plasma corticosterone levels were analysed, along with following blood clinical chemical parameters (BCCPs): albumin, calcium, phosphorus and γ-glutamyltransferase, lactate dehydrogenase, total proteins, aspartate aminotransferase, alanine aminotransferase, alkaline phosphatase, cholesterol, triglycerides, creatine kinase, amylase, glucose, creatinine, bilirubin, potassium and uric acid. Stable carbon (^{13}C, ^{12}C) and nitrogen isotopes (^{15}N, ^{14}N) were analysed in body feathers to evaluate inter- and intraspecific contaminant exposure. Due to the low amount of feather samples, Hg could only be analysed in 13 WTE and 8 NG. Mean ± SD were 0.51 ± 0.34 mg/kg in NG and 3.01 ± 1.34 mg/kg in WTE. The significantly higher levels in WTE than in NG ($T_{(19)} = -7.61$, $p < 0.01$) may be related to different dietary input, as confirmed by stable carbon and nitrogen isotope analysis of body feathers. The marine prey of WTE seem to determine the Hg loads, as Hg is known to be abundant in the marine environment. Partial correlations to assess the relations between Hg and biochemical parameters (corticosterone, BCCPs) showed relations between Hg and aspartate aminotransferase (an enzyme that may increase after liver damage). The effect of mercury on this enzyme seems controversial, as some experimental studies on nestlings of different species have found both positive and negative relations. Moreover, the lack of information on reference values in GH and WTE complicates the interpretation. Acknowledgements: NILS Science and Sustainability.

MO082

Thyroid-related gene expression, hormones, and thyroid gland histology in American kestrels exposed in ovo to two persistent organic pollutants, SCCPs and TBBPA-BDBPE

A. MacLeod, University of Maryland, College Park / Environmental Science and Technology; P.F. Henry, U.S. Geological Survey / Patuxent Wildlife Research Center; K.J. Fernie, Environment & Climate Change Canada / Ecotoxicology and Wildlife Health; N.K. Karouna-Renier, USGS Patuxent Wildlife Research Center /

Patuxent Wildlife Research Center
Highly brominated flame retardants are being replaced by alternative flame retardants such as Tetrabromobisphenol A bis[2,3-dibromopropyl ether] (TBBPA-BDBPE). TBBPA-BDBPE was introduced as a possible substitute for decabromodiphenyl ether (decaBDE), but has shown similar persistency and environmental transport mechanisms. This additive flame retardant is used in plastic products, resins, textiles, paints, and household electronics. Although it is produced only in the US, Israel, and China, TBBPA-BDBPE is detected in environmental samples and wildlife tissues from across the globe. Short-Chain Chlorinated Paraffins (SCCPs) are priority emerging persistent organic pollutants (POPs) identified as chemicals of concern by the Stockholm Convention, Environment and Climate Change Canada, and the U.S. Environmental Protection Agency. SCCPs are used in metal lubricants and coolants in metal cutting, and as plasticizers and flame retardants in plastics and paints. SCCPs are of concern because they bioaccumulate in wildlife and humans, are environmentally persistent, transported globally, and are toxic to aquatic organisms at low concentrations. However, few data are available on the potential adverse effects of TBBPA-BDBPE and SCCPs in birds. A comparative exposure assessment of these two classes of flame retardants was conducted using egg injections in a non-model species, the American kestrel (*Falco sparverius*) to assess survival, molecular, biochemical, and endocrine, growth and reproductive endpoints. Analyses conducted on tissue samples from hatchings included thyroid related gene expression in the liver, thyroid hormone levels, and thyroid gland histology. Preliminary results will be presented from the molecular to biochemical to cellular level.

MO083

Bioaccumulation of metals in bats: non-lethal vs lethal sampling to assess risk

J. Alves, R. Mina, A. Alves da Silva, CFE - Centre for Functional Ecology, / Department of Life Sciences, University of Coimbra; T. Natal da Luz, University of Coimbra / Department of Life Sciences, University of Coimbra; J.A. Cabral, P. Barros, CITAB - Centre for Research and Technology of Agro-Environment and Biological Sciences / Laboratory of Applied Ecology, Unversity of Trás-os-Montes e Alto Douro; C.J. Topping, Aarhus University / Department of Bioscience; J. Sousa, University of Coimbra / Department of Life Sciences

More than 22% of bat species are threatened according to the World Conservation Union. Among the reasons responsible for this decline are the environmental changes due to anthropogenic factors, namely habitat loss through agriculture, forestry, urbanization and industrialization, contamination by pesticides and metals, changes in water quality. There is a growing concern about the possible consequences of environmental contamination in several bat species. Until now, most of the studies on the effects of contaminants in bats are focused on organic contaminants, and the consequences of exposure to other substances (particularly metals), remaining largely unknown. The aim of this study was to evaluate the potential risk of metal contamination in bat species occurring in Portugal, and to evaluate the suitability of non-lethal sampling methods. The concentration of As, Cd, Co, Cr, Cu, Mn, Ni, Pb, Se and Zn was measured in two categories of biological samples (lethal-samples: liver, heart, bone and brain; and non-lethal samples: wing membrane and fur) collected from bat carcasses of four different species (*Hypsugo savii*, *Nyctalus leisleri*, *Pipistrellus pipistrellus*, *Pipistrellus pygmaeus*). Concerning the metal concentration obtained in each sampling tissue, significant differences were found between the concentrations obtained in each species for all the metals ($P < 0.05$), except for Zinc ($P=0.223$). Significant differences were also found between organs ($P < 0.001$), metals ($P < 0.001$) and the interaction between organs and metals ($P < 0.001$). Depending on the metal, the organ/tissue that showed the highest concentrations varied, but even so fur and wing presented the highest concentrations of most of the metals. These results support the hypothesis that non-lethal samples may be useful for studies on wildlife ecotoxicology, and may help to define a protocol capable of being applied at large-scale, to investigate the risk of metal accumulation for bats. For this purpose, non-lethal samples are the best option, and as demonstrated by this study can yield reliable results. Our results therefore provide valuable insights for development of further studies, aiming to understand the importance of metals as a cause for some of the observed declines in bat populations worldwide.

MO084

Metallic element composition of egg contents and eggshells of the Kelp Gull Larus dominicanus

J.D. van Aswegen, North West University (Potchefstroom Campus) / Unit for Environmental Science and Management; L. Nel, N. Strydom, Nelson Mandela University; H. Bouwman, North-West University / Unit for Environmental Science and Management

The Swartkops River Estuary near Port Elizabeth, South Africa, is an important recreational, industrial, residential, and ecological asset, but under severe pressure. Seabirds are good indicators of trace elements within their environments. Seabirds tend to feed at different trophic levels at different distances from the land and they are long-lived. Pollutants that have accumulated in the seabirds can be excreted in various ways from the body, one being deposition into eggs. Sixteen eggs of the Kelp Gull (*Larus dominicanus)* were analysed using ICP-MS (EPA 3050b method) for 30 trace elements, for both the contents and eggshells. We selected five elements (Cr, Sr, Tl, U, Zn) to compare and assess the absolute and relative compositional patterns in egg contents and shells. Mean concentrations for Cr in eggshells and egg contents were 3.8 and 18 mg/kg dm, for Sr it was 880 and 12 mg/kg dm, for Tl it was 0.00017 and 0.00022 mg/kg dm, for U it was 0.000057 and 0.000084 mg/kg dm, and for Zn it was 2.1 and 62 mg/kg dm, respectively. Of the five elements, only Sr ($p = 0.0141$) and Tl ($p = 0.0013$) concentrations showed significant positive regressions between egg contents and eggshells. Chromium and Zn showed a positive association, but the regressions were not significant. Uranium also showed no association. The mean mercury concentration in the contents was 0.38 mg/kg dm, and the maximum was 2.1 mg/kg dm. The Toxic Reference Value for mercury in bird egg contents is 2 mg/kg dm, indicating concern about this element in the Swartkops River Estuary. Additional toxic implications, as well as comparisons with concentrations in other media will be discussed.

MO085

Heavy metals concentrations in Mediterranean Osprey eggs: variations by location, habitat and egg constituent part

F. Monti, University of Siena / Department of Physical Sciences, Earth and Environment; S. Ancora, University of Siena / Physical sciences, Earth and environment; A. Sforzi, Maremma Natural History Museum, Grosseto; N. Bianchi, University of Siena / Department of Physical, Earth and Environmental Sciences; C. Leonzio, University of Siena / Department of Physical Sciences, Earth and Environment

The osprey (*Pandion haliaetus*) has been historically used world-wide as a sentinel species for the biomonitoring of selected contaminants for aquatic ecosystems. In spite of this, occasional and fragmentary information are available for the species at the Mediterranean scale, where relict and vulnerable populations exist. In this study, we analyzed heavy metals concentrations in osprey eggs from three different populations of the Mediterranean basin (Corsica, Italy and Balearic Islands). In total, 21 unhatched eggs were collected, over a period spanning from 2005 to 2016. Mercury (Hg), Cadmium (Cd) and Lead (Pb) in osprey eggs were analyses with the aim to: (1) evaluate geographical patterns of for possible identification of inputs at the regional scale; 2) to evaluate differences in concentrations between samples from different habitats (marine environments and wetlands); and 3) to investigate any differences in concentrations among different parts of the egg (i.e. content, membrane and shell). Samples from the Balearic Islands showed higher Hg concentrations (1.4 ± 1.2 mg/kg on dry weight basis) compared to other samples. Egg shells from marine environments (Corsica and Balearics) had five times greater [Hg] than those from wetlands. Egg content and membrane showed higher Hg concentration values than those of the shell. On the contrary, for Cd and Pb (though with minor differences) higher concentrations were found in the egg shell. Our study represents a first survey at regional scale and provides a first set of data for the long-term biomonitoring of heavy metals for the vulnerable osprey populations of the Mediterranean basin \n

MO086

Interactive effects of vitamin E and BDE-47 yolk supplementation on morphology and oxidative status of yellow-legged gull embryos

M. Parolini, University of Milan / Department of Environmental Science and Policy; C.D. Possenti, B. De Felice, Università degli Studi di Milano; N. Saino, University of Milano

Oviparous mothers transfer to the eggs components that have both independent and combined effects on offspring phenotype. Functional interactions between egg components, such as antioxidants and contaminants, lead to expect that a change in the concentration of one component has effects on offspring traits that depend on the concentration of other interacting components. However, the combined effects of variation in different egg components are virtually unknown. Bird eggs contain vitamin E (VE), a major antioxidant, and also a variable amount of maternally-transferred contaminants. Polybrominated diphenyl ethers (PBDEs) are a family of brominated flame retardants that have been widely used as non-reactive additive compounds diverse commercial products. Many monitoring studies have revealed the presence of PBDEs in the biota, which can induce a plethora of adverse effects at different organisms' life stages, often mediated by the onset of oxidative stress. Although PBDEs have been found in birds and their eggs, the consequences related to the exposure to these chemicals, mainly during early development, are inadequate. In addition, no study has considered that the oxidative stress-related toxicity of these compounds may be counteracted by the presence of antioxidant molecules that mothers allocate to their eggs at the time of laying. The independent consequences of variation in the egg concentrations of VE and PBDEs on offspring phenotype, including morphological and oxidative stress effects, are largely unknown, while no study has investigated their combined effects. Thus, we manipulated the concentration of VE and BDE-47, a PBDE congener having a well-known pro-oxidant activity, in the eggs of wild yellow-legged gull (*Larus michahellis*) by administering a physiological, large (2 standard deviations) dose of VE and 150 ng/g yolk of BDE-47 both independently and in combination. We tested for effects on morphological traits (body mass, skeletal growth) and oxidative stress, as changes in total antioxidant capacity, amount of pro-oxidant species, antioxidant enzyme activity, lipid peroxidation, protein carbonylation and DNA fragmentation, in embryos soon before the hatching.

MO087

Sensitivity of freshwater pearl mussel juveniles (Margaritifera margaritifera) to different environmental and contamination factors

T. BELAMY, University of Bordeaux; A. LEGEAY, University of Bordeaux / UMR EPOC CNRS 5805; B. ETCHEVERRIA, University of Bordeaux / UMR CNRS 5805 EPOC; M. Baudrimont, Université de Bordeaux / UMR EPOC CNRS 5805

Margaritifera margaritifera is a freshwater bivalve mollusk threatened with extinction in Europe. The population of this freshwater pearl mussel has declined by 90% in Europe, during the XIXth century because of pearl fishing and from the XXth century due to habitat degradation, decrease in water quality and pollution. Nowadays, french population of *M. margaritifera* is estimated at 100,000 individuals with the largest population found in the river Dronne (Dordogne - FRANCE) with up to 15,000 individuals. Freshwater pearl musselsare considered as an excellent indicator of aquatic ecosystem health since they require high water quality and they filter up to 50 L of water a day. As a result, they are called «umbrella species», meaning that their conservation will benefit all species living in the same river. With the aim of preserving this pearl mussel, the European project LIFE « Preservation of *Margaritifera margaritifera* and restoration of river continuity of the Upper Dronne river 2014-2020 » has been set up in which a farm was created in order to produce juveniles in captivity. Some of them will be reintroduced into the environment while others will be used for ecotoxicological studies.\nThe aim of this work was to determine the sensitivity of *M.margaritifera* juveniles to different environmental and contamination factors, since they are considered as the most sensitive lifestage of this species. Acute toxicity tests were carried out on one-year-old juveniles in order to determine toxicity thresholds (LC50) of several factors such as temperature, dissolved oxygen, nitrates, phosphates and metals. Those data will allow to target reintroduction areas of juveniles produced in the farm and help the conservation strategies of *Margaritifera margaritifera* in the Upper Dronne river.

MO088

Using population modelling to reduce uncertainty - an example of a herbicide

M. Wang, WSC Scientific GmbH / Dept Efate Modelling; M. Foudoulakis, Dow Agrosciences / RSRA ERS

Uncertainty is perceived as a major acceptability criterion for higher tier risk assessment. While uncertainty does not start to be present at the higher tier (though it is generally not considered in the first tier) it needs to be addressed explicitly when higher tier refinements are applied which make the risk assessment more realistic. The use of standard higher tier refinements or even modelling is sometimes perceived as increasing uncertainty. However, in the present poster we demonstrate that refinements and population modelling help to reduce uncertainty by providing additional insight into the mechanistic understanding of risk and the ecological relevance of effects. This is done based on a risk assessment for a herbicide. It is shown how what-if questions help to address specific questions on uncertainty in the risk assessment and how a margin of safety can be calculated.

MO089

SETAC Wildlife Toxicology Interest Group

J.E. Elliott, Environment Canada / Science Technology Branch

LCIA method developments in a global perspective: Status and outlook (P)

MO090

A tool to integrate consumer and environmental exposure in life cycle impact assessment

O. Jolliet, University of Michigan; L. Huang, University of Michigan / Dept of Environmental Health Sciences; P. Fantke, Technical University of Denmark / Quantitative Sustainability Assessment Division

Direct exposure of consumers to chemical ingredients within our daily products is an important pathway that often dominates environmental performance profiles of these consumer products, but has been currently left aside in LCIA toxicity characterization. The aim of the present study is to update and extend the existing framework to consistently incorporate consumer exposure pathways in way fully compatible with existing LCIA toxicity characterization methods, and to illustrate it via a case study of plasticizer chemicals in building materials. We developed a general a framework and a tool that extends the toxicity assessment to the near-field and consumer exposure assessment and combines it consistently with the USEtox far-field environmental exposures: The chemical mass per functional unit in the consumer product is multiplied by the product intake fraction (PiF) to yields the total exposure expressed. The PiF represents the fraction of the chemical in products that is taken in by the consumer. It is determined by coupling fate processes in consumer environments (near-field) with existing environmental compartments and processes (far-field), via a consistent and mass balance-based set of transfer fractions. The developed tool already enables to calculate characterization factors for 22 types of building products, 8 types of personal care products, 7 contact food materials and multiple cleaning product-chemical combinations. The case study of DEHP plasticizer in a vinyl flooring shows that starting from a mass of DEHP in products of 82 kg, 0.15 kg will be taken in, mostly by the household users via dust ingestion as a dominant pathway. This leads to intake doses of 0.14 $mg/kg_{body}/d$ for an adult and 0.5 $mg/kg_{body}/d$ for a 3 years old child. Performing a full LCA of the vinyl flooring shows that the 16% of DEHP plasticizer in flooring are associated with dominant shares of impact on human health (78%) and on aquatic ecosystem (95%), whereas PVC is the dominant contributor to climate change impacts (59%). This case study illustrates well the importance to account for consumer exposure to chemical in product during their use. Final outcome is a consistent and quantitative framework and directly applicable tool to determine factors based on scientific consensus for assessing life cycle exposure and toxicity impacts of chemicals in LCIA, as an input to the LCIA guidance efforts of the Life Cycle Initiative.

MO091

Towards the integration of an Agent-based Model into LCA framework to assess dynamic indoor air quality

A. Micolier, University of Bordeaux / The Life Cycle Group CyVi; P. Loubet, University of Bordeaux / ISM CyVi; F. Taillandier, University of Bordeaux / I2M GCE; G. Sonnemann, University of Bordeaux / ISM CyVi

The construction sector, representing 44% of the total final energy consumption in Europe, is recognized as a major hotspot of resource use and environmental impacts. Thus, strong efforts are made to encourage the design of environmentally friendly buildings. However, the airtightness of low energy buildings has created particularly confined and polluted indoors. Indoor pollution has been raised as a major public health issue since we spend on average 80% of our time in closed spaces. Designing sustainable buildings with good indoor air quality is even more challenging since this latter is strongly influenced by occupant's lifestyle and behavior. Life-cycle assessment (LCA) is a relevant methodology to account for impacts from indoor air while avoiding potential burden shifting from the life cycle of energy or materials used. Nevertheless, the current use of LCA still faces scientific obstacles such as: (a) the inclusion of the dynamical effects of indoor pollution on human health and (b) the consideration of the behavior of the occupants. In order to address these concerns, a model of autonomous agent has been developed structured around (i) an agent-based model Li-BIM (Live in BIM) which explicitly represents human behavior, (ii) a physical model to capture the building thermal behavior, (iii) the numerical representation of the building (BIM) and (iv) an innovative indoor air quality model Be-BIM (Breathe in BIM). Li-BIM is an operational model which simulates the behavior of the occupants based on an evolved occupational cognitive and social framework. Be-BIM is currently being developed as a dynamic and localized fate model sensitive to users' behavior and the related dynamic of air emissions. Therefore, Be-BIM will (i) generate the inventory data for dynamic pollutant emissions and (ii) assess the local impacts from air emissions. Expected outcomes of our integrated model include characterization factors for human toxicity due to indoor air which are dynamic and spatially differentiated at the scale of the building. Eventually, our model will allow the comparison of life cycle impacts of different building scenarios with a specific focus on indoor air quality suited for residential dwellings.

MO093

Adding the resource dimension to the WULCA framework on assessing freshwater use in LCA

C. Pradinaud, IRSTEA Montpellier / ITAP ELSA; S. Northey, Monash University; B.M. Amor, Universite de Sherbrooke / Département de génie civil; J.C. Bare, U.S. Environmental Protection Agency / National Risk Management Research Laboratory; L. Benini, European Environment Agency; M. Berger, Technische Universitaet Berlin / Chair of Sustaiable Engineering - Office Z1; A. Boulay, CIRAIG - École Polytechnique de Montréal / Chemical engineering department; A.D. Henderson, Noblis Inc / Environmental Science; G. Junqua, Ecole des Mines dAlès / LGEI; M.J. Lathuilliere, University of British Columbia / IRES; M. Margni, CIRAIG - École Polytechnique de Montréal / Mathematical and Industrial engineering; M. MOTOSHITA, National Institute of Advanced Industrial Sci. and; B. Niblick, U.S. Environmental Proctection Agency / Life Cycle and Decision Support Branch; S. Payen, AgResearch; S. Pfister, ETH Zurich; P. Quinteiro, University of Aveiro / Department of Environment and Planning; T. Sonderegger, ETH Zurich; R.K. Rosenbaum, National Research Institute of Science and Technology for Environment and Agriculture - Irstea / UMR ITAP

Effective management of freshwater resources is recognized as being vital. At present, existing LCIA methods for water use do not entirely reflect the state of such a vital resource remaining for future generations. Thus, the objectives of this paper are to (1) identify how freshwater resources can be defined as an entity to protect within the Area of Protection (AoP) natural resources, (2) describe the impact pathways affecting this resource, and (3) propose a characterization framework to assess the impacts from the identified impact pathways. Freshwater resource has a particular status in LCA resource modeling. First, it exists in the form of three types of resources: flow, fund, or stock. Then, in addition to being a resource for human economic activities (e.g. hydropower), it is above all a non-substitutable support for life that can be affected by both consumption (source function) and pollution (sink function). Therefore, both types of elementary flows (emissions and water consumption) should be linked to a damage indicator for freshwater as a resource. In order to clearly define what is to be protected, the freshwater resource is put in perspective through the lens of three safeguard subjects. Considering the current scope of the AoP natural resources, the complex nature of freshwater resources and the dimension of freshwater to safeguard, a definition of freshwater resource is proposed. Also, a wide range of possible impact pathways to freshwater resources is identified, establishing the link between different inventories (water elementary flows, emissions and land use) and their potential to cause freshwater depletion or pollution in the long-term. The concept of recovery period is used to operationalize this framework: when the recovery period lasts longer than a given period of time, impacts are considered as being irreversible and fall into the concern of freshwater resources protection (i.e. affecting future generations). The study shows that it seems relevant to include this concept in the impact assessment stage in order to discriminate the long-term from the short-term impacts, as some dynamic fate models already do. Recommendations are also given for freshwater resource impact indicator(s). Therefore, such an indicator would allow LCIA to capture potential long-term impacts that could transparently advise decision makers about potential safe water supply issues in the future.

MO094

Considering water and soil conservation works in Life Cycle Assessment: focus on contour ridges and erosion impacts

M. Jouini, Montpellier SupAgro / Département de génie rural; R. Ciampalini, IRD, UMR LISAH; S. Follain, Montpellier SupAgro, UMR LISAH; J. Burte, CIRAD / UMR GEAU; N. Benaissa, National Agronomic Institute of Tunisia / Science de la production végétale; C. Sinfort, ITAP, Irstea, Montpellier SupAgro, Univ Montpellier / ELSA Research group and ELSA-PACT Industrial Chair

Soil is a rare natural resource and it is at the center of the main issues in agronomy, environment and land use planning. At global level, erosion is one of the major soil degradation processes and it is responsible for the decrease in agronomic potential of soils and in agricultural land surfaces. Water and soil conservation works (WSCW) are built to protect soil from erosion. The financial and environmental cost the WSCW construction is very high. However, the positive impacts of WSCW are not taken into account in Life Cycle Assessment (LCA). The objectives of this study is to intergrate the impact of WSCW on soil quality in LCA. There are different types of WSCW with different functions and they act differently on erosion process. In this study we focussed on contour ridges as a type of conservation works because they are associated to crop systems. Contour ridges are generally built in upland areas to reduce runoff and erosion, to increase on-site deposition of eroded particles and to increase local water infiltration. Contour ridges modify water and soil flows at catchment scale, so it is necessary to use a model able to calculate the inventory flow at the catchment and not only at the plot level. In this study we present a methodology to integrate the impact of contour ridges on topsoil erosion at the catchment level and to compute characterization factors in presence of such WSCW.The proposed method was applied in a case study in semi-arid context in central Tunisia (Merguellil watershed) which presents the issues of over-exploitation of water resources, accelerated land degradation and a high expansion of conservation works. In order to highlight the impact of WSCW on topsoil erosion, diffrent catchment scenarios (with and without contour ridges) and land use types were tested. For life cycle impact assessment, we focussed on two midpoint impact categories on soil quality of LANCA model : erosion resistance and mechanical filtration. The results showed how contour ridges can modify topsoil erosion process and thus the impact on soil ecological functions for several production systems. In conclusion, It is necessary to integrate the positive impacts of contour ridges in life cycle assessment but usual models are not able to evaluate them. It will be also neccessary to integrate the impact of the other types of water and soil conservation works in topsoil erosion impact modelling.

MO095

Impact of heavy metals on human toxicity using LCA: a case study for Walloon corn

S. Gerbinet, Université de Liège / Chemical Engineering; F. Van Stappen, CRAW Walloon Agricultural Research Centre; S. Belboom, ULiege; E. Pezennec, Knauf Insulation sprl.; S. Groslambert, University of Liège - Chemical Engineering / Dpt of Chemical Engineering - PEPs; A. Leonard, University of Liege

This paper aims to focus on surprising results when assessing the human toxicity of corn farming in Wallonia, Belgium. The USEtox method is applied to the farming of one hectare of corn. Local data are used for farming data and GaBi datasets are used for background data. The field emissions due to farming are calculated by the most used models. The results in human toxicity, cancer effect, underline the large contribution of chromium (Cr) emissions due to the use of organic and mineral fertilizers. But during fertilizers composition characterization only the total chromium is measured and therefore, unspecified chromium is used as emissions. However, it is known that the chromium in natural environment is most probably Cr (III) and this could really decrease the impact as the characterization factor for unspecified chromium, is, in USEtox, the average of the one of Cr (III) (non-toxic) and Cr (VI) (toxic), therefore really larger than the one of Cr (III). Therefore, a test is realized where 95% of the Chromium is Cr (III) and the rest is Cr (VI). In this case, score in human toxicity cancer effect is divided by 7, whereas this has no influence on the other results. The impact for human toxicity, non-cancer effect is mostly related to zinc emissions in soil due to the use of organic fertilizers, especially pig manure. However, zinc is abundant and is an important trace element in the human body. It is useful for growth, bone and brain development, etc. and the European Commission recommends the consumption of 7- 10 mg of zinc by person and per day. Moreover, mammals are able to eliminate zinc, therefore they are able to maintain a constant level of zinc independently of the exposure level. Consequently, only the exposure to high doses can have toxic effects. A test was made with the characterization factor of zinc equal to 0 in the USEtox model. In this case, the corn cropping obtains a human toxicity, non-cancer effect divided by 12 compared to the base case and mostly related to lead and mercury emissions in the soil. In both case, the contribution of pesticide is negligible. In conclusion, although the uncertainties about toxicity categories are well-known, this case study underlines the impact of the user hypotheses and shows that a detailed analysis of the results is essential for a critical view on the toxicity results.

MO097

Comparing ProScale Hazard Factors with USEtox Effect Factors for human toxicity

T. Rydberg, IVL Swedish Environmental Research Institute; H. Holmquist, Chalmers University of Technology

The purpose of this study is to compare side-by-side, the Hazard Factors (HF) of ProScale^{TM}, and the Effect Fators (EF) for human toxicity of USEtox^{TM}, and analyse the results, as both factors have been developed as a metric for adverse human health impacts due to toxic effects. Hazard factors in ProScale are derived based on substances classification in the GHS/CLP classification system, reflecting health effect severity based on H-phrase as basis for substance grouping in five ProScale hazard classes, and an OEL based correction factor has been introduced o account for potency within each class. The effect factor (EF) is a metric of the change in life time disease probability due to change in life time intake of a pollutant (cases/kg). USEtox^{TM} determines effect factors for carcinogenic and noncarcinogenic chemicals separately. Both methods have separate factors for inhalative and oral exposure routes. All the effect factors available in USEtox 2.0 were the starting point. The comparison was only carried out for the inhalative exposure route. The factors were then filtered into two different sets of substances. All substances having a carcinogenic effect factor were compared to the resulting Hazard factor as calculated with the ProScale method. Substances not having a carcinogenic effect factor in USEtox were compared separately. Tendencies of correlation can be identified, but differences are large. Interesting discrepancies, also of principal matters, have been identifed. The results shown are very first results from comparison of ProScale HFs and USEtox EFs for human toxicity. Further work is needed, and under way.

MO098

Integrating the Use Phase Impacts of Building Materials into Near-Field LCA Characterization

L. Huang, University of Michigan / Dept of Environmental Health Sciences; N. Anastas, US Environmental Protection Agency / National Risk Management Research Laboratory; P. Egeghy, D. Vallero, US Environmental Protection Agency / National Exposure Research Laboratory; D.E. Meyer, United States Environmental Protection Agency / National Risk Management Research Laboratory; J.C. Bare, U.S. Environmental Protection Agency / National Risk Management Research Laboratory; O. Jolliet, University of Michigan

Historically, LCA has focused on impacts with far-reaching temporal and spatial scales, and not exposures to near-field goods such as consumer products and building materials. However, with increased use of LCA to support decisions related to chemical alternative assessments, characterization of the near-field exposures to these products is becoming recognized as increasingly important. Therefore, the US EPA has developed a research project to improve such characterization. Several recent papers have suggested that the exposures to these consumer products and building materials may be significantly greater than exposures to far-field emissions, and therefore, not including these exposures may result in decisions which are unknowingly biased in a manner which could lead to increased risk. The difficulty up to this point in time has been to characterize these exposures since the product compositions are often unknown and the pathways to exposure have been poorly characterized. This poster will describe the research project, including the conceptual framework which demonstrates the methods by which the EPA intends to include exposures to these goods, the definition of Product Intake Fraction (PiF), the many exposure pathways being characterized, the methods for development of PiFs, and the data and models being recommended for a variety of populations to support this characterization. Finally, data gaps and other research needs will be discussed along with the future direction of the project. Keywords: LCA, LCIA, building materials, consumer products *Acknowledgement* - This work was conducted under US EPA Contract No. EP-16-C-000070 with the University of Michigan. *Disclaimer* - The views expressed in this abstract are those of the authors and do not necessarily represent the views or policies of the U.S. Environmental Protection Agency. Mention of trade names or commercial products does not constitute endorsement or recommendation for use.

MO099

Combined use of Mixed-Integer Optimisation and Thermodynamic, Molecular and Charge Density attributes for predicting Life Cycle Production

Impacts of Chemicals
R. Calvo-Serrano, G. Guillén Gosálbez, Imperial College London / Chemical Engineering
Process sustainability has become one of the fundamental criteria for decision making in chemical industry, being Life Cycle Assessment (LCA) the most popular method in recent years, currently being one of the most extended sustainability assessment methods. Since LCA is based on the analysis of all interactions for all the stages of the life cycle, it ends requiring large amounts of information. This information, however, can be difficult or impossible to gather, being one of the main obstacles when trying to apply LCA. Chemical industry is particularly affected, easily having thousands of interactions even for small and relatively simple processes and only information of a few hundreds. In these cases, when a full LCA cannot be applied, a simplified version is used instead. These Streamlined LCA (SLCA) follow the same basis as LCA, but generally either simplify the scope of the analysis and/or reduce the amount of information required in the assessment. The precise simplifications to be done (and the assessment discrepancy with the full LCA) have to be specifically considered for the process or activity assessed. Under these principles, we present a novel approach for the estimation of LCA impact categories associated to the production of chemicals using information of their chemical and physical properties. We propose that the physical properties of the products are directly related with the impacts generated in the production process, and that these impacts heavily contribute to the overall impact generated for the production of the chemical analysed. Previous studies demonstrated the prediction capabilities of molecular and thermodynamic attributes. Here we also consider the σ-profile of chemicals as attributes, for a better characterisation of the chemicals and therefore to the generated impacts. In addition, we propose to reduce the whole assessment methodology to linear prediction models, selecting in each model only the attributes that better describe specific impact categories. This approach has been applied to a database consisting of 83 chemicals, considering 15 molecular descriptors, 17 thermodynamic attributes and 8 σ-profile partitions, being able to obtain reasonable LCA impact estimates such as Global Warming Potential (GWP)(33.55%) or Eco-Indicator99 (EI99)(18.34%).

MO100
Development of USEtox characterisation factors for micropollutants in effluents
E. Maillard, ELSA-PACT Industrial Chair
Many substances are increasingly detected in surface waters, after their use by the human population. In most cases, these substances will exert the same effects as desired when they are originally applied, only now affecting different organisms. These effects can occur at concentrations of µg/l, which is why these substances are called micropollutants. In the context of Life Cycle Assessment, there is a need of characterising the toxicity potential of these micropollutants affecting ecosystems and/or the human population. A substance which is not characterised will not be considered in a LCA study, which may result in misguided decisions and the omission of essential environmental issues related to biodiversity and human health. The aim of this project is to develop a database of characterisation factors for the main micropollutants found in the effluent of waste water treatment plants with the USEtox model. In order todevelop this database, the following tasks are needed: identification of a priority list of substances currently missing in USEtox, while being highly relevant in the context of treated and untreated effluents; Literature review and database searches on existing data (required to calculate fate, exposure and effects) for the priority substances identified; Establishment of a database of new characterisation factors for human toxicity and ecotoxicity impact potentials. All these newly developed characterisation factors will be submitted for inclusion to the official USEtox database center.

MO101
Assessment of freshwater ecotoxicity with USEtox
M. LOT, CEHTRA; P. Thomas, CEHTRA SAS; F. Sahigara, KREATiS; M. Jacob, TOTAL SA
USEtox is determined as the reference tool for freshwater ecotoxicity impact evaluation in LCA context. By the way, it is recommended by several institutes: by European Commission for PEF/OEF project, by JRC-IES in ILCD handbook, by WBCSD in Life cycle metrics for chemical products guideline and by US-EPA in TRACI tool manual. An assessment of the relevance of the Characterisation Factor (CF) with its associated factors was realized. We found that CF appears to be significantly influenced by the effect factor (EF), implying that the 2 others factors, XF (Exposure Factor) and FF (Fate Factor), do not intervene, or very little, in the final calculation of CF. This finding is surprising because the XF and FF factors should influence the calculation of CF as they represent, significant fate adjustments: substance bioavailability (XF) and its presence in the medium (FF). Hence, the influence of each factor was analysed in more detail and some inconsistencies were noted. For EF, USEtox includes an extrapolation calculation for acute to chronic toxicity (called Acute-to-chronic Ratio). This extrapolation is not always reliable and will certainly not be applicable for substances with a log Kow>6 where acute toxicity is superior to solubility but chronic toxicity may still occur. Moreover, the value used for the ACR seems inappropriately low and should be different according to the mode of action of the substance. For XF, adsorption starts to reduce XF at log Koc around 5 whereas adsorption of organic substances is generally considered to become highly significant in ecotoxicological studies performed at low concentrations from log Koc of 4. Overall, the XF seems to be overestimated in this model for the majority of substances with a log Kow between 3 and 5. For FF, one of the parameters that most influences its calculation seems to be the biodegradation, which alone can alter the relationship between the EF and the CF by an order of magnitude (between a highly persistent and a highly biodegradable compound). However, the relative influence of biodegradation (and the other FF parameters) on the CF is too limited compared to the EF value. Indeed, the difference between the minimum and maximum biodegradation on CF is approximately 2 orders of magnitude while the EF itself spans at least 8 log units. Thus, because of these inconsistencies, it is crucial to discuss the relevance of each factor with the aim of improving the model providing a more realistic approach.

MO102
Advancing nutrient modelling in eutrophication methods for life cycle impact assessment
B. Niblick, U.S. Environmental Proctection Agency / Life Cycle and Decision Support Branch; B. Morelli, T.R. Hawkins, Eastern Research Group Inc / Franklin Associates; H.E. Golden, U.S. Environmental Protection Agency / National Exposure Research Laboratory; J.E. Compton, U.S. Environmental Protection Agency / National Health and Environmental Effects Research Laboratory; E.J. Cooter, U.S. Environmental Protection Agency / National Exposure Research Laboratory; A.D. Henderson, Noblis Inc / Environmental Science; A. Edelen, ORISE; J.C. Bare, U.S. Environmental Protection Agency / National Risk Management Research Laboratory
Anthropogenic contributions of the nutrients nitrogen (N) and phosphorus (P) threatens the health of freshwater and marine ecosystems around the world [1]. The increase in environmental nutrient availability, known as eutrophication, can lead to harmful algal blooms (HABs) and decreased levels of oxygen (hypoxia) needed to sustain aquatic life [2]. This presentation evaluates the current state of life cycle impact assessment (LCIA) methods for eutrophication in freshwater and marine ecosystems using a criteria-matrixed review of the underlying fate and transport (F&T) models. Results of the review point to several key recommendations for further scientific development of midpoint eutrophication methods in life cycle assessment (LCA). Current eutrophication methods in LCA are limited. Most LCA studies use highly simplified F&T models with continental or global geospatial resolution, characteristics of which tend to be inadequate for regional analyses and complex community-based decisions. Therefore, this critical review examines a set of surface water quality models, watershed models, marine models, and air quality models that each have potential for integration into LCIA. Factors examined include sources of nutrient loading to each environmental compartment (e.g., water compartments, soil, and air), the forms of each nutrient modeled, and the representation of each F&T mechanism. Review results suggest several possible recommendations, including the continuation of the recent trend toward separation of freshwater and marine eutrophication methods, expanded characterization of the freshwater cause-effect chain, and the development of new soil and freshwater fate factors. By incorporating findings of the F&T models into current eutrophication methods, LCA can better inform scientific decisions that affect water quality, nutrient management, and environmental policies across watersheds and global ecosystems. [1] Rockström J, Steffen W, Noone K, Persson Å, Chapin FS, Lambin EF, Lenton TM, Scheffer M, et al. 2009. A safe operating space for humanity. Nature 461: 472-475. [2] Diaz RJ, Rosenberg R. 2008. Spreading dead zones and consequences for marine ecosystems. Science 321: 926-929. *Disclaimer* - The views expressed in this abstract are those of the authors and do not necessarily represent the views or policies of the U.S. Environmental Protection Agency. Mention of trade names or commercial products does not constitute endorsement or recommendation for use.

MO103
Land Use Change comprehensive framework in LCA for microalgae cultivation systems as emerging production option in the bio-economy
L. Compagnoni, D. Marazza, University of Bologna; S. Righi, University of Bologna / Physics; E. Balugani, E. Merloni, University of Bologna
Europe is nowadays facing serious issues about natural resources depletion. Promoting the sustainable growth of bio-economy sectors will enhance the transition from a fossil- to a bio-based society. In this context, algae represent an emerging resource of great importance for their potential applications. Specifically, microalgae are currently promoted not only as fuel-sources, which have been studied since decades, but also as high-value products useful in pharmacy, cosmetics, nutraceutics as well as for aqua- and agricultural uses, hence considered as an untapped resource for a bio-based economy. In the last thirty years, several LCAs of algae-based products have been performed on a wide range of production processes. A Scopus review on “algae LCA”, indeed, reported 228 total papers published in the scientific literature since 1989, experiencing a fast-growing trend from 2010 onwards, mostly regarding biofuels (>77%). However, one impact category of the algal-based product life cycle that is commonly overlooked, while being of high importance for the bio-economy, regards land use change (LUC), with only 8% of algae’s LCA-related studies including it. Land use influences biodiversity as well as the structure and functions of ecosystems, causing damage to many areas of protection through diverse impact pathways, such as Biodiversity

Damage Potential, Ecosystem Services Damage Potential, Biotic Natural Resource Depletion and Climate Change. However, only few of these impact pathways are fully implemented in currently available LCIAs, also due to lack of significant consensus on this novel impact category. Specifically, LUC has to be carefully evaluated when assessing microalgae's cultivation systems, as they may be strongly diverse one each other, hence impacting through diverse paths. Cultivation layouts may range, in fact, from large open ponds to more compact photo-bioreactors; they may be installed either in natural environments, such as fresh water ponds or offshore cultivation systems, either in brownfield lands in an optic of redevelopment of industrial areas, hence even generating a positive effect to the environment, mostly in terms of GHG's fluxes and biodiversity. In this respect, the study aims at providing a consistent framework of the current methodology on LUC impact category and its application to bio-economy and, specifically, to microalgae's production in order to provide support to business and policy decision making.

MO104

Application of LCIA water use methods to renewable energy systems in Spain

I. Sánchez-De Castro, D. Garrain, Y. Lechón, CIEMAT / Energy Dpt Energy Systems Analysis Unit

The topic of 'water' in LCA has emerged as an important approach to quantify the related effects of water use from consumption of goods and services. Several assessment methods have been proposed by the scientific community, encompassing both the computation of water use and its impacts, but differing in the communication of results. After developing a comprehensive state-of-the-art of the methodologies to consider this impact category, this work presents the application of the most relevant methods for quantifying the water use in LCA of several renewable energy systems in Spain: i) Water Footprint standard harmonized by means of ISO 14046, and ii) the AWARE method developed by WULCA working group (http://www.wulca-waterlca.org), a midpoint water use indicator representing the relative Available WAter REmaining per area in a watershed.

MO105

Identification of methodological challenges remaining in the assessment of a water scarcity footprint

P. Quinteiro, University of Aveiro / Department of Environment and Planning; b. ridoutt, CSIRO; L. Arroja, A. Dias, University of Aveiro / Department of Environment and Planning

Freshwater stress and its implications for present and human welfare and the natural environment awaked the need to develop spatially explicit methods to calculate the product water footprint (WF) from a life cycle perspective. In this sense two parallel developments emerged: a water footprint assessment (WFA) developed by the Water Footprint Network (WFN) to map direct and indirect water use along supply chains and its relevance in water resources management; and an impact-based WF following the life cycle assessment (LCA) methodology, according to the ISO 14046. Both WFN and LCA-oriented impact assessment methods considers blue water (fresh surface and groundwater) and green water (rainfall on land that does not run off or recharge the groundwater, but is stored in the soil or temporarily stays on the top of the soil and vegetation). Although these both approaches agree on considering the degradation of water, the WFN defines grey water as the volume of freshwater that is required to assimilate the load of pollutants based on natural background concentrations and existing ambient quality standards, while LCA-oriented impact assessment methods consider the water degradation within the well establish LCA impact categories (e.g. eutrophication, aquatic acidification). This work addresses the state of development of both WFA and impact LCA-based WF approaches, identifying recent impact LCA-based WF methods, and the following methodological challenges that need to be overcome to establish a consensual and comprehensive impact LCA-based WF method: (1) accounting and assessing the potential environmental impacts of green water flows; (2) inventory of actual blue freshwater consumption in agriculture; (3) temporal and spatial variation to establish explicit characterisation factors (CFs); and (4) adequate connection between inventory flows and spatio-temporal explicit CFs. Robust and reliable methods, and guidelines for assessing and reporting WF results are needed. It is crucial to ensure the applicability of the WF by non-academia, enabling a stand-alone WF reporting for a general public audience as a means of assessing the potential environmental impacts of freshwater stress and raising awareness of a sustainable use of freshwater resources. A distorted communication of the WF results, without understanding the effects of the land-use and land cover change and water irrigation on the WF damage, and without considering spatial differentiation, can represent a danger and pitfall for decision-making.

MO106

Filling the Gap of Overfishing in LCIA: Eco-factors for Global Fish Resources

M. Stucki, Zurich University of Applied Sciences / Institute of Natural Resource Sciences; L. Eymann, ETHZ Swiss Federal Institute of Technology; R. Keller, R. Itten, Zurich University of Applied Sciences / Institute of Natural Resource Sciences

There is a lack of LCIA methods to assess the contribution of fish consumption to the global problem of overfishing. Due to this methodological gap, fish is often determined to have a lower environmental footprint than other sources of animal protein in LCA studies. The aim of this publication is to present the development of eco-factors for fish resources and by-catch as an addition to the Swiss Ecological Scarcity Method 2013 and to compare the eco-factors of different fish resources and the impacts of fishing as well as the impacts of dietary alternatives like chicken, pork, lamb, beef and veal. The sustainable use of fish resources ensures that the fish stock for each fish species exceeds the minimum stock needed for a maximum sustainable yield. Therefore, the method developed uses a distance to target approach based on the fish stock for maximum sustainable yield (FS_{MSY}) and the current fish stock (FS) for the characterisation of the use of fish resources. The eco-factors were calculated for each fish species and fishing area. The inclusion of the eco-factors shows the relevant contribution to the total environmental impact of fish compared to fishing and fish processing. Depending on the fish species, fish can have significantly higher overall impacts compared to different types of meat. The contribution of the eco-factors for by-catch is minor compared to fish resources but still relevant for the comparison with dietary alternatives. Distance to target based eco-factors using regionalised data for FS and FS_{MSY} not only follow a comparable underlying approach as suggested in literature but also enable aggregation into a single-score with other environmental impacts. The overexploitation of fish resources is highly variable for different fish species and fishing areas as well as for by-catch. When comparing fish to equivalent dietary alternatives like chicken, pork, lamb, beef or veal, consideration of the overexploitation of fish resources results in some fish species exceeding the environmental impact of dietary alternatives. Therefore, the overexploitation of fish resources is relevant in the Life Cycle Assessment of fish products in different diets. The suggested approach can be integrated in a full single-score LCIA of meals and is able to reflect the regionalised impacts caused by the overexploitation of fish resources.

MO107

Constructing life cycle inventories for the hydroelectric sector in Peru: methodological considerations and environmental impacts

I. Vázquez-Rowe, Pontifical Catholic University of Peru / Civil Engineering Environmental Science; D. Verán-Leigh, Pontificia Universidad Catolica del Peru / Civil Engineering Environmental Science

According to recent reports, hydropower currently accounts for ca. 16% of worldwide electricity generation. Low carbon emissions are usually related to this source of energy, making it an attractive option for nations with hydropower potential to meet increasing electricity demand without relying on burning fossil fuels. However, the new wave of hydropower plant construction is occurring mainly in three tropical river basins: Amazon, Congo and Mekong; therefore, an additional environmental impact must be considered: biogenic greenhouse gas (GHG) emissions due to the degradation of biogenic carbon in reservoirs. Peru is planning on installing up to 2,000 MW of installed capacity in hydropower until 2021, but the input and output flows, as well as the environmental impacts that these generate have not been explored. In this context, a set of three run-of-river hydropower plants built in the past decade located along the Peruvian Andes were analyzed from a life-cycle perspective. The main objective of the study was to generate detailed life cycle inventories for each of these three hydropower plants with the aim of having specific information for real conditions in Peru. This information was used to compute the environmental impacts linked to the generation of electricity at the plants. Although the main aim was to determine the GHG emissions linked to this process, considering the important policy implications of decarbonizing the Peruvian electricity grid, other environmental categories, such as eutrophication or the depletion of abiotic resources, were also considered. The results computed show that GHG emissions per kWh of electricity produced were in the lower range of emissions observed in the literature, in all three cases below 3 g CO_2eq per kWh. Biogenic emissions represented less than 5% of the total GHG emissions despite their location in a tropical nation, due to the arid conditions of the landscape in the Highlands, as well as the mild temperatures that are present in the reservoirs. Results intend to be of utility for an array of applications, including relevance in decision-making in the energy sector, policy-making at a national level, considering the implications in terms of meeting the Nationally-Determined Contributions to mitigate climate change in the frame of the Treaty of Paris, and for the international LCA community in an effort to expand the amount of inventories available for different geographical and technological conditions.

MO108

Global scale characterization factors for freshwater eutrophication from nitrogen and phosphorus emissions to water and soil

F. Brands, Radboud University; R. University, Radboud University Nijmegen / Department of Environmental Science; A. Beusen, PBL; R. Van Zelm, Radboud University / Department of Environmental Science

Eutrophication is a key water quality issue triggered by increasing nitrogen (N) and phosphorus (P) levels and potentially posing risks to freshwater biota. In life cycle impact assessment, spatially explicit characterization factors (CFs) of phosphorus emissions to freshwater causing eutrophication have been derived. However, for nitrogen impacts, no efforts have been undertaken yet. Moreover, neither for agricultural emissions of P nor for N spatially explicit CFs have been derived. Therefore, the goal of this research was to determine spatially explicit CFs for freshwater eutrophication due to nitrogen as well as phosphorus emissions from wastewater treatment plants (WWTPs) and agriculture on the global scale. CFs were defined as the change in potentially not occurring fraction of species (PNOF) due to a change in the river basin-specific emission of P or N to freshwater (via WWTPs) or agricultural soil and consist of a fate and an effect factor. To determine the fate factors, the change in N and P concentrations resulting from agricultural as well as from WWTP emissions in year 2000 compared to year 1900 were separately modeled for every river basin in the world. Effect factors were based on log−logistic relationships between the PNOF (dimensionless) of heterotrophic species and total P (TP) or NO_3^- concentrations. The PNOF − concentration relationships were determined using data on the highest concentration where a species was observed in field surveys. Our work provides the opportunity to quantify worldwide spatially-explicit eutrophication impacts due to nitrogen as well as phosphorus from soil as well as from freshwater emissions, in a coherent way. The factors can be applied to determine eutrophication impacts of products in LCIA, as well as to determine country-specific eutrophication footprints using multi-regional input-output (MRIO) analysis.

Building of large-scale inventories of emissions and resources and applications for environmental footprints of territories, nations and sectors (P)

MO109

Carbon and material footprint of consumption in Flanders - an input-output based assessment

A. Vercalsteren, VITO NV; K. Boonen, M. Christis, VITO; E. Vander Putten, VMM-MIRA

Environmental footprints of a country or region are a measure for the environmental impact that is caused worldwide by national or regional consumption. The Flemish Environment Agency (VMM) asked VITO to assess the carbon and material footprint of consumption in Flanders (region of Belgium) and identify hot spots and bottlenecks, based on the Flemish environmentally extended input-output model. The Flemish EE-IO model is based on specific monetary and environmental data for the Flemish region of Belgium and is part of an interregional IO-model in which trade with the Brussels and Walloon Region is modelled in IO-tables. Imports from outside Belgium are included via the link with the use table of Exiobase. A monetary consumption matrix for households divides the final demand vector of households in different consumption categories. In 2010 the carbon footprint of Flanders amounted to about 20 tonnes per inhabitant. Nearly three quarters of the carbon footprint are linked to household consumption, mainly caused by the production and transport of the goods and services consumed. Three quarters of the carbon footprint of goods and services purchased by households are linked to housing, food and personal transport. Whilst the majority of the greenhouse gas emissions, primary materials and employment is outsourced, the added value linked to Flemish consumption is mainly created in Flanders. The presentation will introduce the overall results of the carbon and material footprint assessment of Flemish consumption in 2010 and go more in detail into the value chain impact of some household consumption activities e.g. food consumption. The relation between carbon and material footprint, geographical and sectoral distributrion of different production chains and consumption activities, and the relation with added value and employment created by Flemish consumption will be discussed. As the model is available for 2003, 2007 and 2010, the evolution over these years will also be presented. Conclusions will mainly focus on methodological issues and policy implications that follow from this analysis. To achieve the greatest possible global environmental benefit, it is not enough to focus on a country or region's boundaries alone. There is also a need for a policy that is aimed at making production chains and consumer behaviour more sustainable, including internationally harmonised calculation methods and targets of footprints to evaluate the results.

MO110

A cross-country analysis of relationship between economic structural change and CO2 emissions.

K. Shironitta, Kyushu University / Economics; S. Kagawa, Kyushu University; Y. Kondo, Waseda University

In this study, we focus on the effects of changes in material and energy input structure on the life-cycle CO2 emissions (i.e., consumption-based emissions). Previous studies demonstrated the connection between domestic structural changes including the shift toward a service economy and the increase in greenhouse gas (GHG) emissions embodied in consumptions of a specific country (United States or Japan) (Suh, 2006; Nansai et al., 2009). This study is an important follow-up research that examines the environmental effects across countries and evaluates whether or not the development levels of countries can explain those environmental effects. Specifically, we employed a multiplicative structural decomposition analysis based on the World Input-Output Database (WIOD) during 1995 to 2008 (Dietzenbacher et al., 2013) and decomposed life-cycle CO_2 emissions of 40 nations into the following four inducement sources: (i) inputs from material goods (including energy) to material goods, (ii) inputs from material goods to services, (iii) inputs from services to material goods, and (iv) inputs from services to services. From the results, we found that the role of inputs from material goods to services and inputs from services to material goods in increasing life-cycle CO_2 emissions have become more important than inputs from material goods to material goods in not only developed countries but also developing countries. Services are sustained by manufactured goods, so manufactured goods are necessary for the continued growth of service sectors. In a developed country, a large proportion of its GDP is generated by tertiary sector activities, and the role of service sectors in economic growth will expand more and more. In developing countries too, since the proportion of production from primary and secondary industries will shrink and that from tertiary industries will rise with their economic growth, the importance of service sectors will be enlarged. For this reason, it is essential to focus on the production systems of service sectors to reduce domestic CO2 emissions. It is especially important to sift to the industrial structure with less emission-intensive material goods.

MO111

Influence of substance coverage on impacts from the electricity sector

A.S. Leclerc, DTU / Management Engineering; M.Z. Hauschild, Technical University of Denmark / DTU Management Engineering Division for Quantitative Sustainability Assessment; R. Wood, Norwegian University of Science and Technology / Biology; A. Laurent, DTU / Division for Quantitative Sustainability Assessment DTU Management Engineering

The electricity sector is a major source of emissions of greenhouse gas, but also heavy metals, dioxins or radioactive isotopes. However, most environmental assessments of the electricity sector at national or global scale focus solely on climate change and do not include other environmental impact categories such as particulate matter formation or toxic impacts on human health. At the national scale, the few available databases are limited to a narrow substance coverage. For example, official reports of pollutants emissions to the European Monitoring and Evaluation Programme (EMEP) should cover 23 substances in 51 countries, but they are not always complete. The Multi-Regional Input-Output database EXIOBASE includes environmental extensions emitted to air in 44 countries and 5 regions but only for 33 substances. In comparison, the database Ecoinvent provides emission data for hundreds of substances in the unit process inventories for electricity and heat generation. Here, we aim to to develop a globally consistent and extensive dataset of airborne emissions from electricity production to get a more realistic coverage of toxicity impacts in large-scale life cycle assessments (LCAs). We thus built the *Ecoinvent-based National Energy-related Emission Inventory (ENEEI)* by upscaling processes from Ecoinvent 3.3 with national production volumes of electricity and complementing it with emission data from external sources. The resulting inventory *ENEEI* covers 229 substances, including 51 radioactive isotopes. By comparing inventories and databases at midpoint level, we show that LCAs using Ecoinvent may underestimate the toxicity impacts associated with electricity production by a factor ranging from 1.4 to 1.9, while Exiobase may cut them off by up to 4 orders of magnitude in some countries. This demonstrates the importance of having an extensive substance coverage to fully represent the environmental impacts of electricity production.

MO113

Mapping the carbon, air pollution, and biodiversity footprints of nations: A GIS + global supply chains

K. Kanemoto, Shinshu University / Faculty of Economics and Law; D. Moran, Norwegian University of Science and Technology

"Spatial footprinting" is an approach for locating the actual hotspots where impacts driven by consumption occur. Spatial footprinting offers the potential to link any remote sensing or earth observatory GIS data that is tagged to an economic sector to any multi-regional input-output (MRIO) based economic model. We present new method for locating at a subnational level the environmental emissions induced by global supply chains. As the world economy becomes more complex it is increasingly difficult to connect consumers and other downstream users to the origins of their GHG emissions and other impacts. Given the important role of subnational actors in GHG abatement and other environmental protection efforts, it is advantageous to connect consumers to the locations where their purchases are driving environmental pressure. We present spatial footprint results for 187 countries showing the footprint of GHG emissions, air pollution hotspots, and biodiversity threats, and discuss our spatial footprinting methodology.

MO114

LCA data machine applied

A. Ciroth, GreenDelta; M. Srocka, GreenDelta GmbH

In any LCA study, finding data sets that are "fit for purpose" is probably one of the aspects which consumes most time and effort, and the data sets which are used have obviously a direct implication on the stability of the result achieved. This high effort for finding and creating data sets is true despite the existence of several comprehensive, generic databases for LCA. It is underlined by the presence of intransparent, outdated, or out-of region and context data set in these databases. For this reason, an "LCA data machine" has been developed at GreenDelta in the last 3 years, and was already presented earlier at conferences. The LCA data machine automatizes creation, update, and to some extent also review of data sets in LCA and sustainability assessment. Data sets are created to meet several specific requirements, e.g. related to region, time, or nomenclature system, but can also be created so that they meet requirement sets, such as, for example, related to PEF. Meanwhile, the "DAMA" has been applied to various sectors, products, and data sources. The presentation will summarise key steps of the development and will demonstrate the DAMA, for specific data sets and also for specific use cases. The LCA data machine will be demonstrated in three different application cases: 1) finding and if necessary creating data sets in situations where no data set is directly available, i.e. for data gaps: Paper machine example 2) creating a data set as copy of an existing process, to adapt to specific, local needs: Creating soy bean production for India from soy bean production US 3) Product comparison, identification of differences between compared products The approach with DAMA will also be compared to approaches currently used in LCA, with examples from the PEF remodeling project and others. The LCA data machine has the potential to truly change the curent approach for data set creation, exchange, and also use, in LCA and related areas, and can be especially useful for creating inventories in a larger scale.

MO115

Static and dynamic modeling of high performance buildings: Comparison of average and marginal electricity mixes, a consequential effect on LCA results

M. Bilec, W. Collinge, University of Pittsburgh / Civil and Environmental Engineering; H. Rickenbacker, University of Pittsburgh / Civil & Environmental Engineering; A. Landis, Clemson University / Environmental Engineering and Earth Sciences; C. Thiel, New York University School of Medicine / School of Population and Public Health

Traditional life cycle assessment (LCA) involves explicit assumptions and major uncertainties associated with the source of electricity across two main dimensions: spatial and temporal. There is a need within the LCA community for an approach that addresses this ambiguity and allocates environmental impacts as a function of marginal and time-specific variations. In our study, particular attention is paid to the dynamic characteristics of two buildings' electrical energy consumption in relation to regional power generation sources. Our LCA models incorporate metered hourly energy use data for on-site renewable production at a net-zero energy building (NZEB), and hourly or sub-hourly electrical energy usage data at a LEED Gold building; both are situated in an energy conservation district located in Pittsburgh, PA. Seven iterations of both static and dynamic life cycle assessment (DLCA) based-models were performed and evaluated based on building energy use (predicted vs. observed), electrical grid time resolution (yearly, monthly, hourly), and region-specific electrical grids and data sources (Environmental Protection Agency, Department of Energy). Our results illustrate that the use of photovoltaics at the NZEB produced excess electricity by on-site renewables which is distributed back to the grid and can be interpreted as avoided upstream emissions (generation at the power plant), which in some cases may offset or erase initial material phase impacts. The marginal consequential model improved the payback period by an order of magnitude (12.5 years to 3.0 years). Additionally, the dynamic scenarios explored in this study were able to effectively account for the growth in natural gas generation, assigning or ignoring emissions based on a marginal increase or decrease load during the building's energy use. The LEED Gold building is solely reliant on the regional electricity grid, making our findings indicative of a market-as-usual scenario and therefore comparable and/or scalable to other building studies. No studies combining time-resolved building electrical usage with time-resolved grid electricity production have been found in a U.S. context. There are notable differences between the European and North American power grids, on that account this approach aides to the advancement of DLCA research domestically.

MO116

Life cycle framework for environmental assessment of public transport systems

A. Shinde, Indian Institute of Technology Bombay, Mumbai, India; A. Dikshit, Indian Institute of Technology Bombay, Mumbai, India / Centre for Environmental Science and Engineering (CESE); R. Singh, Thinkstep Sustainability Solutions Pvt. Ltd., Mumbai, India

Several studies have assessed life cycle environmental impact of public transport systems. However, there is no single platform, software or tool for comparing the environmental impact of urban commuting modes. The objective of this study was the development of an LCA based framework to evaluate, analyze and compare the life cycle environmental impact of public transport systems. The modular and parameterized system of GaBi 6.5 platform has been used for exploring the life cycle environmental performance of commuting trains, public bus transport and intermediate public transport (IPT) modes viz. taxi, auto-rickshaw. The system boundary comprises the life cycle environmental impact construction and maintenance of transport infrastructure, manufacturing and maintenance of the vehicle, energy/fuel production and tail-pipe emissions. This framework is capable to characterize the per passenger km travelled (PKT) and vehicle km travelled (VKT) environmental impacts of an existing and proposed project. Since per PKT environmental impact of public transport systems are highly sensitive to ridership levels, the environmental impact of public transport systems and IPT modes has been compared for off-peak, average and peak hour levels of ridership. This framework has been developed on the basis of extensive data collected for the material and energy required for the construction and maintenance of infrastructure, manufacturing and maintenance of rolling stock, use phase impacts of fuel/energy production and tail-pipe emissions, in addition to the transport of raw materials. Therefore, this framework is highly comprehensive but it is also amenable to future additions and expansions. The regional transport authorities can proactively address the target areas for improving the environmental performance of their transportation system, and ultimately the competitiveness of their network. The regulatory authorities will have the information to improve on their policies to reduce environmental impacts associated with each mode at each stage.

MO117

Environmental impact assessment of rail freight intermodality in Belgium using the Life Cycle Assessment methodology

A.L. MERCHAN, University of Liege / Chemical Engineering, PEPs; S. Groslambert, University of Liege / Chemical Engineering; A. Léonard, Liège Université / Chemical Engineering - PEPs

BRAIN-TRAINS is a project supported by the Belgian Federal Government that deals with the possible development of rail freight transport in Belgium, analysing the current situation of the intermodal freight transport from an interdisciplinary perspective. The objective of increasing the rail freight transport is linked to the European Commission's White Paper on transport (2011), which aims to shift the 30% of road freight over 300 km to other modes of transport more energy-efficient such as rail or waterborne transport by 2030. In the framework of the BRAIN-TRAINS project, the Life Cycle Assessment (LCA) methodology has been chosen to analyse the environmental impact of the intermodal rail freight transport in Belgium. In a first stage we have carried out the LCA of rail freight transport (distinguishing between electric and diesel traction), inland waterways transport and road freight transport independently. In a second stage we have carried out a study of the environmental impacts related to intermodal rail freight transport. For this, we have studied several consolidated intermodal rail-road routes in Belgium. The aim of this analysis is to compare the environmental impacts of these intermodal routes depending on the freight transport mode chosen (rail or road transport) for the major part of the intermodal route. Finally, we have analysed how the increase of rail freight transport in the modal split as a result of the possible development of the intermodal rail freight transport affects the environmental impacts of inland freight transport in Belgium. For this, three divergent Belgian scenarios with a time frame set in the year 2030 have been built for further analysis. These scenarios are directly linked to the third strategic goal of the European Commission's White Paper on transport (2011). As a result, a best, worst and medium case scenarios have been developed, depending on whether the 30% shift will have been successfully accomplished, the status quo will have been maintained or the goal will not have been completely reached by 2030, respectively. The results obtained in this research will be used to build plausible scenarios for rail freight development in order to define the sustainability impact of future intermodal transport. They could help in making optimised policy decisions relative to the development of intermodal transport in Belgium, including environmental aspects and allowing the reduction of emissions in the transport sector.

Modelling and monitoring of pesticides fate and exposure in a regulatory context (P)

MO119

Quantifying visual assessment of kinetics - Development of an objective criterion to support visual assessment of SFO fits of parent soil degradation

J. Witt, Bayer AG / Environmental Safety; S. Beulke, Enviresearch Ltd; S. Ford, JSC International Limited; D. Patterson, Syngenta; B. Erzgraeber, BASF SE; M.A. Thomas, Monsanto Company; I. Hardy, Batelle; R.L. Jones, K. Hammel, Bayer AG, Crop Science Division / Environmental Safety; J.A. Hingston, Chemicals Regulation Division

Kinetic evaluation of soil degradation studies for parent compounds is a key step to derive degradation endpoints. For modelling endpoints, single-first order (SFO) kinetics is preferred when acceptable, because it is implemented in exposure models. In presence of some bi-phasic tendency, acceptability of SFO is a recurrent source of discussions in the regulatory context. FOCUS kinetics guidance proposes Chi2err < 15% and visual assessment as decision criteria. However, the Chi2err may be misleading as it does not account for systematic deviations, while visual assessment is inherently subjective. In the framework of a group led by UK CRD, to update FOCUS kinetics guidance, we aimed at finding criteria to quantify visual assessment. We collected 40+ example soil degradation studies that were assessed separately by 4 experts based on visual assessment, using scores between 0 (clearly SFO) and 10 (clearly bi-phasic). Individual scores showed high variability, confirming the subjectivity of visual assessment. Based on group discussions, we derived group consensus scores. Consensus scores showed little correlation with Chi2err ($R^2 = 0.23$). Among several proposed criteria, the SWARC (scaled and weighted area under the residue curve) criterion showed the best correlation to the consensus scores ($R^2 = 0.77$). SWARC was specifically developed for this task. The residue curve is split into blocks of the same sign (i.e., over- or underestimating the measured data). The absolute area under the curve in each block is weighted depending on the number of residues and summed up. The result is normalised by the study duration; a scaling factor accounts for high deviations from the last data point. Thus, the criterion mimicks the visual assessment process, taking into account the presence and size of systematic deviations, and whether the model adequately predicts the last data point, as a measure for extrapolation capacity. We find that SFO fits with SWARC < 40 can be considered clearly acceptable; for higher SWARC values, SFO may still be acceptable (particularly if SWARC < 65), but DFOP should also be assessed. Testing of the criterion for metabolite fits showed that, in spite of a weaker correlation to consensus score, the criterion can also be useful for metabolites. Taken together, we provide a novel tool that quantifies the visual assessment of SFO fits. This can guide decision making and thus help to reduce subjectivity in regulatory assessments.

MO120

"Southside"- Bridging the hemispheres - Global use of field trials based on ecoregion similarities between New Zealand, Chile and Europe

B. Gottesbueren, BASF SE / Crop Protection, Environmental Fate Modelling; H. Bayer, BASF SE; K. Platz, BASF SE Agrarzentrum Limburgerhof / Environmental Fate Modeling; B. Erzgraeber, BASF SE; F.P. Donaldson, BASF Corporation / APD/EFR; J. Goulet-Fortin, BASF SA; F. Kröger, Eurofins Agroscience Services GmbH

In European regulations degradation rates in soil (DegT50) from terrestial field dissipation studies TFD studies considered for exposure modeling may originate from "any" sites with soil and climatic conditions similar to Europe. An OECD Ecoregion similarity model (ENASGIPS) had been developed (OECD 2016) for gaining acceptance of field studies conducted in North America to Europe and vice versa, and there is no obvious scientific reason why this cannot be extended to other regions of the world. An experimental and GIS/modeling feasibility study ("Southside") was initiated to demonstrate if TFD studies conducted in the Southern hemisphere (i.e. New Zealand, Chile) under climatic, soil and cropping conditions similar to conditions in the Northern hemisphere may deliver similar soil degradation rates and DegT50 endpoints than those from Europe. Similar similarity zones were identified between the New Zealand and Chilean sites and EU / NAFTA using the OECD ENASGIPS tool as well as an adapted GIS crosswalk with JRC-EFSA climate and soil maps for EU. The trial sites had soil types ranging from loamy sands, sandy loam, loam and silty loams. In New Zealand the sites were located on the Northern Island having an average annual air temperature of ~ 12-13 °C and an average cumulative annual rainfall of ~ 780- 970 mm. In Chile the sites were located in the Región del Bío-Bío east of Concepción having an average annual air temperature of ~ 14 °C and an average cumulative annual rainfall of ~ 800-900 mm. The terrestrial field dissipation (TFD trials) were conducted according to OECD 232 (DegT50 module, soil covered with sand) with different pesticides at 3 sites in New Zealand and in Chile, having no historic use of these pesticides. All pesticides were applied in commercial formulations as a tank mix together in the same spraying on the same field plots at the same time The field DegT50 were normalised to reference conditions (20°C, moisture pF2) during kinetic analysis according to FOCUS, considering local soil conditions and weather data to estimate soil temperature and soil moisture with the PEARL model (as had been done with the EU studies). The quality indicator values of curve fit to data (Chi^2-error) were found to be acceptable. The normalized SFO DegT50 in the "Southside" trials in New Zealand were found to be in the range of those from TFD studies in Europe using the same study design.

MO121

Residues of currently used pesticides in Central Europe arable soils: status quo, reasons and consequences

J. Hofman, Masaryk University, RECETOX / Faculty of Science, RECETOX; P. Kosubova, Central Institute for Supervising and Testing in Agriculture; S. Polakova, Central Institute for Supervising and Testing in Agriculture / Official control section; M. Hvezdova, Masaryk University / Research Centre for Toxic Compounds in the Environment (RECETOX); L. Brodský, Charles University in Prague; L. Bielská, RECETOX, Faculty of Science, Masaryk University / Faculty of Science, RECETOX; K. Brandstätter-Scherr, University of Natural Resources and Life Sciences / Institute for Environmental Biotechnology, Department for Agrobiotechnology (IFA-Tulln); P. Dinisová, AQUATEST Inc.; Z. Simek, L. Skulcova, Masaryk University / Research Centre for Toxic Compounds in the Environment (RECETOX); M. Šudoma, Masaryk University; M. Sáňka, Masaryk University / Central Institute for Supervising and Testing in Agriculture; M. Svobodová, L. Krkošková, J. Vasickova, Masaryk University / Research Centre for Toxic Compounds in the Environment (RECETOX); N. Neuwirthová, Masaryk University

Current agricultural management is usually based on high consumption of pesticides which may bring a lot of environmental problems. Alarming results from monitoring pesticide residues in EU groundwater and surface water evoke the question of whether the arable soil can contain significant contamination as a result of the intensive use of pesticides in the present or past. Therefore, in 2014 - 2017, agricultural soil was monitored at more than 100 locations in the Czech Republic for more than 50 representatives of currently used pesticides, their selected transformation products and also banned atrazine and simazine with their transformation products. The results showed that the contamination of the monitored soils with the analysed pesticides is quite extensive. At least one pesticide was detected in 99% soils and in 81% soils the concentration of at least one pesticide exceeded the threshold of 0.01 mg/kg. The soils also frequently contained multiple residues: 85% soils contained 3 or more pesticides and 51% soils 5 or more pesticides. Over half the soils (53%) contained at least 2 pesticides exceeding 0.01 mg/kg. The most frequent compounds were triazine herbicides (present in 89% soils), which were also in significant concentrations (47% soils with triazine sum exceeding 0.01 mg/kg). Based on the association with the occurrence of terbuthylazine and crops, it was confirmed that banned toxic simazine is still introduced significantly to the soils as an allowed impurity of massively applied terbuthylazine. Persistent atrazine residues are still a legacy of the past, even over 10 years after its last use. The second most frequent compounds were conazole fungicides (present in 74% soils; 53% soils with conazole sum exceeding 0.01 mg/kg). Although no health or environmental risk analysis has yet been carried out on the data, the results draw attention for potential impacts, because: (a) foreign limits based on risk calculations have often been exceeded; (b) many of these substances are suspected carcinogens or endocrine disruptors; (c) substances occur in mixtures whose (eco)toxicity may be additive or even synergistic. The research was carried out with the support of the GACR (project 15-20065S).

MO122

Does the TOXSWA model simulate reliable concentrations in FOCUS surface water scenarios for a single segment water layer ?

P.I. Adriaanse, Alterra Wageningen University and Research Centre; W. Beltman, Alterra Wageningen UR

For the aquatic risk assessment of pesticides according to EU Regulation 1107/2009 exposure concentrations are calculated in ponds, ditches and streams in ten FOCUS surface water scenarios distributed across the EU (https://esdac.jrc.ec.europa.eu/projects/surface-water). Currently, these scenarios are based on simulation periods of 12-16 months, so only one application year. However, for more realistic probabilistic assessments a simulation period of about 20 years seems more appropriate. This will result in significantly increased simulation times. For the TOXSWA model simulation times may raise up to approximately 5 minutes for ponds, 15 minutes for ditches and one hour for streams. We investigated whether it would be possible to reduce the simulation time without compromising the accuracy of the predicted concentrations. In the current FOCUS scenarios TOXSWA uses segments of 30 m (ponds, 1 segment), 10 m (ditches, 10 segments) and 5 m (streams, 20 segments) in the numerical solution of the pesticide mass balance describing the concentration in the water layer. This allows to e.g. create concentration profiles as a function of distance in the ditches and streams. To reduce the simulation time rigorously, we cut down the number of segments in the water layer of ditches and streams to one segment. Next, concentrations calculated with a single segment for the water layer were compared to the maximum concentrations in the most downstream segment of the ditch or stream as used in the FOCUS scenarios. We considered maximum and 7 d time-weighted average concentrations both in water and sediment for a range of fictitious compounds. Initial simulations for the 12 and 16 months demonstrated that simulation times greatly reduced by replacing the standard FOCUS segmentation in the water by a single water segment (still coupled to the standard 14 sediment segments). For the water layer we found that instantaneous peaks lowered up to 11% for ditches, but less than 2% for streams. For the sediment peak concentrations changed up to 20 or 30%, indiscriminately for ditches and streams. Consequently, time-weighted average concentrations also changed, up to 7% both in water and in sediment. Based upon these initial calculations replacing the standard 20 segments in FOCUS streams by one segment could be applied to obtain accurate concentrations in water, while significantly reducing simulation times.

MO123

Recent development of approaches for quantitative use of surface water monitoring data in aquatic exposure assessments

W. Chen, Syngenta Crop Protection, LLC; P. Mosquin, J. Aldworth, RTI International

Current pesticide regulatory ecological exposure assessments conducted by the U.S. Environmental Protection Agency are almost exclusively based on standard scenario computer modeling. Monitoring data may exist from targeted (prospective or retrospective) programs and/or general water quality research by industry, governments, and academic organizations. However, use of the monitoring measured data has been limited in the regulatory assessment process to refine/inform modeling. The limited use of water monitoring data is largely due to variability in the monitoring program sampling designs (frequencies, timing etc.) and insufficient information regarding the exposure conditions and the context setting of the vulnerability of the monitoring location relative to a broader regional extent. In this paper, we summarize a set of recently developed approaches to infer and quantify realistic pesticide exposure potential based on monitoring data, including bias factor (BF), universal kriging (UK), and survey statistics. These approaches can be used in a systematic way to provide a useful reality check for comparison with exposure model output in regulatory assessments, thus increasing confidence in decision making. Examples of applying these approaches are provided to demonstrate their usefulness for watershed scale assessments.

MO124

Multi-year evaluations in the FOCUS Surface Water assessment - results of beta testing

D. Weber, M. Brauer, Eurofins Regulatory AG / Environmental and Ecological Modelling; A. Bolekhan, Bayer AG, Research & Development, Crop Science; G. Spickermann, ADAMA Deutschland GmbH; D. Schaefer, Bayer Crop Science / Environmental Safety

The calculations of the predicted environmental concentrations (PEC) of active substances in surface water are based on a "single year" approach with an initial 6 year warm-up phase followed by 16 months of the year selected by the FOCUS group. Unlike in groundwater with a 20 years assessment period, surface water exposure calculations based on a "single year" approach can be strongly affected by individual rainfall events (EFSA, 2013) which was discussed repeatedly by authorities, industry and academia (Klein, 2013, Goerlitz, 2015, Bach *et al.*, 2016, Poulsen, 2016). This presentation provides background on the technical methods and assumptions currently implemented into a software tool (Weber *et al.*, 2017) that allows 20-year simulations of FOCUS surface water scenarios. In addition, results of a beta test including revealed technical issues, problems and assumptions are discussed. The software tool in its current state can easily be adapted to updated technical requirements or changes, *i.e.* any comments from official side (EFSA FOCUS Repair Group) or from other sources can be addressed according to given consensus. The aim is to contribute to the development of an improved and generally accepted approach for surface water calculations representing a realistic worst case based on a robust evaluation. Bach M *et al.* (2016): Pesticide exposure assessment for surface waters in the EU. Part 1: Some comments on the current procedure. Pest Manag Sci 2016; 72: 1279–1284. April 2016 Goerlitz G (2015): Multiyear FOCUS surface water modelling: Options and Proposals for Realisation. XV: Symposium in Pesticide Chemistry. Piacenza, Italy. September 2015 Klein M (2013): Long term surface water simulations using the FOCUS scenarios. Pesticide Behaviour in oils, Water and Air, York, UK. September 2013 Poulsen V (2016): Higher tier assessments of aquatic and terrestrial studies. AGCHEM Forum, Barcelona. September 2016 Weber *et al.* 2017: Multi-Year evaluations in the FOCUS Surface Water assessment. Conference Pesticide Behaviour in Soils, Water and Air, York 2017.

MO125

Spatial and temporal explicit catchment modelling in aquatic risk assessment using the modular framework CMF

S. Multsch, F. Krebs, S. Reichenberger, DR. KNOELL CONSULT GmbH; S. Heine, Bayer Ag / Effect modelling; P. Kraft, L. Breuer, Justus Liebig University Giessen / Chair of Landscape, Water and Biogeochemical Cycles; T. Schad, Bayer Ag / Environmental Modelling

The EFSA Guidance Document on Aquatic Risk Assessment indicates a key role for effect modelling in future aquatic risk characterisation in a tiered risk assessment framework. Such approaches require correspondingly adapted exposure tools and scenarios ranging from simple edge-of-field to spatiotemporally explicit landscape-scale catchment models. These approaches should be sufficiently flexible and transparent in order to design lower- and higher-tiers of consistent protection level. Current models like SWAT or MIKE-SHE come with a fixed model structure which makes adaption to tiers of different complexity difficult. Flexible and modular approaches are needed to provide a spatially and temporally explicit aquatic exposure pattern to investigate effects on organisms according to Specific Protection Goals. A flexible and modular catchment model for water and pesticide transport has been developed which allows for stepwise adaption of model complexity to address tiered risk assessment problem formulations. The approach is based on the hydrological programming library CMF. Core functions of CMF are implemented in C++ and specific catchment setups are designed by Python scripting. The current approach focuses on the following abilities in order to investigate landscape-scale interactions: (a) a modular programming structure that enables replacement of process descriptions and (b) an incorporation of additional models; (c) an up-to-date connection between models at memory level in order to ensure high computing performance. A landscape is represented by the following components: Vertical water fluxes in fields are modelled with Richards equation, with the soil profile discretised into 24 soil layers. Each field holds a surface water, a groundwater and an optional drainage storage which are connected by a kinematic wave with the river bodies. Plant growth (phenology, leaf and root growth, water uptake) is modelled with similar methods and parameters as in the model MACRO 5.2. The setup was tested for a 350 ha catchment in Belgium under intensive arable use with detailed information on farming practice and observed discharge as well as herbicide loads at the catchment outlet for a time period of almost four years. The predicted environmental concentrations were used as input for an effect model in order to investigate the impact of the herbicide loads on the aquatic plant *Lemna* at population level.

MO126

Determination of runoff and drainage triggers for PEC surface water using automated simulation with FOCUS models

B. Kind, A. Guckland, J. Kleinmann, WSC Scientific GmbH

For the zonal registration in the EU predicted environmental concentrations in surface water need to be simulated based on the FOCUS models. Three different entry paths are considered: runoff (simulated in PRZM), drainage (MACRO) and spray drift (SWASH drift calculator). While the latter only depends on the amount sprayed, the distance to the water body and the spray equipment used, runoff and drainage amounts are also triggered by substance properties, e.g. degradation in soil and adsorption to soil. Often, a lot of runs need to be simulated for different crops or application timings to proof a safe use of plant protection products as defined in the Good Agricultural Practice (GAP). For this poster we evaluate the FOCUS scenarios compared to the substance properties DT_{50} and K_{OC}. The idea is to find DT_{50} and K_{OC} values which trigger runoff and drainage events and to distinguish worst-case FOCUS scenarios for different DT_{50} and K_{OC} values. Dummy substances will be created which have different values for K_{OC} and/or DT_{50} in soil. The remaining properties will be identical for each K_{OC}/DT_{50} variation. Using automated FOCUS surface water simulations PECsw values were calculated for different crops at different application times without consideration of spray drift as entry paths to focus solemnly on drainage and runoff. The results for different K_{OC}/DT_{50} values of a single scenario were compared to identify a trigger value for runoff or drainage in this scenario. Furthermore, the results of different scenarios for a single substance will be compared to find the most sensitive scenario for these K_{OC}/DT_{50} values. Finally, the amount of simulations necessary to show a safe use might be reduced to certain worst-case scenarios depending on the DT_{50} and K_{OC} properties of the substance.

MO127

Quantitative exploitation of passive sampler data for pesticide mass flow calculation in catchments and exposure risk evaluation

T. Galle, Luxembourg Institute of Science and Technology; M. Bayerle, D. Pittois, V. Huck, Luxembourg Institute of Science and Technology LIST

Pesticide monitoring remains the blind spot in WFD monitoring schemes because of the episodic occurrence of their emissions following application periods. Full coverage of relevant exposure periods is logistically impossible on a larger scale with classical monitoring methods like grab or automatic sampling. Passive samplers provide a cost-effective solution that is deployable in great numbers allowing thereby a good spatial resolution. However, passive sampling still suffers from a lack of confidence of regulators and investigators with regard to the reliability of the ambient concentrations it represents and the supposed variability of sampling rates in the field. This contribution will show a rational monitoring strategy that has been applied in several catchments in Luxembourg and validated with parallel autosampling of flood events during application periods. It establishes that passive sampling is essentially time proportional and that base- and high flows can be separated for their contribution in terms of time-weighted averages and event mean concentrations. The biases and uncertainties in terms of load calculations are addressed. Based on monitoring in different hydrogeological contexts the approach is then used to derive land and crop use specific loads in catchments and exceedance probabilities of EQS values resulting in a risk map of impacted surface waters in Luxembourg.

MO128

Spatially distributed environmental fate modelling of terbuthylazine in a mesoscale agricultural catchment using passive sampler data

M. Gassmann, University of Kassel / Department Water Quality Management - Modelling and Simulation; T. Galle, Luxembourg Institute of Science and Technology; J. Farlin, Luxembourg Institute of Science and Technology LIST

The impact of agricultural practices on water pollution can be assessed by process-based reactive transport modelling using catchment scale models. Most previous studies only used concentrations at the catchment outlet for model calibration and validation. Thus, even if the applied model is spatially distributed, predicted spatial differences of pesticide loss cannot be directly compared to observations. In this study, we applied the spatially distributed reactive transport model Zin-AgriTra in the mesoscale (78 km²) catchment of the Wark River in Luxembourg in order to simulate concentrations of terbuthylazine in river water. In contrast to former studies, we used six sampling points, equipped with passive samplers, for pesticide model validation. At each sampling point, event mean concentration of six events from May to July 2011 were calculated by subtraction of baseflow-mass from total collected mass assuming time-proportional uptake by passive samplers. Continuous discharge measurements and high-resolution autosampling during events allowed for accurate load calculations at the outlet. Detailed information about maize cultivation in the catchment and nation-wide terbuthylazine application statistics (average of 341 g/ha in the 3rd week of May) were used for a definition of the pesticide input function of the model. The hydrological model was manually calibrated to fit baseflow and spring/summer events. Substance fluxes were calibrated using 1000 Monte-Carlo simulations of physico-chemical substance characteristics as provided by the literature: surface soil half-lives of 10-35 d, Freundlich K_{OC} of 150-330 ml/g, Freundlich n of 0.9 – 1 and adsorption/desorption kinetics of 20 – 80 1/d. A multi-criteria Nash-Sutcliffe efficiency including substance loads and concentrations at all stations was calculated resulting in values up to 0.80. The best 100 parameter sets were evaluated for terbuthylazine pathways and balances. The model simulated overland flow to be the major source (80-95%) of terbuthylazine in the main channel and soil water fluxes to be the most important pathways in the tributaries. Simulation results suggest that 0.07-0.14 % of applied terbuthylazine mass was exported to the river in the Wark catchment. In addition to calibration of substance characteristics, passive sampler data was helpful for verifying model setup of application field connectivity.

MO129

Recalibration and cross-validation of pesticide trapping efficiency equations for vegetative filter strips (VFS) using additional experimental data

S. Reichenberger, DR. KNOELL CONSULT GmbH / Environmental Fate / Modelling / GIS; R. Sur, Bayer AG - Crop Science Division / Environmental Safety; C. Kley, Bayer AG Crop Science Division; S. Sittig, DR. KNOELL CONSULT GmbH / E-Fate Modelling; S. Multsch, DR. KNOELL CONSULT GmbH

Vegetative filter strips (VFS) are widely used for mitigating pesticide inputs into surface waters via surface runoff and erosion. To simulate the effectiveness of VFS in reducing surface runoff volumes, eroded sediment and pesticide loads the model VFSMOD (Muñoz-Carpena and Parsons, 2014) is frequently used. While VFSMOD simulates infiltration and sedimentation mechanistically, the reduction of pesticide load in surface runoff by the VFS (deltaP) is calculated with the empirical multiple regression equation of Sabbagh et al. (2009). This equation uses the following inputs: predicted reduction of total inflow (deltaQ) and eroded sediment load (deltaE), absolute surface runoff volume and eroded sediment load entering the VFS, linear adsorption coefficient Kd of the pesticide, and the clay content of the field soil (as a proxy for the clay content of the eroded sediment). The Sabbagh et al. (2009) equation, the coefficients of which were obtained by calibration against 47 data points, has not been widely accepted by regulatory authorities, on the grounds that its reliability has not been sufficiently established yet. Hence, evaluation against additional experimental data is necessary. Chen et al. (2016) proposed an alternative regression equation with a different structure based on 181 experimental data points. This equation uses fewer independent variables, but has more parameters than the Sabbagh equation. The objective of the present study was to improve the predictive capability of the Sabbagh et al. equation by broadening the underlying experimental data. For this aim, additional experimental VFS datasets were compiled from the available literature and thoroughly checked for their suitability. Moreover, existing errors in the calibration and validation data points of Sabbagh et al. (2009) were corrected. The consolidated experimental dataset (n = 244) was used to recalibrate the Sabbagh and Chen equations. Moreover, a k-fold cross validation analysis was performed to assess the predictive capability of both models. The Sabbagh equation fitted the whole dataset slightly better than the Chen equation ($r^2 = 0.82$ vs. $r^2 = 0.79$) and performed consistently better in the cross-validation exercise (with respect to the prediction performance indicators Q^2, predictive r^2, and RMSEP). Finally, a maximum-likelihood-based calibration and uncertainty analysis were performed for the Sabbagh equation using the DREAM algorithm.

MO130

VandA - Visualize and Assess: a tool for the pesticide risk mitigation in surface water

F. Galimberti, G. Azimonti, ICPS International Centre for Pesticides and Health Risk Prevention / Public Health; A. Moretto, Università degli Studi di Milano

The Directive 2009/128/CE of European Parliament and Council on Sustainable Use of Pesticides introduced a community action framework to protect the Environment of the EU and requested Member States to implement policies and actions in order to reduce the risk of pesticide use. In the Region of Lombardy, in Italy, this Directive was adopted with DGR n. X/3233. The aim of the present work is to develop an innovative, easy to use tool to visualize the pesticide surface water contamination, assess the potential pesticide risk and identify areas where to introduce mitigation measures to reduce the contamination, and consequently to reduce the risk in the surface water compartment. The datasets to start with are the monitored concentrations of pesticides in surface water, produced by the Regional EPA. These values are used in this context as Measured Environmental Concentration - *MEC*. . The ratio MEC/PNEC is proposed in this work as a sort of risk assessment, even though the limitation and the complexity of usage of monitored data is well known. In addition, the ratio MEC/*EQS* - Environmental Qualitative Standard (annual average concentration), is considered, to address the water quality with respect to the regulattory limit for pesticides in surface water (Directive 2000/60/EC). A MS Excel tool has been developed to map the monitored residues of pesticides, assess the potential pesticide risk (MEC/PNEC) and identify "hot spots", thet is areas where mitigation measures should be included. The tool is thought to be an anyone-can-use one, even with no particular knowledge of GIS or database management. Its peculiarity to be built inside MS Excel gives itself the possibility to share and to ease the dissemination of results. For more advanced mapping, the tool can interact with ESRI ArcGIS. The openness of VandA makes it a tool suitable to work with other environmental compartments or other environmental thematics.

MO131

Selecting application dates for UK higher tier drainflow modelling: comparing the FOCUS PAT and CRD PAT rules, and assessing the role of soil trafficability

J. Carnall, G. Hughes, Cambridge Environmental Assessments; J.A. Hingston, J. Evans, Chemicals Regulation Division

Pesticide losses *via* drainflow are strongly dependent on the soil moisture status at the time of application and the rainfall pattern that follows application. For drainflow simulations, the choice of application date can therefore have a significant influence on predicted environmental concentrations. To standardise the selection of application dates, the FOCUS surface water models include a calculator tool, the Pesticide Application Timer (FOCUS PAT), which selects an application date from a window defined by the user by applying a set of rules to the daily rainfall data used in the simulation. Alternative criteria for selecting application dates for drainflow simulations were proposed by Brown *et al.* (2004; Pest Manag Sci. 2004 Aug; 60(8); 765-76), and incorporated into a modelling tool developed recently by the HSE's Chemicals Regulation Division (CRD) for performing UK higher tier drainflow assessments using the MACRO model. Under these rules (referred to herein as CRD PAT) a different set of criteria is applied to the daily rainfall data, with the algorithm selecting the first compliant date on or after a particular target day. Both the FOCUS PAT and CRD PAT algorithms select pesticide application dates based on daily rainfall volumes. In practice, however, application dates can be constrained significantly by the trafficability of the soil. Product GAPs are designed to cover a wide range of application periods to account for seasonal variation, e.g. in dry springs applications might take place in March while in wetter years, when it is more difficult to traffic wet soils, they may take place in April or even early May. When conducting multi-year modelling risk assessments this nuance is often lost from the risk assessment, and applications are forced into months when farmers would not have been able to travel their lands and apply crop protection products. Using a soil moisture deficit based approach to defining soil trafficability, the CRD and FOCUS pesticide application timing algorithms were modified to account for this agronomic restriction. In this poster, the results from the four approaches – namely CRD PAT, FOCUS PAT, CRD Traffic PAT and FOCUS Traffic PAT – are contrasted and compared, with a view to drawing conclusions for the standard and refined UK higher tier drainflow risk assessment process.

MO132

Considering diffuse urban and agricultural sources of pesticides at the landscape and catchment scale

G. Hughes, J. Carnall, Cambridge Environmental Assessments; F. Ericher, CEA

For plant protection products (PPPs), there is a strong move towards landscape and catchment scale risk assessments as this allows for integrated risk assessments that consider multiple sources of pollutants, different exposure pathways as well as different receptors within a single framework. This landscape/catchment approach moves away from realistic worst case scenarios, designed to be protective of a wide range of usage situations, to a more realistic representation of usage environments that the risk assessment is trying to protect. Two important diffuse sources of pesticide residues emitted to surface water bodies are from urban and agricultural uses. At present these sources are risk-assessed using very different scenario-based approaches in isolation. Using a multi-disciplinary approach drawing on landscape implementations of the FOCUSsw scenarios to describe possible agricultural sources and an urban emission model to describe possible hard surface usage, this poster considers the likely spatial and temporal coincidence of these different sources of pesticide residues. The need for these two sources to be considered jointly within landscape and catchment scale risk assessments and the possible implications are discussed.

MO133

Calibration of passive samplers for the monitoring of chlordecone in French Caribbean rivers

N. Tapie, Univ. Bordeaux, CNRS, EPOC UMR 5805 / EPOC UMR 5805; T. Risser, Univ. Pau et des Pays Adour, CNRS / IPREM UMR 5254; A. Haouisse, Univ. des Antilles / UMR BOREA UA-CNRS7208-IRD207-MNHN-UPMC-UCN; P. Pardon, UMR CNRS EPOC Universite Bordeaux / EPOC UMR 5805; B. Lauga, Pau et des Pays Adour, CNRS / IPREM UMR 5254; D. Monti, Univ. des Antilles / UMR BOREA UA-CNRS7208-IRD207-MNHN-UPMC-UCN; H. Budzinski, University of Bordeaux

The uncertainty of the tropical weather in the French Caribbean makes spot sampling of chlordecone obsolete and new approaches should be explored to monitor the fate of this molecule in the aquatic system. Here, three types of integrative samplers, differing by their membrane, were calibrated in laboratory and on field for 14 days for the molecule chlordecone: the classical POCIS (Polar Organic Chemical Integrative Sampler) (with Polyethersulfone membranes), the POCISny 30µm (with nylon membranes), and the POCISny 0.1µm. Calculated sampling rates (Rs) were corrected by a PRC (Performance Reference Compounds) approach. Laboratory calibration was done in triplicates under a continuous flow system, and the field calibration was done in triplicates in river Capesterre (Guadeloupe, French Caribbean). Rs in laboratory calibration were 0.30 ± 0.02 L.day^{-1} for the POCIS, 0.09 ± 0.01 L.day^{-1} for the POCISny 0.1µm and 1.54 ± 1.38 L.day^{-1} for the POCISny 30µm. Two distinct Rs have been calculated for the POCIS and the POCISny 0.1µm: one for the first five days of the experiment (Rs= 0.19 ± 0.01 L.day^{-1} for POCIS; Rs= 0.48 ± 0.50 L.day^{-1} for NOCIS 0.1µm), and one for the overall experiment (Rs= 0.19 ± 0.02 L.day^{-1} for POCIS; Rs= 0.43 ± 0.01 L.day^{-1}). POCISny 30µm followed the same pattern than in the laboratory calibration and reached equilibrium after 3 days, with a Rs significantly higher than in the laboratory calibration (Rs=4.82 ±1.93 L.j^{-1}). POCIS and POCISny samplers can accumulate chlordecone efficiently despite its hydrophobic properties. POCIS 30µm seem to be a useful tool to monitor short flash floods, which happen regularly in this area.

MO134

Temporal patterns of pesticide residues in four major river basins in Korea

C. Kim, K. Son, Y. Ihm, H. Lee, National Institute of Agricultural Sciences / Department of Agro-food Safety & Crop Protection

To evaluate residues of environmental concerned pesticides which mainly include pesticides used for rice cultivation, total ninety four sampling sites were selected through main streams and branch streams of four major river basins. And the water samples at these sites were collected four times per year, April, May-June, July-August, and September-October or November-December in 2012 and 2014. Besides, the water samples at sites of Keum, Mangyung and Dongjin rivers belong to the Keum river basin were regularly collected with a month interval, especially biweekly from May to August in 2013. Of the pesticides monitored, fenoxanil, hexaconazole, isoprothiolane, iprobenfos and thifluzamide as fungicides were mainly detected in rice season. While other fungicides including diniconazole, propiconazole, fenarimol, nuarimol and boscalid, were detected with low frequencies and their average residue levels in positive samples were also fairly low. Of the insecticides monitored, some organophosphoruses, cadusafos, diazinon, fenitrothion, fenthion, phenthoate and prothiofos, two carbamates, carbofuran and fenobucarb, and endosulfan were detected with low frequencies and low residue levels. Of the herbicides monitored, nine pesticides which include alachlor, butachlor, dimethametryn, dithiopyr, ethalfluralin, metolachlr, oxadiazon, simetryn and thiobencarb were detected with frequencies of 1-48% and in their residue level of 0.01-1.9 µg/L. Detection frequencies and residue levels of insecticides and herbicides were the highest in waters sampled in May and June. Almost pesticides detected were for the rice plants and their residue levels were very low to compare with standard values.

MO135

Occurrence of 14 representative pesticides in surface and ground waters of the State of São Paulo, the biggest sugarcane producer in Brazil

R.D. Acayaba, SCHOOL OF TECHNOLOGY UNICAMP; C. Raimundo, UNICAMP / Institute of Chemistry; A. de Albuquerque, G. Umbuzeiro, School of Technology, UNICAMP / LAEG

São Paulo State is the biggest sugarcane producer in Brazil and the second at pesticide consumption. The aim of this project was to develop a method to determine the presence of 14 pesticides representative from sugarcane plantation, 7 herbicides (simazine, atrazine, ametryn, clomazone, diuron, hexazinone and tebuthiuron), 3 fungicides (azoxystrobin, carbendazim and tebuconazole), 3 insecticides (carbofuran, imidacloprid and malathion) and 1 transformation product (atrazine-2-hydroxy) in surface and ground waters using liquid chromatography tandem-mass spectrometry (LC(ESI)MS/MS) and solid phase extraction as sample preparation. Limits of detection (LOD) and quantification (LOQ) were ranged from 0.9 to 22 ng L^{-1} and from 2.8 to 74 ng L^{-1}, respectively, and mean recovery was 66 %, which allowed obtaining a sensitive and accurate method for the determination at trace levels. In total, 196 samples located in the main sugarcane area from São Paulo were analyzed (175 surface waters and 21 groundwaters) between October/2015 to October/2016. The most frequently detected pesticides in surface water were atrazine-2-hydroxy (100%), diuron (94%), carbendazim (93%), tebuthiuron (92%), hexazinone (91%), imidacloprid (96%) and ametryn (81%). The pesticide that presented the highest concentration for this matrix was imidacloprid, reaching 2579 ng L^{-1}. The risk to aquatic life were evaluated dividing the maximum environmental concentration of each pesticide by the lowest water quality criteria found in the literature. The potential risk for aquatic life was observed for imidacloprid, carbendazim, atrazine and malathion. For the groundwaters the most frequently detected pesticides were atrazine-2-hydroxy (24%), imidacloprid (14%), carbendazim (10%), tebuthiuron (10%), atrazine (10%) and diuron (10%). The pesticide that presented the highest concentration for this matrix was tebuthiuron, reaching 107 ng L^{-1}.

MO136

Exposure scenarios for aquatic risk assessment of pesticides in Brazil

B. Jene, BASF SE / Environmental Fate; R.P. SCORZA JUNIOR, Embrapa / EMBRAPA AGROPECUARIA OESTE; D. Máximo, R. Rebelo, IBAMA / DIQUA / CGASQ; A.V. Waichman, Universidade Federal do Amazonas; N. Peranginangin, Syngenta Crop Protection, LLC / Product Safety; A. Tornisielo, BASF SA / GENCS - E-Fate; L. Murakami, Bayer AG Crop Science Division; O. Perez-Ovilla, Bayer CropScience / Environmental Safety; E. Henry, Bayer / Environmental Safety; T. Haering, BASF SE

A tri-partite technical working group consisting of regulators, academia and industry was formed to develop a framework for aquatic risk assessment of pesticides in Brazil. The framework should include a sophisticated science based approach resulting in a comprehensive guidance. The basis of the exposure assessment is the selection of the 90th percentile vulnerability which is seen to represent a reasonable worst case and is used as basis of the exposure assessment in other parts of the world. Surface water scenarios should be identified in six pre-defined climatic zones for the seven most important crops soybean, maize, sugar cane, wheat, cotton, citrus and coffee. Runoff and spray drift were found to be the main entrance pathways of pesticides into surface water bodies. Whereas spray drift mainly depends on technology and local climatic conditions during application, surface runoff is influenced by pedoclimatic conditions that could be assessed in a spatial vulnerability analysis. For this the USDA Runoff Curve Number approach (RCN) which is implemented in PRZM was used. A simple model based on the equations of the RCN approach was developed to calculate daily surface water runoff volumes for the agricultural area of Brazil for 34 climatic years. Calculations were carried out on highest available resolution of soil data resulting in more than 63,000 raster cells. Hydrological soil groups were determined by using a Brazilian specific classification scheme applied to the national soil map. Relative runoff vulnerability for pesticides was estimated with an integrated vulnerability index approach where indices for daily runoff such as average annual number of runoff events and average maximum runoff volume of each year were combined with an index for the expected substance concentration in the runoff water based on organic carbon content of the soil. The 90th percentile relative vulnerability was determined for the relevant crops for each climatic zone to select the relevant surface water scenarios. For the specific crop area, census data on municipality level were used first, but it was decided to switch to satellite images as far as they become available. After discussions in the technical working group the Brazilian environmental authority IBAMA decided to use the US-EPA PWC model for the surface water exposure assessment. Representative flowing and static water bodies which need to be natural and permanent will be defined for each selected scenario.

MO137

Identification of Herbicide Source Areas and Spatial Variability of Dominating Transport Processes in a High Agricultural Intensity Catchment

H. Rathjens, M.F. Winchell, Stone Environmental, Inc. / Environmental Systems Modeling; R. Sur, Bayer AG - Crop Science Division / Environmental Safety; D. Baets, Bayer AG Crop Science Division / Sustainable Operations; F. Krebs, DR. KNOELL CONSULT GmbH; D. Lembrich, Bayer AG Crop Science Division

The occurrence of herbicides in surface waters of intensively cultivated catchments can originate from a variety of sources. These include transport via runoff and erosion during storm events, subsurface transport through lateral flow and through subsurface tile drainages, and from spray drift during applications. The Soil and Water Assessment Tool (SWAT) is widely used in the United States and the EU for catchment scale hydrologic and water quality modeling of non-point source chemicals in the environment. The SWAT model was applied to a 992 ha agricultural catchment in the Flanders region of Belgium to help in better understanding the sources of the herbicide detections observed in daily sampling over 3.5 years at two locations along the catchment's primary stream. The SWAT model was calibrated to observed flow and chemical monitoring data, then used to characterize the relative contributions of herbicides via surface processes, subsurface processes, and spray drift. In addition, very vulnerable fields with significant contributions to surface water exposure were identified. A quantitative comparison between monitoring data and simulated exposure profiles was made to single out those high residue concentrations that could not be attributed to any of these traditionally considered exposure pathways, and could ultimately be only explained by point source contributions . The model results demonstrates that SWAT is capable of simulating streamflow in a small agricultural catchment, and is capable of simulating diffuse source pesticide concentrations. This allowed application of an approach that incorporated model uncertainty analysis in distinguishing between diffuse source dominated high concentrations from those most likely affected by point sources. The SWAT model also proved useful in identifying the spatial variability in the dominant transport processes contributing pesticide residues to the stream. While surface runoff of soluble pesticide was the major non-point source contributor on most fields, lateral subsurface flow was found to be important as well, especially in the western portion of the catchment. Spray drift is likely the least significant contributor at the catchment scale. Overall, the analysis of monitoring data and modeling results shows that the potential for reducing herbicide concentrations in the study catchment can be addressed by mitigating both point source contributions from farmyards as well as diffuse sources.

MO138

Pesticides in water and surface sediments from Douro River estuary (Portugal) - assessment of environmentally relevant mixtures using acute toxicity bioassays

M. João Rocha, ICBAS U.Porto, CIIMAR CIMAR LA; C. Cruzeiro, CIIMAR CIMAR LA, Porto, CEF FCTUC U.Coimbra; S. Amaral, ICBAS U.Porto; E. Rocha, ICBAS U.Porto, CIIMAR CIMAR LA

The Douro River is an international water river that passes through extensive agricultural fields, of both Portugal and Spain, before reaching the estuary at Porto and Gaia cities. Therefore, the presence of pesticides is suspected. Accordingly, the evaluation of 56 pesticides of different categories (insecticides, herbicides, and fungicides) should be viewed as a priority for this habitat due to their negative impacts on the biota. For this purpose, water was collected over one year at six sampling sites in the Douro River estuary. Samples were extracted by solid-phase extraction and quantitatively analysed by gas chromatography–mass spectrometry (GC-MS). Results show that 96% of measured pesticides were detected in 79% of the quantified samples and that twelve compounds showed concentrations well above the limits established by the 2013/39/EU Directive. Individually, the concentrations of the analysed pesticides ranged from 39 to 1265 ng/L. Since the occurrence of these compounds happens in mixtures, we conducted a theoretical hazard assessment considering the average and the maximum environmental mixtures of all measured pesticides. The theoretical approach suggested that invertebrates were the most sensitive group. Therefore, short-time exposure in vivo assays using *Artemia salina* and *Daphnia magna* were done. These results disclosed significant toxic effects of the analysed mixtures - high mortality rate and abnormal swimming behaviour - over the exposed animals. Both approaches (theoretical and experimental) support the analytical results, alerting for the need of interventions on this estuarine environment and of other comparable. **Acknowledgements:** European Regional Development Fund (ERDF) through COMPETE, Framework of the Structured Program of R&D&I INNOVMAR – Innovation and Sustainability in the Management and Exploitation of Marine Resources (NORTE-01-0145-FEDER-000035), Research Line ECOSERVICES, supported by the Northern Regional Operational Programme (NORTE2020), through the ERDF. ICBAS – U.Porto. **Keywords:** monitoring, *Artemia salina*, *Dapnia magna*, pesticide mixtures

MO139

Monitoring programme to investigate the presence of myclobutanil and its soil metabolite in Italian groundwater following use in pome fruit, stone fruit and vineyards

G.L. Reeves, Dow AgroSciences Ltd; A. Finizio, University Milano - Bicocca / Department of Earth and Environmental Sciences; R. Verro, University Milano - Bicocca - Lybra ambiente e territorio S.r.l. / Department of Earth and Environmental Sciences; <u>C. Vaj</u>, Dow AgroSciences Italia s.r.l.; R. Bradascio, Dow AgroSciences Italia srl / RD

Myclobutanil is a fungicide used mainly in pome and stone fruit areas, and in vineyards. For national registration in Italy, FOCUS groundwater modelling showed that the PECgw for its soil metabolite (X11292885; up to 6% of applied) reached up to 2 µg/L. This was accepted at EU level since the metabolite is not toxicologically relevant. However, according to national rules, this triggers a groundwater monitoring study for a metabolite when the PECgw is >0.75 µg/L. For completeness, myclobutanil was also monitored. To allow for a robust monitoring study, it was necessary to identify monitoring areas with the following characteristics; (i) be representative of an intensive use of myclobutanil, and (ii) reflect reasonable worst case scenarios for Italy. To facilitate this, a GIS-based indicator (PLI: Potential Leaching Indicator) was developed to integrate information about sales data, the spatial distribution of the target crops and the distribution of the FOCUS groundwater model scenarios throughout Italy. Use of the PLI allowed five suitable areas to be identified (Bolzano and Trento for apple trees, Verona for stone fruit and grapes, Forlì-Cesena for pear trees and stone fruit, and Matera for stone fruit). These areas are also representative of the FOCUS groundwater scenarios relevant in Italy (Châteaudun, Hamburg, Piacenza and Thiva). In each area, five monitoring wells were identified to cover a range of parameters (depth to groundwater, hydrology, pedology, and presence of the target crops). Subsequently, a three year monitoring campaign was conducted (autumn 2014-spring 2017). From a total of 150 samples analyzed, the concentrations of myclobutanil and its soil metabolite (X11292885) were below the LOD (0.0001-0.0002 µg/L) in 89% and 94% of cases, respectively. For myclobutanil, the number of positive detections was 23 and of these, 20 samples showed concentrations well below 0.01 µg/L. Only in one sample did the measured concentration exceed the trigger value of 0.1 µg/L. However, a number of factors indicate that this is due to point source origin. Even better results were obtained for X11292885 where the number of positive detections was only 13, with concentrations less or very close to 0.01 µg/L. Based on these results it can be concluded that given the actual use conditions, the probability of myclobutanil and its soil metabolite exceeding the threshold value of 0.1 µg/L in groundwater in Italy is very low.

MO140

Identification of areas at risk of groundwater leaching in Italy for the fumigant 1.3-dichloropropene

R. Verro, University Milano - Bicocca - Lybra ambiente e territorio S.r.l. / Department of Earth and Environmental Sciences; R. Bradascio, Dow AgroSciences Italia srl / RD; <u>C. Vaj</u>, Dow AgroSciences Italia s.r.l.; A. Finizio, University Milano - Bicocca / Department of Earth and Environmental Sciences

1,3-Dichloropropene (1,3-D), also known as Telone™, is an active substance used worldwide in soil fumigant products for the control of cyst and free-living nematodes. It is used in a variety of crops including fruiting and vegetable crops. Soil sorption studies have shown that 1,3-D and its metabolites present characteristics of highly mobile molecules with a potential to leach into groundwater when applied in vulnerable areas such as sandy soil and in areas characterized by shallow groundwater. European member states have a wide range of groundwater monitoring activities for plant protection products and their metabolites, but analysis of 1,3-D and its metabolites is not currently part of the routine programme. In this study we illustrate a methodology allowing to identify areas most at risk where monitoring should be focused in priority, taking the example of Italy. The methodology considers three parameters: i) crops distribution, ii) soil properties and iii) 1,3-D use. The data, structured as layers of information are managed within a GIS, and are intersected to get the so called Uniform Geographic Units (UGU) which are areas of uniform in their characteristics and are representative of a specific set of values parameters. The data about the spatial distribution in Italy of crops where the 1,3-D is applied were first gathered from the last agricultural census, which provide information at a provincial scale. Successively, these data were refined at municipality scale. The identification of sandy soil areas in Italy was performed using pedological information extracted from different official sources. Subsequently, the information was processed in order to identify the areas where the percentage of sand in soils falls among three different categories (< 60%, 60-80%, >80%). Also these data were structured as GIS layers, which were processed and represented using the same GIS of the crop distribution. Overlaying the crop distribution and sandy soil areas and by merging the two databases, it was possible to identify sub-communal areas where crops and sandy soils coexist, characterizing the extension in relation to the rest of the municipality and the province. Finally, by considering 1,3-D sales data, it was possible to refine the areas previously identified and quantify the percentage of areas potentially at risk of leaching where Telone™ is applied. ™ of DowAgroSciences

MO141

Development of an European Tier 3+ Spatially Distributed Modelling Framework

<u>G. Hoogeweg</u>, Waterborne Environmental, Inc. / Data Technologies; P. Sweeney, Syngenta

Higher tier groundwater assessment in the European Union (EU28) allow the use of spatially distributed modeling approaches for the assessment of groundwater and exposure of soil organisms. An advantage of a distributed model is that model inputs can reflect local conditions and capture the spatial variability of the landscape and weather patterns. An advanced modelling framework, based on the GeoPEARL 4R model was developed for the EU28. This model fills the niche for higher Tier assessments needs. This modelling framework represents over 1.340.000 km^2 of arable agricultural lands in Europe. Nearly 382.000 unique soil, weather, FOCUS zone combinations represent the variability of the landscape and climate. Datasets to populate the model, included CORINE land cover, soils data (ESDB, ESDB Derived Data for Modelling and HYPRES, EFSA organic matter) and the JRC MARS 25km gridded daily weather data. Agricultural management practices, irrigation, and cropping scenarios are gleaned from the standard FOCUS modelling scenario, but can be updated as needed. This European modeling framework (EMF2014) can be used for EU28, member state, FOCUS zones or crop specific groundwater vulnerability assessments, screening of existing and new plant protection products, context setting of standard scenarios, test sites, and lysimeter, site selection. In this presentation we will show how we developed the framework and several example outputs as well as discuss the implications of conducting large-scale distributed modelling assessment.

MO142

Influence of aquifer parameters on groundwater residue concentrations

<u>F. Hegler</u>, DR. KNOELL CONSULT GmbH; D. Liss, SGS Institut Fresenius GmbH / Agro; W. He, DR. KNOELL CONSULT GmbH; O. Naeb, SGS Institut Fresenius GmbH; S. Reichenberger, DR. KNOELL CONSULT GmbH / Environmental Fate / Modelling / GIS

FOCUS leaching models are used in a regulatory context to calculate pesticide leaching flux concentrations in 1 m depth (PEC_{GW}; "Predicted Environmental Concentrations in groundwater") from the unsaturated to the saturated zone. These values are used in risk assessments in order to evaluate the impact of plant protection products on groundwater. In higher tier groundwater monitoring studies the properties of the saturated zone add additional complexity influencing actual pesticide residue concentrations in shallow groundwater. In this work the impact of groundwater flow velocity and aquifer porosity on groundwater residues for a defined leachate concentration (i.e. decoupled from the unsaturated zone) was determined. In a sensitivity analysis using a realistic range of aquifer parameters the impact on the resulting residue concentrations in groundwater was quantified. For the sensitivity analysis FOCUS model outputs for selected scenarios were combined with realistic aquifer parameters for some representative regions in northern Italy and Germany. In these regions the relevant shallow aquifers are variable in terms of hydraulic conductivity, gradient and effective porosity and provide a representative parameter range.

MO143

Implications of Dataset Selection and GIS Processing on Modelling

<u>G. Hoogeweg</u>, Waterborne Environmental, Inc. / Data Technologies; M. Geuvara, Waterborne Environmental Inc / Modeling

Groundwater assessment guidelines provided by the FOCUS groundwater working group (2009) and EFSA (2014) describe succinctly a multi-tiered modelling framework that includes spatio-temporal assessments in the higher tiers; e.g., tier 3a and 3b. As part of the spatio-temporal assessment several GIS and daily climate datasets were recommended. These recommended datasets, however, have been superseded by new datasets in the past few years. Specifically, daily weather and soils data have undergone significant updates, which are reflective of the considerable effort in Europe to update this spatial information. Not only does dataset choice, but also how datasets are being processed in a geographic information system, impact modeling results. Basic assumptions regarding aggregation of data, data slicing for determining climatic zones and data resolution impact our modelling results. In this poster, we will show the implications of data selection and data processing on a distributed modelling framework centered around GeoPEARL 4R. Specifically we will focus on differences between datasets, data set resolution, capturing variability and ones ability to model at the pan-European level within EFSA's tier 3 guidelines.

MO144

Combining specific and public groundwater monitoring data as higher tier for pesticide regulatory risk assessment

A. Boivin, ANSES

Pesticides risk assessment for groundwater in France is performed according to Regulation 1107/2009. The European tolls are routinely used by considering the same models and same tiered approach.Groundwater monitoring data are identified as higher tier that may supersede modelling. Still, there is currently no agreed guidance available on the use of groundwater monitoring data for regulatory purposes. Work is ongoing on behalf the SETAC-EMAG GW group (www.setac.org/group/SEAGPest).The main issues when dealing with groundwater monitoring data interpretation were related to site selections and related vulnerably, and then of how representative was the groundwater monitoring.In recent case, usefulness of the groundwater monitoring programs submitted at EU level as been questioned. Notably, the groundwater hydrology including its vulnerability and how representative / which situations the test sites might be considered to cover were not seen as not being properly addressed.Monitoring programs have also been submitted and assessed at national level mainly to refine metabolite groundwater risk assessment. Combined information from targeted and public monitoring were keys to address the representativeness of monitoring programs. The French groundwater public network (wells) is vast and the database is available online (www.ades.eaudefrance.fr). This databased (ADES) is owned by the BRGM (French Geological Survey). This database mainly actives substances, more metabolites will be included in the future.Proposals to combine targeted together with public groundwater monitoring dataset were made to enhance the representativeness of the GW monitoring conducted.

MO145

Minimal variation in input parameters highly influences PEARL and PELMO results: how can these results be trustable?

S. Ullucci, ICPS; L. Menaballi, International Centre for Pesticides and Health Risk Prevention

The calculation of Predicted Environmental Concentration of pesticides in groundwater (PEC_{GW}) is a crucial point in the registration and authorization process of plant protection products (PPPs) in Europe. Calculations are usually performed by FOCUS models, in particular PEARL and PELMO models. These models allow a realistic but conservative assessment of the potential leaching of pesticides in the groundwater compartment. Model results are influenced by substance specific parameters such as DT_{50}, K_{om} and Freundlich coefficient (1/n). Great variations in PEC_{GW} values are expected when high variability occurs in one or more of the parameters listed above. In this work, we demonstrate that PECgw outputs are significantly affected also by minimal variation of the same parameters. Considering that a minimal variation is intrinsic in all laboratory studies (es. 25% uncertainty in K_{OM} determination was calculated applying the Horwitz equation), it is questionable whether a corresponding high variation in model is scientifically acceptable. In a previous project (York, 2017), dummy substances with different combinations of DT_{50}, K_{OM} and 1/n values were used in FOCUS PEARL, in order to quantify the influence of each single parameter on the final PECgw. It was verified that the sensitivity of PEARL model can been considered quite excessive. In this follow-up project, further calculations were performed using FOCUS PELMO to compare the sensitivity of these two models, commonly used in a regulatory contest. Leachate concentrations were plotted as a function of K_{OM} and as a function of degradation rate coefficient. PECgw obtained by the simulations of these two models were used to create a classification system for the input parameters K_{OM} and DT_{50} according to models sensitivity. Conservative values for each parameter class, to be used in PEC_{GW} calculations, are proposed for all substances. This approach can minimise the effects of the intrinsic input variability providing a better scientific approach to the assessment of groundwater modelling in the regulatory context.

MO146

European regulatory network on pesticide groundwater monitoring

A. Gimsing, The Danish Environmental Protection Agency / Pesticides and Gentechnology; W. Koenig, UBA Umweltbundesamt; A. Boivin, ANSES; A. Poot, Ctgb; A. Schwen, AGES; M.E. Balmer, Plant Protection Chemistry; A. Massey, Chemicals Regulation Directorate; W. Tüting, German Federal Office of Consumer Protection and Food Safety

Groundwater monitoring data should be included in the assessment of the leaching risk of pesticides and their metabolites. Monitoring data is generated in most European countries, but their use for leaching risk assessment during pesticide authorization is hampered due to the following reasons: (i) The data is often not publically available or available only in an aggregated form in a report, (ii) most often it is available in the national language of the origin country only, which makes it hard for other countries to use it, and (iii) the interpretation of groundwater monitoring data requires detailed knowledge of the local geological, hydrological and climatic conditions, and also knowledge about crops and the use pattern for pesticides. To overcome these difficulties, a network among regulatory authorities in Europe involved in groundwater risk assessment of pesticides is proposed. The aim of the network is to exchange pesticide groundwater monitoring data and frequent updates about the focus of national monitoring, and to assist each other in the interpretation of the data. The network plans to have its first meeting in Copenhagen, Denmark, in September 2018 with an invitation to all interested European countries to participate. The poster will present the thoughts about the network and the status of the start-up, and aims at promoting the network to interested authorities.

MO147

Overview of measured wash-off factors from experiments suitable to derive a refined input for FOCUS modelling

G. Reinken, E. Hellpointner, Bayer AG, Research & Development, Crop Science / Environmental Safety; D. Schaefer, Bayer Crop Science / Environmental Safety

Recent regulatory interest in the wash-off process resulted in a proposal to that the effects of wash-off should be generally considered as additional soil loading for FOCUS modelling of foliar applied pesticides (EFSA 2010, 2016). EFSA also proposed to increase the default wash-off factor for FOCUS modelling from 0.5 cm-1 to 1 cm-1 (EFSA 2012). On the other side, EFSA has stated that effects of wash-off should be not considered as an additional worst case but rather as average effect (EFSA 2015, 2017). The foliar wash-off factor of a compound is a product (formulation) specific modelling input parameter that can be experimentally determined. A generic experimental study design has been derived in a workshop organised by the European Crop Protection Association (ECPA). This study design was used for the experimental determination of wash-off factors for modelling purposes. Overall 25 individual wash-off factors have been determined experimentally, mainly under GLP. The data set comprises 12 formulations, 8 compounds and 6 crops. Experiments consider one heavy rainfall invent of 15 mm over one hour, applied 24 hrs after foliar pesticide spray. The determined wash-off factors are normally distributed. The arithmetic mean of all single values is 0.38 cm-1 with a median of 0.40 cm-1. Just 7 of the 25 values are slightly above the existing default wash-off factor of 0.5 cm-1. The highest measured wash-off factor is 0.57 cm-1 and the lowest 0.14 cm-1. A wash-off factor of 1.00 cm-1 would be clearly outside the 3-sigma range of the experimental data set. This experimental evidence does not support the proposal to increase the existing default wash-off factor from 0.5 cm-1 to 1 cm-1. EFSA 2010: PPR opinion 1442 - Outline proposals on exposure of organisms in soil EFSA 2012: Scientific Opinion 2562 – Science behind the guidance on soil scenarios EFSA 2015: Guidance Document 4093 - Predicting environmental concentrations in soil EFSA 2017: Guidance Document 4982 - Predicting environmental concentrations in soil

MO148

Leaching and plant uptake of trifluoroacetic acid (TFA) under cropped outdoor conditions

G. Reinken, M. Beckmann, Bayer AG, Research & Development, Crop Science / Environmental Safety; K. Hammel, Bayer AG, Crop Science Division / Environmental Safety; U. Kölzer, Bayer AG, Research & Development, Crop Science / Environmental Safety; M. Lamshoeft, Bayer CropScience AG / R&D; S. Sittig, DR. KNOELL CONSULT GmbH / E-Fate Modelling

In recent regulatory discussions about the plant uptake process the question was raised if tracer-like substances (very low or zero sorption, very slow or no degradation) are fully available for uptake by plant roots under dynamic outdoor conditions. An alternative hypothesis is that such substances move so quickly in the soil column that they are taken up by plant roots to a lower extent than indicated by static laboratory plant uptake studies. To address this particular question an outdoor container study was conducted with 14C-labeled trifluoroacetic acid (TFA) and winter wheat plants for a time period of 228 days after application onto soil. Trifluoroacetic acid is a common breakdown product of several chemical product classes (e.g. refrigerants like HFCs and HCFCs, anaesthetics, pharmaceuticals, pesticides, pyrolysis of PTFE (used as coating in may products like Teflon® or GORE-TEX®)). Plant root uptake of TFA under static conditions has been determined in parallel with experiments using plants growing in nutrient solution (hydroponic study design). The translocation and uptake of trifluoroacetic acid observed in the outdoor container study was evaluated with the PEARL model considering root uptake processes as routinely applied in standard FOCUS modelling. A transpiration stream concentration factor (TSCF) for TFA under cereal growing outdoor conditions could be derived. The results demonstrate that mobile, tracer-like substances are taken up extensively by plant roots even *under dynamic outdoor conditions; the hydroponic study design is suitable to determine conservative input parameters for regulatory modelling; uptake experiments with cropped outdoor container may be suitable as higher-tier to derive a refined TSCF. Further experiments will indicate to which extent this study design is also suitable to derive refined TSCF for compounds with other sorption and degradation properties.*

MO149

Investigating the variance of edge-of-field deposits of spray drift

H. Holterman, Wageningen University & Research / Agrosystems Research; J. Michielsen, H. Stallinga, P. Van Velde, J. Van de Zande, Wageningen University and Research / Agrosystems Research

Spray applications in arable crops often lead to off-target spray deposits downwind from the treated field. Throughout several decades, many experiments have been carried out by different researchers to quantify the downwind spray deposits. Relations between downwind spray deposits and parameters like sprayer settings, field conditions and environmental conditions were investigated. Still, there is a large variance in the observed data that cannot be explained satisfactorily by the experimental and environmental conditions. Sprayer boom movements and local fluctuations in driving speed, wind speed and wind direction are the most likely factors affecting variance in downwind spray deposits. In this study variations in downwind deposits of spray drift caused by sprayer boom movements are investigated both experimentally and based on simulations using the spray drift model IDEFICS. Downwind deposits of spray drift were measured alongside a treated potato field, at 2 m and 5 m off the edge. Wind speed and direction were recorded during the experiments. Horizontal and vertical movements of the sprayer boom were recorded as well. Variance of spray deposits at 2 m downwind from the field edge was about 50%. At 5 m downwind variance was about 30%. A quasi-dynamic model was developed based on the IDEFICS spray drift model. In the new model the effect of both horizontal and vertical boom movements on downwind spray deposits was studied. From the above mentioned experiments, the most important frequencies and amplitudes of boom movements were derived. Using these frequencies, the model simulations resulted in variances of spray drift deposits similar to those established experimentally. Effects of fluctuating wind directions are to be investigated in the near future.

MO150

Exposure assessment for edge-of-field watercourses next to tree nurseries regarding spray drift deposits

H. Holterman, Wageningen University & Research / Agrosystems Research; J. Van de Zande, Wageningen University and Research / Agrosystems Research

Pesticide applications in tree nurseries involve spraying techniques that apply the pesticide in upward or sideways direction. Particularly for high avenue trees the downwind loss of pesticides due to spray drift can be relatively large. The upward directed part of the spray that is blown towards the top of the trees may reach heights above the trees, where wind can take the spray cloud and move it far downwind. Usually, the branches and leaves at the lower part of the stems of high avenue trees are cut away. Consequently, the part of the spray that is applied sideways may pass underneath the tree canopies and reach downwind areas easily. Measurements of downwind spray deposits for tree nurseries indicate deposition levels comparable to those occurring for fruit orchards, which are well above those occurring when spraying arable crops. The current paper deals with the countrywide exposure assessment for pesticides applied to tree nurseries reaching edge-of-field watercourses in the Netherlands. Apart from nurseries with high avenue trees, nurseries with the younger spindles and transplanted trees are considered as well. Although the total area of tree nurseries is limited, exposure risk to edge-of-field watercourses is important enough to investigate. Spray drift mitigation techniques are considered and evaluated as well.

MO151

Investigating the exposure of residents to pesticides due to airborne spray drift

H. Holterman, Wageningen University & Research / Agrosystems Research; J. Van de Zande, J. Michielsen, H. Stallinga, P. Van Velde, Wageningen University and Research / Agrosystems Research

In the Netherlands approximately 90,000 people live within 50 m of flower bulb or fruit cultivation. It is unclear how many of these people are exposed to pesticides or whether their health is at risk. Recently, a research project was launched to assess the exposure of residents to pesticides next to flower bulbs fields. This research projects involves both measurements and simulations of airborne spray drift. Volatilization is investigated for several days after spray application. Outdoor and indoor exposure of residents to pesticides is estimated. The current paper deals with the exposures to spray drift only. After application using a conventional boom sprayer ground deposits and airborne distributions of spray drift are measured down to 50 m from the treated area. Airborne spray drift is measured up to 10 m height, using two different sampling techniques. At 50 m downwind, airborne spray drift appear to be up to 100 times higher than ground deposits. Simulations of spray drift are studied using the IDEFICS spray drift model for boom sprayers. The simulations result in downwind ground deposits and airborne spray drift with values in the same order of magnitude as those found in the experiments. The results indicate that potential exposure of residents to pesticides used when treating nearby fields may be significant and further assessment of this exposure route is important.

MO152

Risk assessment for consumers of co formulants used in Plant Protection Products. Case study of polymers.

P. Adrian, M. Liegeois, M. Darriet, B. Journel, CEHTRA SAS

Actually there is no recognized guidance on how to conduct a risk assessment for consumers for co formulants present in plant protection products. One of the reasons is the lack of exposure data when the product containing its co formulants is applied onto crops. To our knowledge only one software is predicting the level of crop residues of chemicals after application i.e. PARDIS [1] (Prediction of Agrochemical Residue Data on fruit using an Informatic Sytem) however its use is limited to orchards. In addition in the case of polymers, from an analytical point of view it may be difficult if not impossible to analyse the crops for residue content of this type of co formulant. The objective of this work is to develop a methodology to be applied under this conditions. As a case study we present this methodology for latex polymers i.e. polymer based on methylene succinic acid with buta-1,3-diene, styrene and methacrylic acid. [1] Prediction of agrochemical residue data on fruit using an informatics system (PARDIS model), Calliera M, Balderacchi M, Capri E, Trevisan M. 2008

MO153

Dietary exposure to pesticide residues: the big picture

L. Villamar Bouza, L. Ferreira, EFSA - European Food Safety Authority / Pesticides Unit

Science-based approaches and integrated risk assessments by using experimental data, models for pesticide residues intake estimations, monitoring data considering real exposure, etc. are working tools to contribute to the mission of the European Food Safety Authority (EFSA) on protecting European consumers' health and the environment in the field of pesticide residues. Maximum residue levels (MRLs) are the upper levels of pesticide residues that are legally permissible in food of plant and animal origin. Before an MRL is established, the EFSA assesses the residue behavior of the pesticide and the dietary exposure resulting from the residues expected in food. The chronic and acute dietary consumer exposure to pesticide residues are estimated by using a calculation model developed by EFSA called PRIMo (Pesticide Residue Intake Model) based on the international agreed methodology. This provides the key information to be interpreted by risk assessors and for risk managers' consideration. Essential input values in risk assessment are toxicological data and residue values subject to many environmental scenarios and considerations that are used to define and characterize the residues to which consumers can be exposed throughout the diet. Complex metabolic pathways in plants and animals, degradation of the compounds in soils and and its transformation, the possible uptake and translocations of the residue to the edible parts of the crop and degraded products as result of industrial and household processing are considered to set the residue definitions for risk assessment purposes. Secondary metabolites characterized by metabolism studies and degraded products may pose a completely different toxicological profile than the parent compound, being more, less or equal toxic than the pesticide under assessment, and showing a new big picture for an active substance and its residues that should be assessed in detail to avoid consumers' concerns. The dietary risk assessment of pesticide residues takes into consideration these possible scenarios in order to protect consumers, reason why residue definitions might be different for monitoring and for risk assessment purposes and where the uncertainty due to missing data might play a fundamental role in risk assessment.

MO154

Exposure and Risk Assessment for Agricultural Applicator to Insecticide Flubendiamide during Cabbage Cultivation using Whole Body Dosimetry

J. Lee, Seoul National University / Department of Agricultural Biotechnology; Y. Shin, Seoul National University; J. Lee, College of Agriculture Sciences Seoul National Univ / Agricultural biotechnology; J. Lee, Seoul National University / Department of Agricultural Biotechnology; B. Kim, Seoul National Univ.; E. Kim, H. Ryu, Seoul National University / Department of Agricultural Biotechnology; D. Jeon, Seoul National Univ.; X. Yuan, Seoul National University / Department of Agricultural Biotechnology; E. Park, S. KIM, Seoul National Univ.; M. Rehan, Seoul National University; J. Kim, Seoul National University / Department of Agricultural Biotechnology

Flubendiamide belongs to diamide insecticide that has been used to control a wide range of insects in fruit and vegetables. Assessment for agricultural applicator's dermal and inhalation exposure to flubendiamide during cultivation of cabbage in field was carried out. For dermal exposure measurement, whole body dosimetry (WBD) was performed, which consists of cotton/polyester outer clothes and cotton inner clothes. Hand exposure was measured by washing of nitrile gloves and hands, while head exposure was monitored by face/neck wipe technique. Inhalation exposure was evaluated with personal air sampling pumps and IOM sampler (glass fiber filter). Analytical limit of quantitation was 5.0 ng/mL with good linearity (R^2 > 0.99) of calibration curve. Recovery (77~117%) of insecticide from various exposure matrices were reasonable including field recovery (77~109%). Field exposure experiments were carried out by 8 replicates. During application, total dermal exposure of flubendiamide was 3635.7 µg, while that of mixing/loading case was 815.3 µg. Hand exposure of flubendiamide (688.7 µg) in mixing/loading was higher than the case of application (680.8 µg). Exposure of body was highest (42.0%) in case of application. Penetration rate of insecticide between outer and inner dosimeter was about 0.5% (upper body) and 5.9% (lower body). Inhalation exposure during application was 20.2 µg while in case of mixing/loading no exposure was observed. Risk index (RI) was calculated to be 0.09 using 6 µg/kg/day of acceptable operator exposure level, suggesting that health risk of agricultural applicator during treatment of flubendiamide for cabbage field would be minimum. Keywords: Flubendiamide, exposure, risk assessment, whole body dosimetry, IOM, cabbage *Corresponding author: kjh2404@snu.ac.kr; Tel, 82-02-880-4644

MO155

Multi-year FOCUS Surface Water calculations: What do they mean for real regulatory cases?

D. Schaefer, Bayer Crop Science / Environmental Safety; G. Reinken, Bayer AG, Research & Development, Crop Science / Environmental Safety; A. Bolekhan, Bayer AG, Research & Development, Crop Science; S. Heine, Bayer Ag / Effect modelling; G. Goerlitz, Bayer CropScience AG / Environmental Safety

The surface water exposure calculations for pesticides according to FOCUS are currently under revision by an EFSA working group. In particular the working group was mandated to extend the calculation period from a preselected single year to full 20 years, with the objective to better capture the effect of variable weather conditions on aquatic exposure patterns. This is meant to provide a more robust and reliable basis for aquatic risk assessments in a regulatory context. The planned revision requires decisions on some technical aspects of the calculations (e.g. evaluation of the original FOCUSsw weather data, filling of data gaps, completion of irrigation data sets, definition of multi-year application dates) and also new rules for the interpretation of the results. The main challenge in this context is the lack of experience with extended FOCUSsw calculations and with their results. Surface water exposure is strongly driven by individual weather events triggering run-off or drainflow, and depends in a complex way on substance properties and use patterns. Since multi-year calculations are time-consuming, a systematic investigation of the consequences of the switch to 20 year calculations and associated changes of the procedures is still missing. In this work we conducted such an investigation by running multi-year FOCUS Surface Water calculations for several substances with a range of realistic properties and use patterns, and by analyzing the resulting exposure patterns. We discuss our experiences with the calculations themselves, and present exposure characteristics of the different test substances. These allow some generic conclusions with regard to the consequences of currently discussed options for multi-year aquatic exposure calculations, and may support the technical and regulatory decisions that the EFSA working group has to take.

MO156

Effectiveness of grass buffer strips in reducing Spinosad runoff

S. Otto, Italian National Research Council; S. Gottardi, M. Pasini, Agrea SRL; R. Bradascio, Dow AgroSciences Italia srl / RD; O. de Cirugeda Helle, Dow AgroSciences

Outcome from FOCUS and a recent Document from Italian Ministry of Health allows vegetated buffer areas as a mitigation measure for runoff to surface water. The modelling framework includes default values relating to the pesticide removal efficiency of such buffers. Recent research suggests that these default removal efficiency values are possibly too low for Spinosad, and that experimentally derived values specific to Spinosad may be useful in demonstrating compliance. A GLP field study was performed in summer 2017 to test vegetated buffer strip removal efficiency, in relation to Spinosad and its major metabolites, and based to the FOCUS Surface Water risk assessment scenarios, but with worse (and prudential) conditions. The selected site was near Verona, in an hilly zone rich in vineyards and famous for high-quality vine production. Runoff containing a known amount of each of the four spinosad components to be tested (the parents – spinosyn A and spinosyn D; and the metabolites – spinosyn B and spinosyn B of D), have been artificially applied to 9 grass plots of 4.2 m width and 12 m length, with sandy-loam soil and slope ranging from 10 to 13%. Natural vegetation cover was 60-90%. The artificial runoff was organized to simulate a runoff generated in a source area of 500 m^2 flowing in a run-on area (buffer area) of 50 m^2, to evaluate its buffer capacity both in runoff displacement and concentration. Runoff event consisted in 2 phases: 1) Irrigation with sprinkler at 14 mm/h for 50 min (total: 12 mm), to simulate rainfall before runoff; 2) Run-on/Runoff event: 200 mm of water were released into the buffer area in 2:20 hours using a runoff generator (flow of 85 mm/h). Water contained a precise amount of the 4 spinosyns and a tracer (KBr); once released into buffer area, the “Run-on” becomes “Runoff”, and Runoff water was sampled at 0.75, 1.5 and 2.20 hours after Run-on start. During Run-on, irrigation continued until the end of run-on (other 33 mm), and a total of 45 mm were applied to buffer area. Given the frequencies of selected rainfall (low, return period of 2 years), the runoff/rainfall rate (high, 45%), the source to buffer area proportion (high, 10 to 1), and the plot slope (from 10% to 13%), conditions of the experiment can be considered highly precautionary, and more prudential that those of Focus R4. First results show that the runoff displacement ranges from 3 to 11 m from Runoff releasing. Analysis of spinosyns concentration are in progress.

MO157

EFSA's innovative guidance on the establishment of the residue definition for dietary risk assessment

R. Leuschner, EFSA - European Food Safety Authority / Pesticides, Regulated Products (REPRO); A. Friel°, EFSA - European Food Safety Authority / Pesticides Regulated Products REPRO

*The positions and opinions presented in this poster are those of the authors and are not intended to represent the views or scientific works of EFSA Commission Regulation (EU) No 283/2013 setting out the data requirements for pesticide active substances provides that the toxicological significance of compounds and their amount likely to be present shall be considered when judging which compounds are to be included in the residue definition for risk assessment. On request of the European Commission, EFSA prepared a guidance[1] on the residue definition for dietary risk assessment which intends to complement the OECD guidance[2]. The EFSA guidance is intended as a practical instrument helping risk assessors, on the basis of factual information (derived from toxicological and metabolism data), non-animal testing methods, by weight of evidence, to transparently: Conclude for which residues of a pesticide on food and feed commodities a hazard identification and characterisation is needed; Perform such a hazard identification and characterisation; Define the compounds that should be included in the residue definition for risk assessment. The innovative aspect of the EFSA guidance is a structured sequence of three modules, each of which addresses hazard characterisation and dietary exposure by selecting genotoxicity as the starting criteria for human health assessment. The modules are: Module 1: Exclusion of genotoxicity; Module 2: General toxicity assessment; Module 3: Decision making for residue definition for risk assessment. The guidance provides as appendices an analysis of ADI and ARfD distribution for pesticide active substances and three case studies illustrating the practical application of this modular approach to derive a residue definition for dietary risk assessment for isoproturon, spiroxamine and epoxiconazole. In September 2016, EFSA organised a technical meeting[3] with stakeholders on its new guidance to exchange views. [1]EFSA PPR Panel (EFSA Panel on Plant Protection Products and their Residues), 2016. Guidance on the establishment of the residue definition for dietary risk assessment. EFSA Journal 2016;14(12):4549, 129 pp. doi:10.2903/j.efsa.2016.4549. [2]OECD (Organisation for Economic Co-operation and Development), 2009. Series on testing and assessment No. 63 and Series on pesticides No. 31 Guidance document on the definition of residue; ENV/JM/MONO(2009) 30; 28-Jul-2009. [3]Info session on applications – pesticides - technical meeting with stakeholders on EFSA GD on residue definition for dietary risk assessment. http://www.efsa.europa.eu/it/events/event/160926

Alternative Approaches to Animal Testing for Ecotoxicity Assessments (P)

MO158

Investigations on the bioconcentration of xenobiotics in the freshwater amphipod Hyalella azteca

C. Schlechtriem, Fraunhofer IME / Department Bioaccumulation and Animal Metabolism; J. Hollender, Eawag / Environmental Chemistry; Q. Fu, Eawag, Swiss Federal Institute of Aquatic Science and Technology / Department of Environmental Chemistry; V. Kosfeld, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology; D. Esser, A. Schulte, Fraunhofer IME; I. Ebersbach, Fraunhofer IME Institute for Molecular Biology and Applied Ecology; I. Bischof, Fraunhofer Institute for Molecular Biology and Applied Ecology IME

Bioconcentration factors (BCF) are needed for regulatory purposes to assess the bioaccumulative characteristics of a substance in the aquatic environment. Traditionally these BCFs are determined in fish flow-through tests according to TGD OECD 305. These fish bioaccumulation studies are time consuming, expensive, and demand many laboratory animals. Accordingly, alternative methods that replace, reduce and refine (3Rs) this test system are needed. Two promising alternative test approaches have been developed as alternative to *in-vivo BCF testing*: I) An invertebrate flow-through bioconcentration test system using the freshwater amphipod *Hyalella azteca* and II) *in vitro* depletion assays with rainbow trout hepatocytes or rainbow trout hepatocyte S9 fractions. Flow-through test with almost 20 compounds showed, that the *H. azteca* bioconcentration test could be an appropriate test to predict bioconcentration in the standard fish test. Bioconcentration studies with *H. azteca* would support laboratory animal welfare considerations using a non-vertebrate species, improve efficiency and reduce costs for BCF-testing. The results of the *in vitro* depletion assays have been successfully applied to improve *in-silico* predictions for BCF values by adding the highly variable aspect of metabolism capacity to the existing BCF prediction models. In this study a total of five substances with different characteristics, four substances with log K_{OW} values ranging from 2.5 to 4.5 and one ionic substance, were tested both test systems. To obtain a more detailed understanding of the metabolic activities in *H. azteca*, its metabolism of the five tested compounds were compared to the metabolites generated in vitro by rainbow trout hepatocytes. The results show that Hyalella BCF testing, in addition to in vitro assays and in silico predictions, may help to reduce, refine, and replace the classic BCF estimation with fish in accordance to regulatory needs.

MO159
Assessing Differences in Sensitivity to Aromatase Inhibitors Among Freshwater Fish Species
J.A. Doering, US EPA / Mid Continent Ecology Division; G.T. Ankley, U.S. EPA / National Health and Environmental Effects Research Laboratory; B.R. Blackwell, USEPA ORD / National Health and Environmental Effects Research Laboratory; J.E. Cavallin, U.S. EPA / US EPA MidContinent Ecology Division; K.A. Fay, CSRA, Inc.; D. Feifarek, SC Johnson / GSARA; K.M. Jensen, U.S. EPA / Mid-Continent Ecology Division; M. Kahl, U.S. EPA / National Health and Environmental Effects Research Laboratory; C. LaLone, U.S. EPA / Mid Continent Ecology Division; S. Poole, US EPA Mid-Continent Ecolgy Div / National Health and Environmental Effects Research Laboratory; E. Randolph, U.S. EPA / National Health and Environmental Effects Research Laboratory; C. Tilton, U.S. EPA / ORD; D.L. Villeneuve, U.S. EPA / National Health and Environmental Effects Research Laboratory
There is significant concern regarding potential impairment of fish reproduction associated with exposure to endocrine disrupting chemicals (EDCs). Aromatase is a steroidogenic enzyme involved in the conversion of androgens to estrogens. Inhibition of aromatase activity by exposure to chemicals can reduce levels of circulating estrogen leading to reduced synthesis of vitellogenin and production of fewer eggs by females. This mechanism has been extensively studied in the laboratory model species, fathead minnow (*Pimephales promelas*). However, differences in sensitivity to inhibition of aromatase among species of fish is largely unknown. This is particularly true for species that are not routinely studied in short-term reproduction assays, including many fishes of significant ecological and economic importance such as catfish (Ictaluridae), eel (Anguillidae), and perch (Percidae). This study investigated *in vitro* inhibition of aromatase by the model inhibitor, fadrozole, across eighteen phylogenetically diverse species of freshwater fish. Concentrations of fadrozole that result in 50 % inhibition of *in vitro* aromatase activity range from 0.0014 to 0.088 nM among these species. This suggests that intrinsic differences in sensitivity to inhibition of aromatase could be greater than 60-fold among fishes. Paddlefish (Polyodontidae), white sucker (Catostomidae), rainbow trout (Salmonidae), and fathead minnow (Cyprinidae) were investigated for sensitivity to *in vitro* inhibition of aromatase by four additional inhibitors. Potencies of letrozole, imazalil, prochloraz, and propiconazole relative to fadrozole were comparable among paddlefish, white sucker, rainbow trout, and fathead minnow despite up to 40-fold difference in sensitivity to fadrozole. This suggests that relative potencies generated for a model species, such as fathead minnow, could be applicable across diverse species, despite great differences in relative sensitivity. Results of this study are being used in the construction of a cross-species quantitative adverse outcome pathway (qAOP) that incorporates *in vitro* sensitivity data as a relative-sensitivity adjustment for prediction of impacts at the individual and population level. This information could guide more objective ecological risk assessments of native species to EDCs that inhibit aromatase. *The content of this presentation neither constitute nor necessarily reflect US EPA policy.*

MO160
Fish scales as a tool for temporal biomonitoring of trace element concentrations
D.A. Vignati, CNRS / LIEC UMR7360; G. Masson, Université de Lorraine and CNRS / LIEC UMR7360
Direct measurement of contaminant concentrations in biological tissues is attractive for monitoring purposes because it accounts, in principle, for the environmental factors controlling their bioaccessibility and bioavailability. In the case of trace elements, the validity of this approach is confirmed by its adoption in the regulatory European framework for mercury measurement in fish. For other elements, the current framework privileges measurements in the dissolved (filterable) aqueous matrix. However, this approach provides only a limited temporal resolution of possible trends in elements' concentrations and, where relevant, neglects uptake via dietary pathways. Furthermore, when fish is the selected matrix for monitoring, one or more individuals have to be sacrificed to collect the material necessary for analysis; a strategy that, apart from the associated ethical problems, may become problematic in situations where the number of resident fishes is limited. In this contribution we examined the use of fish scales as a non-lethal, rapid and efficient alternative for monitoring trends in trace element levels in a reservoir receiving cooling waters from a nuclear power generation plant. The variations in the concentrations of Cu, Zn and lanthanides were followed in fish scales from archived fish material (*Abramis brama*) collected annually between 1990 and 2016. Scales were dried, calcinated and mineralised using concentrated nitric acid. After dilution, Cu and Zn were assayed by atomic absorption spectrometry and lanthanides by ICP-MS. For Cu and Zn, triplicate measurements generally agreed to 10% and accuracy, with respect to the standard reference material TORT2 (lobster hepatopancreas), was better than 90%. In the case of lanthanides, analysis were made on one sample per year and only La, Ce, Pr, Nd, Pm, Sm, Eu and Gd could be systematically quantified. Overall, fish scales proved to be a suitable matrix for the biomonitoring of trace element concentrations, including elements of emerging concerns such as lanthanides. Compared with fish muscle (or whole fish), fish scales are easier to preserve (no need for dissection, freezing and lyophilization) and, in some situations, may even allow repeated surveillance of the same individuals. This methodological validation study paves the way to further research to establish relationships between accumulation in scales, internal organs and biological responses.

MO161
Assessing differences in sea turtle organ sensitivity using cell-based toxicity assays
K. Finlayson, Griffith University - Smart Water Research Centre / Australian Rivers Institute; F.D. Leusch, J. van de Merwe, Griffith University / Australian Rivers Institute
The long-lived nature of sea turtles and their lengthy residence in coastal foraging grounds can result in high exposure to contaminants from urban, industrial and agricultural sources. Organic or inorganic contaminants have been quantified in all species of sea turtles worldwide. However, very little is known about how these contaminants impact turtle health, valuable information for identifying populations at risk. *In vitro* exposure experiments using cell cultures established from turtle tissue provide an ethical, reproducible and cost-effective method to identify threats of environmentally relevant contaminants to sea turtles. In recent years, the majority of sea turtle cells lines have been established from skin samples. However, as ingestion is the main route of exposure for these animals, cell cultures established from organ tissues may be more pertinent. This study used a number of primary cell cultures established from the skin, ovary, heart, liver and small intestine from three individual green turtles to investigate the variation in sensitivity between organ types. Cytotoxicity of five organics and five inorganic compounds was investigated using a total of 13 cell cultures. Differences between the cell cultures were investigated based on their order of sensitivity to all compounds tested. The results were then used to assess risks to sea turtle populations worldwide. The results provide recommendations for further toxicological studies involving turtle cells lines that will allow more robust and meaningful risk assessments to be conducted for sea turtles, assisting conservation and management strategies worldwide. Our results also support the use of sea turtle cell cultures as an ethical and reliable method for investigating toxicological effects of environmental contaminants.

MO162
Comparison of rat liver S9 to an animal-free alternative ewoS9R in the Ames fluctuation assay
J. Brendt, RWTH Aachen University; B. Thalmann, EWOMIS; K. Bluhm, University of Saskatchewan; K. Kauffmann, RWTH Aachen University / Department of Biochemical Engineering; S.E. Crawford, RWTH Aachen University / Institute for Environmental Research, Dept. of Environmental Analysis; A. Schiwy, EWOMIS; J. Büchs, RWTH Aachen University / Department of Biochemical Engineering; H. Hollert, RWTH Aachen University / Institute for Environmental Research
The Ames test is the most important *in vitro* test for mutagenicity performed in many variants. The original agar-plate assay was modified to reduce the amount of assay components like rat liver S9 and the length of time needed for test preparation and evaluation. The Ames fluctuation test was established as a less time and material consuming method and introduced into international guidelines like OECD 471 and ISO 11350. In the Ames test the bacterium *Salmonella typhimurium*, with a lack of histidine-synthesis, is exposed to a sample to determine the mutagenic potential, measured by the ability of revertant bacteria to synthesize histidine. The resulting growth in a histidine-free medium is visible due to a colour change caused by acidification in a pH indicator medium. Nevertheless, it can be further improved and combined with the RAMOS-technique (Respiration Activity Monitoring System). This technique improves the sensitivity and the data output of the fluctuation variant. It enables a precise measurement of the oxygen transfer rate (OTR) and therefore conclusions on growth and metabolism of the bacterial culture. Furthermore, it can be implemented as an online-monitoring system on mutagenicity in applications like drinking water monitoring. However, as the Ames test is conducted with a microorganism, it lacks the metabolic activation of a mammalian metabolism. Hence, the mutagenicity of promutagens would be underestimated. Therefore, the bioassay is supplemented with an animal-derived product the rat liver S9. In animal experiments rats are treated with toxic substances via percutaneous or intravenous injections or feeding. Subsequently, the animals are euthanized to obtain the liver, which is homogenized, centrifuged and frozen. The commonly used rat liver S9 is a necessary component in various *in vitro* tests to increase the information about potential mutagenic substances. However, this product varies in its enzyme consistency and purity from batch to batch. Therefore, in the context of 3R to reduce animal experiments and to obtain more reliable *in vitro* assay components alternatives should be introduced. In the present study, we investigate the applicability of a substitution of rat liver S9 with the biotechnological animal-free *ewo*S9*R* in the Ames fluctuation assay and the Ames-RAMOS system. Therefore, we investigate 26 promutagens with both metabolic systems. Preliminary results suggest that *ewo*S9R is a suitable alternative to rat liver S9.

MO163
QSAR: a predictive approach for electronic cigarettes toxicological assessment
D. Zarini, University of Insubria; E. Papa, A. Sangion, University of Insubria /

Department of Theoretical and Applied Sciences (DiSTA); E. Caruso, University of Insubria / DiSTA; S. Zucchi, S. Sterpone, E. Ferri, TRUSTICERT SRL
Electronic cigarettes (e-cigarettes) are devices that typically deliver nicotine, flavorings, and other additives to users via an inhaled aerosol. They are designed to closely mimic the experience of smoking conventional cigarettes. Nowadays, e-cigarettes are the most commonly used tobacco-related product among youth, surpassing conventional cigarettes in 2014. However, insufficient data are available to accurately predict the risk associated with the particulate aerosols to which consumers are exposed; at the same time studies evaluating whether e-cigarettes are less harmful than cigarettes are inconclusive. Minimal valid chemistry data are available on e-cigarette emissions and no standardized methods and threshold values exist for e-cigarette analysis. To fill the chemical and toxicological data gaps, comprehensive assessments of e-cigarette chemical emissions and toxicological studies are certainly needed. The aim of the present study is to provide, by means of Quantitative Structure-Activity Relationship (QSAR) approaches, a first toxicological screening of several e-liquids ingredients. Different Ordinary Least Squares (OLS) regression-based QSAR models were developed to define the potential acute toxicological profile of 265 molecules contained into the e-liquids. The selected end-points were: inhalation LC50 and oral LD50 in mouse and rat respectively. Theoretical descriptors were calculated by PaDEL-Descriptor software, and the best modelling variables were selected in the software QSARINS. Models were validated for robustness, stability and absence of chance correlation using leave-one-out, leave-more-out and the scrambling of the responses. External validation was performed on multiple external prediction sets. The QSAR models have satisfactory fitting, internal and external predictivity with R^2 values and Q_{LOO}^2 values ranging from 0.7 to 0.9 and Q_{EXT}^2 values ranging from 0.7 to 0.8. These models were used to perform a screening of the acute toxicological profile of the 265 molecules of interest and to compile a priority list of substances of potential toxicological concern. This preliminary study represents a first step toward the hazard assessment of e-liquids and for the identification of safer alternatives to existing and potentially harmful ingredients screened from the chemical structure. However, additional work still needs to be done to make these products safe for human use.

MO164

Evaluation of QSAR models for daphnia and fish chronic toxicities of human pharmaceuticals

T. Yamada, National Institute of Health Sciences; M. Kurimoto, National Institute of Health Sciences / Division of Risk Assessment; H. Shiraishi, National Institute for Environmental Studies; H. Yamamoto, National Institute for Environmental Studies / Center for Health and Environmental Risk Research; N. Tatarazako, Ehime University / Center for Health and Environmental Risk Research; T. Nishimura, Teikyo Heisei University / Faculty of Pharmaceutical Sciences; A. Hirose, National Institute of Health Sciences / Division of Risk Assessment
Recently, medical regulatory agencies require pharmaceutical companies to assess environmental impacts of new pharmaceutical products before marketing. Hence, it would be valuable to predict ecotoxicity of new pharmaceuticals at developmental stage. As a tool for predicting toxicity in daphnia and fish, two QSAR programs ECOSAR by USEPA and KATE by Ministry of Environment in Japan are available, both of which are built using dataset of mainly industrial chemicals. In this study, we evaluated applicability and predictivity of the QSAR models using external dataset of the chronic ecotoxicity of human pharmaceuticals. The chemical structures and toxicity data based on *D. magna* reproduction test (OECD TG211) and fish early-life stage toxicity test (OECD TG210) were gathered from public domain. In order to examine the applicable domain where more reliable prediction results can be obtained, the following criteria were defined in this study; (1) logP values of target substances are within the lowest and highest values of the category chemicals, (2) number of category members is 5 or more, and (3) correlation coefficients of the linear regressions are greater than 0.70. Since KATE equips models for acute toxicity only in both species, Acute-Chronic Ratio of 10 was applied to estimate NOEC values. Then, ratio of calculated NOEC and measured NOEC (C/M) was determined. For ECOSAR daphnia model, 82 out of 126 pharmaceuticals satisfied the criteria. Of these, 44 pharmaceuticals had C/M between 0.1 and 10, some of which were assigned to amides or aliphatic amines. 72 pharmaceuticals had the C/M between 0.01-100. 12 pharmaceuticals had the values greater than 100, half of which have pharmacological action to neurotransmitter receptors in human. For KATE daphnia model, 19 pharmaceuticals met the criteria. The C/M values were between 0.1 to 10 for 15 substances, most of which belong to primary amines aliphatic/aromatic, amides or imides, or neutral organics. For fish chronic toxicity, only 11 and 21 out of 72 pharmaceuticals satisfied the criteria with ECOSAR and KATE models, respectively. Further examination will be needed to expand applicability by modifying the criteria, combined with other approaches including acute-to-chronic extrapolation or daphnia-fish interspecies extrapolation. This work was supported by the Research on Regulatory Science of Pharmaceuticals and Medical Devices from Japan Agency for Medical Research and Development, AMED.

MO165

Optimization and Accessibility of the Eco- Database and the Ecotoxicological Threshold of Concern (ecoTTC) tool

R.R. Otter, Middle Tennessee State University / Biology; M. Embry, ILSI; S.E. Belanger, The Procter & Gamble Company / Environmental Stewardship and Sustainability Organization; M.G. Barron, U.S. EPA / Gulf Ecology Division; A. Beasley, The Dow Chemical Company / TERC; J. Brill, The Procter & Gamble Co. / Environmental Stewardship and Sustainability; H. Chang, FDA / Center for Tobacco Products; D. Chang, United States Environmental Protection Agency; D. De Zwart, DdZ Ecotox / Centre for Sustainability Environment and Health; B. Farr, ILSI Health and Environmental Sciences Institute (HESI) / Health and Environmental Sciences Institute HESI; A. KIENZLER, JRC-EC / F3-Chemical Safety and Alternative Methods Unit-EURL ECVAM; T.J. Norberg-King, U.S. EPA / NHEERL/Mid-Continent Ecology Division; H. Sanderson, Aarhus University / Environmental Science; P.W. Wilson, Sanofi U.S., Inc. / Health, Safety and Environment
The Ecological Threshold for Toxicological Concern, or ecoTTC, has been proposed as a natural next step to the well-known human safety TTC concept. The ecoTTC is particularly suited for use as an early screening tool in the risk assessment process, in situations where chemical hazard data is poor, or when an appropriate QSAR is unavailable. EcoTTCs are developed using statistical distributions of Predicted No-Observed Effect Concentrations (PNECs) to reflect the breadth and depth of the ecotoxicological dataset beneath, and therefore, the diversity and quality of the underlying dataset is crucial to the future utility of the ecoTTC. An eco-database consisting of approximately 110,000 unique ecotoxicological records, 6200 unique CAS numbers and 1900 species from three trophic groups has been created based on recent assessments of published data and international chemical management programs. Stepwise data selection strategies, query systems and curation techniques were applied to ensure a transparent, methodical process towards a final dataset, which also includes reference-sourced toxicity data associated with physical chemistry data and taxonomic information for the tested chemical. In order to make these data accessible and useful to stakeholders, the dataset was transitioned from Microsoft Excel and Access into a modern MySQL format. This allows for a database format that is relational and scalable, facilitating easy access, sharing, and integration with other datasets and tools. This dataset is accessed via a web-based query system that is integrated with PNEC calculator and probability distribution tools. The novel interface allows users to explore the data, upload additional datasets, derive threshold values based on specific criteria, and explore the potential use and application of the ecoTTC concept. This poster will present the architecture, web-interface, and associated tools and a live demonstration of the web interface and associated web tools will be available.

MO166

Using toxicokinetic and toxicodynamic modelling to predict effects of chronic toxicity on rodent growth based on in vitro assays

T.O. Martin, Environment Department, University of York / Environment Department; R. Ashauer, University of York / Environment; P. Thorbek, Syngenta / Environmetal Safety
According to 2011 figures, 80% of the animals used for testing procedures in the European Union are rodents and almost 23% are used in longer term repeated dose tests. Alternative methods to predict the effects of chronic toxicity in rodents can therefore make a significant contribution to the reduction, replacement and refinement (3R’s) of animal testing. Body weight is one of the many endpoints monitored throughout chronic toxicity tests. We aim to develop *in silico* models to extrapolate the effect of toxicant exposure, measured as an appropriate internal dose metric, on the growth of rodents from *in vitro* assays. Initially, *in vivo* toxicokinetic-toxicodynamic models will be used to predict the selected internal dose metric and its effect on growth over the duration of repeated dose toxicity studies. These models will be developed using data from regulatory toxicity testing of pesticides. Experiments will then be designed to assess the effects of known intracellular pesticide concentrations on cell population growth *in vitro*. Cell number can be converted to cell mass, after which it should be possible to model the effects of matching internal doses on growth over time, *in vitro* and *in vivo.* The weight normalised effect on growth ($mass_{dose\ group}$ / $mass_{control}$ at a given time point) can then be calculated at various points along the predicted *in vitro* growth curves.These predictions can then be compared to corresponding *in vivo* observations. The predictive ability of this extrapolation will be explored for 10 pesticides, which will provide a good indication of the reliability and repeatability of the methods. Should predictions prove to be consistently accurate, this will provide a fast and inexpensive *in vitro* screen for body weight effects in rodents. Initially this may be applied as an alternative to range finding studies which are not a regulatory requirement but are commonly carried out prior to regulatory testing. In the longer term this may form part of a suite of *in vitro* and *in silico* alternatives to *in vivo* chronic toxicity testing.

MO167

Screening of metabolic- and neurotoxicity of environmental chemicals using C. elegans and transgenic zebrafish models

Y. Kim, S. Lee, University of Seoul; H. Lee, University of Seoul / School of Environmental Engineering; N. Chatterjee, University of Seoul / Environmental Engineering; J. Choi, University of Seoul / School of Environmental Engineering
Metabolic and neurodevelopmental disease have been attracting attention as

environmental disease. Epidemiological evidences show that the disease is associated with exposure to hazardous chemicals. However, causal relationship has not been clearly understood. In the present study, we aimed to elucidate the link between occurrence of metabolic or neuro disease and exposure of environmental chemicals. We first screened potential of environmental chemicals on the disease model organisms, *C. elegans* and Zebrafish. To maximize the advantage of these model organisms, we conducted the chemical screening using *C.elegans* mutant; oga-1(ok1207), ogt-1(ok1474), nlg-1(ok259), transgenic zebrafish; Tg(T2Kins:nfsB-mCherry)^{jh4} and Tg[elavl3:EGFP]knu3.The highly conserved O-GlcNAc transferase; OGT and O-GlcNAcase; OGA genes are related to type 2 diabetes and null mutations cause alterations in *C. elegans* carbohydrate and lipid metabolism. Neuroligin NLG-1 control synaptic function, which is conserved from nematodes to mammals. It is related to attention deficit hyperactivity disorder (ADHD). Tg(T2Kins:nfsB-mCherry)^{jh4} fish express insulin nitroreductase(InsNTR) mcherry fusion protein in the pancreatic β-cell and Tg[elavl3:EGFP]knu3 fish express GFP in most post-mitotic neurons. Various category of environmental chemicals, such as, heavy metals (i.e. arsenic, lead, cadmium), EDCs (i.e. Nonylphenol, Bisphenol-A,E,D,F,S) and biocides (i.e. Chloropyrifos, CMIT/MIT, PGH), were screened using *C. elegans* reproduction assay and zebrafish transgenic assay. The preliminary results showed CMIT/MIT and BPA reduced fluorescence intensity of insulin gene on zebrafish, suggesting possible involvement of these chemicals on metabolic pathways. In brief, our results suggest multi-model approach could complement the limitations of a stand-alone model organism and thus more accurately identify chemical hazard on human health. Therefore, the outcome of the current study could be utilized for efficient chemical screening and better risk assessment of the chemicals.
Acknowledgement: This work was supported by the Mid-career Researcher Program (2017R1A2B3002242) through the National Research Foundation of Korea (NRF) funded by the Ministry of Science, ICT and Future Planning.

MO168

In vitro effects of two pesticides on the motility and viability of bovine spermatozoa

I. Bulhosa, University of Aveiro / Biology department; M. Lopes, ICBAS-University of Porto / Department Veterinary Clinics; I. Lopes, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro

The use of plant protection products has exponentially increased in the agricultural sector over the past decades. Copper sulfate and glyphosate are two commonly used pesticides, the former as fungicide and the latter as herbicide. Farm animals may be exposed to this type of products through different ways: i) the drift of pesticides during their application may lead to inhalation or dermic exposure or ii) through the ingestion of contaminated food. This exposure may lead to adverse effects in the reproduction of those animals. Actually, spermatozoa are extremely sensitive to slight variations in the organism. The interaction between chemicals and sperm may alter its mobility; velocity and/or viability depending on which cell structures are affected. This work aimed at assessing the toxicity of ecological relevant concentrations of copper sulfate and glyphosate on bovine spermatozoa. Commercial frozen semen from five different bulls was exposed to three concentrations of the two pesticides, diluted in phosphate-buffered saline (PBS), plus a control (PBS). For each bull, three replicates were made. Motility and velocity endpoints were measured with a sperm analyzer computer program and viability was measured using an eosin-nigrosin staining procedure. Endpoints were measured at 0, 30 and 90 minutes. Copper sulfate did not induce significant effects on the monitored parameters, but for the first observation period (time=0 minutes) the lowest concentration showed higher motility and velocity than the other treatments, suggesting that copper may enhance motility at low concentrations. Glyphosate significantly reduced the motility and viability of spermatozoa. *In vitro* results are limited, but they are a good starting point for dose calculations and for unveiling primary mechanisms of toxicity without the need to use living beings.

MO169

Assessing the bioaccumulation potential of several pharmaceuticals using fish S9 and hepatocyte assays

L.A. Constantine, Pfizer, Inc. / PDM; M. Embry, ILSI; R. Sharma, Pfizer / PDM

As per the EMA Guideline for Environmental Risk Assessment of Medicinal Products for Human Use, a fish bioconcentration study is triggered in Phase I for pharmaceuticals having a log K_{ow} > 4.5 to support the Persistence, Bioaccumulation and Toxicity (PBT) assessment and in Phase II, Tier A for pharmaceuticals having a log K_{ow} > 3. The recommended protocol for bioconcentration is OECD Test Guideline 305: Bioaccumulation in Fish, Aqueous and Dietary Exposure. However, in the interest of reducing the number of animals used in environmental testing, data from *in silico*, *in vitro* and *in vivo* assays have been developed to support a weight of evidence approach to assess bioaccumulation potential in fish. A draft guideline entitled, Determination of *in vitro* intrinsic clearance using cryopreserved hepatocytes (RT-HEP) or liver S9 sub-cellular fractions (RT-S9) from rainbow trout and extrapolation to *in vivo* intrinsic clearance is currently undergoing OECD review. The procedures as outlined in this draft guideline were used to determine measured *in vitro* intrinsic clearance rates. These rates were then used to predict fish BCF values for several active pharmaceutical ingredients for which *in vivo* clearance and fish BCF values have been determined as per the OECD 305 Guideline. The outcome of these *in vitro* assays will be presented along with the *in vivo* BCF data.

MO170

Chemoavailability of Organic Electrophiles - A Nonanimal Approach to Identify Candidates for Reactive Toxicity

A. Böhme, UFZ - Helmholtz Centre for Environmental Research / Ecological chemistry; G. Schuurmann, Helmholtz centre for environmental research - UFZ / Department of Ecological Chemistry

Organic electrophiles are important components within the exposomes of humans, flora and fauna. Their toxicity toward aquatic organisms is driven by two molecular initiating events (MIE): the hydrophobicity-triggered disturbance of cellular membranes and the chemical reaction with nucleophilic sites of proteins, peptides or the DNA. The toxicity enhancement T_e, which indicates the ratio of narcosis baseline (hydrophobic MIE) vs. experimental in vivo or in vitro bioassay toxicity, has been used as a measure for the reactive MIE for many years. However, very early studies already show that T_e does not solely depend on reactivity, but also decreases with increasing hydrophobicity. This indicates that the relevant nucleophilic targets are located in aqueous compartments and that the hydrophobic and the reactive MIEs do not contribute independently to overall toxicity. In this communication, we employ our concept of chemoavailability[1,2] to a set of 58 Michael acceptors, in order to analyze the impacts of reactivity and hydrophobicity on the overall toxicity as well as on T_e. To this end, reactivity was quantified by the second order rate constant for the reaction of the Michael acceptors with glutathione (GSH) k_{GSH},[3] hydrophobicity through the octanol/water partition coefficient and toxicity through the 48-h-effect concentration yielding 50 % growth inhibition of *Tetrahymena pyriformis.* The results demonstrate that the decreasing T_e with increasing K_{ow} is caused by a rate-determining transfer-step of the electrophile from lipophilic compartments into the aqueous cytosol. Finally, chemoavailibilty, as a trade-off between log k_{GSH} and log K_{ow}, is shown as a promising nonanimal tool to analyze whether aquatic toxicity is predominantly driven by the hydrophobic or the reactive MIE, or by both MIEs working in parallel. The authors thank the EU-funded project OSIRIS (GOCE-CT-2007-037017) and the BMBF-funded project ProHapTox (FKZ 031A422A and 031A422B) for financial support. [1] Mulliner D, Schüürmann G 2013. *Mol. Inf.* 32: 98-107. [2] Böhme A, Laqua A, Schüürmann G 2016. *Chem. Res. Toxicol.* 29: 952-962. [3] Böhme A, Thaens D, Paschke A, Schüürmann G 2009. *Chem. Res. Tox.* 22: 742-750.

MO171

Local Electrophilicity Describes Experimental Glutathione Reactivity and Aquatic Toxicity toward Tetrahymena pyriformis

D. Wondrousch, G. Schuurmann, Helmholtz centre for environmental research - UFZ / Department of Ecological Chemistry

Electrophilic compounds such as *α,β*-unsaturated carbonyls are valuable reactants in organic synthesis and commonly used as industrial intermediates and products. At the same time, their electrophilic reactivity may become critical in case of uptake into organisms because of the ubiquitous presence of nucleophilic sites in proteins and DNA, resulting in reactivity driven excess toxicity. Therefore, exposure to electrophiles is of high toxicological concern. Thus, identification of toxicologically relevant compounds is desired. A step forward would be to predict – rather than measure – the electrophilic reactivity of compounds directly from molecular structure. This would enable screening with regard to their intrinsic toxicity potential. In this work, local electrophilicity parameters were developed based on quantum chemistry. Their performance to describe electrophilic reactivity was investigated. For this purpose, their correlation with logarithmic reaction rate constants toward Glutathione (GSH) was analyzed. GSH is a small tripeptide which acts as a protector against electrophiles in the cytosol. The dataset of electrophilic chemicals contained 97 *α,β*-unsaturated esters, ketones and aldehydes. In the context of aquatic toxicity toward *Tetrahymena pyriformis*, reactive toxicity is assumed to be the primary mode of action of the aforementioned compound classes. Therefore, the descriptive power of calculated and experimental GSH reaction rate constants was compared: Both models perform equally well and yield root mean squared errors of about 0.4 log units in modeling the toxicity enhancement as deviation from narcosis level based on 48h-inhibition-growth concentrations.

MO172

Using mechanisms of toxic action to classify and predict ester ecotoxicity

P. Bicherel, F. Bauer, KREATiS; P.C. Thomas, CEHTRA SAS / Ecotoxicology and Risk Assessment

Even though esters are often used and released into the environment, little is known about their mechanisms of action in relation to their toxicity. Most of the simple esters are considered to exert a specific narcosis, while some other esters can exert toxicity related to their potential reactivity. Therefore the critical step, before predicting the toxicity of an ester, is to determine its mechanism of toxic action (MechoA). For this purpose the classification of Bauer *et al*., (2018) is used in combination with an accurate modelling approach which is derived from empirical data specific to the MechoA. The acute toxicity of esters to aquatic flora and fauna may be regressed against a hydrophobicity descriptor (i.e. log K_{OW} or water solubility), and compared with similar regressions for non-polar narcotics. The similarity between these regressions confirms non-reactive esters are simple narcotic compounds for algae but not for fish or daphnids. For the animal species, the regressions for esters are not the same as for narcotic compounds. The most likely explanation for this difference is the balance between hydrolysis rate and the toxic action of the parent and the degradation products. That is why they are considered as pronarcotics. A toxicity to algae in line with a narcotic mode of action suggests that the enzyme responsible for hydrolysis found in fish and daphnids is absent and therefore hydrolytic activity of esters by algae is negligible. Thus, di-esters appear more toxic than mono-esters for fish and daphnids because they can produce two times more metabolite than mono-esters. The more reactive esters are usually unsaturated, like allyl/vinyl-esters and alpha,beta-unsaturated esters, whose double bond can be activated by the carbonyl group. For these compounds the substitution around the double bond plays a decisive role in effective reactivity. Thus, methacrylates which have an alkyl substituent in position alpha of the carbonyl are not more toxic than aliphatic esters for any of the three aquatic species. On the other hand, Acrylates clearly express excess toxicity and have to be considered as acting through yet another mechanism of action relating to a model dedicated to soft electrophiles. Rather than model the toxicity only according to structural analogy, a modelling approach is used to develop QSARs for esters based on three pillars: structure, mechanism and species metabolism.

MO173

Nanosecond pulsed electric field incorporation technique to predict molecular mechanisms of teratogenicity and developmental toxicity on fish embryos

K. Arizono, Prefectural University of Kumamoto / Faculty of Env Symbiotic Science; A. yamaguchi, National Institute of Technology, Ariake College; M. Uchida, Ariake National College of Technology / Department of Chemical and Biological Engineering; H. Ishibashi, Ehime University / Faculty of Agriculture; S. Kono, National Institute of Technology, Ariake College; N. Tominaga, Ariake National College of Technology / Department of Chemical and Biological Engineering

We developed and applied the nanosecond pulsed electric field (nsPEF) treatment condition and assessed the teratogenicity and embryonic developmental toxicity by chemicals using fish embryos. Furthermore, we analyzed gene expression profiles in fish embryos using DNA microarray and performed pathway and network analyses to understand the molecular mechanisms of chemicals in teratogenicity and embryonic developmental toxicity. Our findings suggested that nsPEF technique is a powerful tool for assessing teratogenicity and embryonic developmental toxicity of chemicals and predict their molecular mechanisms in fish embryos.

MO174

Moving 3D in vitro intestinal models forward: transcriptomic characterization of the RTgutGC cell line.

L.M. Langan, Plymouth University / Biological and Marine Sciences; S. Owen, AstraZeneca / Safety Health Environment; A.N. Jha, Plymouth University / Biological Sciences

Intestinal derived cell lines are useful *in vitro* models which allow for focused investigations of metabolism and other responses in the intestine. The development of the first immortalized intestinal cell line derived from the rainbow trout (RTgutGC) offered an opportunity to explore intestinal uptake without the need for the use of numerous animals. Recent work using numerous compounds has acknowledged its potential as a replacement tool for animal based laboratory studies, there is still a lot to be explored before its widespread incorporation as a toxicity tool. Cell lines are known to acquire additional mutations or modifications while in culture, and it is important to understand to what extent this cell line retains the genetic landscape of primary intestinal tissue. In this study, RNA-Seq sequencing of the RTgutGC cell line was used to establish gene expression in this potential animal replacement model. Over 84% of the sequences were mapped to the genome. Following filtering for transcript abundance using TPM (transcripts per million), 24,890 contigs were identified and blasted against the NR database. InterProScan was run in parallel to blast annotation and later merged with annotation to confirm. Over 43 genes were shown to be differentially expressed in the cell line compared to the native tissue while 229 were shown to be down regulated. KEGG pathway analysis revealed the presence of significant metabolism pathways still active in the model. This study provides the first in-depth sequence data of any rainbow trout cell line and identifies many commonalities between the 3D model and native tissue. Characterization of the RTgutGC transcriptome and genes and enzymes expressed in this model will greatly help in building realistic *in silico* models of exposure when integrated with other available chemical data.

MO175

Impact of test concentration on the in vitro intrinsic clearance using trout liver S9 fractions to predict the bioaccumulation potential of fragrance chemicals

H. Laue, Givaudan Schweiz AG / Fragrances S & T; L. Hostettler, Givaudan Schweiz AG; K. Jenner, Givaudan / Global Regulatory Affairs & Product Safety; G. Sanders, Givaudan International SA / Regulatory Affairs and Product Safety; A. Natsch, Givaudan Schweiz AG / Fragrances S &T

Bioaccumulation in aquatic species is a critical endpoint in the regulatory assessment of chemicals. The bioconcentration factor (BCF) is usually determined in fish (OECD TG 305). *In vitro* systems measuring biotransformation rates of chemicals to refine BCF model estimates have been established as alternative methods to refine predictive models which are based on hydrophobicity (i.e. log K_{ow}). Fragrance chemicals frequently contain different isomers complicating its analysis especially at low concentrations. Thus, they have been commonly tested at 1 µM. Results reported recently indicate that first order depletion rate constants (k_{dep}) measured at test concentrations of 1 µM could underestimate the in vitro intrinsic clearance resulting in an overestimation of the BCFs. However, these observations were mainly reported for substances from one chemical class (polyaromatic hydrocarbons, PAHs). For pyrene, chrysene and benzo[*a*]pyrene, k_{dep} determined at lower concentrations were 4- to 12-fold higher than k_{dep} measured at 1 µM. However, the effect of test concentration of industrial chemicals is lacking. The goal of this study was to compare k_{dep} values using different concentrations (e.g. 0.2, 1 and 5 µM) for four fragrance chemicals. These chemicals represent a diverse class of high log K_{ow} (4.3-6.5) industrial chemicals. Rainbow trout liver S9 fractions from different sources were used and their enzymatic activity characterized using commonly used fluorescence assays (EROD, *p*-nitrophenol glucuronidation and CDNB-glutathione conjugation) and substrate depletion assays with testosterone, 7-hydroxycoumarin, pyrene and Cyclohexyl salicylate as reference chemicals. Decrease of the parent chemicals was analysed by GC-MS or LC-MS and k_{dep} values determined. For the lowest concentration (0.2 µM) ca. 2-fold higher k_{dep} values were observed for Polysantol, Ambrofix, Cyclohexyl salicylate and Karanal compared to k_{dep} values determined with 1 µM. Measured k_{dep} values were 2-fold lower with 5 µM except for a 4-fold lower rate for Polysantol compared to 1 µM test concentration. The biotransformation rates of the fragrance chemicals tested seem to be less affected by the test concentration (0.2-5 µM) compared to PAHs indicating that their K_M may be substantially higher. Thus, for fragrance chemicals which are moderately to rapidly biotransformed, the use of 1 µM as start concentration seems to be a suitable approach to estimate the bioaccumulation potential.

MO176

Biological effects of 3 metals on "D" larvae of japonese oyster Crassostrea gigas

A. Sobrino-Figueroa, Universidad Autonoma Metropolitana Iztapalapa / Hidrobiology; C. Cáceres-Martínez, Universidad Autónoma de Baja California Sur

The Japanese oyster is an introduced species from Asia, which is cultivated in the coastal systems of the Mexican Pacific. Due to the fact that in the last 10 years the populations have had problems in their survival, in this work the evaluation of 3 biomarkers was performed in "D" larvae of this species, exposed to the metals Cd, Cr , Pb and its mixture, because these xenobiotics, are in high concentrations in the sites where the oysters are grown. Bioassays (72 hrs) were conducted where the "D" larvae were exposed to 5 concentrations of metal and their mixtures in proportion: 1:1. With the obtained data, the LC_{50} was calculated and the evaluation of 3 biomarkers was measured in the surviving organisms: the degree of lipoperoxidation (Tbars: Buege & Aust. 1978), the activity of the AchE enzyme (Ellman et al., 1961) and genetic damage (Comet Test: Singh et al., 1988). The toxicity of metals according to the calculated LC_{50} values was: (from most to least toxic): Pb = Cd> Cr. The most toxic metal mixture was Cd + Cr. The Kruscal-Wallis test indicated that there are significant differences in the degree of lipoperoxidation, inhibition of AchE activity and genetic damage between the exposed organisms and the control group. The metal with the highest oxidative effect was Chromium (32 ± 8.97 nM Tbars mg^{-1}). And the metal mixture: Cd + Cr + Pb (45 ± 11.89 nM Tbars mg^{-1}). In the evaluation of genotoxicity it was observed that Cadmium had the highest effect (91% cells with damage) and Lead the lowest (43%). Cadmium and the mixture of metals caused inhibition in the activity of AchE (56% 38% respectively). The results of this study show that the Cd, Cr and Pb metals in sublethal concentrations have deleterious effects on the "D" larvae of *Crassostrea gigas.*

MO177

Toxicity effects caused by exposure to Dichlorvos in organisms of different trophic levels

A. Sobrino-Figueroa, Universidad Autonoma Metropolitana Iztapalapa / Hidrobiology; A. Perez-Rojas, Universidad Autonoma Metropolitana Iztapalapa / Lab. Limnology and geology. Department of Hydrobiology

Dichlorvos (DDVP) is an organophosphate insecticide considered by the EPA as highly toxic. Because there are few studies of the effects of DDVP in aquatic organisms The objective of this study was to evaluate the toxicity of Dichlorvos in organisms of different trophic levels Cladocerans: *Daphnia magna*, *Daphnia exilis*, *Daphnia pulex* and *Simocephalus mixtus*. The ostracod *Cypris* sp. and fishes: juvenile charal (*Chrirostoma jordani*) and juvenile zebrafish (*Danio rerio*). In addition their sublethal effects were evaluated by means of assessment of four biomarkers (growth rate, O:N index, lipoperoxidation and inhibition of acetylcholinesterase enzyme). Acute bioassays were performed, the organisms were exposed to 6 pesticide concentrations to determine the LC_{50}. Subsequently tests with duration of 15 days were made where the organisms were exposed to a sublethal concentration (LC_{10}), for assessment of 4 biomarkers (growth rate, O:N index, lipid peroxidation and inhibition of acetylcholinesterase enzyme). The LC_{50} values obtained in the bioassays varied from 5,300 to 0.021 mg L^{-1} In the tests it was evident that the cladoceran *Daphnia exilis* was more sensitive to DDVP compared to other species. The O:N index had values below 9 fact indicates that organisms were in a high degree of stress. Growth rates of intoxicated organisms were between 19 to 49% lower than those observed in the control group. The average concentrations of Tbars registered organisms varied from 2.5 to 25.6 nM Tbars mg^{-1} and show a direct dose-response relationship, since when increasing the time of exposure to DDVP increased the degree of lipid peroxidation in the tissues. A decrease in AChE activity was observed in cladóceros between 22 and 45% and fish from 22 to 35%. The results of this study indicate that the effects of the pesticide DDVP are likely irreversible in some species.

MO178

Characterising estrogenic activity of arctic char tissue extracts in two fish in vitro bioassays

K. Petersen, NIVA - Norwegian Institute for Water Research; M. Hultman, Norwegian Institute for Water Research; J. Bytingsvik, Akvaplan-niva AS; M. Harju, NILU   Norwegian Institute for Air Research; A. Evenset, Akvaplan-niva AS; K. Tollefsen, NIVA / Ecotoxicology and Risk Assessment

Contaminants from anthropogenic activities find their way to the Arctic through long-range atmospheric transport, ocean currents and via transport by living organisms (migrating fish or seabirds). Although the levels of POPs in arctic fish are generally low, local hot-spots of contamination have been demonstrated in freshwater systems affected by seabird guano, such as Lake Ellasjøen at Bear Island (Norway). High concentrations of organic halogenated compounds have been measured in resident populations of Arctic char. Accumulation of dioxin-like compounds of up to 8 times higher levels than the lowest observed effect level for egg mortality in temperate salmonid fish raise concern that residential Arctic char might be at risk for adverse effects at the individual and population level. In this study the aim was to compare the toxic potency of pollutants in Arctic char from the contaminated Lake Ellasjøen with those from the less contaminated Lake Laksvatn at Bear Island. This was done by *in situ* sampling, extraction, and fractionation of liver tissues from the two fish populations. The following fractions (F) were produced: F1- nonpolar POPs such as PCBs, PBDEs and most of the nonpolar pesticides, F2- polar pesticides and metabolites of POPs, and F3- polar POPs (phenolics such as chlorinated phenols and hydroxylated metabolites of PCBs and PBDEs). A method for isolation, cultivation and exposure of primary hepatocytes from Arctic char was developed and used together with the established method for primary hepatocytes from rainbow trout to investigate cytotoxic and estrogenic effects of the fractions. The estrogenic potency, measured as induction of the estrogen receptor, (ER)-mediated production of vitellogenin (Vtg), was higher in liver extracts from Lake Ellasjøen than Lake Laksvatn. Although primary hepatocytes from both species displayed estrogenic activity in response to the F3 from Ellasjøen fish, higher Vtg induction was observed in rainbow trout hepatocytes than in hepatocytes from Arctic char. However, the Arctic char hepatocytes were more susceptible for cytotoxic effects than rainbow trout hepatocytes. F3 and F2 from both fish populations (Lake Ellasjøen and Lake Laksvatn) appeared to have similar effect on cell viability with F3 having largest effect. Chemical analysis was preformed to identify potential contributors to the observed effects. *Acknowledgements:The project was funded by the Norwegian Research Council, project. No. 221373.*

MO179

Ultrasound: A novel approach to non-lethally measure hepatosomatic index in sentinel fish for environmental monitoring programs

A. Manek, University of Saskatchewan / Veterinary Biomedical Sciences; V. Palace, IISD-Experimental Lakes Area; P. Borrett, University of Saskatchewan; L. Hrenchuk, IISDExperimental Lakes Area; M. Murdoch, Stantec Consulting Inc; L.P. Weber, University of Saskatchewan / Veterinary Biomedical Sciences

Canada's environmental effects monitoring (EEM) program studies impacts of metal mining and pulp mill effluents on aquatic receiving environments. The EEM regulations recommend lethal sampling of 20 fish (male and female) of different species to study body condition, liver size (hepatosomatic index-HSI), and gonad size (gonadosomatic index-GSI) during every monitoring cycle. Developing and implementing non-lethal methods for environmental monitoring programs provides an opportunity to protect sentinel endangered fish which might be threatened by repeated lethal sampling. Also, non-lethal methods could protect sentinel fish that are part of systems with low productivity. Ultrasound is a non-invasive tool that has been tested to assess gonad size in fish. Currently, its potential as a non-lethal tool in environmental monitoring programs is not well explored. We conducted feed withdrawal studies in the laboratory to test the accuracy and sensitivity of ultrasound to measure HSI in sentinel fish with a compact liver such as rainbow trout (*Oncorhynchus mykiss*). With the ultimate goal of providing empirical evidence of the applicability and ease of this technique in the field, we also tested the accuracy of ultrasound method to measure HSI in lake trout (*Salvelinus namaycush*) at IISD-experimental lakes area. Our laboratory studies provide a significant correlation for the accuracy (HSI, r^2=0.73, n=16, p< 0.05) and evidence for the sensitivity of ultrasound method (p=0.06, n=7) versus traditional lethal gravimetric method (p< 0.05, n=7) to measure HSI within the acceptable critical effect size for HSI mandated by EEM. Our field ultrasound method testing also revealed a significant correlation between the traditional lethal and ultrasound method in measuring HSI (r^2=0.81, n=9, p< 0.05) in lake trout. Our field analyses provide evidence for the ease and potential application of this technique for future environmental monitoring programs. Our ongoing method refinement and exploration of this technique in sentinel fish with diffuse livers across different size ranges will strengthen our goal of proposing ultrasound as a viable non-lethal alternative to measure HSI on a global scale.

MO180

Weight of evidence for fish acute toxicity: a Bayesian network modelling approach

J. Moe, Norwegian Institute for Water Research (NIVA) / Section for Catchment Processes; A. Lillicrap, NIVA / Ecotoxicology and Risk Assessment; W.G. Landis, Western Washington University / Institute of Environmental Toxicology; S.E. Belanger, The Procter & Gamble Company / Environmental Stewardship and Sustainability Organization

Reduction of animal testing wherever possible is requested by EU Directive 2010/63/EU. Fish Embryo Toxicity (FET) testing can be an alternative to using juvenile fish in acute toxicity testing. However, FET data are currently not accepted as a replacement to juvenile fish acute toxicity data for regulatory purposes such as REACH, without sufficient weight of evidence (WoE). The development of a WoE approach for FET data has been recommended by the European Chemicals Agency to significantly reduce the number of animals required for hazard assessments of chemicals. We propose a Bayesian network (BN) modelling approach for quantifying the weight of evidence. BN is a probabilistic modelling methodology which is an increasingly used in ecological risk assessment as well as in environmental research and assessment more generally. The purpose of the proposed BN model is to integrate information from large and varied ecotoxicological and physico-chemical datasets, and apply it in a WoE approach to predict fish acute toxicity of chemicals from data on fish embryo toxicity testing in combination with other relevant information. The planned steps of the model development and application are: (1) Identification of suitable chemical properties and endpoints as nodes for the BN. (2) Compilation of data on ecotoxicity data for fish embryos and other relevant information. (3) Construction of a BN model for integrating the data and other information in a probabilistic framework. (4) Evaluation of the WoE approach using ecotoxicity data for fish embryos in combination with data on (juvenile) fish acute toxicity. (5) Application of the BN model to assess the risk of various contaminants based on chemicals registered in the European Chemicals Agency database (http://echa.europe.eu).

MO181

Divergent immunomodulatory effects of cadmium between two murine innate immune cell models in vitro, macrophages and mast cells.

D. Garcia, Wageningen University & Research / Toxicology; H.H. van den Berg, N. van den Brink, Wageningen University / Dept of Toxicology

Cadmium can induce toxic effects via different mechanisms, e.g. depletion of cellular antioxidants leading to increase of ROS levels and induction of apoptosis. Such effects have been addressed in different types of tissues and cells, including the immune system. However, these mechanisms of toxicity may have differential impacts among physiological functions, depending on the ability of the targeted cells to deal with these toxic effects. Considering the multitude of different types of immune cells and cell-subsets with different functions, cadmium could impair immune functions, such as the immune responses against infections, through cell-type specific effects. Macrophages and mast cells are two types of innate immune cells part of the first line of defence, able to initiate fast inflammatory responses. Each cell type acts via different mechanisms in the two main types of inflammatory responses, type 1 and type 2. Type 1 or cell-mediated immunity is involved in the defence against intracellular bacteria and infected cells, carried out especially by phagocytes like macrophages. In contrast, mast cells are associated with type 2 or humoral/antibodies-mediated immunity, concerned with extracellular pathogens and parasitic infestations. In order to study the immunomodulatory effects of cadmium on macrophages and mast cells we carried out a mechanistic *in vitro* study. Exposure to cadmium depleted glutathione in the four cell lines tested, potentially modulating functional parameters in macrophages mainly as a result of activation of redox-sensitive pathways leading to pro-inflammatory effects. Mast cell showed steeper GSH-depletion, compared to macrophages, prior to the onset of cytotoxicity, indicating increased ROS levels, resulting in potentially increased oxidative stress. A dose-response inhibition in the secretion of histamine was shown, suggesting that mast cell function could be impaired by cadmium. In this way, cadmium may modulate the function of the innate immune system, in such a way, that favours to a type 1 response by enhancing macrophages responses and at the same time affecting the functioning of mast cells.

MO182

Changes in protein expression of primary sea turtle cells exposed to contaminants indicate the potential for in vitro proteomics as a high throughput tool to support biomarker discovery.

S.J. Chaousis, Griffith University - Smart Water Research Centre / Australian Rivers Institute; F.D. Leusch, Griffith University / Australian Rivers Institute; A. Nouwens, The University of Queensland / School of Chemistry and Molecular Biology; J. van de Merwe, Griffith University / Australian Rivers Institute

The development of biomarkers of chemical exposure and effect in threatened wildlife is challenging because traditional methods for biomarker discovery that involve *in vivo* testing or destructive sampling cannot be utilised. These challenges have long since been acknowledged and the development of non-destructive methods for the detection of biomarkers in wildlife has been examined for many decades. However, despite these efforts, progress in this field has been slow and the development or confirmation of non-destructive biomarkers of exposure in threatened wildlife has been limited. The usefulness of cell lines along with non-targeted omics has not been well explored in this context despite the potential for these methods to greatly enhance non-destructive biomarker development. In order to assess the potential of these methods for the development of biomarkers of exposure and effect, we optimised exposure and extraction methods and aimed to initially examine the effect of time and dose on global protein expression. Primary green sea turtle (*Chelonia mydas*) skin cells were exposed to two contaminants known to accumulate in sea turtles - a polychlorinated biphenyl (PCB153) and perfluorononanoic acid (PFNA). The exposure was performed over 24 or 48 hours to three environmentally relevant concentrations (1 µg/L, 0.1 µg/L, and 0.01 µg/L). Global protein expression was then measured using quantitative LC/MS resulting in over 1000 unique protein identifications. Our results show that a large number of proteins, over 700, were significantly differentially expressed by cells under exposure conditions and that time and concentration had significant effects on overall differential expression as well as on the expression of individual proteins. Most significantly, a biomarker of PCB exposure that has previously been identified in sea turtles (superoxide dismutase) was expressed by cells exposed to PCB indicating that these methods can potentially reflect biomarkers measured in whole organisms. Overall, the results from this study provide insight into the effects that time, dose and treatment have on global protein expression of green sea turtles cells, as well as preliminary evidence for the usefulness of non-targeted proteomics for biomarker discovery. This indicates the potential of the methods described here to support higher throughput confirmation of biomarkers of exposure and effect while reducing the need for invasive sampling on threatened wildlife.

MO183

Baseline vs. Reactive Toxicity toward the Nematode C. elegans as Alternative Bioassay

M. Amposah-Offeh, University of Duisburg-Essen; S. Saleem, E. Büttner, A. Bier, A. Paschke, UFZ   Helmholtz Centre for Environmental Research / Department of Ecological Chemistry; G. Schuurmann, Helmholtz centre for environmental research - UFZ / Department of Ecological Chemistry

The nematode *Caenorhabditis elegans* is the first multicellular organism with a completely sequenced genome. Since more than 20 years, it has been employed as bioassay for assessing the environmental toxicity associated with sediments.[1] More recent work indicates that this worm may have a toxicity-relevant metabolic capacity.[2] This finding makes *C. elegans* attractive as alternative bioassay for sensing the toxicological potency of compounds that become activated through biotransformation. A prominent example are organic pro-electrophiles that may be biotransformed to reactive toxicants and then covalently attack nucleophilic targets of proteins and the DNA. Despite a large amount of studies with *C. elegans* in human and environmental toxicology,[3] characterization of the narcosis-level toxicity toward *C. elegans* was still lacking. The latter, however, would be useful for identifying reactive toxicants through their toxicity enhancement (T_e) over baseline narcosis.[4-6] In the present communication, organic narcotics have been employed to calibrate a respective regression line with log K_{ow} (octanol/water partition coefficient). Additional tests with electrophiles demonstrate the capability of *C. elegans* to sense reactive toxicity, which is a prerequisite for identifying the toxicological potency of pro-electrophiles. Financial support from the HEC-DAAD scholarship no. 91649208 for Sumaira Saleem is gratefully acknowledged. [1] Traunspurger W et al. 1997. *Environ. Toxicol. Chem.* 16: 245-250. [2] Leung MWK et al. 2013. *The Worm Breeder's Gazette* 19: 28-29. [3] Tejeda-Benitez L & Olivero-Verbel J 2016. *Rev. Environ. Contamin. Toxicol.* 237: 1-35. [4] Blaschke U et al. 2012. *Chem. Res. Toxicol.* 25: 170-180. [5] Schramm F et al. 2011. *Environ. Sci. Technol.* 45: 5812-5819. [6] Böhme A et al. 2016. *Chem. Res. Toxicol.* 29: 952-962.

MO184

Oxidative Activation of Pro-Electrophiles Mediated by an Fe-loaded Zeolite - A Nonanimal Tool for Mimicking Phase I Metabolism

J. Moldrickx, Helmholtz centre for environmental research - UFZ / Ecological chemistry; A. Böhme, UFZ - Helmholtz Centre for Environmental Research / Ecological chemistry; A. Becker, Leipzig University; G. Schuurmann, Helmholtz centre for environmental research - UFZ / Department of Ecological Chemistry

The electrophilicity of a chemical substance can have a significant impact on its toxicological profile, as this property determines the chemicals` ability to form adducts with electron rich sites of proteins, lipid components, DNA and RNA.[1] In many cases, this type of adduct formation is the molecular initiating event (MIE) of a complex adverse outcome pathway. Kinetic chemoassays, which address this type of MIE, are used for the quantification of a test chemicals` reactivity towards typical biological targets, and have facilitated the development of *e.g.* models for the prediction of the aquatic excess toxicity or skin sensitization potential.[2,3,4] Apart from chemicals that possess electrophilic substructures in their initial form, there are compounds that can become electrophilic only after abiotic or biotic oxidation. The reactive toxicity of these chemicals, also described as pro-electrophiles, is often difficult to characterize with simplified model systems like chemoassays or *in vitro* bioassays because these methods typically do not sufficiently include an activation step. Our presentation introduces a new tool, based on an Fe-loaded zeolite, that is able to mediate the transformation of pro-electrophilic phenol and dihydroxybenzene derivatives into potent electrophiles. The reactive oxidation products were trapped by coincubation with the tripeptide WCG (tryptophan, cysteine, glycine) and analyzed using high performance liquid chromatography coupled to tandem mass spectrometry. Profiling of the obtained adduct patterns enable the identification of formed electrophiles, and provides new insights into the oxidation pathways causing the reactive toxicity of pro-electrophiles. The authors thank the EU-funded project OSIRIS (GOCE-CT-2007-037017) and the BMBF-funded project ProHapTox (FKZ 031A422A and 031A422B) for financial support. [1] Böhme A, Laqua A, Schüürmann G 2016. *Chem. Res. Toxicol.* 29: 952-962. [2] Mulliner D, Schüürmann G 2013. *Mol. Inf.* 32: 98-107. [3] Chipinda I, Ajibola RO, Morakinyo MK, Ruwona TB, Simoyi RH, Siegel PD 2010. *Chem. Res. Toxicol.* 23: 918-925. [4] Böhme A, Thaens D, Paschke A, Schüürmann G 2009. *Chem. Res. Tox.* 22: 742-750.

MO185

Integrated assessment of aquatic ecotoxicity for regulatory purposes

c. cappelli, IRCCS   Istituto di Ricerche Farmacologiche Mario Negri; C. Toma, IRCCS   Istituto di Ricerche Farmacologiche Mario Negri / Department of Environmental Health Sciences; A. Manganaro, Kode s.r.l.; D. Gadaleta, IRCCS Istituto di Ricerche Farmacologiche Mario Negri / Department of Environmental Health Sciences; J. Arning, German Environment Agency UBA; A. Biegel-Engler, German Environment Agency - UBA / Chemicals; E. Benfenati, IRCCS Istituto di Ricerche Farmacologiche Mario Negri / Department of Environmental Health Sciences

The REACH regulation requires the assessment of the CMR and PBT properties of the chemicals produced or imported in EU in amounts exceeding 10 t/a in order to protect the human health and the environment. The JANUS project aims at the development of a JAVA application to prioritize and assess the chemicals according to the PBT, CMR and endocrine disruption properties with uncertainties estimation. To assess the ecotoxicity part of the T property, we developed six continuous QSAR models for acute and chronic aquatic endpoints for the main trophic levels: EC_{50} 96h and NOEC 96h algae (*Raphidocelis subcapitata*), EC_{50} 48h and NOEC 21d *Daphnia magna*, LC_{50} 96h fish (*Oryzias latipes*) and NOEC fish (more fish species). We used gaselect and VSURF to select the DRAGON descriptors and the tree ensemble (random forest) method to derive the models, obtaining good performance (R^2 up to 0.96 on the training set and up to 0.78 on the validation set when the applicability domain is considered). We used the three new QSARs on chronic endpoints to perform the assessment of chemicals; we used the three new QSARs and the seven ones implemented in the VEGA platform (https://www.vegahub.eu/) on acute endpoints for screening purposes (two QSARs for *Daphnia magna*, two generic QSARs for fish, three QSARs for specific fish species). The ecotoxicity workflow is divided in three parts: algae, *Daphnia magna* and fish. For each part and each endpoint, the workflow integrates the experimental values (if any), the QSAR predictions and their reliabilities. The experimental values have a higher reliability than the predictions. The reliability takes into account the intra- and interspecific variability, the most sensitive species and the applicability domain index of the predictions. The users can choose a consensus or a worst-case approach. The experimental values and the predictions are compared to the regulatory thresholds to verify if the ecotoxicity criterion is fulfilled for each trophic level. The final assessment for ecotoxicity is based on the integration of the assessment of the trophic levels (the values and their reliabilities) and the number of trophic levels that fulfil the ecotoxicity criteria. The scheme will be applied to other categories of chemicals, such as the biocides within the LIFE COMBASE project. The authors thank the projects JANUS (contract Z 6 - 80 710/20 - 3716 65 414 0) by Umweltbundesamt (UBA) and LIFE COMBASE (LIFE15 ENV/ES/416) for the financial support.

MO186

An integrated testing strategy to fill data gaps for environmental risk assessment of iso-alcohols

G.E. Bragin, ExxonMobil Biomedical Sciences, Inc. / Toxicology and Environmental Sciences; B. Hedgpeth, ExxonMobil Biomedical Sciences, Inc.; C.A. Sutherland, ExxonMobil Biomedical Sciences, Inc. / Toxicology and Environmental Science; B. Kelley, D. Letinski, ExxonMobil Biomedical Sciences Inc; J. Butler, ExxonMobil Biomedical Sciences, Inc / Environmental Toxicology and Chemistry Laboratory; M. Lampi, ExxonMobil Biomedical Sciences, Inc.

Environmental risk assessments require quality data to provide defensible environmental quality benchmarks. Quantitative Structure Activity Relationship (QSAR) endpoint estimates are often appropriate for alcohols with a very strong correlation to aquatic toxicity test data. However, QSAR estimates require comprehensive justification to demonstrate applicability, and still may not fully meet regulatory requirements, leading to extensive long-term toxicity testing. Here, limited, strategic environmental testing was used to support QSAR predictions, thereby reducing animal testing while still meeting regulatory requirements.

Aquatic toxicity testing with algae, daphnids and fathead minnows (OECD 201, ISO 20665 and OECD 210) was performed with isooctanol and isoundecanol. The study objective was to employ a testing program consisting of long-term fish (limit test), invertebrate and algal toxicity tests to demonstrate that QSAR estimations accurately predict aquatic effects from long-term continuous exposure to these substances, further supporting the use of QSAR models across a range of iso-alcohols. The data demonstrate that the QSAR model employed accurately characterized the hazard of iso-alcohols and is protective of these endpoints. Moreover, this combined information, by demonstrating a regular and predictable pattern of toxicity amongst these substances, further justifies read-across between substances for other endpoints (such as bioaccumulation) and supports efficient use of data for general purpose risk assessments.

MO187

Looking for an alternative to glyphosate-based herbicides

V. Liousia, K. Eisner, S. Limbeck, D. Rünzler, University of Applied Sciences Technikum Wien / Department of Biochemical Engineering

Glyphosate-based herbicides are widely used in agriculture. When these products were originally introduced to the market they were considered to affect only target species i.e. plants. However, over the past decades there is growing evidence on the toxicity and genotoxicity of glyphosate on non-target species. On 27th November 2017 the EU member states agreed on a five-year renewal period for the use of glyphosate based herbicides. However, in case glyphosate-based herbicides become prohibited eventually, the availability of alternative active substances will become an urgent need. Nonanoic acid (a.k.a. pelargonic acid) is a biological derived substance considered as an environmental friendly herbicide. Its toxicity level to mammals is low and is also not expected to have adverse effects on non-target organisms. The aim of the present study was to compare the toxicity levels of glyphosate and a glyphosate based herbicide against pelargonic acid and a pelargonic acid-based herbicide on aquatic ecosystems using zebrafish as a model organism. In order to do so, we investigated the effect of both active substances and their formulations on the developmental stages of zebrafish embryos (OECD Guideline, Test No 236). The corresponding values of LC_{50} were calculated. The larvae that hatched from the acute toxicity tests were recorded in the observation chamber DanioVision and their swimming behavior was estimated in EthoVison software. The potential effect of the tested substances on the respiratory system of aquatic organisms was investigated *in vitro* by performing the Neutral Red Uptake assay on the trout-derived gill cell line RTgill-W1. Results of our *in vivo* and *in vitro* tests indicate that pelargonic acid and its formulation are more toxic (acute toxicity) than glyphosate and its based-herbicide. Furthermore, the behavioral assay indicates a potential for a neurotoxic effect of pelargonic acid on zebrafish larvae. To our knowledge, so far there are no available data for a neurotoxic induced effect of pelargonic acid on aquatic organism. Hence, this outcome has to be further investigated. *Financial support from the City of Vienna project Ökotoxikologie (MA 23 - Project 15-06) is gratefully acknowledged.*

MO188

Chemoassay Profiling of Salicylates to Assess Their Reactive Toxicity

A. Werner, Leipzig University; A. Böhme, UFZ - Helmholtz Centre for Environmental Research / Ecological chemistry; G. Schuurmann, Helmholtz centre for environmental research - UFZ / Department of Ecological Chemistry

Salicylates are widely used as fragrance additives or UV light absorbers in cosmetics and consumer care products, and thus can contribute to the human exposome. Moreover, they are released into the aquatic environment where they may act as constituents of the exposomes of waterborne flora and fauna. As organic electrophiles, salicylates are able bind to nucleophilic sites of proteins, peptides or the DNA, thus triggering the reactive molecular initiating events of aquatic excess toxicity or dermal sensitization. For assessing the toxicological hazard of organic electrophiles, chemoassays have turned out as promising nonanimal approaches and employ simple chemicals or model peptides as surrogates for the nucleophilic sites of biomolecules to profile the reaction behavior in terms of kinetic rate constants[1,2] and adduct patterns.[3] For this communication, the chemoassay reactivity of selected salicylates toward model peptides featuring the SH group of cysteine and the N terminus as nucleophilic target sites is analyzed in terms of adduct patterns by high performance liquid chromatography coupled with tandem mass spectrometry.[3] The discussion of the results includes a structure-reactivity analysis to provide new insights into the reaction behavior of salicylates. Augmented by in vitro bioassay analyses addressing the aquatic toxicity of salicylates, it is shown how their chemical reactivity, as one component of an electrophiles´ chemoavailability,[4] translates into aquatic excess toxicity, i.e. toxicity enhancement (T_e) over baseline narcosis. The authors thank the BMBF-funded project ProHapTox (FKZ 031A422A and 031A422B) for financial support. [1] Böhme A, Thaens D, Paschke A, Schüürmann G 2009. *Chem. Res. Tox.* 22: 742-750. [2] Thaens D, Heinzelmann D, Böhme A, Paschke A, Schüürmann G 2012. *Chem. Res. Toxicol.* 25: 2092–2102. [3] Slawik C, Rickmeyer C, Brehm M, Böhme A, Schüürmann G 2017. *Environ. Sci. Technol.* 51: 4018-4026. [4] Böhme A, Laqua A, Schüürmann G 2016. *Chem. Res. Toxicol.* 29: 952-962

MO189

Membrane-water partition coefficients to aid ionogenic surfactant risk assessment

S. Droge, University of Amsterdam/IBED Institute / IBED

Surfactants are important ingredients in various household products, personal care products and industrial processes. Many surfactants are technical mixtures of ionogenic head groups and linear or branched hydrophobic alkyl chain lengths ranging between C_{12}-C_{18}. As for many ionogenic compounds, the environmental fate assessment of ionogenic surfactants is complicated because it is not clear how to parameterize critical partition coefficients such as K_{ow}, K_{oc}, or how to model uptake in organismal tissue. For example, for one of the most common anionic soap ingredients, SDS, the entry for logP in the REACH registration dossier of ECHA provides a range from -2 (calculated, and recommended) to 1.6 (experimental, but considered erroneous), ranging more than a factor of 3000. Various techniques to derive logP for surfactants are described in these REACH dossiers, with conflicting argumentations on which is most relevant. It is well known that K_{ow} is a problematic parameter for surfactants, but this means that to reduce animal testing for bioconcentration factors and (baseline) toxicity of surfactants, insight in the key parameters of pure surfactant components driving uptake in biota is highly needed, alongside better understanding of elimination rate processes for such compounds. The BIONIC model could apply such key parameters for ionogenic surfactants. The phospholipid-water partition coefficient is considered to be the dominant contributor to the overall tissue-water partition coefficient for ionogenic surfactants, because membranes lipids allow for both ionic interactions at the zwitterionic head groups and hydrophobic interactions at the membrane core. Sorption experiments verify orders of magnitude higher affinities of ionogenic surfactants for membrane lipids than storage lipids. With data for 19 cationic surfactant structures recently published, we will now present membrane water partition coefficients (K_{mw}) for 15 anionic surfactants, using an optimized solid supported lipid membrane (SSLM) assay. For the anionic surfactant SDS, a $\log K_{mw}$ of 4.6 was determined, a factor of 1000 higher than the highest reported logP in the REACH dossier, and 5.000.000 times higher than the recommended logP. Our aim is that K_{mw} data from these SSLM assays will be sufficient to validate the accuracy of quantum-chemistry based molecular software calculations of K_{mw} for ionic structures using COSMOtherm, or to calibrate QSARs for K_{mw} of specific types of surfactants.

MO190

The Xenopus Embryonic Thyroid Signalling Assay (XETA) for assessment of effluents contamination in thyroid active molecules.

D. Du Pasquier, Watchfrog S.A.; S. Guerin, V. Rocher, SIAAP; J. Mougel, AQUIRIS; A. Tindall, G.F. Lemkine, Watchfrog S.A.

The Xenopus Embryonic Thyroid signalling Assay (XETA) was designed as a screening assay to provide information on the potential of a test substance or a sample to alter the normal functions of the thyroid system. The XETA provides a rapid (< 72h) way to measure the response of embryonic stage tadpoles to potential thyroid disrupting chemicals, allowing a efficient method for screening thyroid disruptors. XETA could provide an alternative to complex *in vivo* tests. It can be used for screening large number of molecules or testing environmental samples that couldn't be stored or sampled in large quantities. OECD is currently validating this *in vivo* assay, the final stage of validation has been completed in 2017 and it is expected that the XETA may be approved as an OECD Test Guideline by 2019. OCDE validation focus on using the XETA to test pure chemicals but this test could be particularly useful for the hazard assessment of effluents. During the 12 past years we applied this assay to effluents including municipal wastewater, treated wastewater, hospital wastewater, water from industrial processes. A part of our studies focussed on performances of wastewater treatment plant (WWTP). Assessing the quality of the WWTPs outlets for endocrine active molecules is a major challenge for reaching the good ecological status of the natural water bodies defines by the EU Water Framework directive. An evolution of the water treatment process is required to remove the endocrine effect presents in the wastewater. The evolution should include measurement of the endocrine effect to allow the control of the treatment performance. Only Bioassays have the capacity to integrate the effect of all compounds present into a global hormonal potential and are therefore promising tools for future development of in-line assessment. Our results using the XETA on WWTP effluents showed 1) Daily variations of the thyroid effect in wastewater linked to economic activities and rainfall 2) Most WWTP effluent still contains thyroid active molecules, results from chemical analysis performed on the same samples indicted a correlation between the total micropollutant load, and the thyroid effect 3) A minor part of the thyroid effect removal occurs during and decarbonification process. The major removal of the thyroid active molecule occurs during the nitrification step of the water treatment.

MO191

Advances on locomotion detection of Daphnia magna, Artemia franciscana and Paramecia caudatum

E.M. Salzer, V. Liousia, X. Monforte Vila, D. Rünzler, University of Applied Sciences Technikum Wien / Department of Biochemical Engineering

Animal behavior is complex and multidimensional. Over the past decades researchers tried to qualify and quantify it, in order to understand and predict it. The advances in this field are so radical that they actually formed a new scientific domain called "computational ethology". A major gap in this field is that most studies focus on terrestrial and aerial species in comparison to aquatic organisms. A reason for this discrepancy is that compared to terrestrial species, additional technical challenges need to be overcome when studying aquatic species e.g. light refraction and reflection interferences at the air/water boundary, positioning of the light source and suitable body marking techniques. However, a deeper understanding of the movement patterns of small-sized aquatic invertebrates and planktonic organisms is urgently needed, as locomotion and/or swimming behavior can be used as a stress sensitive indicator for a wide range of environmental contaminants. Furthermore, changes in their locomotion could be used as an endpoint when studying neurotoxic effects. As a result, the aim of the present study was to improve the current tracking techniques of *Daphnia magna, Artemia franciscana and Paramecium caudatum.* In order to do so,custom-made, polydimethylsiloxane/glass or polymethylmethacrylate plates were constructed. The tested organisms were placed in the costume-made plates and recorded under the microscope or in the observation chamber DanioVision. The horizontal and/or vertical tracking of the tested species were performed with the software EthoVision. The results of the present study showed that our custom-made plates had a higher tracking efficiency and a higher reproducibility score compared to the comercially available multi-well plates. Therefore, these easy to fabricate and cost efficient plates can be implemented on behavioral and ecotoxicological studies on small-sized aquatic invertebrates and planktonic organisms in any lab with an access to a tracking system. *Financial support from the City of Vienna project Ökotoxikologie (MA 23 - Project 15-06) is gratefully acknowledged.*

MO192

Validation of the in silico prediction tool for toxicity of Algae by pharmaceuticals in environment

A. Hirose, M. Kurimoto, National Institute of Health Sciences / Division of Risk Assessment; H. Shiraishi, National Institute for Environmental Studies; H. Yamamoto, National Institute for Environmental Studies / Center for Health and Environmental Risk Research; N. Tatarazako, Ehime University / Environmental Risk; T. Nishimura, Teikyo Heisei University / Faculty of Pharmaceutical Sciences; N. Kobayashi, Y. Ikarashi, T. Yamada, National Institute of Health Sciences

There are some concerns for environmental impacts of the pharmaceuticals due to the unintended environmental effects, which may be different from biological medicinal effects. Therefore, medical regulatory agencies require the assessment reports of environmental impacts by new drugs before marketing. It would be useful to predict the ecotoxicity of the new drug at the developmental stage, because the ecotoxicity studies are usually conducted at the final drug developmental stage just before submission. To validate the current *in silico* prediction tool, we evaluated the applicability for ecotoxicity prediction by the ECOSAR software, which is well known for ecotoxicity prediction of industrial chemicals. In the last year, we evaluated the prediction performance of Daphnia magna reproduction and of the fish chronic toxicity. In this study, we evaluated prediction performance of the acute and chronic toxicity for algae. We used the ecotoxicity test data sets of about 100 pharmaceuticals. The EC50 values for the acute toxicity and the ChV for the chronic toxicity were compared with the prediction values estimated by the ECOSAR. The percentages of the pharmaceuticals of which the predictive values are different in less than one digit from the actual measured values are 43% and 44% for acute and for chronic toxicity, respectively. Overall applicability of toxicity prediction for algae was similar to that of toxicity for Daphnia magna or fish. In the case of Daphnia magna or fish, some antibiotics, anti-cancer, central nervous system agents with lower LogPow were underestimated. However, there is little dependency of LogPow in the case of Algae toxicity. Most of chemicals with aliphatic amines had tendency to be underestimated. This difference may reflect on the difference of the mode of actions between Daphnia/fish and Algae. In order to improve the predictability of the *in silico* ecotoxicity QSAR tool, more researches on discovering the structure dependent toxicological profile of Algae would be needed. This work was supported by the Research on Regulatory Science of Pharmaceuticals and Medical Devices from Japan Agency for Medical Research and Development, AMED.

MO193

SeqAPASS to Evaluate Conservation of High-Throughput Screening Targets Across Non-Mammalian Species

C. LaLone, J.A. Doering, US EPA / Mid Continent Ecology Division; G.T. Ankley, U.S. EPA / National Health and Environmental Effects Research Laboratory; S.J. Degitz, U.S. Environmental Protection Agency / ORD, NHEERL, Mid-Continent Ecology Division; D.L. Villeneuve, U.S. EPA / National Health and Environmental Effects Research Laboratory

Cell-based high-throughput screening (HTS) and computational technologies are being applied as tools for toxicity testing in the 21st century. The U.S. Environmental Protection Agency (EPA) embraced these technologies and created the ToxCast Program in 2007, which has served as a screening and prioritization tool for thousands of chemicals. The rapid and automated screening methods take advantage of hundreds of (primarily) mammalian-based HTS assays for identifying biological activity suggestive of potential toxic effects. The data can aid in identifying chemicals that are most likely to impact biological pathways that lead to adverse health effects. To realize the full potential of the ToxCast data for predicting adverse effects to both humans and wildlife, it is necessary to understand how broadly these data may plausibly be extrapolated across species. Therefore, the U.S. EPA Sequence Alignment to Predict Across Species Susceptibility (SeqAPASS) tool was used to evaluate conservation of the 460 protein targets represented in the ToxCast assay suite. The SeqAPASS query sequence was selected based on the model organism used in the ToxCast assay (e.g., human, cattle, chimpanzee, guinea pig, rabbit, rat, mouse, pig, or sheep). Similarity of primary amino acid sequences and sequences from appropriate functional domains were compared across species to understand conservation of each assay target across taxa. To demonstrate application of the SeqAPASS data for extrapolation of ToxCast targets, case studies were developed that focus on the extrapolation of targets being evaluated as part of the Endocrine Disruptor Screening Program, including the androgen receptor, enzymes involved in steroidogenesis, and proteins in thyroid axis function. These case studies demonstrate the utility of SeqAPASS for informing the extrapolation of HTS data and identification of model organisms likely to be suitable for follow-up or complementary in vivo toxicity tests. *The contents of this abstract neither constitute nor reflect official US EPA policy.*

MO194

In silico site-directed mutagenesis informs species-specific predictions of chemical susceptibility derived from the Sequence Alignment to Predict Across Species Susceptibility (SeqAPASS) tool

J.A. Doering, US EPA / Mid Continent Ecology Division; S. Lee, ORISE/USEPA; K. Kristiansen, UiT The Arctic University of Norway; L. Evenseth, The Arctic University of Norway; M.G. Barron, U.S. EPA / Gulf Ecology Division; I. Sylte, The Arctic University of Norway / Department of Medical Biology; C. LaLone, U.S. EPA / Mid Continent Ecology Division

The Sequence Alignment to Predict Across Species Susceptibility (SeqAPASS) tool was developed to address needs for rapid, cost effective methods of species extrapolation of chemical susceptibility. Specifically, the SeqAPASS tool compares the primary sequence (Level 1), functional domain sequence (Level 2), or individual amino acid residues at key positions (Level 3) of the protein target of a chemical in a known sensitive species to sequences of other species and calculates sequence similarity metrics to predict potential cross-species chemical susceptibility. Level 3 analyses offer the greatest resolution for extrapolation of chemical susceptibility across specific species, but uncertainties into the role of specific amino acid substitutions at key positions of proteins and whether they affect interaction with chemicals made manual interpretation of Level 3 analyses time consuming and potentially inconsistent. Therefore, this study used *in silico* site-directed mutagenesis coupled with docking simulations of computational models for acetylcholinesterase (AChE) and ecdysone receptor (EcR) to investigate how specific amino acid substitutions impact protein-chemical interaction. This study found that substitutions in identities of key amino acids cause no change in chemical interaction with a protein if residues share the same side chain functional properties and have comparable molecular dimensions, while differences in side chain functional properties or molecular dimensions can reduce protein-chemical interaction. These findings were considered in the development of automated Level 3 analyses and enabling automatically generated species-specific predictions of chemical susceptibility. These predictions were shown to agree with Level 1 and 2 predictions of AChE and EcR for more than 90 % of investigated species, but also identified dramatic species-specific differences in chemical susceptibility that align with results from standard toxicity tests. The consistency of automated predictions of susceptibility across Levels 1, 2 and 3 and agreement with results of standard toxicity tests provides a compelling line-of-evidence for use of SeqAPASS in deriving screening level species-specific chemical susceptibility predictions across broad taxonomic groups applicable to addressing challenges in species extrapolation for human and ecological hazard assessment. *The content of this presentation neither constitute nor necessarily reflect US EPA policy.*

MO195

Survival and Teratogenic Evaluation of 91 compounds with environmental impact.

S. Calzolari, ZeClinics

ZeClinics (www.zeclinics.com) is a biotech company interested in developing efficient and reliable zebrafish screening tests to predict compound toxicity (general and organ related). Our aim, as part of the NTP consortium, is to define a universal set of rules – incubation time, chorion/no chorion, analysis timing, type of end phenotypes, analysis procedure, etc. – that can be applied by all the zebrafish toxicology community (SOP like protocols) and, eventually, to become the base for applying towards regulatory approval for the standardized test. In this study, we have performed a Developmental Toxicity Test on the NTP 91 compound list. For each molecule, 20 zebrafish embryos per condition were exposed to five different concentrations (Log3 dose/response curve: 100µM, 33 µM, 10µM, 3.3 µM and 1 µM) for a single biological replicate. Experiments were performed in chorionated embryos from 3 hpf to 96 hpf. Endpoints were analysed at 24, 48 and 96 hpf. Quantified phenotypes include mortality rate and teratogenic endpoints such as body deformity, scoliosis, pigmentation, heart edema and motor behaviour. It is important to note that the majority of the provided compounds were already dissolved in DMSO at 10 mM. This fact limited the range of maximum concentrations tested, which might have impacted in a lower-than-expected correlation between zebrafish and human data. In fact, 49/91 compounds did not show any toxic phenotype at the maximum evaluated concentration. On the other hand, 39 displayed mortality and teratogenic phenotypes. Among them, the most toxic compounds were Saytex CP-2000, 4,4-hexafluoroisopropylidene diphenol, 3-lodo-2-propynyl n-butylcarbamate, diethylstilbestrol, hexachlorophene, methylmercury chloride, rotenone and tetraethylthiuram disulfide.

MO196

MPA - an alternative for the standard procedure of Ames Test

J. Rossetto Martins Zwarg, School of Technology, UNICAMP; D.A. Morales, State University of Campinas / Faculty of Technology; W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; G. Umbuzeiro, School of Technology, UNICAMP / LAEG

The Salmonella/microsome assay (Ames Test) is the most widely used mutagenicity test for evaluation of pure chemicals and environmental samples. There are several protocols available in the literature, including those that reduce the amount of sample needed for testing with liquid and agar media. There is miniaturized version using liquid media called Microplate Fluctuation Protocol (MPF) that has been extensively used specially in Europe. It is has similar sensitivity with the standard Ames as well as other protocols and good performance in interlaboratory studies. However, the MPF has some disadvantages such as being difficult to apply with strains with low and high spontaneous mutation frequencies. Another miniaturized version of the Ames test is the microsuspension assay, which is 13 to 20 times more sensitive than the standard protocol. It is performed 5X concentrated bacteria and less sample and S9 mixture but still uses conventional petri dishes (90 x 15 mm). It has been extensively used for environmental samples testing, including in Effect Directed Analysis (EDA). The objective of this study was to miniaturize of the microsuspension Salmonella/microsome assay using agar microplates under the concept of the 3R principle. The conventional plates were replaced by plates with 12 micro wells. For validation of this miniaturization, we selected 13 known more or less potent mutagenic compounds. Six were tested only without metabolic activation (S9) and the other 7 were tested only with S9 using three Salmonella tester strains that were selected based on their different spontaneous reversion frequencies (low, mean and high). The miniaturization procedure conditions were made as similar as possible to the Microsuspension protocol, using the same testing design, metabolic activation, and data interpretation. Each test was conducted in parallel. MPA and Microsuspension protocols showed 100% agreement, qualitatively and quantitatively. MPA is less laborious, uses less sample, materials, and reagents reducing overall costs. The amount of sample required for testing is at less 20 times less in comparison with the standard Ames assay. We conclude that MPA is a promising tool and could be used in substitution of the standard Ames procedure especially in situations where sample quantity is a limitation such as impurities, drugs in development and environmental monitoring studies.

MO197

SETAC Animal Alternatives Interest Group

A. Lillicrap, NIVA Norwegian Institute for Water Research / Ecotoxicology

Bioavailability and realistic risk assessment of organic chemicals (P)

MO198

The necessity of OASIS bead and polyethersulfone membrane extraction for the Polar Organic Chemical Integrative Samplers (POCIS) calibration: a case study for alkylphenol monitoring in produced water

L. Silvani, Norwegian Geotechnical Institute; C. Riccardi, INAIL; E. Eek, Norwegian Geotechnical Institute; M.P. Papini, Università La Sapienza / Chemistry; N. Morin, Environmental and Food Laboratory of Vendee / Chemistry; g. cornelissen, Norwegian Geotechnical Institute; A.M. Oen, Norwegian Geotechnical Inst. / Environmental Technology; s.e. hale, Norwegian Geotechnical Institute

Produced water (PW) is one of the largest discharges from the oil and gas industry and includes formation and injected water. It contains several toxic compounds such as polycyclic aromatic hydrocarbons (PAHs), alkylphenols (APs), heavy metals, etc. PW is usually treated on the platform and then directly discharged into the sea. Despite the low content of toxic compounds that remain in PW after treatment, the large volume of PW lead to high total amounts of toxic compounds discharged every year into the sea; thus an effective tool for monitoring the hydrophilic organic compounds (HpOCs) is necessary. Passive sampler devices (PSDs) are the most common tools for monitoring a wide range of organic contaminants in water. By this regard, several PSDs have been used to monitor hydrophobic organic compounds (HOCs) in PW including semipermeable membrane devices (SPMDs). However, SPMDs are not suitable for monitoring HpOCs in PW owing to the nature of these chemicals. It is therefore necessary to develop and standardize a passive sampler for HpOCs, such as APs. Polar organic chemical integrative sampler (POCIS) is a PSD that has been used to monitor a wide range of HpOCs. It allows the time weighted average (TWA) concentration to be measured and thus assess fluctuation in discharge concentrations. POCIS is composed of a sorbent (OASIS beads), two polyethersulfone (PES) membranes and two stainless steel rings. POCIS is calibrated by evaluating the sampling rate (R_s), which is correlated with the contaminant concentration in the water and in the sampler, and is usually assessed by extracting the OASIS beads alone. We evaluated the effect of the PES membranes on AP uptake and, for the first time, calculated the R_s following the extraction of both the sorbent and the PES membranes. This study demonstrated that there was a lag phase in uptake for APs, and that APs with log K_{ow}ow>5 were accumulated more efficiently in the PES membranes. The extraction of both the PES membranes and the OASIS beads is thus needed when working with the POCIS in order to capture low contaminant concentrations and allow the detection of the less hydrophilic APs. This can be very useful in environmental applications because it may justify the use of only one passive sampler to monitor a wider range of contaminants.

MO199

In situ passive sampling methods to measure freely dissolved concentration of PAHs in contaminated soil: comparison with ex situ measurements and evaluation over one year

N. Bartolome, Agroscope Reckenholz-Tänikon Research Station ART / Environmental Analytics; I. Hilber, Agroscope / Environmental Analytics; R. Schulin, ETH Zurich / Department of Environmental System Science; P. Mayer, Technical University of Denmark / Department of Environmental Engineering; T. Bucheli, Agroscope ART / Environmental Analytics

Bioavailability studies can be used to improve risk assessment and legislation relating to soil and sediments contaminated by hydrophobic organic contaminants (HOC). Over the past decade, researchers have successfully developed various passive sampling (PS) methods to assess the freely dissolved concentrations of HOC in soil pore water or suspensions (C_{free}). The C_{free} play a key role for environmental fate and toxic effects of these compounds. Field conditions such as temperature, ionic strength or soil water content may influence the distribution of HOCs, and are accounted by using PS methods *in situ*. While *in situ* PS methods are providing promising results to measure C_{free} in the pore water of sediments, there is still very little information on the suitability of these methods for their application to soils, particularly under unsaturated water conditions. Here, we present the results of *in situ* PS concentrations of polycyclic aromatic hydrocarbons (PAHs) in six different PAH contaminated field soils. Three of them were located in peat bogs and hence permanently water-saturated, while the other three were located in grassland and thus not saturated. Low density polyethylene (LDPE) was used as PS method. The samplers were deployed *in situ* covering a depth of 20 cm below the soil surface. Concentrations were assessed at all sites after six, nine and twelve months of exposure. For comparison, soil samples from the same locations were analyzed using a conventional *ex situ* soil suspension method. The main objectives of this study were (1) to compare the measurements obtained with the two sampling methods, (2) to assess the influence of soil water saturation of the measurements (unsaturated versus saturated), and (3) to determine the role of seasonal variation (temperature and precipitation variation) and exposure time on the results of the *in situ* PS method. To our knowledge, this is the first experiment where PS methods were tested to determine PAH concentrations in the pore water of soils under field conditions *in situ* and to study the impact of soil water saturation. This study will help to find out whether *in situ* PS methods in soils are a tool to be potentially included in risk assessment and legislation.

MO200

Bioaccumulation of native and spiked p,p´- DDE by Eisenia andrei in γ-sterilized and non-sterilized soils

L. Skulcova, Masaryk University / Research Centre for Toxic Compounds in the Environment (RECETOX); K. Brandstätter-Scherr, University of Natural Resources and Life Sciences / Institute for Environmental Biotechnology, Department for Agrobiotechnology (IFA-Tulln); J. Hofman, Masaryk University, RECETOX / Faculty of Science, RECETOX; L. Bielská, RECETOX, Faculty of Science, Masaryk University / Faculty of Science, RECETOX

The fate of organic chemicals and their metabolites in soils is often investigated in model matrices having undergone various pretreatment steps that may qualitatively or quantitatively disturb results. Presently, effects associated to γ-irradiation, spiking and dwelling of earthworms were studied in field-contaminated (sterilization after contamination) and freshly spiked (sterilization prior to contamination) soils for the case of 1,1-dichloro-2,2-bis(p-chlorophenyl) etylene (*p,p´*-DDE). Changes in its sorption and bioavailability were linked to the changes in soil organic matter (SOM) chemistry measured by Diffuse Reflectance Infrared Fourier Transform (DRIFT) spectroscopy. Qualitative interpretation of obtained DRIFT spectra revealed changes in SOM chemistry manifested in a reduction of relative intensities of aliphatic moieties (sterilization), in bands of hydroxyl, aromatic, and aliphatic moieties (spiking), and of reduction in bands of aromatic accompanied by an increase of aliphatic moieties (earthworms dwelling). Using DRIFT, changes induced by spiking and earthworms were noted to be more pronounced compared to sterilization. The variation of bioaccumulation factors (BAFs) of native and spiked *p,p´*-DDE in sterile and non-sterile soils was limited to a factor of 1.5., depending on the incubation time and the particular approach used for BAF calculation. Despite the absence of quantitative effects of γ-irradiation on *p,p´*-DDE bioaccumulation, the uptake kinetics were shown to vary between non-sterile and sterile soils. Sterilization appeared to increase uptake rates and reduce the influence of *p,p´*-DDE-soil contact time on bioaccumulation. These effects might be attributed to the effects of γ-irradiation on SOM chemistry alone or in combination with earthworms. Following our findings, γ-irradiation can be recommended as a relatively non-destructive method that is not expected to significantly affect risk assessment of bioaccumulative chemicals. However, in mechanistic studies the possible side-effects brought about by γ-irradiation should be taken into consideration.

MO201

Dissipation in soil and bioavailability to earthworm of two fungicides: comparison of laboratory and field experiments

S. Nélieu, G. Delarue, J. Amossé, S. Bart, INRAAgroParisTech; C. PELOSI, INRA (Institut National de la Recherche Agronomiqu

The pesticide fate in soil is currently studied through laboratory experiments, using homogenized soil and controlled incubation conditions. However, the representativeness of such experiments is questionable regarding to field conditions, i.e., soil heterogeneity, vegetation cover and climatic variations. Upon the few studies performed simultaneously under field and laboratory, the pesticides persistence and/or sorption on soil were evaluated as different; thus bioavailability could also be different. This study aimed at comparing the dissipation and the bioavailability of dimoxystrobin and epoxiconazole (two fungicides used in the commercial formulation of Swing Gold®) in a loamy soil and in earthworms under laboratory and field conditions. Field experiments were conducted in a meadow located in Versailles (France) on 100 m^2 plots. The field soil and earthworms were regularly sampled after the treatment, over one year (April to April). Laboratory experiments were performed by mixing the fungicide solution with surface soil issuing from the same field, and incubated in dark, at 15°C and constant humidity. In both cases, four replicates were performed using the recommended Swing Gold® dose, extra doses adapted to observe ecotoxicological effects and controls. The dimoxystrobin and epoxiconazole concentrations in soil were determined by an exhaustive extraction method and, to evaluate their availability, with a mild method engaging hydroxypropyl-β-cyclodextrin. At the same time, the bioavailability of the two fungicides was evaluated by determining their concentrations in exposed earthworms *Aporrectodea icterica* and *Aporrectodea caliginosa*. All analyses were performed by UHPLC-MS/MS. Under field conditions and five days after pesticide application, only 10% to 45% of pesticide residues were measured in topsoil, with high heterogeneity between replicates. After one month, the concentrations in soil increased, probably due to a plant-soil transfer. By contrast, applied dose was observed at initial time under laboratory conditions. For later dates and in both cases, dissipation was observed. The available fraction showed homogeneous rates under controlled conditions but highly heterogeneous ones in the field. However, the ratio of available/total concentrations showed the same trend of fate for the two tested substances. The bioavailability of the two fungicides was also different between field and laboratory conditions in terms of heterogeneity.

MO202

Experimental assessment of specific plant uptake factor of 1,2,4-triazole with different concentrations in wheat

R. Faraldo-Alonso, Innovative Environmental Services (IES) Ltd / Plant Metabolism; E. Esteban, Universidad Autónoma de Madrid / Agricultural Chemistry and Food Science; S. Höger, Innovative Environmental Services (IES) Ltd / Environmental Toxicology; D. Williams, Innovative Environmental Services IES Ltd; R. Piskorski, Innovative Environmental Services IES Ltd / Plant Metabolism

Environmentally significant risk assessment decisions in EU for Plant Protection Products (PPP) regulation are based on information obtained from Environmental Fate models. Such models (e.g., FOCUS PEARL and PELMO) attempt to quantify the potential for movement of pesticides and their metabolites to ground water and require data derived from experimental studies based on validated guidelines. Currently, guidelines detailing degradation and absorption processes of pesticides and their metabolites in soil do not accurately account for uptake by plants via root system, with plant uptake compartments only being given a default value. As plant uptake affects the environmental availability of these substances, accurately quantifying this contribution may result in more accurate prediction models. The plant uptake describes the process of translocation of dissolved compounds in the soil pore water to the plant via the transpiration stream and it can be described using the plant uptake factor (PUF) – uptake into shoots and roots – or the transpiration stream concentration factor (TSCF) – uptake into shoots. Recent work by the ECPA/IVA Working Group “Plant Uptake Factor” has produced a draft working protocol designed to experimentally determine the uptake of active substances as well as metabolites via plant roots. The purpose of the present study was to obtain reliable substance-specific plant uptake data – with different root zone exposure concentrations – using the study design proposed in draft working protocol. The set-up of the experiment was chosen to enable optimal growth of the test plants – wheat seedlings – grown in a hydroponic system under controlled environmental conditions. At BBCH 13 (3 leaves unfolded) ^{14}C-labelled 1,2,4-triazole was spiked into the hydroponic solution at different concentrations and the plant root system was exposed for 8 days. Mass balance – calculated from the sum of radioactivity found in the hydroponic solution, root wash plus roots and shoot tissue – and transpiration – calculated gravimetrically – were determined. The experimental data obtained were used to calculate uptake parameters – PUF and TSCF – according to the formulas mentioned in the literature.

MO203

LFER Models for Partition Coefficients of Environmental Concern

R. Kühne, Helmholtz centre for environmental research - UFZ / Department of Ecological Chemistry; S. van der Heijden, T.L. Sinnige, Utrecht University / Institute for Risk Assessment Sciences; S. Droge, University of Amsterdam/IBED Institute / IBED; J. Hermens, M. Jonker, Utrecht University / Institute for Risk Assessment Sciences; A. Paschke, UFZ Helmholtz Centre for Environmental Research / Department of Ecological Chemistry; G. Schuurmann, Helmholtz centre for environmental research - UFZ / Department of Ecological Chemistry

Traditionally, partition coefficients of environmental concern, including bioconcentration factors (BCF) and soil or sediment sorption coefficients, are predicted using approaches based on simple linear relationships with the octanol/water partition coefficient (K_{ow}). Recently, more sophisticated prediction models have been developed and applied, including LFER approaches. Such approaches allow distinguishing between separate sorption compartments. However, this requires data on distinct partitioning processes, which are rather scarce. For theoretical models without the need for additional experiments, the partition coefficients for these separate processes have to be estimated. While these coefficients basically could be roughly estimated from K_{ow}, the more sophisticated LFER equations, known as Abraham models, are preferred for such predictions. The present study presents new models of this type for the prediction of equilibrium partition coefficients of neutral organic compounds between water and immobilised artificial membranes (IAM), liposomes (membrane lipids), triolein (storage lipids), and sediment. The development of the models was based on new experimental data, as determined by the authors. Possible sources of the required Abraham parameters are examined, compared and discussed. Particular attention is given to the applicability domain of the models. Acknowledgment: This study was financially supported by the European Union 7th Framework Programme SOLUTIONS (FP7-ENV-2013) of the under grant agreement no. 603437.

MO204

Influence of grain size on the bioavailability and bioaccumulation of sediment-associated cypermethrin to benthic invertebrates

H. Li, J. You, Jinan University / School of Environment

Sediment particle-size distribution is an important factor influencing the bioavailability and toxicity of hydrophobic organic contaminants (HOCs) in sediment. Cypermethrin, a pyrethroid was used as an example in the current study to investigate the effect of particle size distribution on the desorption kinetics and bioaccumulation potential of sediment-associated HOCs. Bioaccumulation test with oligochaete *Lumbriculus variegatus* and two chemical techniques, namely Tenax extraction and matrix-solid phase microextraction (SPME) were applied in the current study. A field sediment was collected and wet sieved to obtain five particle-size fractions: i.e., < 20 μm, 20-63 μm, 63-180 μm, 180-500 μm and >500 μm, and the respective ratios of the five size fractions were 81.2%, 0.96%, 14.7%, 2.86% and 0.31%. The results of sediment characterization showed that the material, composition, surface area and adsorption capacity were significant different among sediments with different particle size, and adsorption capacity increased with decreasing particle size. In addition, the desorption rates of cypermethrin measured by Tenax extraction decreased with decreasing particle size, which supported the hypothesis that HOCs bound stronger to fine sediment particles than coarse sediment. The different desorption rates of cypermethrin in different particle-size sediments may influence the freely dissolved concentrations in sediment porewater, and subsequently bioaccumulation potential and toxicity.

MO205

Effect of suspended particle on polycyclic aromatic hydrocarbon (PAH) bioaccumulation by zebrafish (Danio rerio)

Y. Zhai, X. Xia, School of Environment, Beijing Normal University; X. Xiong, School of Environment, Beijing Normal University / School of Environment; L. Xia, x. guo, School of Environment, Beijing Normal University

Hydrophobic organic compounds (HOCs) tend to be associated with suspended particles in surface aquatic systems, however, the bioavailability of HOCs on suspended particles to fish is not well understood. In this study, a passive dosing device was used to control the freely dissolved concentration of polycyclic aromatic hydrocarbons (PAHs) including fluoranthene and pyrene, and the influence of particle-associated PAHs on their bioaccumulation by zebrafish was investigated. Results showed that the body burden of PAHs in the zebrafish including the digestive tract (without the head part) were higher than that in the zebrafish excluding digestive tract at the beginning of PAH bioaccumulation, and lower after two day bioaccumulation. The difference may be caused by the effect of PAHs associated with particles in the digestive tract. When PAHs on suspended particles were ingested and they were mainly stored in the digestive tract of zebrafish in the beginning of the bioaccumulation; because the bioaccumulation factors of PAHs in zebrafish were higher than the partition coefficients of PAHs in suspended particles, the concentrations of PAHs in zebrafish excluding digestive tract were higher than that in zebrafish including digestive tract in the later bioaccumulation process. Suspended particles promoted the uptake and elimination rate constants of PAHs to zebrafish body excluding head and digestive tracts. The uptake rate constants with 0.5 g/L suspended particle were approximately twice that without suspended particles, and the body burden in zebrafish increased by 16.4% - 109.3% for pyrene and 21.8% - 490.4% for fluoranthene during the first 8-d exposure. The findings from this study indicate that PAHs on suspended particles are partly bioavailable to zebrafish and particle ingestion is an important route in PAH bioaccumulation. Therefore, it is important to consider the bioavailability of HOCs on suspended particles to improve ecological risk assessment.

MO206

Methods for Deriving Site-Specific Relative Bioavailability Factors from Animal Bioavailability Data

B.H. Magee, ARCADIS; N.D. Forsberg, Arcadis U.S., Inc. / Environmental and Molecular Toxicology; A.K. Meyer, United States Army Corps of Engineers / Huntsville Center

The U.S. Department of Defense is responsible for the environmental restoration of properties that were formerly used for firearm training ranges. Remnants of spent skeet targets were determined to be a source of polycyclic aromatic hydrocarbons (PAHs) at these sites based on results from surface soil analyses and historical information demonstrating that skeet targets were commonly prepared using coal tar pitch as a binding agent. It was hypothesized that the nature of the coal tar pitch/limestone matrix of the skeet fragments reduces the oral bioavailability of PAHs compared to that seen in animal studies using pure benzo(a)pyrene in solvents added to diets. To test this hypothesis, soil samples were collected from the two sites to provide a range of PAH concentrations. Female B6C3F1 mice were fed diets amended with soil or soil extracts at a rate of 5% in the diet for fourteen days. For benzo(a)pyrene (BaP), the fraction of total dose excreted in the urine (FUE) was determined for the soil- and soil extract-amended treatment groups. The Relative Bioavailability Factor (RBAF) is the ratio of the FUE in animals treated with soil over the FUE in animals treated with solvent extracts of soil. Because each soil sample was tested in four cages of mice (two for soil and two for soil extract), there are different ways of computing the RBAF of each soil and the grand RBAF for the site. Pairwise RBAFs can be determined and averaged, but the more robust way to determine a site-wide RBAF from multiple sample points is to determine a linear regression of metabolite excretion rates versus daily dosing rates. The FUEs produced coefficients of determination (r2) that were greater than 0.83 and typically greater than 0.95, showing that the rate of BaP metabolite excretion was directly proportional to the daily dose rate of BaP. RBAFs were determined using Monte Carlo simulations to calculate the 95% upper confidence limit on the ratio of the soil and soil extract FUEs. The site-wide RBAF was equal to 14% for BaP. Pairwise RBAFs will be compared to the RBAF resulting from the regression approach, and the regulatory precedent for the regression approach will be presented.

MO207

Accurate determination of adsorption coefficients for low adsorbing compounds - from experiment to result evaluation

T.J. Richter, BASF SE, Agrarzentrum Limburgerhof / APD; T. Richter, BASF SE Agrarzentrum Limburgerhof / Global Product Safety and Registration; K. Platz, BASF SE Agrarzentrum Limburgerhof / Environmental Fate Modeling; A. Irmer, Eurofins Agroscience Services EcoChem GmbH; M. Traub, Eurofins Agroscience Services EcoChem GmbH / Environmental Fate

The determination of accurate adsorption parameters is a critical key parameter for the assessment of the leaching properties of low adsorbing compound's through the vertical soil profile. In case of expected low adsorption of a chemical compound, several experimental preconditions need to be considered to enable accurate adsorption parameter determinations: (1) An initial soil/solution ratio of 1/1 and (2) the liquid phase needs be removed as completely as possible from the soil phase upon completion of the equilibrium. An experimental approach was developed and optimized allowing the efficient separation of the soil and liquid phase by centrifugation through the soil itself and a filter/frit system. Determination of distribution coefficients is done based on the direct method, hence extraction and analysis of the soil phase as well. Apart from the optimized experimental approach the data evaluation is addressed. This includes the elimination of any apparent sources of experimental random errors e. g. by suitable outlier tests. Possible systematic errors have been addressed by the experimental design/data evaluation itself leading always to an underestimation of obtained adsorption parameters. The data evaluation includes the calculation of adsorption coefficients (e. g. Kf) and of p-values with p=Kf * (msoil/msolution); note: msoil/msolution after phase separation. If p>0.3, reliability of obtained Kf values is given according to "EFSA, 2017. Technical report on the outcome of the pesticides peer review meeting on the OECD 106 evaluators checklist". If p< 0.3, additional considerations are necessary, e. g. suitable statistical tests, in order to evaluate data quality and to demonstrate significance of the adsorption coefficients. Finally, fit quality as well as upper and lower 95 % confidence intervals of Kf and Kfoc from isotherms are derived. By reference to examples, data evaluation for cases with p values > and < 0.3 are presented indicating opportunities of that whole approach.

MO208

Evaluation of the swimming behavior and tactic response to atrazine of the Pseudomonas sp. strain ADP

L. Rolando, Instituto de Recursos Naturales y Agrobiología de Sevilla / Water Research Institute; A. Barra Caracciolo, National Research Council / Water Research Institute; J. Ortega-Calvo, Instituto de Recursos Naturales y Agrobiologia / Agroquimica y Conservacion del Suelo

Atrazine is an herbicide used to control grassy and broadleaf weeds in sugarcane, wheat, conifers, sorghum, nuts and corn crops. Although since 1992 in European Union Countries it has been banned owing to its toxicological effects, it remains one of the most consumed worldwide pesticide with annual consumption of about 70,000–90,000 tons. Atrazine removal from the environment depends on abiotic (photolysis and hydrolysis) and above all biotic degradation. The latter can be significantly affected by the herbicide bioavailability. The behavioral reactions of bacteria are rarely included in the biological assessment of contaminants ecotoxicity. For this reason, we investigated the swimming behaviour and tactic response of the motile atrazine-mineralizing bacterium *Pseudomonas* sp. strain ADP to different concentrations of the herbicide in a laboratory experiment. The tactic response was assessed by a chemical-in-capillarity method and an inverted capillarity assay for the repellent reaction in association with microscopic observations. The swimming behaviour was evaluated by a computer motion analysis software (CellTrack). We observed attraction responses at relatively high concentrations of the chemical, including at water-saturating concentrations. We also noticed that atrazine can elicit a negative tactic response at low concentrations. We also observed that the swimming patterns of *Pseudomonas* sp. strain ADP was influenced because of these low concentrations, by increasing twisting motility. The physiological relevance of the chemioattraction to pollutants lies in the fact that these compounds serve as carbon and energy sources. The overall results suggest that we can use the behavioral responses of motile bacteria as a useful method to estimate pollutant toxicity at ecologically relevant concentrations. The bacteria behavioral assay on pollutants can be an alternative or complementary method to the current ones, because it is a high sensitivity and visualization method.

MO209

The influence of biochar on the toxic effects of imidacloprid to the lifecycle parameters of Eisenia fetida

N.W. Nyoka, University of the Free State / Zoology and Entomology; P.M. Leeto, P. Voua Otomo, University of the Free State / Department of Zoology and Entomology

Imidacloprid is an organic active ingredient for various insecticides used to kill a number of biting and sucking insect pests. As a soil amendment, it can enhance sorption and reduce the bioavailability of organic toxicants. Earthworms are important soil promoters and provide various benefits to plants and they are too sensitive to various pollutants, thus they are relevant indicators of environmental change. The current study aimed to assess if biochar has the ability to decrease the effects of the toxicity of the agro-insecticide imidacloprid (IMID) to earthworms. Cocoons laid by earthworms that had never been in contact with any toxicant were exposed to biochar-amended and non-amended aqueous solutions of IMID for a period of 28 days. The data was non-parametric and it was analyzed using the Kruskal-Wallis ANOVA followed by Dunns' test. The level of significance was set to be $p < 0.05$. The results revealed that the effects of IMID on hatchling success were only significant at the highest concentration of 400 mg/L. These findings indicate that the recommended field concentration of IMID (400 mg/L) has the potential to prevent cocoons of earthworms such *E. fetida* from hatching. This suggests that IMID could lead to decrease population growth rate or complete population collapse in such invertebrates.

MO210

Chlordecone elimination kinetics in ewes

M. Saint-Hilaire, Université de Lorraine UL / URAFPA INRA; A. Fournier, Université de Lorraine UL; J. Thomé, Université de Liège ULg / LEAE-CART; C. Adam, University of Liege / LEAE-CART; J. Parinet, C. Inthavong, ANSES / Unité PBM; C. Feidt, Université de Lorraine UL / URAFPA INRA; J. Parinet, ANSES / Unité PBM

Chlordecone (CLD) is an organochlorine pesticide used from 1972 to 1993 against the black banana weevil (*Cosmopolites sordidus*) in the French West Indies and is persistent in the soils (concentrations are above 1 $mg.kg^{-1}$ of dry matter). Consequently, animals can be directly contaminated by involuntary soil ingestion. Previous studies showed a CLD absorption of 100% in goats and its metabolization in humans, gerbils and pigs CLD is reduced into chlordecol (CLDOH). Then CLD and CLDOH can be conjugated. No data are available about CLD metabolism and elimination in ewes, species usually consumed in the French West Indies. The objective of this study was to characterize the CLD elimination in ewes (linearity of the toxicokinetic, half-life in serum, metabolism, excretion forms and excretion routes). Three groups of 5 ewes received an intraveinous single dose of CLD (0.04, 0.2 or 1mg/kg body weight (BW)). Blood, urine and feces samples were taken at defined times up to 84 days after CLD administration. CLD analysis in serum (analysis for each dose) was performed at the CART (Belgium) and CLD and its metabolites were analyzed in urines and feces (for the 1 mg/kg BW dose) at ANSES (France). For 1 mg/kg BW, 0.2 mg/kg BW and 0.04 mg/kg BW the half-life was respectively of 28.5 ± 3.0 days, 24.0 ± 6.3 days and 27.7 ± 5.0 days. These three values were not significantly different ($P< 0.05$).Thus, It was possible to conclude that CLD toxicokinetic of CLD in ewe is linear. In urines, CLD and conjugated CLDOH were quantified. In feces, CLD and CLDOH were quantified. By comparing the two way of CLD excretion, feces appears to be the principal route of CLD elimination. Almost 60% of the administrated dose was found in feces and only 2% was found in urines. To conclude, the elimination of CLD in serum of ewe is non-dependant with the dose. In consequence, the different results obtained of CLD transfer in ruminant can be extrapolated for different real levels of exposure in the range of 0-1 mg/kg BW. This study reveals the CLD metabolism in ewes which was never shown before. The principal route of CLD elimination is via the feces. These results clearly indicate the CLD elimination kinetic in ruminants and will help to decontaminate exposed animals in the French West Indies.

MO211

Development and validation of QuEChERS extraction methods with or without enzymatic pretreatment to analyze chlordecone and its metabolites by HPLC-MS/MS in urine and feces of ewes

M. Saint-Hilaire, Université de Lorraine UL / URAFPA INRA; T. Bertin, C. Inthavong, G. Lavison-Bompard, T. Guerin, ANSES / Unité PBM; A. Fournier, Université de Lorraine UL; C. Feidt, G. Rychen, Université de Lorraine UL / URAFPA INRA; J. Parinet, ANSES / Unité PBM

Chlordecone (CLD) is an organochlorine pesticide used from 1972 to 1993in the French West Indies to fight against the banana black weevil.It is known to increase the riskof prostate cancer and affect development of the foetus and after birth. Nowadays, this pesticide is no longer used but, because of its high persistence, it still remains in soil. Consequently, farm animals can be contaminated by soil ingestion and this is key issue for French West Indies breeding. Thus, this work tempts to collect data about the CLD elimination in ewe so as to propose a decontamination strategy. CLD is mainly eliminated in feces and low amounts of CLD can be found in urines. CLD can be metabolized into chlordecol (CLDOH) in humans, pigs and gerbils livers. Then CLD and CLDOH can be conjugated by the glucuronyltransferase. In feces, CLDOH was found but no conjugated metabolites were present. In urines, no conjugated metabolites were found although the authors thought they would. Actually, no information about the CLD elimination in ewe is available.These findings results were based on a former extraction method developed in 1980 to analyze CLD and its metabolites in urines and feces. The extraction was performed by liquid-liquid extraction.Quantification of the conjugated metabolites was calculated by difference between a sample with and without enzymatic treatment. The analysis was then performed by gas chromatography hyphenated with mass spectrometry. In order to have an update and more sensitive method, a new development was carried out with this work. The extraction was based on the QuEChERS methodology which is more and more used in the pesticides field. As no conjugated standards were available an update enzymatic pretreatment was set up. The analysis method was performed by liquid chromatography with tandem mass spectrometry using isotopic dilution given a reliable method. The methods were then validated according to the French standard *NF V03-110* and the European Union guidelines. At the outcome of the method development, urines and feces samples of contaminated ewes were analyzed. According to the literature, CLD and CLDOH were present in ewe feces. In urines, CLD and conjugated CLDOH were quantified. These results highlighted a better sensitivity of the new method and allow proving the CLD metabolism in ruminants which was never made before.

MO212

Organic Contaminants in High Mountain Areas: Where and When to find them??

O. Machate, Helmholtz centre for environmental research - UFZ / Plant and Environmental Science; W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; D. Schmeller, Helmholtz Centre for Environmental Research UFZ / Conservation Biology; M. Krauss, Helmholtz centre for environmental research - UFZ / Effect-Directed Analysis

Semivolatile organic contaminants (SOC) are well known to undergo atmospheric long-range transport and enrich in remote high mountain ecosystems. To predict the risk for high mountain ecosystems it is necessary to have knowledge of the present concentrations of the SOCs and their bioavailability. It has been shown that the chemical concentrations of SOCs differ vastly in magnitude and spatial distribution within these areas. Mentioned as important drivers of these variations are the change in precipitation and temperature with increasing altitude and the locally prevailing wind patterns. Despite this knowledge, the task to interpret the results gained within studies on the spatial distribution of organic contaminants within high mountain areas remains difficult. To aid future researchers with their assessment this poster tries to condense the key information on fate and behavior of organic contaminants in high mountain areas. Therefore, fate determining variables will be named and the movement and bioavailability of organic contaminants throughout the seasons be described.

MO213

Pesticide occurrence in different apicultural matrices (honey bees, wax and pollen)

P. Calatayud-Vernich, M. Andrés Costa, Universitat de Valencia / Environmental and Food Safety Research Group; F. Calatalyud, E. Simó, Agrupación de Defensa Sanitaria Apícola (apiADS); Y. Pico, University of Valencia / Medicine Preventive

Sprayed crops with pesticides are visited by honey bees during pollen and nectar collecting process. Pesticides are transported inside the hive, where both, agrochemicals from agriculture and compounds used in-hive against varroosis by beekeepers are accumulated in wax, pollen and honey bees. Samples of honey bees (45), wax (43) and pollen (45) were obtained from 45 different apiaries located in Spain. The samples were extracted by a slightly modified QuEChERS procedure depending on the matrix, and then screened for 58 pesticides and its degradation products by liquid chromatography mass spectrometry (LC-MS/MS). The target analytes were chosen based on their potential toxicity to honey bees and their widespread use in plant protection or in the beehive agains varroa mite. Wax and pollen were the most contaminated matrices and exhibited a wide contamination by pyrethroids and organophosphates. Beeswax lipophilic nature and its lowest replacement rate in the hive are responsible of its highest pesticide content. Acaricides used in beekeeping such as coumaphos, chlorfenvinphos, amitraz and fluvalinate were the most frequently detected pesticides in wax. Some pesticides used in crops as organophosphate chlorpyrifos were detected in lower frequencies and concentrations. Pollen contamination pattern was similar to wax matrix. Acaricides applied in beekeeping were the most frequent and with the highest concentrations. Neonicotinoid acetamiprid and organophosphates chlorpyrifos and dimethoate were detected in pollen samples. Both insecticides are sprayed in crops and deposited on the pollen grains, which are transported to the hive during the foraging activity of the honey bees. Honey bee samples were less contaminated, although some acaricides and insecticides were found in this matrix. Given the concentrations detected in the matrices analyzed, honey bee colonies health could be compromised. Assessing pesticides content in these three different apicultural matrices at the same time is a useful tool to understand the magnitude of honey bee colonies exposure to toxic compounds, which is one of the main causes of the progressive decline in honey bee colonies around the world.

MO214

Adaptation requirements for the use of measured BCF for a realistic risk assessment of organic chemicals.

N. Pucheux, INERIS; S. ANDRES, INERIS / Toxicological Ecotoxicological Assessment of chemical Substances

One of the main factor in the secondary poisoning risk assessment is the bioavailability of potentially hazardous organic chemicals, especially in the case of soil contaminated with persistent organic pollutants. In the context of the TROPHé project, the transfer of PCBs and PCDD/Fs to plants and invertebrates has been studied: BCF in several plants and in earthworm had been measured and different models to calculate predator exposition have been used. One of the conclusions drawn is that there is no match between available guidelines to produce measured BCF in terrestrial organisms and the BCF needed with the REACH regulation guidance for ecological risk assessment. This guidance states that the exposure concentration for terrestrial predators can be calculated in taking in account the quantity of soil contained in the earthworms guts and the contaminant fraction bioaccumulated in its flesh. This fraction is calculated as the product of the contaminant concentration in interstitial water and the BCF. But this BCF, relatable to interstitial water, is not comparable with BCF measured with available guideline as OECD 317 – Bioaccumulation in Terrestrial Oligochaetes, relatable to total concentration in soil. Data obtained in the context of the TROPHé project allow for the comparison between PCB-PCDD/F $BCF_{earthworm}$ measured with the OECD 317 guideline and PCB-PCDD/F BCFearthworm extrapolated from the Kow of the substance. It was also possible to illustrate the impact of these differences on the results of the secondary poisoning exposure modeled concentrations. A screening on the ECHA registration site also provides an approximation of the number of registered substances that have a $BCF_{earthworm}$ measured with guideline relatable to total concentration in soil and therefore unusable as such in the recommended methodology according to REACH.

MO215

Assessing risks from PBT substances in surface waters: possible alternatives to biota monitoring?

E. Valantine, B. Bradford, Environment Agency / Environment & Business; M. Gardner, Atkins Ltd.; P.K. Curtis-Jackson, Environment Agency (England and Wales) / Chemical Assessment Unit; P. Whitehouse, Environment Agency / Evidence

The Water Framework Directive (WFD) requires waterbodies to be at ‘good chemical status’ by meeting Environmental Quality Standards (EQSs). Normally these EQSs are expressed as concentrations in water but in recent years standards expressed as critical concentrations in the flesh of aquatic biota (biota EQSs) have been developed for some chemicals that are persistent, bioaccumulative and toxic (PBT) with the aim of protecting predators and humans from chemical exposure via the foodchain. Biota standards are now set for 11 PBT substances or groups of substances, requiring Members States to set up monitoring regimes to assess the risks to surface waters. Biota monitoring (fish or invertebrates, depending on the substance of interest) is the most relevant sampling matrix but it is destructive, and suitable biota cannot always be found where sampling is required. As a result, the coverage offered by biota sampling programmes is much less extensive than sampling of water. This means that extrapolation to unsampled waterbodies is needed but this is highly uncertain, so national risk assessments are difficult to achieve. This study explores alternative matrices to biota sampling, focussing on sampling of (a) whole water and (b) the dissolved fraction estimated from passive sampling. We describe studies in which chemical analyses of whole water and passive samplers for a range of PBT substances are compared with water thresholds back-calculated from their respective biota standards. These risk assessments are compared with those made using biota samples taken from the same locations in UK surface waters. The utility of these matrices as possible alternatives to biota monitoring is examined, and their implications for future risk assessment is discussed.

MO216

Risk Associated with Alternative Cleaning Method for Carrot

P. Abara, Federal University of Technology Owerri / Department of Biological Sciences; L.A. Adjeroh, C.O. Ezea, Federal University of Technology Owerri / Biology; A.C. Udebuani, Federal University of Technology / Department of Biotechnology

ABSTRACT Risk Associated with Alternative Cleaning Method for Carrot **Introduction** Carrot is a nutritional root vegetable which is loaded with beta carotene, a precursor of vitamin A. It is necessary to wash carrots in order to remove soil and other foreign materials before eating either raw or in processed form (Moos *et al*., 2002). It is common practice nowadays to soak carrot in detergent solution before washing to achieve better cleaning. Some components of detergent are toxic (HERA, 2013; Chuku *et al*., 2015). The aim of this study is to evaluate the detergent residue accumulated in carrot exposed to detergent. **Methodology** The first stage involved distribution of questionnaires to determine the popularity of the use of this chemical substance in washing carrot before selling to consumers. The second stage involved soaking 2 kg of fresh carrots in five increasing concentrations of Detergents 1 and Detergent 2.The carrots were soaked for 20, 40 and 60 minutes, after which they were grounded and analyzed using the titrimetric method described by IPAN (2005). **Results a.** 64.29% of the respondents agreed to the use of detergent in soaking before washing, 25.14% do not use detergent in washing their carrots before selling to consumers while 10.57% were indifferent. Anionic Surfactant Residue in Exposed Carrots There was a concentration and time dependent increase ($P < 0.001$) in the percentage anionic surfactant in the exposed carrots Figure 1: Anionic Surfactant present as Residue in Carrot Washed with Detergent. **c. Percentage Cationic Surfactant Residue in Exposed Carrot** The percentage cationic surfactants residue increased with concentration and lenght of exposure. Figure 2: Percentage Cationic Surfactant Residue in Exposed Carrot. Figure 29: Quantity of Detergent Residue in Exposed Carrot. **Conclusion** The presence of residual amount of detergent in the exposed carrot raises a public health concern as this food item is daily consumed by unsuspecting public.

REFERENCES Chuku, E. C., Ogunka,-Nnoka, C. U. and Chuku, O. S. (2015). Effect of washing carrot with Omo detergent on the nutrient composition, shelf life, associated fungi and health hazards. PacesettersJournal of Scientific Research, 1(1): 1 - 5 Institute of Public Analyst of Nigeria IPAN (2005). Training Manual for 2005 pre admission workshop training. pp 287-288

Environmental risk assessment in time and space - new approaches to deal with ecological complexity (P)

MO218

Uncertainty concepts and misconceptions for landscape scale risk assessment

P. Thorbek, Syngenta / Environmetal Safety; M. Hamer, Syngenta / Environmental Safety; K.Z. Travis, Syngenta / Product Safety; A. Raybould, Syngenta

In Europe there appears to be two opposing views on the future of ecological risk assessment of pesticides. One aims to improve ecological realism and move towards landscape scale risk assessments whereas the other aims to simplify and focus on lower tier exposure and effect assessment. To some extent this dichotomy is based on differences in uncertainty perception. Thus, one camp focuses on what we here term “in-study uncertainty” and therefore holds that higher tiers are more uncertain than lower tiers. The other camp focuses on what we term “extrapolation uncertainty” and therefore holds that higher tiers are closer to real uses in real landscapes and therefore less uncertain than lower tiers. A purely statistical view of uncertainty often assumes different sources of uncertainty are independent and when this is combined with a focus on high percentiles, uncertainty measures quickly multiply up and inflates the perceived uncertainty. However, biological systems are controlled by a range of feedback and regulating mechanisms aimed at maintaining homeostasis and ecosystems normally have at least some redundancy; therefore uncertainties are typically not multiplicative. Further compounding the different views of uncertainty, is the natural variability in real landscapes. One view holds that the effects of pesticides should be isolated from the natural variability to describe the “true” effect and, since this is difficult, holds that landscape scale risk assessments increases uncertainty. The other view holds that the effect of pesticides should be related to the natural variability and hence landscape scale risk assessments reduce uncertainty. Here we describe different components of uncertainty, what role they play in landscape scale risk assessment and we propose a way forwards for making uncertainty analysis more useful for decision making.

MO219

Concept for a regional geospatial landscape analyses to predict site specific vegetation covers

A. Toschki, Research Institute gaiac; G. Lennartz, Research Institute gaiac / gaiac - Research Institute for Ecosystem Analysis and Assessment; T. Schad, Bayer Ag / Environmental Modelling; T. Preuss, Bayer Ag / Environmental Safety

The spatial pattern of plant communities in the agricultural landscape is depending on land management and the site specific environmental and soil conditions. In vegetation science the composition of plant species and their abundances in dependence on these factors are extensively described. This project follows the hypothesis that if the decisive environmental and soil parameters (soil type, soil texture, moisture etc.) and the determining management drivers (meadow or pasture, extensive or intensive etc.) of a specific site are known it should be possible to predict the vegetation cover and subsequently the composition of plant species on this site. As a starting point the main grassland types of North Rhine-Westphalia and Mecklenburg-Western Pomerania (Germany) were considered and data for vegetation communities, plant species and their frequency and abundance were imported in a PostGis database. Additionally geospatial data (shapes of grasslands, soil types etc.) were imported in this spatial database. As a second step a matrix of combinations of soil and environmental parameters was built and calibrated in `if-then´ steps with the main preferences of the different vegetation communities. The poster show first prediction results and discuss pro and cons of the concept as well as possible refinements in the future. The supply of data originated from these predictions could be helpful in many facets of risk assessment on a regional scale.

MO220

B-Rice: bird focal species identification in rice paddy

A. Caffi, ICPS International Centre for Pesticides and Health Risk Prevention / Public Health; F. Marchetto, ICPS / Public Health; F. Galimberti, A. Riva, ICPS International Centre for Pesticides and Health Risk Prevention / Public Health; L. Bani, V. Orioli, Università degli Studi Milano Bicocca / Dipartimento di Scienze dellAmbiente e della Terra; S. Ubbiali, ICPS International Centre for Pesticides and Health Risk Prevention / Public Health

Pesticide registration in EU (Reg. 1107/2009 EC) requires appropriate risk assessment for non-target organisms including birds. The European Food Safety Authority developed a Guidance Document (GD) to conduct the risk assessment considering a series of exposure scenarios from a combination of crops and growth stages, selecting relevant species at the lower steps of a tiered approach. The actual GD doesn’t include scenarios for pesticide applications on rice; nowadays bird risk assessment is generally performed considering rice as the other cereals. Rice paddy is characterized by two cultivation conditions: the dry one, comparable to bare soil scenario (as common cereal), for which groups of species are equivalent to those identified for the actual risk assessment and already reported in GD; the flooded one, typical of aquatic environments and wetlands, representative of a unique exposure scenario not yet considered in the employed GD. The aim of this work is to characterize areas of rice growing in Northern Italy, which are representative for humid scenarios (via GIS approach), identify and link the relevant focal species to them. A review of the grey literature will be performed in order to estimate presence, abundance, dominance and diet of species associated to North of Italy rice paddies. Indicator and generic focal species will be proposed for the lower tiers of a Specific Rice Pesticide Risk Assessment and suggested as potential model for the Southern European Zone.

MO221

A process-based population model for algae

L. Azevedo, BASF SE, Agrarzentrum Limburgerhof / Global Toxicology; M. Habekost, BASF Corporation; P. Janz, BASF SE Agrarzentrum Limburgerhof

EFSA’s guidance document for the risk assessment of edge-of-field aquatic organisms recommends a maximum of 8 weeks for the ecological recovery option (ERO) of aquatic organisms in a risk assessment for plant protection products (PPP). Here, we propose a process-based model for algal abundance to simulate effects and recovery of algal populations over time following exposure to PPPs. The model integrates the main processes driving algal cell growth, such as (1) toxicity of the PPP and (2) growth limitation due to suboptimal climate conditions, natural mortality, grazing and competition for resources, and density dependence. This model also makes a significant step towards full compliance with EFSA good modelling practices, whereby models for regulatory risk assessments should include validation, and sensitivity and uncertainty analyses. In this poster, the formal model as well as sensitivity and uncertainty analyses are depicted. We also employ empirical data from mesocosm studies conducted for a selective herbicide for model validation.

MO222

Population dynamics of a soil arthropod simulated using an individual based population model and established fate model data

D. Nickisch, T. Wittwer, Rifcon GmbH

The prediction of concentrations of plant protection products in soil, surface and ground water using chemical fate modelling is establisehd since decades and applied in European environmental risk assessments (ERA). Many issues, concerns and limitations were discussed and adequately addressed by scientific communities, applicants and authorities. As a result, a variety of accepted tools are now available for exposure assessment. In contrast, population modelling approaches used in ERA for effect assessment in ecotoxicology are still at a relatively early establishment stage. In this respect, although effect modelling is recognised as a beneficial tool for adding ecological realism to ERAs, EFSA has so far not published any guidance document. Population modelling is at least mentioned in several EFSA (draft) guidances and opinions as a refinement option. However, so far, outcomes of submitted effect modelling approaches in ERAs are not often considered by authorities. Many of the available models deal with protection goals that adress field populations. Therefore, effect models are often stochastic and spatially explicit. This however makes these models more complex in comparison to the established deterministic exposure models and therefore considerable effort is needed for their verification, validation and comprehensive communication. Since effect modelling for ERA aims to predict effects on populations of the model organisms which arise from environmental exposure, we find it meaningful to use, in ecological models, the relevant data from the established fate models. This can make the modelling approaches more harmonised and probably would enhance their acceptability. We illustrated the usability of data on environmental conditions which agree with the established fate models and could as well demonstrate the implications of different environmental conditions on springtail populations. For this purpose, we used an individual based population model which represents the life-cycle of springtails in a tempreature dependent framework. Specifically, we calculated soil temperature series with the groundwater model Pearl. Further, we used weather data given for available FOCUS scenarios and different weather time series from JRC databases. Those series characterised the environment in the springtail model and drove the temperature-dependent model processes.

MO223

Dynamic modelling of fluxes of weathered polychlorinated biphenyls (PCBs) in soil: column experiments vs. modelling approaches in realistic environmental conditions

C. Vitale, University of Insubria; E. Terzaghi, University of Insubria (Como) / Department of Science and High Technology, Como; M. Morselli, A. Di Guardo, University of Insubria / Department of Science and High Technology

A column leaching experiment was performed to simulate realistic conditions that may be representative of different environmental scenarios and evaluate their influence on mobility and transport associated to dissolved organic carbon (DOC) and fine particles of aged polychlorinated biphenyls (PCBs) (PCB 28, 52, 101, 153, 138, 180, 209) in soil obtained from the Brescia-Caffaro contaminated site. The concentrations measured in leachates were compared to the results of simulations performed with a dynamic air-litter-soil model (SoilPlus model) to investigate the predictive ability and the improvements needed to obtain better predictions. The variables taken into account were: 1) contact time between soil and water, 2) DOC content and quality in leaching solutions and in soil pore water, 3) fine particle-mediated transport, 4) temperature (and its influence on endogenous DOC production), 5) soil saturation conditions. These conditions were evaluated collecting consecutive fractions of leached samples after variable pre-equilibration times (2, 5, 7, 48 days), using leaching solution with different DOC content (tap water vs. a solution prepared with commercial humic acid), at different temperature (25 ° C vs. 15° C) and in saturated vs.pseudo field capacity conditions. Results indicated that equilibration time determined differences in measured PCB concentrations up to a factor of 8, probably due to the lack of equilibration with the endogenous DOC or differences in DOC quality among fractions. The addition of exogenous DOC incremented mobility (up to a factor of 4) especially for brief contact time (non-equilibrium conditions) and within the Log Kow range 6-7.5. Samples leached at room temperature showed concentrations up to a factor of 9 higher in comparison to samples collected at lower temperature probably because of the different amount of endogenous DOC produced. Samples kept in pseudo field capacity conditions for seven days and then flushed resulted in about double the concentrations of the samples flushed in saturated conditions with a brief contact time, showing that drying-wetting cycles may determine concentration peaks. These trends were not caught by the model predictions as well as the relevance of the transport associated to fine particles, pushing for incorporation of this dynamic in models.

MO224

Assessing the trait-based ecological vulnerability of aquatic invertebrates for phenol

J. Park, Gwangju Institute of Science and Technology / School of Earth Science and Environmental Engineering; K. Kim, Gwangju Institute of Science and Technology; J. Kim, S. Kim, Gwangju Institute of Science and Technology / School of Earth Science and Environmental Engineering

Recently, there has been considerable concern about the chemical accidents as usage and manufacture of phenol have increased. Phenol is harmful to living organisms and its exposure can cause ecological and economical damages. Therefore, it is necessary to prepare for possible chemical accident of phenol. The Korean government designated phenol as the accident preparedness substance and required to assess and manage the ecological risk of phenol. This study aimed to find the ecological risk at the scenario that phenol exposed to freshwater bodies in Korea. In particular, the vulnerability of aquatic invertebrates was explored to consider not only individual organisms also their populations. Vulnerability integrated the exposure, sensitivity, and recovery of the ecosystem by considering various traits (e.g., body length, food preference, toxicological sensitivity, recovery strategy, etc.). The traits were reviewed by published data or open sources, and respective scores were assigned by using multi-criteria analysis which transformed the traits to numerical expression. The toxicological sensitivity was derived by indirect prediction based on traits because enough toxicity data was not possible. The results figured out the vulnerable invertebrates for phenol in Korean freshwater. In addition, the vulnerable species showed that the consideration of only sensitive species would not be great ecological risk assessment and management. This work was supported by Korea Environmental Industry & Technology Institute (KEITI) through "The Chemical Accident Prevention Technology Development Project", funded by Korea Ministry of Environment (MOE) (No. 2016001970001).

MO225

Assessing and managing food-web effects of Plant Protection Products

K. Swarowsky, German Federal Environment Agency (UBA) / Department IV plant protection products; H. Hötker, Nature And Biodiversity Conservation Union (NABU) Germany / Michael-Otto-Institute; R. Oppermann, Institute for Agro-ecology and Biodiversity (IFAB); C. Bruehl, University of Koblenz-Landau / Institute for Environmental Sciences; S. Matezki, German Environment Agency UBA; J. Wogram, German Environment Agency UBA / Department IV plant protection products

Assessing impacts on biodiversity needs to integrate indirect effects (trophic chain interactions, also referred to as food-web effects or effects on biodiversity). Plant protection law requires protecting biodiversity and data requirements for Plant Protection Product (PPP) active substances (Regulation EC 283/2013) also acknowledge this aspect by mentioning indirect effects to be considered in the assessment of the impacts on biodiversity. The relevance of indirect effects of PPPs has been well documented and recent scientific opinions of the European Food Safety Authority (EFSA) confirm the need for their inclusion in the environmental risk assessment of plant protection products. As a first step towards this direction, the currently renewed approval of glyphosate includes an obligation to the EU Member States to assess and manage the risk to diversity and abundance of non-target terrestrial arthropods and vertebrates via trophic interactions in the course of authorization procedures of glyphosate products. Therefore, we consider it necessary to develop an extension of the risk assessment to evaluate the indirect effects of specific PPPs in addition to the standard risk assessment and provide suggestions to risk managers on how to mitigate them. Due to the large variation in food web compositions and spatial and temporal implications, we do not consider it possible to achieve a representative and realistic estimate of indirect effects by means of mechanistic models. Instead, we suggest a simple empirical model to complement current risk assessment. However, implementing a risk assessment scheme for an additional subject of protection would not be feasible without offering solutions on how to manage the assessed risk. Otherwise, an adverse outcome of the assessment would inevitably lead to non-authorisations. To solve this conflict, we put forward an approach to manage risk by means of compensating food web effects. In practice, compensation is established by ecological compensation areas such as flowering margins, set-asides and beetle banks infield. To make most out of existing types of suitable measures and to enable a maximum of freedom of choice to farmers, we provide a points rationing scheme to categorise the individual measures with regard to their value for supporting in-field biodiversity (and thus to compensate for indirect effects of PPPs).

MO226

Compensating for ecological risks of pesticides

S. Matezki, K. Swarowsky, German Environment Agency UBA; J. Wogram, German Environment Agency UBA / Department IV plant protection products

Current environmental risk assessment (ERA) of pesticides overlooks a considerable part of existing risks and consequently fails to protect the environment from pesticide effects in toto. Examples of such blind spots are risks to field-dwelling species including wild pollinators, amphibians and farmland birds as well as indirect (food web alternations) or cocktail effects. Although scientifically well described and highly relevant for the achievement of the legally defined environmental protection goals, eliminating these blind spots in the risk regulation has failed so far. The ´indirect effect´-issue is an illustrative example for what we would actually regard as a crisis in environmental risks regulation of PPPs. What we recognize is that progress in ERA notably seems to be hampered for types of risk for which no effective risk mitigation measures are established, so that an assessment of such risks would inevitably lead to non-authorizations. To solve this conflict, we wish to put forward a radically new approach in risk management: Compensating adverse effects of pesticides where established methods of risk mitigation fail to prevent them. Once implemented into the iterative process of risk assessment, such new risk mitigation approaches would allow to manage actual risks more adequately than currently possible, thereby preventing an increase of non-authorizations. It has not escaped our notice that our proposal could also make excessive higher tier assessments dispensable, thereby helping to solve the problem of the increase of complexity in ERA.

Fish model species in human and environmental toxicology (P)

MO228

Historical control data of the optimized Zebrafish Embryo Developmental Toxicity Assay (ZEDTA)

D. van den Oetelaar, Charles River Laboratories Den Bosch / GIT; M.A. Tobor-Kaplon, Charles River Laboratories Den Bosch / GET; M. Beekhuijzen, H. Emmen, Charles River Laboratories Den Bosch / GIT; B. van de Waart, Charles River Laboratories Den Bosch / GET

The ZEDTA is a promising and innovative method with a potential to replace the screening of teratogenicity in animals (rats and rabbits) and is gaining acceptance among scientists and regulators. However, so far no harmonized and validated protocol exists for this test. Therefore, a protocol based on the OECD guideline No. 236, has been developed and optimized by Charles River Laboratories Den Bosch, the Netherlands. Multiple studies were performed using the optimized protocol, which allowed for collection of historical control data on the frequency of malformations, mortality and development of the embryos/larvae exposed to the control treatment. In our protocol, embryos in the blastula phase (2-4 hours post fertilization (hpf)) were selected and exposed to adjusted ISO medium. Twenty four embryos (one embryo per well in a 24 wells plate), were exposed at a temperature of 26°C for a period of 96h. Exposure medium was renewed after 48 hours. Development were assessed daily using the Extended General Morphology Score (GMS). Teratogenic endpoints were scored as 'present' or 'absent' after 96 hours of exposure. The Extended GMS grades the normal development of a zebrafish embryo up to 100 hpf. Assessed endpoints consisted of, but were not limited to: detachment of tail, somite formation, eye development, heartbeat and movement. The maximum score at 100 hpf was 18. Teratogenic endpoints scored during the assessment comprise of malformations of saccule/otoliths, head, hart, tail, yolk, pectoral fins and entire body. Data of twenty experiments were analysed. In total 400 embryos were exposed to control treatment (i.e. adjusted ISO medium). The average mortality rate in the control treatment was 2.5% which was considered acceptable. In only three experiments a maximum mortality of 10% was reached, which was still considered acceptable. Sixty percent of surviving larvae scored the maximum of 18 points for development, whereas 32% scored 17 points at the end of exposure (100 hpf). The most frequently observed findings were malformations of yolk (3.3%) tail (3.1%), heart (2.3%) and head (1.3%). These findings were observed in 6.4% of surviving larvae only. Analysis of the historical control data shows that the used optimized protocol produces an optimal development rate of exposed embryos and larvae, with minimal mortality and a minimal background malformation rate. This indicates a low level of confounding factors and high reliability of results produced with our protocol.

MO229

Optimization of the Zebrafish Embryo Developmental Toxicity Assay (ZEDTA)

D. van den Oetelaar, Charles River Laboratories Den Bosch / GIT; M.A. Tobor-Kaplon, Charles River Laboratories Den Bosch / GET; M. Beekhuijzen, H. Emmen, Charles River Laboratories Den Bosch / GIT; B. van de Waart, Charles River Laboratories Den Bosch / GET

The ZEDTA is a promising and innovative method with a potential to replace the screening of teratogenicity in animals (rats and rabbits) and is gaining acceptance among scientists and regulators. However, so far, no harmonized and validated protocol exists for the ZEDTA. The aim of this research was to optimize the protocol, i.e. examine which combination of exposure parameters is optimal for embryonic and larval development and is at the same time most cost-effective. An optimal condition should yield normal growth and development with minimal mortality and/or malformations. The OECD guideline No. 236 was used as base. In our protocol embryos in the blastula phase (2-4 hours post fertilization (hpf)) are exposed to adjusted ISO medium. The following factors and their combinations were investigated: temperature (26 vs. 28°C), exposure vessels (24 vs. 96 well plates), renewal periods (static (no renewal) vs. semi-static (24 or 48 h renewal)), and use of solvent (0.05% v/v DMSO vs. adjusted ISO medium). Development was scored daily, using the Extended General Morphology Score (GMS). This system grades the normal development of a zebrafish embryo up to 100 hpf. Assessed endpoints consist of, but are not limited to: detachment of tail, somite formation, eye development, heartbeat and movement. The maximum score at 100 hpf is 18. Teratogenic endpoints such as malformations of saccule/otoliths, head, hart, tail, yolk, pectoral fins and entire body were scored as 'present' or 'absent' after 96 hours of exposure. Mean developmental and teratogenic scores were calculated and used to select the most optimal condition for each factor. Our experiments showed that exposure in 24-well plates at a temperature of 26°C in combination with renewal of exposure medium after 48 hours of exposure produced the most optimal results with the lowest incidence of malformations. Daily renewal of medium provided similar results, but this was less cost-effective. Use of 0.5% v/v DMSO did not induce more malformations or mortality than exposure to adjusted ISO medium.

MO230

Reliability of ecotoxicological studies in fish

H. Wünnemann, Bavarian Environment Agency; H. Ferling, Bavarian Environmental Agency; G. Dembek, W. Schmidt, W. Koerner, Bavarian Environment Agency; J. Schwaiger, Bavarian Environment Agency / Aquatic Toxicology and Pathology

For substance evaluation in ecotoxicology valid bioassays are essential for deriving Environmental Quality Standard (EQS). The generally established biotests using the three trophic levels - algae, invertebrate and fish - according to OECD Guidelines provide in particular the baseline data for the derivation of the EQS. To obtain the most accurate EQS by use of a low assessment factor of 10 data from all three trophic levels including long-term results are required. Depending on the test substance growth inhibition of algae, immobilization of daphnia as well as deformation and death of fish embryos are not necessarily the most sensitive organisms and endpoints. Fish are in many cases the most suitable test organisms to demonstrate effects of e.g. pharmaceuticals with a specific mode of action in vertebrates. Therefore, prolonged toxicity tests with fish are of great importance. However, the study design has to be adapted to specific endpoints according to the pharmacodynamics of the tested drugs. Thus, in planning and implementing this type of study special care must be taken to ensure that the generated data can be used for derivation of EQSs. Even though several reporting and evaluation criteria for ecotoxicological studies have been published (e.g. Klimisch, Cred) up to date still numerous studies are available which do not fulfill these criteria and thus have to be excluded in the assessment process. Frequent failure sources include e.g. not statistically significant number und unknown origin of test fish, insufficient number of concentrations tested, missing chemical analysis of test compound concentration in the test water, calculation of toxicological endpoints on the basis of nominal and not real concentrations, or insufficient quality of endpoints. The aim of the presentation is to outline, from our point of view, optimal experimental conditions of prolonged fish tests which can be adapted as a model for other scientific studies, thereby increasing the significance of results and considering so far neglected aspects such as possible background contamination of commercially available fish feed frequently used in fish studies.

MO231

Assessment of the relationship between heavy metal bioaccumulation and biomarker responses in Japanese dace inhabit in heavy metal contaminated river

H. Takeuchi, Toyo University / Research Center for Life and Environmental Sciences; Y. Iwasaki, National Institute of Advanced Industrial Science and Technology AIST Japan / Research Institute of Science for Safety and Sustainability; D. Kitamura, Toyo University / Graduate School of Life Sciences; Y. Kato, Toyo University / Faculty of Life Sciences; Y. Shimizu, Toyo University / Graduate School of Life Sciences; H. Tatsuta, University of the Ryukyus / Faculty of Agriculture; S. Kashiwada, Toyo University / Graduate School of Life Sciences

The Watarase River, running in the northern Kanto region of Japan, had been severely polluted by heavy metals due to Ashio mining activities from late 1800s to early 1900s (*e.g.* 20 mg-Cu/L in river water in 1897). Although the heavy metal concentrations remarkably decreased since 1960s, the concentrations are still higher than those in a unpolluted river, the Omoi River. In previous study, our group investigated the heavy metal accumulation status (Cu, Zn, As, Pb, Cd and Fe) in organs of Japanese dace *Tribolodon hakonensis* captured from mid reach sites in the Watarase and Omoi River, and also analyzed those river water and sediment concentrations. Water and sediment concentrations in the Watarase River were generally higher than those in the Omoi River, interestingly, whereas Zn, As, Pb and Cd concentration in liver of Japanese dace in the Watarase River were lower than those in the Omoi River. Additionally, although there were no significant genetic differences between both riverine dace in microsatellite analysis, bile metallothinein (cysteine-rich protein involved in metal detoxification) concentration of dace in the Watarase River was significantly higher than that in the Omoi River. It suggests that the dace inhabit in the Watarase River may have been adapted to metal contamination by biological responses not depending on genetic characteristics. In the present study, for understanding biological response mechanism of dace to the metal contamination, we analyzed multiple biomarkers (such as erythrocyte δ-aminolevulinic acid dehydratase activity, blood protoporphyrin and hemoglobin concentration, and bile metallothinein concentration) in dace captured in the Watarase and Omoi River, as well as metal accumulation status of those fish. In this presentation, we will show detailed results, and discuss about the relationship between heavy metal bioaccumulation and biomarker responses in riverine fish inhabit in metal contaminated river.

MO232

Micronucleus test to evaluate effects of 4 metals on DNA damage of zebrafish Danio rerio

A. Sobrino-Figueroa, Universidad Autonoma Metropolitana Iztapalapa / Hidrobiology; A. Perez-Rojas, Universidad Autonoma Metropolitana Iztapalapa / Lab. Limnology and geology. Department of Hydrobiology

Danio rerio is a species of importance since it is used as a test organism for ecotoxicological studies at the International level. In our country the tests with this organism are limited since they are only used in medical research, for this reason the objective of this study was to determine the toxic and genotoxic effect of the metals Cd, Cr, Cu and Pb which are present in the freshwater systems of the valley of Mexico, on juveniles of *D. rerio*, to evaluate the use of these fish as biosensors in environmental monitoring studies. Static bioassays were carried out with a duration of 48 hours, with each metal. Five toxic concentrations were used in duplicate, plus a non-toxic control. The LC_{50} was determined and with the surviving organisms the evaluation of genetic damage was carried out, by means of the evaluation of frequency of micronuclei in blood cells (1000 cells). The results obtained showed that the toxicity of metals and their mixture, based on the LC_{50} calculated was: Cu> Pb> Mix > Cr > Cd. The Kruscal-Wallis test indicated that there are significant differences between the degree of genetic damage in exposed organisms to different metals and controls (0.04%). The metal with the highest genotoxic effect was lead (0.83%), followed by cadmium (0.65%). Copper showed the lowest genotoxicity (0.37%). The metal mixture had a micronucleous frequency of 1.23 %. The juveniles of *D. rerio* had deleterious effects in concentrations of metals lower than the LMP (Maximum Permissible Limits) that marks the NOM 001 Semarnat for water discharges to natural systems, so it is possible that they can be used as biosensors in the studies of environmental monitoring.

MO233

Endocrine disruption effects of bisphenol S and bisphenol SIP in adult zebrafish (Danio rerio)

K. Ji, j. Lee, Yongin University

As alternative compounds of bisphenol A (BPA), bisphenol S (BPS) and 4-hydroxyphenyl 4-isoprooxyphenylsulfone (BPSIP) are widely used in thermal paper products. These compounds have been detected in human urine samples; however limited information is available on their endocrine disrupting effects. Adult zebrafish pairs (*Danio rerio*) were exposed to environmentally relevant concentrations (0, 0.5, 5, and 50 μg/L) of BPS and BPSIP for 21 days, and the adverse effects on egg production, levels of sex steroid hormones, and transcription of genes related to hypothalamus-pituitary-gonad (HPG) axis were investigated. The estrogenic (increase in 17β-estradiol/testosterone [E2/T] ratio) and anti-androgenic (decrease in T) effects were commonly observed in zebrafish exposed to BPS and BPSIP, and males were more sensitive to the adverse effects than females. Although the effective concentration for endocrine disruption was greater than that of BPS, the actions of BPSIP on the steroidogenic pathway were similar to the effects of BPS exposure. The commonalities and differences in the toxicity of BPS and BPSIP can be explained by their chemical structure: the phenolic hydroxyl group is the key structural component responsible for the estrogenic and anti-androgenic activities of bisphenol analogues. The results of the present study showed that exposure to low level BPS and BPSIP could affect regulatory systems of HPG axis in zebrafish at environmentally relevant concentrations.

MO234

Oxidative Stress Induced by PAH Metabolism: Comparing Three Exposure Routes in Red Drum, Florida Pompano, and Southern Flounder to DWH surrogate oil

D. Wetzel, Mote Marine Laboratory / ELF; R. Medvecky, C. Miller, K. Main, T.A. Sherwood, Mote Marine Laboratory

The magnitude of the oil and dispersant released during the *Deepwater Horizon* blowout caused significant immediate, and often lethal, damage to exposed organisms. However, the sub-lethal impacts of the chronic spill on offshore and nearshore biota are still not fully characterized. To help understand one of the most significant responses in important Gulf fish species, four different exposure-route experiments, DWH surrogate oil contaminated feed, sediments, and seawater, were designed and carried out to examine biological responses of aquaculture reared red drum, Florida pompano, and southern flounder. Environmental pollutants, like polycyclic aromatic hydrocarbons (PAHs) found in crude oil, have the potential to unbalance the antioxidant system of marine organisms. Oxidative stress occurs when there is an imbalance between the production of reactive oxygen species (ROS) and the organism's ability to detoxify reactive intermediates, such as those generated by metabolism of PAHs by cytochrome P450 (CYP1). Depending on the severity of oxidative stress, this imbalance can lead to DNA damage in a variety of ways, such as oxidized bases, apurinic/apyrimidinic sites (AP sites), single or double strand breaks and DNA adducts. Exposure to PAHs can lead to increase DNA damage, such as those created by AP sites (purine loss) and the formation of DNA adducts, in which PAH metabolites intercalate into the DNA. Total PAH concentrations were analyzed in exposure matrices, as well as fish livers and whole bodies to determine specific dosages. Multiple assessments have been carried out to examine oxidative stress; including, total antioxidant power analysis, 2-Thiobarbituric Acid Reactive Substances analysis, GSH/GSSG ratio determination, AP site quantitation, and 8-OHdG quantitation. Evidence of oxidative stress will be discussed comparing multiple pathways of exposure, and resulting impacts in terms of biological and ecological implications.

MO235

Impact of PAH/oxy-PAH mixtures on heart development in zebrafish

V. Cunha, K. Dreij, Karolinska Institutet

Polycyclic aromatic hydrocarbons (PAHs) are ubiquitous environmental pollutants that have been widely studied. Oxygenated PAHs (oxy-PAHs) are also found in the environment and are emitted from the same primary sources as PAHs but also can be formed through secondary oxidation of PAHs. However, relatively little is known about their environmental fate and toxicity. The aim of this work was to determine the effects of binary PAH/oxy-PAH mixtures on cardiac development in zebrafish (*Danio rerio*) embryos (ZFEs). ZFEs (24 hpf) were exposed to a dose range of single PAH (benzo[a]pyrene, BP), oxy-PAH (the ketones 4*H*-cyclopenta[*def*]phenanthrene-4-one (4H-CPO), benzo[*a*]fluorenone (BFLO) and 6*H*-benzo[*cd*]pyren-6-one (6H-BPO)) or their binary mixture for 4 days. After exposure, ZFEs were observed for effects on heart development, heart rate and blood flow. Samples were also collected for gene expression analysis. The results showed abnormal cardiac development, such as formation of string hearts where exposure to 6H-BPO and BFLO in combination with BP was more potent than single exposures. The heart rate and blood flow was significantly decreased, in a dose-dependent manner, in ZFE exposed to 6H-BPO alone and in combination with BP. With the other oxy-PAHs, a decrease in the heart rate was observed, however in a non-monotonic response to the treatments. Blood flow was also decreased but only for BFLO and 4H-CPO in mixture with BP. Gene expression analysis showed significant up-regulation of genes involved in cardiac function (*kcnh6*) and development (*tbx5*), especially for ZFE exposed to the combination of oxy-PAHs with BP. Notably, the up-regulation of these two genes correlated with the formation of string heart. In summary, the binary mixtures were more potent then oxy-PAHs alone in inducing cardiotoxicity, except in the case of 6H-BPO which seems to be a very potent oxy-PAH. The oxy-PAHs and PAHs interact and thereby increase the toxic effect emphasizes the importance of monitoring the presence of oxy-PAHs in the environment.

MO236

Induction of developmental cardiotoxicity in rainbow trout (Oncorhynchus mykiss) following PAH mixture exposure - new insights using an integrated OMICS approach.

A.N. Eriksson, C. Rigaud, University of Jyvaskyla / Department of Biological and Environmental Science; A. Krasnov, NOFIMA; J. Lihavainen, University of Helsinki; A. Ronkka, S. Saraei, T. Suomi, A. Laiho, L. Elo, University of Turku and Åbo Akademi University; E. Vehniäinen, University of Jyvaskyla / Department of Biological and Environmental Science

Worldwide, environmental levels of polycyclic aromatic hydrocarbons (PAHs) have increased over the last century, mainly due to anthropogenic release from incomplete combustion of organic material and oil spills. These compounds are present as a complex mixture in the environment and are known to cause developmental abnormalities, cardiotoxicity, immunosuppression, tumors and altered genetic expression patterns *in vivo*. PAH toxicity has been studied for over 100 years and it is currently known that different PAHs have different modes of action (MoA). PAHs like retene and pyrene are aryl-hydrocarbon receptor agonists that up-regulate CYP1a expression and thereby induce their own metabolism, while other PAHs like fluoranthene directly inhibit CYP1a activity. \n We exposed newly hatched rainbow trout fry (*Oncorhynchus mykiss*) semi-statically to retene and fluoranthene either as a single compound or as a mixture treatment for 1, 3, 7 and 14 days. Body length and yolk (energy) consumption were measured and the hearts collected and pooled at the end of every exposure period. Using transcriptomics, proteomics and metabolomics, cardiotoxicity was investigated at molecular level. Our results shows that fry exposed to retene and the mixture treatments, in relation to control, became shorter and in retene's case, had used more yolk by day 14. Fluoranthene exposure did not affect growth or energy consumption by day 14. Microarray analysis showed that the different treatments caused very different alterations in the transcriptome, both in terms of the number of changed genetic expressions and when in time. The only gene (up-)regulated at all sampling times and treatments was *cyp1a*. In addition, *cept* and *c1q and tnf-like-domains* expression were found similarly changed across all treatments but not at all sampling occasions. Using over-representation analysis revealed several biological processes affected, such as blood vessels and heart development following mixture treatment. Proteomic analysis is underway but protein expressions are suspected to show a low transcript to protein correlation (based upon literature). Heart tissue metabolomic analysis revealed that across all treatments only two out of 33 metabolites were found similarly affected by day 14. Thus combining several methods, our study discovered several pathways affected by PAH exposure, together with phenotypical alterations, highlighting the unique MoA of different PAHs and as a mixture.

MO237

Assessment of the developmental cardiotoxicity of individual PAHs using integrated OMICS

C. Rigaud, A.N. Eriksson, University of Jyvaskyla / Department of Biological and Environmental Science; A. Krasnov, NOFIMA; M. Keinänen, University of Eastern Finland; A. Rokka, University of Turku; S. Saraei, T. Suomi, A. Laiho, University of Turku and Åbo Akademi University; L. Elo, University of Turku; J. Lihavainen, University of Helsinki; E. Vehniäinen, University of Jyvaskyla / Department of Biological and Environmental Science

Fish early life stages (ELS) are among the most sensitive organisms to developmental toxicity caused by polycyclic aromatic hydrocarbons (PAHs), which includes detoxification enzymes induction (CYP1A), hemorrhaging, cardiovascular defects, pericardial and yolk sac edemas, craniofacial deformities or growth attenuation. The cardiovascular tissue is one of the most sensitive to PAHs, and all the aforementioned symptoms are caused by the activation of the aryl hydrocarbon receptor (AhR). However, the mechanisms involved downstream of the AhR activation by PAHs are still unclear. Some weak AhR agonists such as phenanthrene can also produce cardiovascular defects (e.g. arrhythmia) via unknown AhR-independent mechanisms. In this study, we aimed to explore the mechanisms of toxicity of individual PAHs in the rainbow trout (*Oncorhynchus mykiss*) ELS by the use of an integrated OMICS approach, i.e. the combined use of transcriptomics, proteomics and metabolomics. The use of OMICS can lead to evidence of which pathways are altered by PAHs, and thus help choosing candidate genes or proteins involved in their mechanisms of toxicity. Newly hatched rainbow trout larvae were exposed to three different PAHs (retene, pyrene or phenanthrene) at sublethal doses. The heart of each larva was sampled after 1, 3, 7 or 14 days of semi-static exposure, and RNA, proteins and metabolites were extracted. Morphometric parameters such as larval length and yolk sac area were also monitored, but were barely affected by PAHs. Preliminary data from transcriptomics and metabolomics showed different signatures of gene expression alteration as well as different metabolite profiles between treatments, suggesting specific mechanisms of toxicity. Overall, all compounds induced more changes in cardiac gene expression during the very first days of development, with the exception of pyrene which was also very potent after 7 days. Preliminary enrichment analysis (over-representation analysis) revealed that differentially expressed genes were linkable with metabolism of xenobiotics by cytochrome P450, cation transport, muscular contraction or steroid hormone biosynthesis in the case of retene. Some of those processes were shared by pyrene. Phenanthrene appeared to alter collagen biosynthesis, as well as the glutamate release cycle, but only at one sampling point and with very few genes involved. Proteomic analyses are underway to further highlight the mechanisms of toxicity.

MO238

Developmental Toxicity of a Non-steroidal Anti-inflammatory Drug (Acetaminophen), in African Catfish (Clarias gariepinus) embryos.

L.I. Ezemonye, University Benin / Animal and Environmental Biology; N.O. Erhunmwunse, University of Benin, Benin City, Nigeria / Animal and Environmental Biology; I. Tongo, University of Benin / Laboratory of Ecotoxicology and Environmental Forensics, Faculty of Life Sciences, Department of Animal and Environmental Biology,University of Benin, Nigeria; P. Adebayo, University of Benin / Animal and Environmental Biology

The study investigated the developmental toxicity of Acetaminophen, a non-steroidal Anti-inflammatory Drug on the early life stage (0 to 96 hpf) of African Catfish (*Clarias gariepinus*). The 96 hrs fish embryo acute toxicity (FET) test was carried out according to the modified OECD 236 guidelines. Newly fertilized embryos were exposed to different concentrations (0, 0.5, 1, 5 and 10 μgL^{-1}) of the drug in triplicates and observations of embryo development were made at different developmental stages. Morphological, physiological and behavioural alterations were assessed. Exposure of the embryo to acetaminophen significantly altered the morphological and behavioural properties of the fish. The effects were observed to be dose and time-dependent, as more poisoning symptoms were recorded at higher dose. Exposed embryos were observed to have poorly formed somite's, coagulated embryos, non-detached tail, altered spontaneous movement and inhibited swimming performance. Observed physiological alterations include cardiac edema, sac yolk edema, pericardial edema, tail malformation and lordsis. The result demonstrated that acetaminophen has the potential to alter the development of the early life stage of the African catfish.

MO239

In vitro approach for the identification of early warning biomarkers, related to exposure to PBDEs, in human and marine systems: oxidative stress, toxicity and cell cycle modulation

C. Espinosa, S. Manuguerra, M. Morghese, UniPa / DiSTeM; A. Cuesta, M. Esteban, University of Murcia / Fish Innate Immune System Group, Department of Cell Biology and Histology,; A. Santulli, Consorzio Universitario della Provincia di Trapani, / Istituto di Biologia marina; F. Cibella, CNR / Istituto di Biomedicina e Immunologia Molecolare (IBIM); A. Cuttitta, M. Sprovieri, CNR / IAMC-CNR Capo Granitola, Mazara del Vallo, Trapani, Italy; C. Messina, UniPa / DiSTeM

The contaminated Sites of National Interest (SIN) in Italy, are characterized by environmental degradation, determined by the impact of industrial activities during the last decades. The primary objective of the project CISAS "International Centre of advanced study in environment, ecosystem and human health" is to understand the mechanisms underlying the interaction between conventional (heavy metals, POPs, radionuclides, etc.) and emerging contaminants (e.g., PBDE, antibiotics, pharmaceutics, antitumorals, etc.,) from the environment to ecosystem, through a multidisciplinary approach. The goal of our study was to investigate, *in vitro*, ecotoxicological and biochemical analyses, in order to identify biomarkers for early warning, in relation to exposure to selected contaminants. The first experiments were carried on PBDEs, a class of brominated compounds extensively used as flame retardants, that are ubiquitous, toxic and persistent in the environments and for which the molecular mechanism, responsible of cytotoxicity remain unclear. Human and fish cell lines were exposed to different doses of PBDEs until 72 hours. After these experiments, sub-lethal doses were chosen for long term treatments. Expression of genes related to cell cycle, stress, biotransformation, apoptosis and oxidative stress, were analyzed by enzymatic assay, spectrophotofluorimetry, immunoblotting and real time PCR. The preliminary results revealed that fish cell lines are more sensitive to the PBDE than human cells. A condition of oxidative stress, assessed by the presence of reactive oxygen species (ROS) and relative modulation of scavenger molecules/enzymes, seems to be the crucial event that influences the expression of some biochemical markers related to toxicity, inflammation, cell cycle control, angiogenesis, indicating the possible stimulation of pathways responsible of cancer promotion. **Acknowledgemen**ts: the project CISAS "Centro Internazionale di Studi Avanzati su Ambiente, ecosistema e Salute umana" (CUP B62F15001070005) is funded by CIPE- MIUR.

MO240

In silico estimate of affinity constants for perfluorinated compounds in rainbow trout (Oncorhynchu mykiss) proteins.

D. Degli Esposti, Irstea / UR RIVERLY Laboratoire Ecotoxicologie; A. Vidal, Irstea Lyon; R. Casadio, University of Bologna / Department FaBit; M.P. Babut, Irstea / Water

Perfluoroalkyl substances (PFASs) represent an important class of environmental contaminants which have been widely detected in humans and wildlife as well as in surface waters and aquatic sediments. PFCs have been shown to accumulate in aquatic species and some of them have displayed reproductive and development toxicity, hepatotoxicity and behavioral effects. Numerous studies in fish and mammalians have demonstrated higher PFC concentrations in liver and blood compared to other organs. Such a distribution could be explained by PFAS binding to specific proteins, in particular the L-FABP (Liver-Fatty Acid Binding Protein) and the serum albumin. However, the data concerning the binding affinities of PFASs to these specific proteins are rare, refer mainly to mammalian proteins and to date, constitute one of the limitations in modeling the toxicokinetics of PFCs in fish. Moreover, biochemical *in vitro* approaches are often not possible due to the lack of purified proteins for most common fish species. The use of *in silico* approaches such as protein structure modeling and molecular docking between the chemicals and the proteins of interest, may improve our ability to evaluate chemical-protein interaction and allow the extrapolation of biochemical parameters, such as binding constants. This kind of data would be helpful in building more refined toxicokinetics model in aquatic organisms. Here we present a simple two-step method based on protein modeling followed by molecular docking using free online tools. We inferred dissociation constants for 3 different perfluoroalkyl acids (Perfluorooctane Sulfonate, PFOS, perfluorohexane sulfonate, PHxS, and perfluorononanoate, PFNA) and L-FABP in rainbow trout and in human homologue protein. Comparison with experimental data on the human protein showed that this approach provides estimates that range in the same magnitude as those obtained by experimental approaches, such as ligand displacement assays.

MO241

Impact of metformin on zebrafish (Danio rerio) embryos

S. Mieck, University of Heidelberg / Aquatic Ecology and Toxicology; T. Braunbeck, University of Heidelberg / Centre for Organismal Studies

The biguanide metformin is an insulin-sensitising agent through its characteristics to increase peripheral glucose uptake and to decrease hepatic gluconeogenesis and insulin secretion. Through its antihyperglycemic effect, metformin is one of the most abundantly prescribed pharmaceutical treatment of diabetes mellitus type II. At the same time, metformin is also used as therapy agent for women with polycystic ovary syndrome (PCOS), a reproductive abnormality disease, and is being screened as a potential anti-cancer drug. Therefore, just in Germany metformin usage has almost tripled in the last 10 years to 1,100 tons (2010) and it is still increasing. As a consequence of the high consumption, the pharmaceutical is detectable at relatively high concentrations in both waste water treatment plant effluents and surface waters around the world, even though most of the substance gets removed during conventional active sludge treatment. Since most of its active form (up to 100%) gets excreted through urine and faeces, the poor metabolisation rates of metformin in humans add to this outcome. As a consequence, metformin poses a potential risk for aquatic organism and ecosystem within the water cycle. In order to determine potential adverse effects on aquatic organisms, zebrafish *(Danio rerio)* embryos were exposed to metformin hydrochloride ($C_4H_{11}N_5 \times HCl$) according to OECD test guideline 236 for up to 120 hours post-fertilisation and analysed histologically with respect to acute and sublethal effects. \n

MO242

Pyrogallol and its structurally related compounds on animal cytochrome c oxidase activity

Y. Kim, K. Kim, H. Jeon, H. Kim, Y. Choi, S. Lee, Kyungpook National University

Pyrogallol is a benzenetriol being a brownish solid, and is used for hair dyes after mixing with copper sulphate. A recent report on mutagenicity of acid pyrogallol-containing hair gels has demonstrated that there was no 2-fold increase in reverents relative to the controls. However, it still needs to be determined its safety to the living organisms, when it is introduced to the environment. In this study, we evaluated its inhibitory effect on cytochrome c oxidase (COX) activity, which is vital for energy production and is located in mitochondrial membranes. COX activities from zebrafishes (*Danio rerio*), Corydoras (*Corydoras aeneus*), earthworms (*Eisenia fetida*), and the lesser rice weevil (*Sitophilus oryzae*) were placed to be inhibited by pyrogallol and its related chemicals such as gallic acid, 1,2,4-benzenetriol, pyrocatechol, caffeic acid, quinic acid, and chlorogenic acid. For the inhibition of zebrafish COX, pyrogallol was the strongest chemical among the tested compounds with a complete inhibition rate at the concentration of 100 ppm. Gallic acid and 1,2,4-benzenetriol showed potent inhibition on the COX activity with the concentration of 100 ppm. At a 10-times diluted concentration, these three compounds showed moderate inhibition on the enzyme activities. These phenomenon were applied all of the tested animals. Pyrocatechol, caffeic acid, quinic acid, and chlorogenic acid did not show any inhibitory effect on the COX activity. Taken together, benzenetriols including pyrogallol may be caused unexpected inhibitory effects on the animal COX activity, referring fluctuation of the energy production, and the benzenetriol moiety is essential for the inhibition on the COX activity.

MO243

Exposure to environmental concentrations of Triclosan induces oxidative stress and genotoxicity on zebrafish (Danio rerio) embryos

C. Parenti, A. Ghilardi, M. Mandelli, University of Milan; C. Della Torre, State University of Milano / Biosciences; S. Magni, University of Milan / Department of Biosciences; L. Del Giacco, University of Milan; A. Binelli, University of Milan / Department of Biosciences

Triclosan (TCS, 5-chloro-2-(2,4-dichlorophenoxy) phenol) is the most common antibacterial agent used in personal care products, including soaps, body lotions, laundry detergents, toothpastes and deodorants. For its properties it is also added to several household items such as food packaging materials, toys and textiles. Since TCS is not completely removed by WasteWater Treatment Plants (WWTPs), it is becoming a potential worldwide pollutant and it is frequently detected in surface waters, with concentrations ranging from ng/L to µg/L. There is evidence that TCS is acutely and chronically toxic to aquatic organisms and it was already demonstrated that this chemical severely affects both zebrafish adult and embryos, causing embryotoxicity, hatching delay and biomarker alterations. Thus, the European Union (EU) has disapproved in 2016 the use of TCS in biocidal products, due to its unacceptable environmental risk. Meanwhile, consumer antiseptic wash products containing TCS can no longer be marketed in US. The aim of this study was to investigate the adverse effects of TCS at environmental concentrations on zebrafish embryos up to 120 hours post-fertilization (hpf). It is the first time that environmental levels of this contaminant were taken into account, instead of evaluating the effects of sub-lethal or lethal concentrations. The experimental plan consisted in the exposure to two different environmental concentrations of TCS (0.1 and 1 µg/L) for 5 days following fertilization, under semi-static conditions. A suite of biomarkers was applied to evaluate the potential mechanisms underlying the toxicity of TCS such as the generation of oxidative stress and DNA damage. The activity of antioxidant and detoxifying enzymes, namely catalase (CAT), superoxide dismutase (SOD), glutathione peroxidase (GP_x) and glutathione-*S*-transferase (GST), where measured, while the genetic damage was evaluated as the occurrence of the micronucleated cells (MN test). Results show a significant increase in all biomarkers measured, indicating that this chemical is dangerous for aquatic species also at environmental concentrations.

MO244

Comparative study of acute toxicity of a Microcystis aeruginosa bloom containing microcystin-LR on common carp Cyprinus carpio and Wistar rat

Z. Hadjer, R. Bordj, H. Nasri, Laboratory of Biodiversity and Ecosystem Pollution, University of El Tarf, Algeria.; N. Bouaicha, UNIVERSITE PARIS

Microcystins (MCs) are hepatotoxins produced by several groups of cyanobacteria in water bodies throughout the world. Their mechanism of toxicity consist of a potent inhibition of protein phosphatases 1 and 2 A, which causes disruption of the cytoskeleton and consequent cell death. They can also alter the antioxidant system and induce oxidative stress in various organs of many species. Microcystin-LR (MC-LR) is the most studied variant due to its high toxicity and frequent occurrence in surface waters. In this study, we used a *Microcystis aeruginosa* bloom extract containing mainly the variant MC-LR (>95%) to compare the acute toxicity of this extract on two models: the common carp *Cyprinus carpio*, an omnivorous fish living in permanence in fresh waters in the presence of cyanobacteria and their toxins and the Wistar rat used usually as toxicological model. Two doses, 20 and 100 µg equivalent MC-LR/kg body weight (BW), were administered by gavage to male and female of both models weighing approximately the same mass of 200 g. After 48 hours of exposure, the effects on the hepatopancreas/liver, kidneys, intestine, lungs and gills have been assessed by histological observations and analysis of oxidative stress biomarkers: lipid peroxidation (LPO), reduced glutathione (GSH) level, glutathione-s-transferase (GST) and glutathione peroxidase (GPx) activities. The results obtained showed that the two sublethal doses of MC-LR cause in both species remarkable histological abnormalities characterized by a hemorrhage and inflammatory infiltrate in all organs. The analysis of the oxidative stress biomarkers in all organs of both models have shown a very significant increase in the lipid peroxidation level and the activity of the GST with a significant decrease in the concentration of GSH and the GPx activity. In addition, the most affected organs in the rat are the lungs but for the carp are the kidneys. **Key words**: Microcystin-LR, acute toxicity, oxidative stress, histological study

MO245

Subchronic toxicity of a Microcystis aeruginosa bloom extract containing mainly the microcystin-LR congener on the common carp Cyprinus carpio

R. Bordj, Z. Hadjer, H. Nasri, Laboratory of Biodiversity and Ecosystem Pollution, University of El Tarf, Algeria.; N. Bouaicha, UNIVERSITE PARIS

The occurrence of harmful cyanobacterial blooms in surface waters is often associated by a production of variety of cyanotoxins that represent a hazard for human and animal health. Microcystins (MCs) are the most common cyanotoxins and may be expected wherever blooms of cyanobacteria occur in surface water with more than 100 variants have been characterized. Among these cyanotoxins, microcystin-LR (MC-LR) is the most studied congener due to its high toxicity and frequent occurrence in surface waters. The purpose of the present study was to investigate the effects of 12-week gavage of a *Microcystis aeruginosa* bloom containing mainly the congener MC-LR (>95%), in male and female of juveniles (200 g weight) common carp (*Cyprinus carpio*). The fishes were been randomly assigned to three groups. Group I, is the control group, received daily physiological serum (500 µL), groups II and III were daily exposed by gavage (5 days per week) to lyophilized *Microcystis aeruginosa* bloom dispersed in physiological serum (500 µL) containing 2 and 10 µg equivalent MC-LR/kg body weight (BW) for 12 weeks, respectively. The effects on the hepatopancreas, kidneys, intestine and gills have been assessed by histological observations and determination of some biomarkers of oxidative stress: lipid peroxidation, reduced glutathione (GSH) level, glutathione-s-transferase (GST) and glutathione peroxidase (GPx) activities. The histological study showed for the two doses the presence in the male and female carps: signs of hemorrhage and lymphocytic inflammatory infiltrates in the hepatopancreas, renal glomerular deformity with lymphocytic infiltrate in the kidneys, epithelial cell hyperplasia leading to fusion of intestinal villi and a hypertrophy in some cases, with malformations of the lamellae within the gills. The exposure of cyanobacterial bloom containing the two doses of MC-LR resulted in a significant increase of lipid peroxidation and GST activity in both male and female group. However, a significant decrease in both GPx activity and the GSH level have been observed. In addition, the results of the histological study and biomarkers of oxidative stress have shown that male fish are much more sensitive to the bloom of cyanobacteria containing microcystins than females. **Key words**: MC-LR, *Cyprinus carpio*, oxidative stress, histological study.

MO246

Diluted bitumen vs. conventional crude oil: effects of developmental exposure on first- and second-generation zebrafish

D. Lyons, University of Alberta; D. Philibert, K.B. Tierney, University of Alberta / Biological Sciences

The Canadian oil sands industry produces a heavy, viscous crude oil called bitumen. Due to its viscosity, bitumen must be diluted with natural gas condensates to create diluted bitumen or ‘dilbit’ to facilitate pipeline transport. The composition of dilbit differs greatly in chemical composition compared to conventional crude oils and the impact of dilbit exposure on aquatic organisms has not been well characterized, despite its widespread transport across North America. In this study, the effects of developmental exposures on breeding success and next generation embryos were compared between dilbit and two conventional crude oils (mixed sweet blend and medium sour composite). Zebrafish embryos were exposed to water accommodated fractions of these oils from 0-7 days post fertilization (dpf) and gene expression and DNA methylation were measured at 7dpf. Exposed embryos were then grown to adulthood in clean water. These fish were bred and their embryos were collected and reared in clean water (unexposed second-generation embryos). Breeding success of the first-generation developmentally exposed fish was determined by measuring the number of pairs that spawned, number of eggs spawned, fertilization rate, and survival of unexposed offspring. Gene expression and DNA methylation were also measured in 7dpf offspring. Developmental exposure in the first generation did not affect the survival of embryos and also did not affect breeding success when compared to control, but differed among exposure groups. Some target genes were differentially expressed in the unexposed second-generation embryos when compared to control, indicating a heritable change in basal gene expression. This change in gene expression could potentially be due to changes in DNA methylation caused by the developmental exposure in the first-generation. Understanding what changes in DNA methylation mean for fish survival will require further study. Overall, it appears that developmental exposures to dilbit and conventional crudes have varying effects on first- and second-generation zebrafish embryos. Though second-generation endpoints are often overlooked, they are important to consider when evaluating the overall risk of oil exposure.

MO247

Effect of skatole and its metabolites on piscine Phase I metabolism

V. Zlabek, University of South Bohemia in Ceske Budejovice / Faculty of Fisheries and Protection of Waters, LECHB; V. Burkina, University of South Bohemia in Ceske Budejovice / Faculty of Fisheries and Protection of Waters South Bohemian

Research Center of Aquaculture and Biodiversity of Hydrocenoses Vodnany Czech Republic; G. Zamaratskaia, Swedish University of Agricultural Sciences / Department of Food Science
Cytochrome P450 (CYP) enzymes are dominant players in metabolism of xenobiotics and a wide range of endogenous compounds. Skatole is well known mammalian metabolite, however rarely considered as environmental relevant pollutant. In fact, presence of skatole in aquatic environment is common and represent risk to aquatic organisms due to its biological effects. To the best of our knowledge, no studies attempted to investigate the effect of skatole and its major metabolites on piscine CYPs. The aim of this study was to identify weather skatole and its metabolites, 2-aminoacetophenone, indole-3-carbinol, 3-methyloxindole, and 3-hydroxy-3-methyloxindole, can interact with fish CYP isoforms. Enzyme activities for CYP1A and CYP2A in rainbow trout hepatic microsomes were measured in the presence or absence of skatole and its metabolites. Following concentrations of tested inhibitors were used: 0.5; 5 and 50 µM. Skatole and indole-3-carbinol showed no inhibition potency on either CYP1A or CYP2A. 2-Aminoacetophenone, 3-methyloxindole and 3-hydroxy-3-methyloxindole reduced CYP1A enzyme activity by approximately 25-35%, whereas CYP2A activity remained unaltered. Physiological consequences of such inhibition for fish ability to detoxify xenobiotics remain to be elucidated. *Keywords*: rainbow trout, cytochromes, EROD, COH *Acknowledgement* - The study was financially supported by the Ministry of Education, Youth and Sports of the Czech Republic, projects CENAKVA (No. CZ.1.05/2.1.00/01.0024), CENAKVA II (No. LO1205 under the NPU I program), by the Czech Science Foundation (No. 18-15802S) and Swedish University of Agricultural Sciences.

MO248

Linkage of gene expression patterns with in vivo endpoints: gaining deeper insights

A. Zenker, University of Appl. Sc. Northwestern Switzerland / Institute of Ecopreneurship; N. Munz, University of Applied Sciences and Arts Northwestern Switzerland
The herbicide glyphosate and the pharmaceutical lisinopril are among the most popular chemicals that have been detected in many streams at low concentrations. Nevertheless, the ecotoxicological properties, especially of lisinopril, are largely unknown. The objective of the study was to find early detection markers by comparing in vivo effects and genexpression patterns in the fish embryo acute toxicity (FET) test and early-life stage toxicity test (ELS). Per substance 480 individuals of zebrafish were exposed to five different concentrations (lisinopril: 0.03 – 0.5 mg/L; glyphosate: 0.006 – 0.1 mg/L) and analysed for different morphological endpoints such as spontaneous movements, heart rate, hatching success and malformations. Gene expression patterns were determined by transcriptome analyses using real-time polymerase chain reaction. It was concluded that lisinopril is potentially carcinogenic to the zebrafish by affecting the antioxidant defence system. In addition, lisinopril was associated with the formation of angioedema and induced cardiac toxicity in zebrafish by the downregulation of NPPB gene expression. Finally, it should be noted, that sudden death of the zebrafish were observed depending on the dose of lisinopril. A possible reason could be the reduced expression of the ACE2 enzyme. On the other hand, glyphosate slowed the heart rate and significantly increased the HAVCR1 (KIM-1) expression. The SOD1 gene expression was significantly increased because of glyphosate exposure whereas SOD2 and NPPB gene expression were not affected.

MO249

New insights on cross-species differences in the modulation of human and zebrafish nuclear receptors by single chemicals and environmental mixtures

N. Creusot, Eawag - Swiss federal Institute of Aquatic Science and Technology / Environmental Chemistry; C. Garoche, INERIS; A. Boulahtouf, INSERM / IRCM - U1194; F. Brion, INERIS / Ecotoxicology Unit; W. Bourguet, CBS CNRS UMR5048 - INSERM U1054; A. Escande, Université de Montpellier; M. Grimaldi, INSERM / IRCM - U1194; S. Ait-Aissa, INERIS / UMRI SEBIO ECOT; P. Balaguer, INSERM / IRCM - U1194
In the context of contamination of aquatic ecosystems by endocrine disrupting chemicals (EDCs), this work aims to provide new insights on cross-species differences in the modulation of nuclear receptors (NRs) and the aryl hydrocarbon receptor (AhR) by individual chemicals and environmental mixtures, in order to help further cross-species extrapolation in the frame of the environmental risks of EDCs. To this end, a panel of individual ligands and environmental mixtures from an urban waste water treatment plant (WWTP), were screened on a set of recently developed *in vitro* reporter cell lines based on both human (h) and zebrafish (zf) NRs and AhR. Our results revealed that for some of the receptors marked cross-species differences occurred (PXR, PPARγ, PR) while for other receptors the differences were lower (ER, AR, GR, MR) or almost absent (AhR, ERRγ). For instance, promegestone acts as a full agonist of the hPR but as partial agonist of the zfPR whereas the dihydroxy-4-pregnen-3-one -reference ligand of the zfPR- antagonizes the hPR. In the same way, none of the reference ligands of the hPXR (T0913117) modulates the zfPXR whereas the clotrimazole -reference ligand of zfPXR- modulates also the hPXR but with lower potency. Then the hAR was more sensitive to the agonist mifepristone and the antagonist OH-flutamide than the zfAR whereas the dexamethasone was a more potent agonist of the zfGR than the hGR. Also significant differences in selectivity were noted among h and zf ER subtypes. Finally, the *in vitro* profiling of an urban WWTP confirmed these cross-species differences in terms of level, type (agonist vs antagonist), distribution along the WWTP. For instance, h and zf estrogenic activity was differentially detected in the sludge and the suspended material. In the same way, strong zf anti-androgenic activity was detected in the effluent while no human one can be detected. Also, strong zf mineralocorticoidic activity was detected in both influent and effluent whereas only h anti-mineralocorticoid activity was detected. Alogether, our results showed that h and zf NRs are, for some of them, differentially modulated by individual chemicals and environmental mixtures. Also, interaction of EDCs towards NRs cannot always be extrapolated between these species highlighting the need to further document NRs modulation between human and fish and associated responses, to improve human health and environmental risk assessment of EDCs.

MO250

Combining acute toxicity, toxicokinetics and metabolomics approaches to assess the effects of triclosan in zebrafish embryos

D.E. Damalas, National and Kapodistrian University of Athens / Chemistry; M. Agalou, Biomedical Research Foundation Academy of Athens / Developemental Biology; D. Beis, Biomedical Research Foundation Academy of Athens / Developmental Biology; M. Lamoree, VU University, Department Environment & Health / Department Environment & Health; P. Leonards, VU University, Institute for Environmental Studies / Department of Environment and Health; N.S. Thomaidis, National and Kapodistrian University of Athens / Department of Chemistry
Triclosan (TCS) constitutes a common household product ingredient, given its antimicrobial activity, and has been widely used over the past decades. It enters the sewer system and can be transported to wastewater treatment plants (WWTP), seawaters and rivers, resulting in the contamination of the aquatic ecosystem. Consequently, it is urgent to evaluate its potentially toxic effects to aquatic organisms.The zebrafish has emerged as a powerful model organism to study various aspects of developmental and cell biology, while it provides an alternative model for toxicological studies. The objectives of this study were to assess to what extent TCS induce toxicity in zebrafish embryos. In addition, we evaluated the uptake and biotransformation of TCS by zebrafish and examined whether biotransformation data could be used complementary to the concentration of the parent TCS to interpret the induced toxicity. The final goal was to establish an wide-scope targeted metabolomics screening workflow to investigate the induced toxicity in a biochemical perspective and associate the observed toxicity/phenotype with changes in molecular level. Overall, the aim was to highlight a highthroughput testing strategy, incorporating data from different approaches, for a comprehensive toxicity assessment of environmental stressors in aquatic organisms. The zebrafish embryo toxicity assay was used to calculate the LC50 value of TCS as well as to perform the morphological phenotyping. In addition, a liver specific fluorescent transgenic line (Tg:LFABP:GFP) was used, to evaluate TCS liver toxicity potential. Concerning the toxicokinetics and the metabolomics experiment, 96 hpf zebrafish embryos were used. Samples were collected at 5 different time intervals, from 30 s up to 24 hours post exposure (hpe). Detection and identification of tentative TCS-bio-TPs was performed through in-house developed suspect and non-target screening workflows. Bio-TPs arising from both oxidative and conjugative metabolic reactions were identified. Regarding the metabolomics part of the study, a database of over 600 endogenous metabolites (carboxylic acids, amines, nucleotides etc.) was established, covering a broad range of primary metabolism. This approach is an alternative to the classic targeted methods, as it did not focus on a few metabolic pathways, for which we already know that are affected by the specific stimulant and enables to unravel the involvement of unexpected metabolic pathways.

MO251

Isoprostanes in fish mucus - a non-lethal biomarker for oxidative stress

P. Bulloch, University of Manitoba / Chemistry; S. Schur, D. Muthumuni, Z. Xia, University of Manitoba; W. Johnson, University of Manitoba / Chemistry; V. Palace, IISD-Experimental Lakes Area; G. Tomy, Department of Fisheries & Oceans / Department of Chemistry; O. Francisco, University of Manitoba
Prostaglandin F2-like derivatives have emerged as a reliable biomarker for oxidative stress in mammals and fish namely the F2-isoprostanes (F2-isoPs). F2-isoPs are the free-radical catalyzed products of non-enzymatic lipid-peroxidation of arachidonic acid, a fatty acid found in brain tissue and cell membranes. Fish mucus has been investigated in several studies as a potential biological matrix for the analysis of oxidative stress as it is minimally invasive. It is composed mainly of glycoproteins, but notably contains immunoglobulins, pheromones, lysozyme and proteolytic enzymes. Mucus is known to have important biological functions for fish, ranging from communication and reproduction to osmotic regulation. To date, no method for the isolation and quantification of F2-isoPs in fish mucus has been reported. The aims of this study was to develop an efficient method for the extraction of F2-isoPs from fish skin mucus and to optimize the resolution and quantification of F2-isoPs by high performance liquid chromatography tandem mass spectrometry. The method was based on acidification of mucus with HCl and extracting with ethyl acetate. The

extract was then centrifuged, filtered and reconstituted in methanol. Separations were performed on C18 (2.1mm x 50 mm, 3.5um particle size) using methanol (0.1% formic acid) and water as the mobile phase. Negative ion electrospray ionization and specific multiple reaction monitoring ion transitions were used to detect F2-isoPs in mucus. Mass labelled internal standards were used to monitor recovery of native compounds during sample work-up and also to quantify native F2-isoPs. Native isomers of the Class III and VI Fs-isoPs were measurable in Crappie (Pomoxis). This work demonstrates that mucus has the potential to be used a non-invasive, non-lethal matrix for F2-IsoPs analysis in fish.

MO252

Validation of in ovo embryo microinjections to simulate maternal transfer of selenomethionine in the fathead minnow (Pimephales promelas)

T. Lane, University of Saskatchewan; D. Green, K. Raes, University of Saskatchewan - Toxicology Centre / Toxicology; K. Bluhm, University of Saskatchewan / School of Environment and Sustainability; D.M. Janz, K. Liber, L.E. Doig, University of Saskatchewan / Toxicology Centre; M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre

Selenium (Se) is a naturally occurring trace element that is recognized as a contaminant of concern in Canadian aquatic ecosystems because of its high potential to bioaccumulate and persist even after its source has been removed. All animal classes are susceptible to the effects of Se due to the narrow range between dietary essentiality and toxicity; however, oviparous vertebrates, especially fish, are at particular risk because of maternal transfer of Se. The objective of this study was to develop and validate an embryo injection approach to model maternal transfer of selenomethionine (SeM), the primary form of Se in the diet. This model could then be applied to any egg-laying species of interest and could provide insight regarding differences in species sensitivity to Se toxicity during early life stage development. Initially, the maternal transfer of dietary SeM and its effect on the F1 generation were characterized in a short-lived fish species native to North American freshwater systems, the fathead minnow (*Pimephales promelas*). 20 breeding groups (3 females:2 males) were fed a SeM-spiked diet of either 0, 3, 9, or 27 mg Se/kg bloodworms dry weight (dw) and bred for 28 days. Embryo Se concentrations increased immediately upon onset of exposure and Se concentrations reached approximately a 1:1 ratio in food:embryo after 28-days on the diet. There was a significant difference in mean embryo Se concentrations from the control (1.18 mg/kg embryo dw) in the medium (8.75 mg/kg embryo dw) and high (29.58 mg/kg embryo dw) treatment groups. Embryos collected on days 26, 27 and 28 were reared to swim-up and assessed for morphological abnormalities. Preliminary assessment revealed an increasing, although not significant, trend in the frequency of deformities between the control and high treatment groups (p=0.057); however, a more robust analysis is on-going. Average Se embryo concentrations from this study will serve as the basis for subsequent embryo injection studies in fathead minnow. Developmental endpoints from both studies (e.g. mortality, frequency of deformities, types of deformities, severity of deformities) will be compared to determine if the embryo injection model is an appropriate proxy for studying the maternal transfer of SeM. The embryo injection model could also support mechanistic and omic-based research in long-lived species of concern, such as white sturgeon, or in recreationally fished species such as walleye, brook trout and northern pike.

MO253

Preliminary characterization of the rainbow trout intestine using omics based approaches.

L.M. Langan, Plymouth University / Biological and Marine Sciences; S. Owen, AstraZeneca / Safety Health Environment; A.N. Jha, Plymouth University / Biological Sciences

Intestinal function is central to the physiology, health and disease of numerous organisms. However, little is known about its gene or protein profile in trout, a widely studied and environmentally relevant model laboratory organism (*Onchorhynchus mykiss*). In this study, two omics based tools focused on characterizing RNA and protein expression were used to establish the ontology of each intestinal region viz. the pyloric, anterior, mid and posterior intestine. RNA-Seq was carried out on intestinal regions and mapped back to the rainbow trout genome (84 %). Following filtering for transcript abundance using TPM and a p-value cut off, 23,635 – 25,435 contigs were identified over the 4 regions and included enzymes involved in metabolism of chemicals such as the cytocrome P450 family (CYPs). Differential expression of genes between regions did not vary significantly between the pyloric, anterior or mid intestine (~6 genes), however this changed markedly between the pyloric and posterior region (~29) highlighting their differences. Proteomic characterization established over 3,899 proteins present in the intestine with annotated proteins varying from 3,100 to 3,899 dependent on intestinal region. Significant differences in proteins were observed between intestinal regions further confirming trends observed in the parallel transcriptomic study. These data represent the first thorough characterization of the rainbow trout intestine, and will allow the identification of enzymes present in this organ which may be responsible for xenobiotic metabolism.

MO254

Persistent organic pollutants alter the expression patterns of epigenetic factors in the Zebrafish Liver (ZF-L) Cell line.

M. Blanc, Örebro Universitet / MTM Research centre; N. Scherbak, Orebro University / School of Science and Technology, Life Science Centre; S. Keiter, Orebro University / MTM Research centre

Several studies demonstrated that exposure to persistent organic pollutants can induce epigenetic modifications in human and other vertebrates including fish. It is of particular interest since epigenetic changes were reported in pathologies with increasing incidence such as cancer. Besides, epigenetic disruption was suggested to be one mechanism responsible for multigenerational effects of chemical exposure. Epigenetic pathways in zebrafish are similar to mammalians; therefore, it was proposed as an alternative model for epigenetic research. The focus of the present study was set on the investigation of epigenetic effects in the Zebrafish Liver (ZF-L) cell line after 48 h of exposure to 8 selected compounds. The cells were exposed to the LC_{10} values of pesticides (methoxychlor (MXC), permethrin (PER)), plastic additives (bisphenol A (BPA) and S (BPS)), perfluorinated compounds (perfluorooctane sulfonic acid (PFOS), perfluorobutane sulfonic acid (PFBS)), a whitening agent, 7-diethylamino-4-methylcoumarin (DEMC); and to the water solubility value of the pesticide vinclozolin (VCZ). Expression of genes encoding enzymes and factors involved in DNA methylation and histone modifications was monitored using RT-qPCR. The DNA methyltransferases were selected to target DNA methylation (*dnmt1, dnmt3aa, dnmt3ab, dnmt3ba*). They were analyzed together with 2 histone deacetylases (*hdac1, hdac3*), one demethylase (*jarid1b*), and one chromatin remodeling factor (*spt6*). At the selected concentrations, all compounds induced changes in expression of at least one or several of the investigated genes. The most potent compound was BPS, which reduced the expression of all genes. Ranked by decreasing incidence, it was followed by PFBS>MXC>PER>BPA>DEMC>VCZ>PFOS. VCZ induced selective changes in genes involved in histone modifications. Interestingly, industrial alternatives BPS and PFBS induced greater expression changes of epigenetic factors than the well-known BPA and PFOS. Overall, the present results showed that ZF-L cells were responsive to epigenetic disruption. They brought further evidence on the potential of chemicals to interfere with both DNA methylation and chromatin accessibility. However, further studies are required to investigate to which extent the observed changes are reflected in DNA methylation and chromatin accessibility themselves, together with their correlation in *in vivo* models.

MO256

Cross-species applicability of the adverse outcome pathway "deiodinase inhibition leading to impaired swim bladder inflation in zebrafish"

E. Stinckens, University of Antwerp; L. Vergauwen, University of Antwerp / Zebrafishlab Dept Veterinary Sciences SPHERE; H. Witters, VITO / Applied Bio & molecular Systems; R. Blust, University of Antwerp / Department of Biology (SPHERE Research Group); G.T. Ankley, D.L. Villeneuve, U.S. EPA / National Health and Environmental Effects Research Laboratory; D. Knapen, University of Antwerp / Zebrafishlab Dept Veterinary Sciences

The adverse outcome pathway (AOP) framework can be used to help support the development of alternative testing strategies aimed at predicting adverse outcomes caused by triggering specific toxicity pathways. Previously, we developed an AOP describing how inhibition of deiodinase (DIO) enzyme activity leads to impaired swim bladder inflation in fish. Next we assessed the feasibility of selecting alternative *in chemico* assays targeting specific key events along the AOP and evaluated the potential of these *in chemico* data for predicting higher biological *in vivo* endpoints. We were able to demonstrate that the *in chemico* dataset can be used to effectively predict effects on swim bladder inflation. For a limited number of compounds however, zebrafish responded differently than what was expected. In this presentation, we will assess these outliers by examining (1) the cross-species applicability of our AOP-based assays, (2) toxicological mechanisms other than thyroid disruption that could result in effects on swim bladder inflation. We performed *in vitro* DIO assays for 20 compounds using porcine, rat and fish liver homogenates to characterize similarities and differences among species. Results show that the DIO1 inhibitory potential is nearly identical between the selected species. However, a set of bisphenol A derivatives showed lower inhibition in fish and rat compared to pig. In addition, we performed qPCR analysis of a set of 29 genes related to thyroid metabolism and swim bladder inflation after exposing zebrafish to 4 compounds for which false negative predictions were observed. These results suggest that PFOS affects surfactant properties which could impact swim bladder inflation. SMX affected genes related to the development of the 3 cell layers of the swim bladder, suggesting that this compound inhibits swim bladder development and subsequent inflation. Our results suggest that for most compounds, tissue originating from different vertebrate species can be used in the DIO assay to predict apical outcomes in fish. However, it is expected that any predictive model based on measuring only few molecular initiating events could be refined as knowledge on the involvement of other specific thyroid related processes in swim bladder inflation grows. In addition to the fact that differences in predicted effects may be observed as a result of cross-species differences, many different toxicological mechanisms can lead to swim bladder inflation effects as well.

MO257

Zebrafish responses to the fourth-generation progestin drospirenone exposures

C. Quintaneiro, Department of Biology & CESAM - University of Aveiro; A.M. Soares, University of Aveiro / department of Biology & CESAM; M. Monteiro, Aveiro University / Biology

Synthetic progestins (PGs) represent an important class of active ingredients of hormonal medicines/pharmaceuticals. Their widespread use has led to environmental contamination by these substances. Although the knowledge about their environmental concentrations and effects on fish is still scarce, PGs are rapidly uptaken through fish gills and can cause deleterious effects even at low concentrations, such as the inhibition of fish reproduction. Drospirenone (DRP) arises as one of the most used fourth-generation PGs in hormonal pharmaceuticals. In addition to its endocrine activity, it is known that DRP can interfere with other processes in fish, such on regulation of circadian rhythm. Thus, the present work aims to evaluate *Danio rerio* early life stages responses to DRP exposures at physiological and biochemical level. Zebrafish embryos were exposed to 0.01 – 100.0 µg/l of DRP during 96h to evaluate lethal and sublethal parameters. Survival, heartbeat, length and impairments on normal development such as malformations and hatching were evaluated as apical and physiological endpoints. Alterations on enzymes related with neurotransmission (acetylcholinesterase, AChE), energy production (lactate dehydrogenase, LDH) and oxidative stress (catalase, CAT and glutathione S-transferase, GST) were assessed. The oxidative damage was also assessed by alterations on lipid peroxidation levels (LPO). Exposure to DRP did not affect hatching rate, growth and development of zebrafish embryos, however, there was a decrease on heart rate with increasing concentrations of DRP. Several biochemical processes were affected by DRP exposure and oxidative damage was observed. Overall, despite not having affected zebrafish early life stages apical endpoints, our study showed that DRP might exert adverse effects at both physiological and biochemical levels at concentrations similar to those found in environment for PGs. Furthermore, our results highlight the need to assess PGs toxicity at different levels of biological organization.

MO258

Fish caging experiment as a tool for detection of in situ effects of untreated wastewaters: General stress and endocrine disruption

D. Tenji, University of Novi Sad Faculty of Sciences / Biology and Ecology; B. Mićić, Petnica Science Center/Faculty of Sciences, University of Novi Sad / Department of Biology and Ecology; S. Sipos, Faculty of Sciences University of Novi Sad / Biology and Ecology; V. Knezevic, Faculty of Sciences / Department of Biology and Ecology Laboratory of Ecotoxicology LECOTOX; S. Kaišarević, Faculty of Sciences University of Novi Sad / Department of Biology and Ecology, Laboratory of Ecotoxicology (LECOTOX); I. Teodorovic, University of Novi Sad / Department of Biology and Ecology, Laboratory of Ecotoxicology (LECOTOX)

A direct discharge of untreated municipal wastewaters from the city of Novi Sad into the River Danube made a location a focal pollutant hotspot within the framework of FP 7 funded Solutions project. A study conducted previously at this site provided detailed chemical characterization of water samples. Relatively high concentrations of a number of chemicals were identified and their toxic potency was confirmed with a battery of *in vitro* assays. As a follow-up, a fish caging experiment was conducted with the aim to check if the biological effects observed *in vitro* can be seen *in situ*, through a battery of biomarkers in liver and brain of caged fish. Ten specimens of common carp (*Cyprinus carpio* (L.), Cyprinidae), both male and female, were exposed for nine days at three sites in the River Danube: upstream (reference site), cca. 250 m and 7 km downstream of the major discharge point of the untreated sewage into the River Danube. Certain detected chemicals are recognized causative agents of endocrine disruption and stress in general with a potential to lead to adverse physiological effects. Therefore, stress marker enzyme assays (catalase, carboxylesterase and glutathione s-transferase) were performed, and the expression of stress, endocrine disruption, immune response and autophagy related genes was analysed using qRT-PCR. Selected genes included cytochrome oxidase subunit 1 (*cox1*), metalallothioneine (*mt*), heat shock protein 70 (*hsp70*) as general stress related genes; estrogen receptor α (*erα*), estrogen receptor β (*erβ*), androgen receptor (*ar*), cortisol receptor (*cr*) and vitellogenin (*vtg*) as endocrine disruption related genes; interleukin1β (*il1β)* and tumor necrosis factor α (*tnfα*) as immune response related genes, while light chain 3 β (*lc3II*) and dynein (*dyn*) were selected as autophagy related genes. Enzyme activities were higher in specimens caged at downstream locality, while catalase was lower at sewage discharge point. General stress and endocrine disruption representative genes at downstream site follow the trend of overexpression vs control (reference site), while the *vtg* was down-regulated at discharge point. Expression of *tnfα* was strongly up-regulated at downstream site, while no significant difference was noted among expressions of other immune response and autophagy related genes. The results indicate induction of adaptive stress responses and endocrine disruption and are in line with the results observed *in vitro*.

MO259

Gene transcription ontogeny of hypothalamic-pituitary-thyroid axis development in early-life stage fathead minnow and zebrafish

L. Vergauwen, University of Antwerp / Zebrafishlab Dept Veterinary Sciences SPHERE; J.E. Cavallin, U.S. EPA / US EPA MidContinent Ecology Division; G.T. Ankley, U.S. EPA / National Health and Environmental Effects Research Laboratory; C. Bars, University of Antwerp / Applied Veterinary Morphology Dept Veterinary Sciences; I. Gabriëls, E.D. Michiels, University of Antwerp / Zebrafishlab Dept Veterinary Sciences; K. Nelson, U.S. EPA / Toxic Effects Branch; J. Periz Stanacev, University of Antwerp / Zebrafishlab Dept Veterinary Sciences; E. Randolph, U.S. EPA / National Health and Environmental Effects Research Laboratory; C. Robinson, Marine Scotland Science / National Health and Environmental Effects Research Laboratory; T. Saari, U.S. EPA / ORD, NHEERL; A. Schroeder, University of Minnesota-Crookston / Math, Science and Technology; E. Stinckens, University of Antwerp; J. Swintek, Badger Technical Services; S.J. Van Cruchten, University of Antwerp / Applied Veterinary Morphology, Dept Veterinary Sciences; E. Verbueken, University of Antwerp / Applied Veterinary Morphology, Dep of Veterinary Sciences; D.L. Villeneuve, U.S. EPA / National Health and Environmental Effects Research Laboratory; D. Knapen, University of Antwerp / Zebrafishlab Dept Veterinary Sciences

The hypothalamic-pituitary-thyroid (HPT) axis is known to play a crucial role in the development of teleost fish. However, knowledge of endogenous transcription profiles of thyroid-related genes in developing teleosts remains fragmented. We selected two model teleost species, the fathead minnow (*Pimephales promelas*) and the zebrafish (*Danio rerio*) to describe the gene transcription ontogeny of the HPT-axis. Control embryos were sampled at several time points between fertilization and hatching, and larvae were sampled approximately every other day until 33 days post-fertilization. Total RNA was extracted from pooled, whole fish, and thyroid-related mRNA expression was evaluated using quantitative polymerase chain reaction. Gene transcripts examined included: thyrotropin-releasing hormone receptor (*trhr*), thyroid-stimulating hormone receptor (*tshr*), sodium-iodide symporter (*nis*), thyroid peroxidase (*tpo*), thyroglobulin (*tg*), transthyretin (*ttr*), deiodinases 1, 2, 3a, and 3b (*dio1*, *dio2*, *dio3a* and *3b*), and thyroid hormone receptors alpha and beta (*thra* and *β*). A loess regression method was successful in identifying maxima and minima of transcriptional expression during early development of both species. Overall, we observed great similarities between both species, including maternal transfer of almost all transcripts (confirmed in unfertilized eggs), increasing expression of most transcripts during hatching and embryo-larval transition, and indications of a fully functional HPT-axis in larvae. By making these data available to the community, we aim to aid in the development of hypotheses on the role of certain genes and pathways during development. Furthermore, it can function as a background reference dataset for designing and interpreting targeted transcriptional expression studies both for fundamental research and for applications, such as ecotoxicology.

MO260

Skin vitellogenin and estrogen receptor as sensitive biomarkers of estrogenicity in a sub-Antarctic fish.

M. Ferreira, F.L. Lo Nostro, Universidad de Buenos Aires. Facultad de Ciencias Exactas y Naturales - Consejo Nacional de Investigaciones Científicas y Técnicas / Departamento de Biodiversidad y Biología Experimental - Instituto de Biodiversidad y Biología Experimental Aplicada. Laboratorio de Ecotoxicología Acuática; D. AURELIANO, Centro Austral de Investigaciones Científicas (CADIC-CONICET) / Laboratorio de Ecología, Fisiología y Evolución de Organismos Acuáticos; D.A. FERNÁNDEZ, Centro Austral de Investigaciones Científicas (CADIC-CONICET) - Universidad Nacional de Tierra Del Fuego / Laboratorio de Ecología, Fisiología y Evolución de Organismos Acuáticos - Instituto de Ciencias Polares, Ambiente y Recursos Naturales; G. Genovese, Universidad de Buenos Aires. Facultad de Ciencias Exactas y Naturales - Consejo Nacional de Investigaciones Científicas y Técnicas / Departamento de Biodiversidad y Biología Experimental - Instituto de Biodiversidad y Biología Experimental Aplicada. Laboratorio de Ecotoxicología Acuática

The fast expansion of socio-economic activities on coastal areas has increased the presence of anthropogenic pollutants from industrial and domestic sources over the last decades. Recent studies have reported the presence of many persistent organic pollutants in water, sediments and in vertebrates from Antarctic areas; however, information about their potential impact on fish physiology is still scarce. The southernmost city of the world, Ushuaia (Tierra del Fuego, Argentina), and its bays are not the exception to the decline of the environmental quality. Notothenioid fish are the dominant group of the coastal icthyofauna of Antarctic and sub-Antarctic regions playing a key role in these ecosystems. The black southern cod, *Patagonotothen tessellata* is widespread in the Beagle Channel, lives in the intertidal zone, and possess paternal care. The aim of the present work is to validate vitellogenin (vtg) and estrogen receptor (Reα) as biomarkers of estrogenicity in skin samples of this species in order to provide a helpful tool to develop and perform periodic biomonitoring in sub-Antarctic waters and evaluate environmental risk in places where contamination already exists. Male fish were injected with 17β-estradiol (i.p, single dose of 10 mg/g or vehicle). Vitellogenic females were used as positive controls. Samples of skin and liver were obtained to assess vtg and ERα mRNA expression and physiological responses were studied through different endpoints: histological analysis, vtg detection in plasma samples, and sex steroid levels (estradiol (E2) and testosterone (T)). Seventy-two hours post-injection histological analysis showed normal unrestricted testis and intense cytoplasmic basophilia in hepatocytes. No vtg was detected in plasma samples of control males or before E2 injection; however, three days after treatment, males showed plasma vtg-ir bands similar to those of females. Likewise, plasma E2 concentration in males was significantly increased but the opposite was observed in T levels. These changes were also reflected in E2/T ratio. Furthermore, E2 levels in treated males were even higher than those of females. Vtg and ERα gene expression was up-regulated both in liver and skin after E2 treatment. Potential impact in parental behavior is discussed. We conclude that vtg and ERα expression in skin are sensitive and non-harmful biomarkers of estrogenicity in this Sub-Antarctic fish.

MO261

Thyroid disruption and its effects on neuronal development of zebrafish

A. Haigis, Institute for Environmental Research, RWTH Aachen / Department of Ecosystem Analysis ESA; C. Di Paolo, RWTH Aachen University / Ecosystem Analysis (ESA); J. Legradi, Vrije Universiteit Amsterdam; H. Hollert, RWTH Aachen University / Institute for Environmental Research

The endocrine disrupting effects of estrogenic compounds on humans and different species in the environment are well studied. But despite the growing numbers of patients diagnosed with thyroid disorders, thyroid hormone disrupting effects of compounds are less investigated. Furthermore, it is estimated that many substances found in our environment can influence the thyroid system and act as thyroid hormone disruptors. Thyroid hormones play a critical role in brain development and it has been shown that a lack reduces cognitive development. But the connections between thyroid disruption and developmental neurotoxicity are rarely studied and the basic mechanisms remain unknown. Because the thyroid system is well conserved among vertebrates, effects observed in humans can also be expected in wildlife. Our preliminary results have shown that potential thyroid disrupting compounds cause behavioural alterations in zebrafish (*Danio rerio*) larvae. Thereby, substance and concentration dependent effects were observed, indicating differing toxic modes of action. Within this study we aim to further investigate thyroid disruption in zebrafish early life stages and elucidate a possible link to (developmental) neurotoxicity. Therefore, embryos and larvae of zebrafish are exposed to different potential thyroid disruptors. The methodological approach proposes to assess the neurotoxic potential of the test substances based on different behaviour assays, the mechanistic link between thyroid and neurotoxicity will be made using transcriptomics, proteomics and metabolomics. This work will be conducted within the scope of the "NeuroBox"-project and in collaboration with the "Misse" project. In NeuroBox novel bioassays are developed, with the objective to assess the neurotoxic potential of water contaminants and improve water quality, ultimately aiming to reduce the exposition of humans and the environment to these substances. In this context, the project is expected to further contribute to the understanding of basic mechanisms of neurotoxicity, its connection to thyroid disruption and to identify novel endpoints. This knowledge may then be integrated in a bioassay battery and used for the improvement of water quality guidelines.

MO263

Identification of toxicity pathways predicting adverse outcomes of chlorpyrifos in fathead minnows

K. Bluhm, University of Saskatchewan / School of Environment and Sustainability; A. Alcaraz, University of Saskatchewan - Toxicology Centre / Toxicology Centre; D. Green, University of Saskatchewan - Toxicology Centre / Toxicology; T. Lane, University of Saskatchewan; M. Brinkmann, University of Saskatchewan / School of Environment and Sustainability and Toxicology Centre; J. Taghavimehr, A. Masse, N. Baldwin, University of Saskatchewan / Toxicology Centre; D. Crump, Environment and Climate Change Canada / National Wildlife Research Centre; N. Basu, McGill University / Faculty of Agricultural and Environmental Sciences; N. Hogan, University of Saskatchewan / Toxicology Centre and Department of Animal and Poultry Science, College of Agriculture and Bioresources; M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre

Chlorpyrifos is an organophosphate insecticide that acts as a neurotoxicant through inhibition of the enzyme cholinesterase. The mode of action of organophosphates in target and non-target organisms, including mammals, is similar. The aim of the project is to develop an early life-stage gene expression assay (EcoToxChip) that captures critical toxicity pathways of chlorpyrifos for the prediction of apical outcomes of regulatory relevance. As this assay is intended to use early life-stages that are not feeding independently, it would not be considered as a live animal test, and therefore, would address the need for alternative approaches in chemical screening. As part of the project, critical toxicity pathways and associated core genes will be identified following exposure of fathead minnows (*Pimephales promelas*) at early life-stages to three sub-lethal concentrations of chlorpyrifos. Specifically, sequence-by-synthesis-based whole transcriptome (RNASeq) and high-resolution mass-spectrometry-based shotgun proteomics will be used to characterize key molecular toxicity pathways. Pathways will then be correlated with downstream biological responses of ecological and regulatory relevance, and critical genes linked to apical outcomes will be identified for inclusion on EcoToxChips. Chlorpyrifos concentrations were selected based on a preliminary test as well as concentrations in published data. These tests revealed a threshold level of mortality between 1 and 10 µg/L chlorpyrifos. To ensure the determination of solely sub-lethal effects in at least two of the tested concentrations, 0.5, 1.5 and 4.5 µg/L chlorpyrifos solutions were investigated in the fathead minnow early life-stage assay with larvae samplings after 7 and 32 days of exposure. None of these concentrations affected survival or growth, resulting in a sub-chronic NOAEC and LOAEC of 4.5 and 10 µg/L chlorpyrifos, respectively, in fathead minnows. Samples are currently being further analyzed for molecular and physiological endpoints to gain insight into critical toxicity pathways. This study is part of the EcoToxChip project (@ecotoxchip).

MO264

Evaluation of the deleterious effect of 2 pesticides on juveniles of the zebrafish Danio rerio

G. Geraldo Morales, Universidad Autonoma Metropolitana Iztapalapa / Departamento de Hidrobiologia; A. Sobrino-Figueroa, Universidad Autonoma Metropolitana Iztapalapa / Hidrobiology

In this study an evaluation of the toxic effect of 2 pesticides: Dichlorvos which is an organophosphorus insecticide, used for the control of ectoparasites in fish, and insecticide Imiprotrin belonging to the group of pyrethroids, used as a main component in products for domestic use, was carried out. Because the previous studies with these products are scarce, the objective of this work was to evaluate its toxicity and its effects in 3 biomarkers: peroxidation of lipids (lipoperoxidation), the activity of the enzyme acetylcholinesterase (AchE) and the production of macromolecules (proteins, lipids and carbohydrates). Initially a test (96 hrs) was made, where zebrafish juveniles were exposed to 5 concentrations of pesticides (10, 1, 0.1, 0.01, 0.001, 0.0001 mg L^{-1}) to determine the 50 lethal concentration (LC_{50}). Subsequently a bioassay with a duration of 15 days it was carried out where zebrafish juveniles were exposed to 2 sublethal concentrations (LC_1 and LC_5). The results obtained show that Imiprotrin was more toxic ($LC_{50} = 1.67 \pm 0.873$ mg L^{-1}) than Dichlorvos ($LC_{50} = 5.3$ mg L^{-1}). In the sublethal bioassays it was observed that the toxicity of these xenobiotics increased with the time of exposure. The degree of lipoperoxidation in the imiprotrin tests varied from 64.7 to 147.5 nM Tbars mg^{-1} and was higher than that observed in the bioassays with Dichlorvos (22.6 to 93.8 nM Tbars mg^{-1}). In the fish exposed to Dichlorvos a decrease of 43% to 86% in the activity of the AchE enzyme was observed and from 14% to 64% in the juveniles exposed to imiprotrin. The juveniles of zebrafish that showed a decrease in the activity of the AchE greater than 35% had changes in their swimming behavior and in their feeding. The energy content of the fish exposed to pesticides decreased by 64% in the Imiprotrin tests and 81% in the Dichlorvos bioassays. The insecticides Dichlorvos and Imiprotrin are little persistent in the environment, their half-life is 5 to 8 days, but the results of this study indicate that their effects on organisms are probably irreversible.

MO265

Effects of Omeprazole on zebrafish embryos (Danio rerio)

A. Sobrino-Figueroa, Universidad Autonoma Metropolitana Iztapalapa / Hidrobiology; J. Salazar Hernández, Universidad Autonoma Metropolitana Iztapalapa / Biologia

Omeprazole is a proton pump inhibitor, it is used for the control of gastritis, for this reason it is one of the most prescribed drug, consumed by millions of people in the world. This drug has been associated with acid rebound hypersecretion, decreased absorption of nutrients, osteoporosis and neurological disorders. Its toxic effects have been evaluated in mice and rats only, for this reason in this work was made an evaluation of the toxic effects of Omeprazole in zebrafish embryos. Bioassays were performed (OECD test 236) where the embryos were exposed to 5 concentrations of the drug (200, 100, 50, 25, 12.5 mg L^{-1}) plus a negative control, to determine the LC_{50} (24 hours). the embryos were subsequently exposed to the LC_1 LC_5 and LC_{10} for 72 hours to evaluate the degree of lipoperoxidation, by means of the evaluation of Tbars (Buege and Aust, 1978), the activity of the enzyme AchE as an indicator of effects neurotoxic (Ellman et al., 1961) and the frequency of malformations (OECD test 236). In the lethality tests, the LC_{50} value of 193.87 ± 18.48 mg L^{-1} was obtained. In the sublethal bioassays at 72 hours of exposure it was observed an increased in the degree of lipid peroxidation (52%) in in embryos exposed to LC_{10}. In the evaluation of the AchE activity, significant differences were obtained between the control and the embryos exposed to omeoprazole ($p < 0.05$), in the concentrations LC_1 and LC_5 a decrease in the activity of this enzyme was observed. The percentage of inhibition of AchE varied from 9 to 66.7%. A higher frequency (22%) of deformed embryos was observed in the LC_{10} concentration. The results of this study showed that omeoprazole has a neurotoxic and possibly genotoxic effect in sublethal concentrations in zebrafish embryos.

MO266

The neurotoxic effects of Venlafaxine on zebrafish larvae - Omics technologies in the focus of global environmental challenges

M. Gundlach, RWTH Aachen University / Department of Ecosystem Analysis ESA; C. DI PAOLO, Instit for EnvironmResearch RWTH Aachen Univ / Department of Ecosystem Analysis; J. Legradi, Vrije Universiteit Amsterdam; A. Haigis, Institute for Environmental Research RWTH Aachen University; H. Hollert, RWTH Aachen University / Institute for Environmental Research; I. Werner, Eawag, Swiss Federal Institute of Aquatic Science and Technology; M. Fenske, Fraunhofer Insitute for Molecular Biology and Applied Ecology IME

The identification, analysis and evaluation of neurotoxic chemicas are a worldwide challenge. \nThe societal costs for neurological disorders caused only by endocrine disrupters in Europe was \nestimated to amount to hundreds of billion euros per year. Considering the ecosystem services\nprinciple, effects on single species, communities and whole ecosystems would increase that up \nto hundred times, similarly to when considering other chemicals such as neuroactive pharmaceuticals. \nAntidepressants such venlafaxine are of increasing environmental neurotoxic convern. Venlafaxine is one of the most prescribed antidepressants in Europe and the U.S. and a know aquatic pollutant. \nIt is a serotonin-norepinephrine reuptake inhibitor, increasing serotonin and norepinephrine synaptic\nconcentrations in brain regions. It was also shown to affect monoamine levels and cause behavioral alterations\nin fish. The aim of this study was to analyze the neurotoxic potential of Venlafaxine on zebrafish \nlarvae by evaluating transcriptomic profiles and behavioral alterations. The locomotor activity in \nthe light-dark transition test and thigmotaxis were evaluated in 5 dpf larvae exposed to 24 h to \n1 nM, 100 nM and 10 uM venlafaxine using DanioVision(R) and EthoVision. A significant difference\nin the swimming behavior concerning the different concentrations could be detected. Effects on the transcriptome level were verified in zebrafish continuously exposed to Venlafaxine\n1 nM, 100 nM up to 120 hpf. RNA was extracted from pooled samples (n = 25 fish) and submitted \nto Sybr Green quantitative real-time chain reaction (qPCR). Literature-based target gene selection \nconsidering targets involved in circadian rhythm regulation, muscle processes and responeses \nto abiotic stimulus. Behavioral results indicate decreased swimming distance and increased thigmotaxis \nin exposed fish, in agreement with previous own data for continuous venlafaxine exposure. Results \nfor qPCR indicated modulation of some of the pre-selected target genes such as *skbp5*, and currently \nconfirmatory qPCR is being conducted. Further investigations in this project plan to include proteome \nand metabolome analysis. This study is expected to be part of a bigger overview and understanding of \nthe different effects of chemicals and pharmaceuticals on neuronal development.

MO267

Acute effects of the ayahuasca infusion (Banisteriopsis caapi and Psychotria viridis) on zebrafish and rodent models

T.S. Andrade, Universidade de Brasilia / Laboratory of Genetics and Toxicology; W. Melo Junior, University of Brasilia; R. Oliveira, State University of Campinas / SCHOOL OF TECHNOLOGY - UNICAMP; A.D. ANDRADE, University of Brasilia / Laboratory of Toxicology, Faculty of Health Sciences; M.L. da Silva, Universty of Brasilia / genetic toxicologic; J.A. Morais, University of Brasilia / Laboratory of Toxicology, Faculty of Health Sciences; C. Koppe Grisolia, University of Brasilia / Department of Genetics and Morphology; I. Domingues, University of Aveiro / CESAM Department of Biology; E.D. Caldas, University of Brasilia / Laboratory of Toxicology, Faculty of Health Sciences; A. Pic-Taylor, University of Brasilia / Laboratory of Embryology and Development Biology

Ayahuasca is a psychoactive concoction prepared with the plants *Banisteriopsis caapi* and *Psychotria viridis* and used ancestrally by Amazonian Indian populations, and recently, by Christian religions in Brazil and other countries. The present study aimed at identifying the ayahuasca effects in early fish development and compares its neurobehavioral effects in the zebrafish embryo and rat models. Toxicity and developmental endpoints for zebrafish embryos were assessed at 0 to 1000 mg/L during 96 h of exposure. The effects on locomotor activity of zebrafish larvae were assessed using the automated video tracking system Zebrabox at 0 to 20 mg/L and after 120 and 144 hours of exposure. The ayahuasca infusion was administered once by gavage to *Wistar* rats at 1, 5 and 15 times the dose taken during a religious ritual, and neurobehavioral effects evaluated after 2 hours in the open field (OFT), elevated plus-maze (EPM) and forced swimming (FST) apparatus. The LC_{50} of ayahuasca in zebrafish was estimated to be 236.3 mg/L. Ayahuasca exposure caused significant developmental anomalies in zebrafish embryos, mainly at the highest concentration tested, including hatching delay, loss of equilibrium, edema and accumulation of red blood cells. The behavior of embryos was also significantly affected, with a decrease in locomotor activity at the highest tested concentration. Decreased locomotion was also observed in the rats treated at the highest dose in the OFT and EPM, and a higher swimming time in the FST, suggesting a possible antidepressant effect. These results indicated that the effects of ayahuasca correlated well for zebrafish embryos and rodents, showing that zebrafish may provide a useful model to study ayahuasca and other hallucinogenic drugs. Further research focusing on the molecular pathways affected by ayahuasca administration in both zebrafish and rat models could provide additional information on the potential of ayahuasca as an antidepressant. Acute effects of the ayahuasca infusion (*Banisteriopsis caapi* and *Psychotria viridis*) on zebrafish and rodent models

MO268

Chronic exposure to fluoxetine affects growth, feeding, swimming behavior and tissue organization of zebrafish.

N. de Farias, University of Brasilia / Departamento de Genética e Morfologia; R. Oliveira, State University of Campinas / Department of Genetics and Morphology; T.S. Andrade, Universidade de Brasilia / Laboratory of Genetics and Toxicology; J.M. Pinto, University of Brasilia / Departamento de Genética e Morfologia Instituto de Biologia; C.K. Grisolia, University of Brasilia UnB / Department of Genetics and Morphology

Fluoxetine (FLX) is among the top 100 drugs prescribed annually worldwide. This selective serotonin reuptake inhibitor is highly detected in aquatic ecosystems and has the potential to modulate levels of serotonin of non-target organisms. The present study aims to evaluate the effects of chronic exposure to FLX on fish. Zebrafish juvenile were exposed to FLX during 30 days following the OECD protocol (no. 215). The assays were performed in triplicate with six treatments of FLX, 0; 0.01; 0.1; 1; 10 and 100 µg/L. A total of 60 fish per treatment (20 per replicate) were used. Growth and feeding behaviour were analysed at the end of the test. Histological analyses of liver followed standard H&E routine. Video recordings (10 min) were analysed for swimming behaviour by measuring the time spent by each fish in each of the layers of the aquarium (bottom: 0–5 cm, middle: 6–10 cm and upper: 11–15 cm). Our results showed decrease in growth rate and erratic feeding behaviour at 100 µg/L. Also, in concentrations as low as 0.1 µg/L were observed histological alterations in liver microstructure such as decrease of glycogen and progressive loss of hepatic architecture. The pattern of swimming behaviour of fish changes significantly, fish spend more time at the upper part of the aquarium in concentration above 10 µg/L. Altogether, the present study demonstrated that chronic exposure of zebrafish to FLX can affect multiple endpoint such as growth tissue organization, feeding and swimming behaviour. These results emphasize the relevance of an integrated approach in the ecotoxicological assessment of psychiatric drugs.

MO269

Mitochondrial Disorders of Zebrafish Embryos Exposed to Individual Organochlorine Pesticides and Their Mixtures

H. Lee, Seoul National University of Science and Technology / Environmental toxicology and health; S. Lee, Seoul National University of Science and Technology / Enviornmental enginnering; K. Kim, Seoul National University of Science and Technology / Environmental Engineering

Organochlorine pesticides (OCPs), prohibited compounds in the 1970s, are still being detected in human and environmental samples. Mitochondrial dysfunction caused by chemical exposure have attracted great attention on toxicological studies. We evaluated mitochondrial dysfunction in dechorinated zebrafish embryos exposed to individual 5 OCPs (i.e., *p,p*-DDT, Chlordane (mixture), Heptachlor, Hexachlorobenzene (HCB), and beta-hexachlorocyclohexane (beta-HCH)), and their mixtures from 4 to 120 hpf (hours post-fertilization). We measured oxygen consumption rate (OCR) at the embryonic sublethal concentrations of 0.05, 0.1, and 0.5 mg/L, by using Seahorse XFe Extracellular Flux Analyzer at 24 hpf. The OCR results are compared with the activity of mitochondrial complex I~ IV after isolating mitochondria from embryos at 48 hpf. In addition, we analyzed mRNA expression of transcription factors (i.e., *PGC-alpha*, *Acox1*, *SDHA*, *MCAD*, and *CS*), associated with mitochondrial metabolism, at 120 hpf. This comprehensive results could suggest the flexibility of the embryonic zebrafish model on the methodology and a set of research scheme to determine mitochondrial disorders in the exposure of individual OCPs and their mixtures.

MO270

The NeuroBox Project

J. Legradi, Vrije Universiteit Amsterdam; A. Haigis, Institute for Environmental Research RWTH Aachen University; M. Gundlach, RWTH Aachen University / Department of Ecosystem Analysis ESA; C. Di Paolo, RWTH Aachen University / Ecosystem Analysis (ESA); M. Spehr, B. Kampa, RWTH Aachen University; H. Hollert, RWTH Aachen University / Institute for Environmental Research

The societal impact of neurological disorders like Alzheimer’s disease or neurological deficits like reduced IQ is immense. For patients and their relatives, the effects, e.g. severe mental and physical problems, are often devastating. There is mostly no cure available and even treatments to reduce or stop the progression of the diseases are limited. The number of people diagnosed with neurological disorders is increasing. This increase cannot be explained by improved diagnostics and increased age. Exposure to neurotoxic chemicals is suspected to play a role in the development and progression of these diseases. It has been estimated that alone in Europe, exposure to solely endocrine disrupters that lead to neurological disorders, costs society €150 billion per year. This does not include costs due to exposure to known neuroactive substances like pesticides and pharmaceuticals. Currently, testing for neurotoxicity is not required within the EU (REACH) as it is not clear *how to assess neurotoxicity*. Considering the increasing numbers of chemicals and the physiological and morphological complexity of the nervous system, it is a major challenge to test all substances for their neurotoxic potential, new advanced neurotoxicity assessment strategies need to be developed to fulfil these demands. The bmbf funded project NeuroBox ((02WRS1419; coordination UBA, T. Grummt) aims to develop novel assessment strategies for neurotoxicity assessment of anthropogenic substances in water samples. The work is split over six subprojects. In our sub-project, we use zebrafish embryos to identify neurotoxic mode of actions of commonly found water contaminants. Based on our findings novel screening assays will be developed to easily screen water samples for neurotoxic effects. In combination with mouse models and clinical researchers we will also link our results to mammalian neurological diseases like ADHD and autism and neurodegeneration studies, to identify potential exposure-disease relationships. Furthermore, we will study the link between endocrine disrupting compounds and neurotoxic effects. We could see behavioral as well as metabolomics and transcriptomic changes after exposure to compounds. These changes were observed ad concentrations bellow any phenotypic changes could be observed. Our results so far show that assessing neurotoxicity is complex and a tiered approach covering behavioral tests in combination with OMICS techniques seem to be a cost and time efficient way.

MO271

Understanding the correlation between behavioural inter-individual variability and physiology/morphology in zebrafish larvae

K.T. Kirla, C.M. vom Berg, Eawag Swiss Federal Institute of Aquatic Science and Technology / Environmental Toxicology

Background: Zebrafish are widely used in biomedical research as they offer several features such as the fully sequenced genome, availability of a wide range of mutants, generation of large numbers of offspring throughout the year and conserved vertebrate drug targets to a great extent. Early stages of zebrafish are called as non-protected life stages and are therefore used for testing chemicals for their toxicity as an alternative to conventional animal testing. Moreover, zebrafish larvae are amenable to test neurotoxicity and behavioural effects of chemicals as their small body size allows tracking a large number of individuals with full control over the environment. **Aim:** The aim of the project is to investigate behavioural inter- and intra-individual variability in zebrafish larvae as a basis to better estimate effects of chemicals on behavioural responses. Analysis of inter-individual differences might offer new insights into mechanisms of toxicity considering that every individual's response to a chemical differs based on their genetic make-up. **Hypothesis:** We are testing whether inter-individual variability is constant over time and whether levels of locomotor activity correlate with physiological and morphological properties of the larvae. **Methods:** At first, spontaneous locomotor activity is measured for 40 min in continuous light at different timings of the day from 5-7 days post fertilization. Heart rate, body size and other physiological properties of the same individuals are analysed at different time points. **Results:** From the preliminary results of the locomotor activity analysis, we could assign the larvae to three categories based on their activity levels compared to the average activity: highly active, less active and the individuals close to the average activity, which are also the ones less variable over time. To attribute this variability in the individual's activity to its physiology and phenotype, the analysis of heart rate, length and blood flow are on-going. **Outlook:** The variability of each individual will be taken into account to better evaluate effects of the chemicals on behavioural responses. Inter-individual differences will be explored as a source of information on mechanisms of toxicity of chemicals with unknown targets and mode of action.

Environmental fate of emerging contaminants in the water cycle: analytical challenges and engineered solutions (P)

MO272

Effect of iodinated X-ray contrast media in the formation of disinfection byproduts during chlorination and chloramination of water

C. Postigo, IDAEA, CID-CSIC / Environmental Chemistry; M. Armstrong, University of North Carolina at Chapel Hill / Department of Environmental Science and Engineering; H.K. Liberatore, K. Lamann, S. Kimura, A. Cuthbertson, S.D. Richardson, University of South Carolina; T. McDonald, Y.M. Sey, U.S. Environmental Protection Agency / National Health and Environmental Effects Research Laboratory; S. Duirk, University of Akron; J. Simmons, U.S. Environmental Protection Agency / National Health and Environmental Effects Research Laboratory

Iodinated X-ray contrast media (ICMs), used in medical imaging, are poorly metabolized by humans and enter wastewater. As they are incompletely removed during wastewater treatment, ICMs are released to the aquatic environment and have been detected in drinking water sources. ICMs have been identified as iodine sources that may enhance the formation of iodine-containing disinfection byproducts (I-DBPs) during drinking water disinfection. This work investigated the effect of different ICMs, iopamidol (IPAM), iopromide (IPR), diatrizoate (DTZ) and iohexol (IHX), in the formation of different classes of DBPs during source water disinfection by either free chlorination or chloramination. To do this, we performed large-volume (~120 L each), laboratory-controlled, headspace-free disinfection reactions with 5 µM ICM and 100 µM as Cl_2 disinfectant concentrations. The resulting DBP mixtures were chemically characterized for 21 targeted non-I-DBPs, 11 targeted I-DBPs, and non-targeted I-DBPs by means of gas chromatography coupled to low- and high-resolution mass spectrometry. The presence of ICMs in source water had no apparent effect on either the concentration or speciation of the four regulated trihalomethanes (chloroform, bromodichloromethane, chlorodibromomethane, bromoform). IPAM, but not other ICMs, enhanced formation of dichloroacetic acid, bromoacetic acid and dibromoacetonitorile during chlorination. I-DBPs formation was slightly enhanced in the presence of ICMs, particularly in chlorinated water containing IPAM, where the highest levels of I-DBPs were formed, and in chloraminated water containing IPR or IHX. The presence of DTZ did not appear to affect I-DBP formation. Non-targeted analysis of the DBP mixtures revealed the formation of novel I-DBPs in chlorinated IPAM-containing water including iodoacetonitrile, chloroiodoaceontirile, trichloroiodomethane and several iodo-acids. Our results indicate that ICMs enhance the formation of both I-DBPs and non-iodinated DBPs when present during chlorination and that IPAM, in particular, is a relevant iodine source in water undergoing chlorination or chloramination. Acknowledgments: CP acknowledges support provided by EU FP7 (No. 274379, Marie Curie IOF) and the Government of Catalonia and the COFUND programme (Marie Curie Actions, EU FP7) (2014 BP_B00064). This abstract does not represent EPA policy. This work was also partially supported by the National Science Foundation, under Award NSF1124865 to SDR.

MO273

The use of a polymer inclusion membrane for the determination of arsenate by gas-diffusion flow analysis with spectrophotometric detection

R. Vera, University of Girona / Chemistry; C. Fontas, University of Girona / Department of Chemistry; M.G. Almeida, The University of Melbourne / School of Chemistry; E. Antico, University of Girona / Department of Chemistry; R.W. Cattrall, S.D. Kolev, The University of Melbourne / School of Chemistry

Arsenic is a naturally occurring toxic element, which is present in waters in different areas around the world, including South Asia, South America and to lesser extent Europe [1]. Therefore, the World Health Organization has set the guideline concentration for arsenic in drinking water at 10 µg L^{-1} [2]. The most frequently encountered arsenic species in environmental and drinking waters is arsenate (As(V)). Therefore, highly sensitive analytical techniques are required for its determination in water samples.**In the present work a novel flow analysis (FA) system has been developed for the determination of As(V) in environmental and drinking waters at the low µg L^{-1} level. The system uses a polymer inclusion membrane (PIM) based on poly(vinylidenefluoride-co-hexafluoropropylene) as the polymer and Aliquat 336 as the extractant, for the online preconcentration and separation of As(V) in a PIM cell. The sample solution is propelled for a predetermined period of time through the PIM cell where a PIM separates the sample stream and an acceptor stream which is stopped during the sample passage through the PIM cell to allow preconcentration of As(V) in the static acceptor solution located in the acceptor channel of the cell. The analytical procedure involves a 15 min stop-flow time and sample solution flow rate of 2.5 ml min^{-1}. After the stop-flow time the acceptor stream is re-started and As(V) is reduced to arsenite (As(III)) by merging the acceptor stream with a reagent stream containing 4 M HCl, 1% KI and 0.5 % ascorbic acid. This is followed by arsine generation using another reagent stream incorporating 0.5% $NaBH_4$ and 0.05 M NaOH. The generated arsine is transported across the hydrophobic membrane of a gas-diffusion cell into a solution containing 0.02 mM $KMnO_4$ and 0.05 M NaOH where it is oxidised thus producing a decrease in the $KMnO_4$ absorbance, monitored continuously at 528 nm. Under optimal conditions the FA system offers a limit of detection of 3 µg L^{-1} As(V), a sampling rate of 2.8 h^{-1} and a repeatability, expressed as RSD of 1.8% (n=5, 25 µg L^{-1}) and 2.8% (n=5, 50 µg L^{-1}). The FA method has been successfully applied to the determination of As(V) in tap water in the µg L^{-1} concentration range. <strong>References** [1] Villaescusa I, Bollinger JC. 2008. Arsenic in drinking water: sources, occurrence and health effects (a review). Rev Environ Sci Biotechnol 7:307–323. [2] World Health Organization (WHO). 2011. Guidelines for drinking-water quality, 4th edition

MO274

Balancing environmental quality standards and infrastructure upgrading costs for the reduction of microcontaminants loads in rivers

V.P. Gimeno Melia, Catalan Institute for Water Research (ICRA); J. SEVERYNS, AQUAFIN; J. Comas, L. Corominas, Catalan Institute for Water Research ICRA

Investments for upgrading wastewater treatment plants (WWTPs) with tertiary treatment to reduce microcontaminants loads in surface waters at a catchment scale can be daunting. Our hypothesis was that these investments seriously change upon selection of the Environmental Quality Standards (EQS) for unregulated microcontaminants, and hence there is a trade-off between EQS selection and investment which needs to be considered in decision-making. We used a customized Microcontaminant Fate and Transport Model coupled to an optimization algorithm to validate this hypothesis. We used the Llobregat river basin as a case study and diclofenac as the unregulated microcontaminant. The algorithm optimized the number of WWTPs in this catchment requiring an upgrade to minimize the EQS exceedance of diclofenac in all river sections and the total cost. We simulated 40 scenarios representing a combination of 4 potential EQS which are currently being discussed in the European Union (10, 30, 50 and 100 ng/l), 5 levels of uncertainty bounds in the predictions of river concentrations and 2 hydrological scenarios (average flows and low flows). The results showed that the optimal cost of WWTP upgrades with tertiary treatment for the Llobregat river basin was 8 M€/year (upgrading 8 WWTPs out of the existing 56 for fullfilling an EQS of 30 ng/l in the entire catchment). Such an investment seriously changed upon selection of EQS. The cost varied from 6 M€/year (upgrading 3 WWTPs for fullfilling an EQS of 100 ng/l) to 13 M€/year (upgrading 18 WWTPs, for fullfilling an EQS of 10 ng/l). We observed that the selection of catchment hydrological conditions during the upgrading analysis also plays a key role. The cost of the upgrades when considering low surface water flows (minimum environmental flows that ensure compatibilization of environmental needs and human water consumption) was 50% higher than the cost obtained with average flows (average hydrological conditions in the Llobregat). Finally, we demonstrated that the reduction of uncertainty in the modelling process (through R&D activities) provides transparency in the decision-making process.

MO275

Calibration of passive samplers for the monitoring of drugs in French Caribbean

N. Tapie, Univ. Bordeaux, CNRS, EPOC UMR 5805 / EPOC UMR 5805; D. Devault, Univ. Paris Sud / ESE UMR 8079; S. Karolak, Univ. Paris Sud, CNRS, AgroParisTech / ESE UMR 8079; Y. Levi, Univ. Paris Sud / ESE UMR 8079; H. Budzinski, University of Bordeaux

Drugs are more and more consumed worldwide (ONUDC, 2017). The French Caribbean is a hub of world cocaine trafficking and an important place of consumption of cocaine in the form of crack. The local population is particularly affected by this scourge. The consumption of illicit drugs induces the excretion of parent compounds or metabolites (markers of drugs uses) in waste water, and in the end their release into the aquatic environment. So, one of the ways to evaluate the local use of illicit drugs is to track drug residues in waste water treatment plants (WWTP). The present study that takes place in the SENEUR Project and explores the use of passive sampling techniques to monitor illicit drugs in WWTP. Polar Organic Chemical Integrative Samplers (POCIS) were exposed in situ in a Waste Water Treatment Plant in Martinique (French Caribbean) during ten days. First an analytical development by ESI-LC/MS/MS was done in order to be able to analyze 17 compound as markers of drug uses (cocaine, heroin, amphetamine, cannabis, their main metabolites and some substitute products such as methadone) in effluent and in POCIS (LOQ from 0.01 to 0.1 pg.inj) . Secondly triplicates of POCIS were exposed in WWTP for calibration during 10 days. POCIS were collected at different times $T_{0\ days}$, $T_{1\ days}$, $T_{2\ days}$, $T_{3\ days}$, $T_{5\ days}$, $T_{7\ days}$, $T_{10\ days.}$. Water samples were also daily collected. The first result of the calibration show a good capacity of the POCIS to sample cocaine markers (cocaine, benzoylecgonine, cocaethylene, ecgonine methyl ester), cannabis markers (11-nor-9-Carboxy-THC) and morphine over short exposure time (3 to 5 days). The calculated sampling rate (Rs) vary from 0.004 for benzoylecgonine to 0.2 $l.J^{-1}$ for cocaine.

MO276

Passive sampling in surface water as an immission-based approach to extrapolate waste-water-related pressures and potential EQS exceedence in Luxembourg

T. Galle, Luxembourg Institute of Science and Technology; D. Pittois, M. Bayerle, Luxembourg Institute of Science and Technology LIST

The pressure on surface waters that is exerted by emerging pollutants depends on the loads arriving at treatment plants and the elimination capacity of the latter. Both can be variable depending on the compound, the contributors of the sewer network as well as the design and operation of the treatment plant. Several emerging compounds have mixed uses and can therefore stem from domestic as well as from industrial sources. Regulators have an interest in knowing immission situations that will probably lead to EQS exccedences without needing to monitor emerging substances in the whole hydrological network. Here we used passive samplers to monitor the immission situation in 15 surface waters under low-flow conditions and different sanitary pressures in Luxembourg. We define sanitary pressure as the number of population equivalents divided by the surface of the catchment for a monitoring point. For that purpose, a catchment delineation is performed for each measurement point and the PE of discharging treatment plants within the catchment are summed up. PE/ha gives a hint on the dilution of WWTP emissions by natural flow. Since WWTPs are relatively constant sources sampling rates of passive samplers can easily be calibrated with grab samples over all monitoring locations. The data evaluation uses the conservative behaviour of carbamazepine as a tracer for (treated) wastewater input. Carbamazepine concentrations proved to be correlated to the sanitary pressure (PE/ha) in a catchment. The plotting of other compound concentrations against carbamazepine holds useful information: it shows the variability of the WWTP influents as well as elimination capacities in the catchment. According to these hypotheses recalcitrant pharmaceuticals showed very strong and narrow linear correlations with carbamazepine while intermediately degradable compounds displayed higher variability. Complete outliers make it easy to detect industrial sources as was the case for triazoles for instance. Finally the data set made it possible to extrapolate expected concentrations of emerging compounds for different sanitary pressure levels and by integrating EQS values, to define a threshold of 2.5 PE/ha above which EQS exceedance for diclofenac and clarithromycine is expected. This makes it easy to design a map of the river network with segments at risk with basic population equivalent information.

MO277

Determination of Perchlorate by U.S. EPA Method 332.0 Using a Compact Ion Chromatography System Coupled with Mass Spectrometry (IC-MS)

B. Huang, Thermo Fisher Scientific / marketing; T. Cross, Thermo Fisher Scientific; J. Rohrer, Thermo Fisher Scientific / Chromatography and mass spectrometry division

Perchlorate has been used as an oxidizer in rockets, munitions, and fireworks since the 1950s. It has been found to cause thyroid dysfunction, and has been linked to tumors in humans. Perchlorate is regulated under the Safe Drinking Water Act (2011). Massachusetts and California have established standards for drinking water of 2 μg/L and 6 μg/L respectively. Determination of perchlorate in environmental samples has also gained the attention of the International Standards Organization (ISO). U.S. EPA Method 332.0 — Ion Chromatography with Suppressed Conductivity and Electrospray Ionization/Mass Spectrometry is one of the most sensitive and robust characterization methods available for perchlorate determinations. Mass spectrometry (MS) provides lower detection limits in high-ionic-strength matrices than conductivity detection alone. These low detection limits are achieved without sample preparation. Our study updates the IC-MS method published in U.S. EPA Method 332.0 for determination of perchlorate in environmental waters. The method uses a Thermo Scientific™ Dionex™ IonPac™ AS20 column set, on a recently introduced compact IC system coupled with a recently introduced single quadrupole mass spectrometer. The selectivity of the mass spectrometer allows the quantification of perchlorate in high-ionic-strength samples at well below currently enforced action levels. Ionization improvements to the electrospray source eliminate the need to add organic solvent to enhance detection. Method detection limit (MDL) values in deionized water are 20- 60 ng/L, and MDLs in high-ionic-strength matrix are 30- 60 ng/L. The calibration curves for perchlorate in high-ionic-strength matrix at 101 m/z over the range of 125-5000 ng/L using the internal standard and external methods showed good linearity with the coefficient of determination being 0.9993, and 0.9998 respectively. Single laboratory precision in drinking waters, as measured by RSD, was < 5% at concentrations >150 ng/L perchlorate, and accuracy, was 95.6-102% for concentrations >150 ng/L perchlorate, and 111% for concentrations < 150 ng/L perchlorate. Single laboratory precision in high-ionic-strength matrix, was < 5% at concentrations >150 ng/L perchlorate, and accuracy, was 100-103.5% for concentrations >150 ng/L perchlorate.

MO279

NEW OPPORTUNITIES FOR THE NON TARGETED ANALYSIS OF ENVIRONMENTAL CONTAMINANTS USING GAS CHROMATOGRAPHY- ORBITRAP MASS SPECTROMETRY

P. Silcock, Thermo Fisher Scientific / GC-MS; D. Cardona, Thermo Fisher Scientific / Environmental Analysis

Since the middle of the 20th century GC-MS has made a long journey towards its current status as one of the major analytical techniques used in a diverse range of applications. Despite this, GC-MS has had more than four decades to wait for a new type of mass analyzer with the potential to advance capability over previously applied technology. Almost two years on from the first commercial introduction of Orbitrap GC-MS in 2015, in this presentation, we explore how this technology has been applied specifically to the analysis of environmental contaminants and how highly selective non-targeted acquisition can be used to explore changes in our approach to routine environmental analysis. Primary applications to be highlighted are the discovery of new disinfection by-products (DBPs) resulting from water treatment processes, using a non targeted approach, as well as the potential for addressing the difficult analytical challenges for a complex class of emerging persistent organic pollutants: short chain chlorinated paraffins (SCCPs).

MO280

HILIC workflow strategy for the hidden target screening of very polar compounds in surface waters

S. Veloutsou, Technical University of Munich; S. Bieber, Technical University of Munich / Chair of Urban Water Systems Engineering; S. Grosse, T. Letzel, Technical University of Munich

Trace Organic compounds (TrOCs) in water can be biogenic or anthropogenic. These compounds can range a lot in terms of polarity. Reverse Phase Liquid Chromatography (RPLC) is the most common and widely used tool for the separation of non-polar and mildly polar compounds. However, for the separation of very polar compounds, techniques like the Hydrophilic Interaction Liquid Chromatography (HILIC) are needed. HILIC has been established since years as an analytical tool, capable to separate effectively very polar molecules. Using a serial RPLC-HILIC system coupled with ToF-MS the analytical screening of samples comprised of solutes with variability in structure and polarity can be achieved. Full-spectrum acquisitions in non-target screening approaches are producing large datasets with the detected features of the samples. Different workflows have been published, proposing ways to cope with the collected amount of data in an automatic, time efficient and reproducible way, which can be applied to samples with various matrices. These workflows in a form of general steps can be summarized as: a) filtering and prioritizing the detected features (peak picking), b) molecular formula assignment, and c) a search in one or more compound databases. A relatively young compound database for water relevant compounds is STOFF-IDENT. In order to achieve a comprehensive identification of the water's organic content, Non-target screening strategies have become increasingly popular. This study was realized by analyzing river water samples with the established RPLC-HILIC-ToF/MS system and by using the STOFF-IDENT compound database. Its main aim is to demonstrate and discuss an efficient strategy for the non-target screening of aqueous environments, as a mean to facilitate the process of identification of very polar compounds.Three 24h composite samples were collected using time proportional samplers; upstream and downstream of a WWTP and wastewater effluent. The samples were analysed by the established

RPLC-HILIC-ToF/MS system. The analysis data were then processed following a non-target screening workflow for very polar compounds. After importing the data in STOFF-IDENT database, a proposed list of possible compounds in the samples was created. Using reference standards of the proposed compounds and MS/MS fragmentation data, it was possible to positively identify nine very polar compounds, of which six have not been reported previously in water surface samples.

MO281

Analysis of Per/Polyfluoroalkyl Substances (PFAS) in Drinking Water using LC/MS/MS to meet USEPA 537 requirements

T. Anumol, L. Toelgyesi, T. Sosienski, Agilent

Per/polyfluoroalkyl substances (PFASs) are organic molecules that have a C-F bonds in the alkyl chain and have a range of industrial uses and can be found in various household items and consumer goods. PFASs however can have adverse health effects while longer (C-chain >7) PFASs are bioaccumulative. PFASs are extremely persistent and have been detected in various environmental matrices. Consequently, the USEPA has public health guidelines in drinking water for two PFASs, namely perfluoroctane sulfonate (PFOS) and perfluorooctanoic acid (PFOA) at 70 ng/L. However, several other PFASs are also used in manufacturing and need to be monitored. To respond to this, a method was developed for the detection of PFAAs in drinking water using a newly developed triple-quadrupole mass spectrometer and a modified high-performance liquid chromatography system. The USEPA has developed a method for analysis in drinking water for 14 PFAS and this method expands on that method with lower detection limits, and more QA/QC data. The water samples were extracted with solid phase extraction using a novel weak anion exchange cartridge which was optimized to achieve good recoveries for all compounds and will be presented. Several different classes of PFASs including perfluorocarboxylic acids (PFCAs), perfluorosulfonic acids (PFSAs), sulfonamides (FOSA), sulfonamide acetic acids (FOSAAs) and others were separated on a liquid chromatograph (LC) using a reversed phase C-18 column. Since fluoropolymers are used in all LC systems, special precautions including replacing solvent lines and addition of a delay column were employed to avoid PFAS background contamination. The compounds were analyzed in negative electrospray ionization using a tandem quadrupole mass spectrometer in dynamic multiple reaction monitoring (DMRM) mode. Water samples were extracted using both an offline and automated online solid phase extraction techniques and the data was compared. All PFASs were analyzed and method performance parameters such as method detection limits, inter- and intra-day repeatability, matrix spike recoveries and other QA/QC criteria were evaluated. All recoveries were with 70-125% with %RSDs well below 15% that are needed to meet USEPA 537 requirements. Several PFASs were detected in a variety of groundwater, surface water and wastewater samples that were analyzed in the ng/L range. All this was done on a triple quadrupole mass spectrometer that is fully stackable with the HPLC system.

MO282

Optimisation of solid phase extraction parameters for the isolation and characterisation of benzodiazepines in wastewater

S. Nzube, Cape Peninsula University of Technology / Department of Chemistry; C. Van der Horst, University of The Western Cape / SensorLab Department of Chemistry; V.S. Somerset, CPUT / Chemistry

Pharmaceutical pollutants entering the aquatic environment have become a growing environmental concern. These pharmaceuticals are unique pollutants because of their special characteristics and behaviour that cannot be simulated with other organic pollutants. The untreated wastewater effluent that contains pharmaceuticals poses a considerable threat to the aquatic ecosystem because of the negative effects of non-target organisms in the water. Recent years have seen a growing concern about the benzodiazepines, as emerging pollutants, and their effects on the aquatic environment. These compounds are nowadays widely detected in sewage wastewater. It is important to increase the emphasis on the characteristics of the benzodiazepines in order to differentiate them from industrial chemical compounds. In this study, various solid phase extraction techniques have been employed focussing on isolation of benzodiazepines in wastewater matrices. Employing this methodology has shown improved detection and analysis of clonazepam and lorezapam as benzodiazepines. **Keywords:** Benzodiazepines; Emerging pollutants; Solid Phase Extraction; Spectroscopy; Wastewater

MO283

Monitoring source and drinking waters for Microcystins using online LC/MS/MS method.

J. Westrick, Wayne State University / Lumigen Instrument Center; D. Cardona, Thermo Fisher Scientific / Environmental Analysis

In 2015 the USEPA announced an age-dependent drinking water Health Advisory (dwHA) for the natural freshwater toxins, microcystins (MCs). For pre-school age children and adults, the MC dwHA values are 0.3 mg/L and 1.6 mg/L, respectively. Although the dwHA values are non-regulatory values, this announcement provides compelling health information that cannot be ignored. In parallel, EPA Method 544, a solid phase extraction/liquid chromatography tandem mass spectrometry (LC/MS/MS) method was released. Our goal was to create an online concentration LC/MS/MS method with 12 MCs that meets the EPA's quality assurance/quality control (QA/QC) criteria. MC concentrations were measured in samples from freshwater lakes and drinking water. Samples were prepared by three freeze/thaw cycles, centrifuging, and filtering through a 0.25 μm polycarbonate filter. Our LC/MS/MS platform included an online sample concentrator with UHPLC for separation and a triple quadrupole mass spec for MS/MS analysis. This method included 12 MCs with calibration curves from 0.5 – 500 ppt with R^2 values greater than 0.996. The MCs eluted between 2.2 – 5.2 minutes allowing for the analyses time to be 3] MC-RR, [Asp^3] MC- LR, MC-HilR, and MC-WR at concentrations above the low health reference level of 21 ng/L. Our data suggests that 1) by not including 12 MCs in Method 544, the true risk potential of exposure to MCs in drinking and recreational waters will be underestimated greatly, and 2) an untargeted microcystin occurrence study needs to be performed in the USA. Finally, our LC/MS/MS method reduces sample preparation, chemical usage, and instrument and preparation time while meeting EPA quality assurance criteria.

MO284

Development of a LC-MS/MS-based method for screening of non-targeted chemicals of potential concern in northern pike.

L. Tian, McGill University; J. Reinling, Université du Québec à Montréal / Departement des sciences biologiques; J. Verreault, Université du Québec à Montréal / Department of Biological Sciences; M. Houde, Environment and Climate Change Canada / Aquatic Contaminants Research Division; S. Bayen, McGill University / Singapore-Delft Water Alliance

Fish and seafood contaminant monitoring traditionally relies on the surveillance of known chemicals, an approach referred to as targeted analysis. However, very few tools are available to monitor "unexpected" or "unknown" compounds. Non-targeted approaches are particularly useful to detect emerging contaminants in items related to the human diet such as fish and seafood. The non-targeted approach is however quite challenging for trace contaminant analysis as it involves isolating relatively small signals in complex matrices, and this, in absence of a priori knowledge about the analyte identity. In the past decade, the coupling of liquid chromatography, high-resolution tandem mass spectrometry (HRMS/MS) and advanced data processing algorithms has proved to be a robust approach for the analysis of unknown molecules in biological samples. In this study, a non-targeted workflow was developed with the objective to detect/identify unexpected organic contaminants in a predator fish from the St. Lawrence River (QC, Canada), the northern pike (*Esox lucius*), with a focus on chemicals originating from plastic materials. An optimized method was applied to pike tissue sampled upstream and downstream of the Montreal's wastewater treatment plant. The two sampling sites (upstream vs downstream) were then compared using Mass Profiler Professional Software for the presence of other unexpected contaminants. The final confirmation of various substances of interest (e.g. PFOS) was investigated through the comparison with analytical standards. Results indicated that the non-targeted workflow optimized in this study can successfully identify unexpected chemical residues in fish matrices.

MO285

Prioritising site-specific emerging contaminants in surface water based on LC-HRMS nontarget screening data

M. Krauss, C. Hug, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; R. Bloch, Helmholtz Centre for Environmental Research GmbH / UFZ; T. Schulze, W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis

LC-HRMS allows for the non-targeted detection of chemicals in water samples. However, the majority of the several thousand compounds detected in a typical surface water sample remains unknown. Despite we can expect further progress in HRMS-based screening approaches and compound identification, it seems unlikely that monitoring efforts will ever exceed several hundreds of compounds due to financial and time restrictions. Thus, a prioritisation is necessary, guiding decisions on the selection of compounds to monitor and to study at specific sites. Here, we propose an approach to prioritize site-specific compounds solely from LC-HRMS data based on automatically retrieved information and a rarity score derived from signal intensity and frequency of occurrence. The approach was applied to a set of 31 samples from rivers and streams of different size and a different fraction of wastewater from the Saale and Mulde catchments in Germany. These were solid-phase extracted and analysed using LC-HRMS using an LTQ Orbitrap in ESI+ and ESI- mode. After peak picking using the MZmine 2 software, blank peaks were removed and isotopologue peaks, adduct peaks, and homologue series were detected using the R package "nontarget". Rarity scores were calculated for all detected peaks as ratio of maximum and median peak intensity across all samples divided by the ratio of the number of positive detections and the total number of samples. The distribution of rarity scores was similar for ESI+ and ESI- mode, with about 80% of the detected peaks (about 31,000 in ESI+ and 15,000 in ESI- mode) showing values between 10 and 100, while roughly about 1% of peaks had values above 1000 which might be considered as a threshold level for "rare", site-specific compounds in our dataset. The occurrence of these rare peaks at the individual sites differed considerably from 0 to 91 in ESI+ and 0 and 48 in ESI- mode. At two sites, the presence of a high number of rare peaks (48 in ESI- mode) coincided with the largest number of sulfur-containing compounds as indicated by isotopologue annotation. These sulfur-containing compounds could be identified as various derivatives of naphthalene sulfonic acids and have to be considered as a site-specific contaminants, as they were not present at any other sampling site. Thus, the proposed approach is suitable to rapidly characterize surface water samples and allows for a prioritization of sites or compound groups for further in-depth studies.

MO286

Analysis of Phenanthrene Transformation Products Using High-Resolution Mass Spectrometry Coupled to High-Performance Liquid Chromatography

M. Leonard, Oregon State University / Environmental & Molecular Toxicology; J. Schrlau, Oregon State University / Environmental and Molecular Toxicology; S.L. Massey Simonich, Oregon State University / Department of Environmental and Molecular Toxicology

Polycyclic aromatic hydrocarbons (PAHs) are environmental contaminants generated by the incomplete combustion of organic compounds. Several PAHs have been identified as toxic, mutagenic and/or carcinogenic, which has led the United States Environmental Protection Agency (US EPA) to list 16 PAHs as priority pollutants. PAHs can be metabolized by mammals and some microbes to form hydroxylated PAHs (OHPAHs) and a variety of other transformation products (TPs). These TPs have the potential to be more toxic than their parent PAHs, but they are not included on the EPA priority pollutant list. Hence, they are often not screened for in environmental samples. Non-targeted screening based on high-resolution mass spectrometry (HRMS) coupled to high-performance liquid chromatography (HPLC) has made it possible to identify unknown TPs in complex environmental samples. An HPLC-HRMS method was developed for analysis of phenanthrene TPs detected in bioremediated water. C_{18}, phenyl-hexyl, and fluoro-phenyl HPLC columns were evaluated for their ability to resolve hydroxyphenanthrene (OH-Phe) isomers. Baseline resolution of 2-, 4-, and 9-OH-Phe was achieved with the C_{18} and phenyl-hexyl columns using a gradient of water (mobile phase A) and a mixture of acetonitrile and methanol (mobile phase B). Gradient elution beginning with a relatively high percentage of organic solvent ($\geq 70\%$) yielded satisfactory separation and peak shape without the use of an additive or buffer. Detection was carried out with a high-resolution time-of-flight MS employing electrospray ionization (ESI) in negative ion mode. The high organic solvent composition of the eluent enabled optimal ESI performance. Consequently, spectrometric sensitivity was preserved throughout each analysis. Further investigation will determine whether the fluoro-phenyl column is suitable for separation of OH-Phe isomers. The method will be used for separation of phenanthrene metabolites and other PAH TPs in non-targeted screening of bioremediated mixtures.

MO287

Strategies to monitor transformation products in the water cycle

A. Brunner, D. Vughs, W. Siegers, C. Bertylkamp, R. Hofman-Caris, A. Kolkman, S. Kools, T. ter Laak, KWR Watercycle Research Institute

Transformation products (TPs) are formed in the water cycle through both biological and technological processes. Data available showed that some TPs can be more persistent and toxic than their mother compounds. Well-known examples are bromate and NDMA that generate toxic TPs after ozonation. Despite the TPs potentially increased toxicity compared to their parent compounds, transformation processes are not routinely monitored, and in particular those induced by drinking water treatment remain elusive. This lack of information is mainly due to the technical challenges in analyzing TPs, which are often unknown, polar compounds occurring in low concentrations. Candidate analysis methods are bioassays to assess potential effects or advanced chemical analysis to elucidate TPs, such as non-target high-resolution tandem mass spectrometry (HR MS/MS) methods combined with novel data analysis approaches. Here, we addressed the challenges of TP analysis and the scarcity of TP research concerning studies in drinking water in particular, building on the insights gained from previous work. In a recent project, we assessed the relevance of transformation products as specific for the drinking water sector through interviews with the concerned parties. Based on the sector's reported needs, we then performed a lab-scale pilot to monitor TP formation of the three organic micropollutants carbamazepine, clofibric acid and metolachlor during the rapid sand filtration and ozonation, two readily applied biotic and abiotic drinking water treatments, respectively. The experimental results show that degradation of the parent compounds and TP formation are treatment and compound specific. *In silico* TP prediction and literature mining significantly facilitate TP identification, yet a number of TPs remains structurally unidentified, and for the majority of identified TPs toxicological risk assessment is missing.

MO288

Application of high-resolution mass spectrometry to identifying chlorinated transformation products of aromatic emerging contaminants in wastewater

X. Lin, TUNGHAI University; W. Chen, J. Cheng, TUNGHAI University / Department of Environmental Science and Engineering

Chlorination could remove some aromatic emerging contaminants (ECs) in wastewater, but may also convert the ECs into unknown transformation products (TPs). This study developed a method to systemically identify the TPs of multiple aromatic ECs using high-resolution mass spectrometry (HR-MS) and traced the parent aromatic ECs of the TPs. We spiked ten aromatic ECs (5000 ng/L) into 100-mL Milli-Q water. The water was chlorinated at an initial chlorine of 0.7 mg/L for ten minutes. The full-scan mass chromatograms of both the chlorinated (n = 6) and the untreated (n = 6) water samples were acquired using ultra-performance liquid chromatography-quadrupole-time-of-flight mass spectrometry. By comparing the compound profiles, we evaluated the removals of aromatic ECs and discovered the signals of suspect TPs. We also characterized the molecular formulae of the TPs using database searching and isotope-pattern comparison. The parent aromatic ECs of the TPs were then traced back by spiking each aromatic EC to one 100-mL Milli-Q water. Eight of the aromatic ECs were partly removed by chlorination, where triclosan showed the highest removal (99.4%), followed by bisphenol A (72.5%). Nine of the features that were present in the chlorinated and absent in the untreated water samples were indicated as TPs. The results of database searching and isotope-pattern comparison showed that the molecular formulae of all of the nine TPs contain at least one chlorine. Each chlorinated TP was then successfully traced to one aromatic EC. The nine TPs were transformed from five aromatic ECs, including all of the four parabens and triclosan, by replacing one or two hydrogens with chlorine atoms. The HR-MS method successfully identified nine chlorinated TPs. The results of this study demonstrated that parabens and triclosan could be transformed into more persistent, bioaccumulative, and toxic chlorinated compounds. The proposed method will be applied to the systemic identification of TPs in real water samples containing multiple ECs.

MO289

Unravelling the potential of a partial nitritation/anammox biomass towards micropollutants biodegradation

E. KASSOTAKI, ICRA (Catalan Institute for Water Research) / Technologies and Evaluation; M. PIJUAN, Catalan Institute for Water Research ICRA / Technologies and Evaluation; L. Gusmaroli, Catalan Institute for Water Research ICRA; I. RODRIGUEZ-RODA, Universitat de Girona and ICRA / LEQUiA; A. Joss, Eawag Swiss Federal Institute of Aquatic Science and Technology; G. Buttiglieri, Catalan Institute for Water Research ICRA

In the past few years, anammox-based processes have attracted a lot of attention for their implementation at the mainstream line of wastewater treatment plants, due to the possibility of leading to energy autarky if combined with anaerobic digestion. However, little is known about the potential degradation of micropollutants by the microbial groups responsible of these processes and the few results available are inconclusive. This study aims to assess the degradation capability of biomass withdrawn from a partial nitritation/anaerobic ammonium oxidation (PN/A) pilot plant towards five pharmaceutically active compounds (ibuprofen, sulfamethoxazole, metoprolol, venlafaxine and carbamazepine). Batch experiments were performed under different conditions by selectively activating or inhibiting different microbial groups: i) regular PN/A operation, ii) aerobic (optimal for nitrifying bacteria), iii) aerobic conditions with allylthiourea (an inhibitor of ammonia oxidizing bacteria), iv) anoxic (optimal for anammox bacteria), v) aerobic with acetate (optimal for heterotrophic bacteria) and vi) anoxic with acetate (optimal for heterotrophic denitrifying bacteria). Ibuprofen was the most biodegradable compound, being significantly degraded under all conditions tested except heterotrophic denitrification. Sulfamethoxazole and metoprolol showed good percentages of removal under certain conditions (up to 70% and 62%, respectively), suggesting the specificity of different microbial groups towards the degradation of these compounds. Finally, carbamazepine and venlafaxine were hardly removed ($\leq 10\%$ in the majority of cases). Results demonstrate that the activation of different microbial groups in combination with alterating operational parameters can actually enhance the removal of some of the studied micropollutants.

MO290

Removal of pharmaceuticals in a biofilm reactor: effects of manipulating co-degradation by carbon feeding on system performance

L. Zhang, Aarhus University / Department of Bioscience; P. Carvalho, U.E. Bollmann, H. EI-taliawy, Aarhus University / Department of Environmental Science; H. Brix, Aarhus University / Department of Bioscience; K. Bester, Aarhus University / Department of Environmental Science

Pharmaceuticals are frequently detected in the effluent of municipal wastewater treatment plants as conventional activated sludge systems are unable to completely remove these compounds. Biofilm reactors are a promising biotechnology to remove pharmaceuticals from treated wastewater. However, it is currently unclear whether depleting the reactors from degradable carbon (enhancing the need of the microorganisms to go after difficult to degrade carbon) or increasing the load of organic carbon (assuming co-degradation) might be more favourable for removing pharmaceuticals. Therefore, in this study, we built up a saturated sand filter based biofilm reactor to investigate the effects of intermittent acetate feeding on the removal of indigenous pharmaceuticals from treated wastewater. Presently, the sand biofilter was operated at 12 h of hydraulic retention time (HRT). In order to prevent adaption of the species composition of the biofilm to the presence of acetate, the system was intermittently fed with influent without carbon addition or with carbon addition. Ten acetate concentration levels were tested in this study, 5, 10, 20, 30, 60, 90, 120, 150, 200 and 300 mg C/L. For each feeding condition (without or with the different carbon concentration), the system was continuously operated for 1.5 HRTs, after which four samples were taken over 6 hours. The results showed that with the different acetate additions, the effluent oxygen concentration decreased, reaching the lowest value of 0.98 mg/L at 300 mg C/L acetate addition. However, the oxygen levels in the effluent increased always to initial conditions (4.7 mg/L) in each starving phase between the feeding phases with acetate. The acetate addition resulted in three different compound dependent removal patterns considering the pharmaceuticals. Briefly, atenolol and iohexol removal was attributed to co-metabolism (enhanced with acetate). Metoprolol, iomeprol, diclofenac, propranolol and sulfamethizole removal were removed 1) at lower acetate concentrations by co-metabolic degradation dependent on aerobic turnover, and 2) at higher acetate concentrations limited by suboxic conditions. Moreover, sulfadiazine, sulfamethoxazole and trimethoprim were removed independently of oxygen and acetate concentration, which could be interpreted as catabolism. Biofilm reactors can be employed for polishing treated wastewater, and the addition of primary carbon source can enhance the bioreactor's performance.

MO291

Investigating inhibitory effect of anti-inflammatory pharmaceuticals on activated sludge

M. GREEN, E. Topuz, G. Yuksek, E. Ubay Çokgör, D. Okutman-Tas, Istanbul Technical University / Environmental Engineering

The consumption of pharmaceuticals increase annually due to a variety of reasons involving affordability, population ageing and population growth. Due to the obstacles in monitoring all these micropollutants regularly, focusing on priority mixtures and determining their toxicity can be practically convenient. Moreover, there is still need comprehensive research to understand the toxicity mechanism of pharmaceuticals as well as to advance the modelling approaches. A laboratory-scale fill&draw 10L aerobic reactor (sludge age of 5 days; @22°C) was initiated with sludge sample taken from a municipal wastewater treatment plant located in Istanbul. The culture was fed daily with a synthetic wastewater (ISO 8192) (600 mgCOD/L) and methanol (40 µl). To assess acute inhibitory effect of micropollutants respirometric assays were conducted with pharmaceutical mixture (PMx) as dissolved in MeOH (10, 50, 75µg/L of each; Naproxen, Diclofenac, Ketoprofen, Mefenamic Acid, Ibuprofen, Indomethacin). Modelling studies were performed using modified Activated Sludge Model No.1 and Aquasim2.0 software. Pharmaceuticals were quantified with LC-MS/MS . Culture amendment with 10µg/L PMx did not result in considerable change compared to control culture, but upon addition of a higher concentration (i.e., 75µg/L) there was an increase in the initial substrate consumption rate and decrease in the OUR curve. A removal efficiency in the range of 33-55% was observed for tested pharmaceuticals at the end of the respirometric assays. Measurements showed that pharmaceuticals were not accumulated in sludge phase (< 0.2%). Modelling studies reflected that maximum hydrolysis rate of slowly hydrolysable COD (k_{h2}) decreased from 0.84 to 0.72 1/day when the concentration of pharmaceuticals increased from 10 to 50µg/L. When the concentration of PMx increased from 10 to 50µg/L, an increase in the initial COD value of slowly hydrolysable COD was noticed. Furthermore, 75µg/L PMx resulted in differentiation in organic matter structure which caused a change in the maximum hydrolysis rate (k_{h1}) and hydrolysis half saturation constant (K_X) for readily hydrolysable COD (S_{H1}). The results of this study will help to clarify toxic effects of micropollutants on microbial systems as well as will provide valuable data for the discharge of these chemicals into the environment.This work is partially supported by TUBA-GEBIP Award of Turkish Academy of Sciences and BAGEP Award of Science Academy Society of Turkey.

MO292

Elimination of tramadol and methadone in model ozonation experiments: removal kinetics and identification of transformation products

P. Kostanjevečki, Rudjer Boskovic Institute; J. Ćurko, Faculty of Food Technology and Biotechnology; M. Matosic, Faculty for Food Technology and Biotechnology; M. Ahel, S. Terzic, Rudjer Boskovic Institute

Since the conventional wastewater treatment has proven to be ineffective for a number of pharmaceutical compounds, there is a high concern about their ever-increasing release into the aquatic environment. To mitigate this problem, advanced wastewater treatment technologies, such as advanced oxidation and membrane filtration, are often necessary to reduce the emissions to acceptable levels and/or to minimize the possible overall ecotoxicity of the effluents. Therefore, the aim of this work was to examine the removal of two opioid analgesics, tramadol and methadone, using ozonation. The experiments were performed in three different matrices, including pure water, phosphate buffer and secondary effluent from the Central wastewater treatment plant of the city of Zagreb. The removal rate of opioid analgesics was systematically studied as a function of ozone concentration, pH and matrix used to dissolve target compounds. The determination of the remaining concentration of selected compounds as well as identification of transformation products formed during the experiment were performed by ultra-performance liquid chromatography/quadrupole-time-of-flight mass spectrometry. The experiment showed that ozonation at an ozone dosage of 0.05 - 0.5 mg/L completely removed both opioid compounds in less than 5 min in pure water and phosphate buffer solution, providing that pH of the ozonation medium was higher than 7. The elimination of opioids was significantly slowed down at acidic conditions, which indicated the importance of the amino group deprotonation for an efficient reaction with ozone. Elimination of selected compounds in secondary effluent was much slower than in organic-free water matrices, reaching 91.1% and 99.1% in the time period of 10 minutes for tramadol and methadone, respectively. Reason for the lower elimination percentage is ozone depletion by reaction with other organic compounds present in the secondary effluent. The removal of parent compounds was associated with formation of two main transformation products characterized by *m*/*z* values of 250 and 280 for tramadol and 278 and 294 for methadone. The most abundant transformation products of tramadol and methadone were tentatively identified as tramadol N-oxide and EDDP, respectively.

MO293

Fate and transformation of persistent priority contaminants during potable water reuse: the challenge of producing safe water

C. Raimundo, UNICAMP / Institute of Chemistry; K.H. Cochran, B. Fryer, University of South Carolina; S. Kimura-Hara, University of Calgary; W. Abdelraheem, Y. Huang, University of Cincinnati; S.L. Coffin, University of California, Riverside / Environmental Toxicology; D. Schlenk, University of California-Riverside / Department of Environmental Sciences; D. Dionysiou, University of Cincinnati / Department of Biomedical, Chemical and Environmental Engineering (DBCEE); S.D. Richardson, University of South Carolina

Potable reuse of wastewater is becoming more common as populations increase and freshwater resources become more scarce. Producing safe drinking water from treated wastewater is challenging due to the presence of contaminants that are not completely removed in conventional wastewater treatment and due to transformation products that can be formed with advanced oxidation technologies (AOTs) that are used in potable reuse treatments. These contaminants can be harmful for human and ecological health. In 2013, two Science Advisory panels determined two lists of priority emerging contaminants (ECs) to be monitored in aquatic ecosystems and human potable water reuse. The ECs were determined based on toxicity, persistence through wastewater treatment, and environmental water concentrations. This project is investigating the removal and/or transformation of 21 of these priority ECs through UV-C/H_2O_2, microfiltration and reverse osmosis in three samplings from Full-Scale Advanced Wastewater Treatment Plant (Orange County GWRS) in order to make indirect potable reuse of wastewater safer. Ultra performance liquid chromatography-tandem mass spectrometry (UPLC-MS/MS) was used to quantify bisphenol A, ρ-nonylphenol, bis (2-ethylhexylphthlate), butylbenzyl phthlate, perfluorooctanoic acid (PFOA), perfluorooctane sulfonate (PFOS), diclofenac, ibuprofen, erythromycin, triclosan, 17α-ethinylestradiol, 17β-estradiol, estrone and chlorpyrifos. Gas chromatography mass spectrometry (GC-MS/MS) was used to quantify permethrin, galaxolide (HCCB), polybrominated diphenyl ether (PBDE)-47, polybrominated diphenyl ether (PBDE)-99, bifenthrin and N-nitrosodimethyl-amine (NDMA). Transformation products (TPs), disinfection by-products (DBPs), and unknown compounds are being identified via high resolution-time-of-flight GC(TOF)MS/MS and UPLC(Q-TOF)MS/MS. Some compounds were not removed even after UV treatment. Controlled laboratory chlorination/bromination reactions have been performed on some of the ECs in our list to mimic drinking water and wastewater disinfection, and many TPs and DBPs were identified, including chlorine- and bromine-containing by-products. Toxicity studies on these reacted samples were also done, and the results show that many of the TPs are more cytotoxic after being chlorinated/brominated. Mass spectra obtained from these identified TPs and DBPs are being added to a user library for use in determining TPs in our sampled waters.

MO295

Evaluation of a nano-adsorbent for the removal of metallic carcinogens from wastewater

C. Van der Horst, University of The Western Cape / SensorLab Department of Chemistry; B. Silwana, Durham university; M. Makombe, Cape Peninsula University of Technology / Chemistry; E. Iwuoha, University of The Western Cape / SensorLab Department of Chemistry; V.S. Somerset, CPUT / Chemistry

South Africa is experiencing the worst drought in recent history, with water becoming a scarce commodity. However, the supply of good quality water is becoming increasingly difficult in view of large-scale pollution caused by agricultural, domestic and industrial activities. Many technologies including coagulation, membrane process, dialysis, foam flotation, osmosis, photocatalytic degradation and biological methods have been employed for the removal of toxic pollutants from water and wastewater. These technologies are effective but have some disadvantages such as expensive equipment, high operational and maintenance, high energy requirements, generation of toxic residual metal sludge and incomplete metal removal. On the other hand, adsorption offers high efficiency, cost-effectiveness, easy handling and recovery of metals and other adsorbed species. Heavy metals are often found in wastewaters and the removal of these inorganic pollutants using bimetallic iron-based nanoparticles is still unclear. In this study, bimetallic iron-silver nanoparticles were chemically synthesized and impregnated with chitosan to form chitosan bimetallic iron-silver nanoparticles (CS/Fe-AgNPs) to remove heavy metals from wastewaters. In this study, chitosan iron-silver nanoparticles beads have been successfully prepared and its efficiency in the removal of Cd(II) under ambient temperatures has been evaluated. The removal rate of total Cd(II) from actual wastewater was 89.25%. Furthermore, the monolayer adsorption capacity of Cd(II) based on the Langmuir model was measured to be 90 mg/g. Results were satisfactory when employing the adsorbent for removal of Cd(II) from wastewater samples. **Keywords**: Adsorption, Bioavailability, Monitoring, Wastewater.

MO296

WATER JPI Project FRAME: A novel framework to assess and manage contaminants of emerging concern in indirect potable reuse

K. Jewell, N. Hermes, C. Dietrich, German Federal Institute of Hydrology, Bfg; J. Drewes, Technical University of Munich, TUM / Chair of Urban Water Systems Engineering; U. Hübner, J. Müller, Technical University of Munich, TUM; M. Pettenati, W. Kloppman, G. Picot-Colbeaux, French Geological Survey, BRGM; S. Valsecchi, F. Cappelli, Water Research Institute - Italian National Research Council IRSA-CNR; M. Rusconi, Water Research Institute IRSA-CNR / Water Research Institute; S. Polesello, Water Research Institute- CNR / Water Research Institute; G. Mascolo, Water Research Institute IRSA-CNR / Water Research Institute; S. Murgolo, Water Research Institute IRSA-CNR; M. Carere, Italian Institute of Health ISS; I. Lacchetti, Istituto Superiore di Sanità / Environment and Health; K. Thomas, Norwegian Institute for Water Research, NIVA / Product Metabolism; A.L. Macken, K. Petersen, S. Samanipour, Norwegian Institute for Water Research, NIVA; D. Pierre, Geohyd; A. Aurouet, Antea Group; T. Ternes, German Federal Institute of Hydrology

Indirect Potable Reuse (IPR) provides options to maintain sufficient water quantities for communities in the future. Efficient and affordable strategies for IPR are needed to minimize impacts from a broad range of contaminates of environmental concern (CECs) and to preserve ecosystem services and human health. The project FRAME (A novel framework to assess and manage contaminants of emerging concern in indirect potable reuse) is funded by the European research initiative "Water JPI (Joint Programming Initiative, Water Challenges for a Changing World). Principal project aims are: i) to evaluate treatment processes with combined analytical, toxicological and microbiological approaches; ii) to evaluate advanced treatment options in a multiple barrier approach to improve removal of CECs and inactivation of pathogens; iii) to integrate the experimental results in treatment process models and groundwater models to describe the fate of CECs; iiii) to provide a decision support tool for stakeholders, considering process performance and feasibility assessment for treatment scenarios. Advanced treatment options are applied in a multiple-barrier approach at laboratory and full-scale, specifically to improve the removal of CECs, inactivation of pathogens and improvement of other health-related parameters. Detailed fate studies are included to elucidate transformation pathways of CECs and to identify previously unknown transformation products (TPs) formed in biological processes. Multi-residue analysis methods were developed for sensitive analysis of 176 CECs, including 12 PFAS. The majority of quantitation limits are in the range of 0.5 ng/L to 50 ng/L. Sorption of charged CECs onto Fe-oxides and other minerals was simulated with the goal to create a sorption model for complex soil compositions. Treatment process models using kinetic modelling of CEC and pathogen removal were implemented—designed for integration into a decision support system for stakeholders. Results at laboratory and full-scale showed that a sequential biofilter approach at pilot scale shows higher efficiency than conventional single-stage biofilters; the monitoring of full-scale secondary effluent infiltration sites reveals attenuation of certain CECs, while others require further treatment, highlighting the need for a multi-barrier approach to IPR.

MO297

Evaluation of Rainwater collected from Concrete underground tank and other stroage tanks in Owerri Imo State, Nigeria

A. Okeke, University / Chemistry

ABSTRACT Due to the lack of public pipe borne water supply in Owerri municipality and its Environs in Imo State, South-Eastern Nigeria, many individuals have developed different techniques for rainwater storage-harvesting for drinking water and domestic use. Consequently, it is very important to evaluate the quality of rainwater harvested and stored in these different storage tanks so as to ascertain their impacts on rainwater quality. In this study, samples of harvested rainwater were collected from four different storage facilities commonly used by general populace in Owerri (Metal drum tank, concrete underground tank, PVC tank and coated basin for rainwater). The physiochemical and microbiological analysis of these rainwater samples were carried out using standard method. The trace metals in the water samples were relatively below the maximum permissible limit by WHO standard except for lead which was present at low concentration with the value (1.60±0.04mg/L) in metal drum tank based on the heavy metal content. For bacteriological analysis, the concrete underground tank recorded the presence of *pseudomonas* which exceeds the WHO standard stipulated for portable water. The results further explained that concrete underground tank and metal drum tank were more contaminated in terms of physiochemical and microbiological compositions. However, the study shows that harvested rainwater may not be suitable for direct drinking without treatment, but could be used for domestic purposes. **Keynote:** *Harvested Rainwater, microbiological analysis, physiochemical analysis, storage facilities, trace metals*

MO298

Sewage Epidemiology: Investigating the Impact of Phthalates on Human Health

C. Allen, L. Jones, Dublin City University / School of Biotechnology and DCU Water Institute; F. Regan, Dublin City University / Chemical Sciences; R.U. Halden, Arizona State University / Biodesign Center for Environmental Security; A. Staines, Dublin City University / School of Nursing and Human Sciences and DCU Water Institute; J. Lawler, Dublin City University / School of Biotechnology and DCU Water Institute

Phthalates are synthetic organic chemicals commonly used as plasticisers in polyvinylchloride and as additives in personal care products. Over 213M kg of phthalates are produced globally each year with end use products including food packaging, flooring, paints, tubing and medical devices. Due to their high production volume and continuous release, phthalates are emerging contaminants, ubiquitous in the environment. Research has shown that the widespread exposure to these chemicals has been associated with numerous adverse health effects including impaired reproductive health in males, decreased neurological development in children, cancer and obesity. As a result, some phthalates including DBP, BBP, and DEHP have been banned or limited in manufacturing (in particular for items such as children's toys). As new research emerges indicating that substitute plasticizers also contribute to adverse health effects, these restrictions are likely to increase. The present project constitutes the first application of sewage epidemiology to determine phthalate exposure in an Irish population. Phthalate levels in influent, effluent and sewage sludge (biosolids) are being monitored by GC-MS and LC-MS/MS analysis, tracking the cycle of phthalates throughout the wastewater system. Phthalate biomarkers are being analysed in influent to assess phthalate exposure. A meta-analysis on health risk data serves to relate the level of exposure to an associated risk, providing the first step in phthalate risk assessment within the Irish environment. Results will inform on the feasibility of using sewage biomarkers for future compliance monitoring. Metabolites from the following phthalates are considered for investigation: benzylbutylphthalate (BBP), dibutylphthalate (DBP), diethylhexylphthalate (DEHP), diisobutylphthalate (DIBP), di-n-octylphthalate (DNOP), diisononylphthalate (DINP), and diisodecylphthalate (DIDP). This study is part of a large-scale project representing an international collaboration between three research centres Dublin City University (DCU), Arizona State University (ASU), and the Norwegian Institute for Water Research (NIVA). With support from Irish utilities, the team is assessing the sources, environmental fates and human exposure profiles of priority phthalates in Ireland. Study results on some eleven priority phthalates will be leveraged to inform risk assessments and environmental policies concerning the phthalate safety and usage.

MO299

Phthalates and their metabolites in the environment

L. Jones, Dublin City University / Biotechnology and Chemical Sciences; C. Allen, Dublin City University / School of Biotechnology and DCU Water Institute; F. Regan, Dublin City University / Chemical Sciences; A. Staines, Dublin City University / School of Nursing and Human Sciences and DCU Water Institute; R.U. Halden, Arizona State University / Center for Environmental Health Engineering; J. Lawler, Dublin City University / School of Biotechnology and DCU Water Institute

Phthalates, or phthalate esters, are esters of phthalic acid, and their chemical structure consists of one benzene ring and two ester functional groups linked with two consecutive carbons on the ring. These compounds are stable, liquid in ambient temperature, while the ones of higher molar mass have low volatility and are slightly soluble in water. They are a group of synthetic organic chemicals that are used as additives, or plasticisers, to enhance the flexibility, transparency, stability, longevity, and durability of plastic materials and as non-plasticisers in consumer products. They are most commonly used as plasticisers in polyvinyl chloride (PVC) and phthalates have been used in the plastics industry for more than 80 years. As phthalates are so commonly used, their impact on the environment and human health has been extensively studied. Phthalates have been found to be recalcitrant, ubiquitous within the environment, and in many cases, detrimental to human and animal health. This project represents an important collaboration between three research centres (DCU, ASU, & NIVA) with support from Irish Water, Panda and Fingal Co. Co., to assess the potential sources and environmental fates of priority phthalates in Ireland. This project is supported by several by studies carried out in Ireland already (DCU) and a vast array of literature in the area of priority pollutant monitoring. The impact of such study would be the analysis of these eleven phthalates from source to fate, in order to inform environmental policy on the risks posed by phthalate usage. The phthalates included in this study are Benzylbutylphthalate (BBP), Dibutylphthalate (DBP), Dipentylphthalate (DPP), Diisopentylphthalate (DIPP), Diethylhexylphthalate (DEHP), Dihexylphthalate (DHP), Diisobutylphthalate (DIBP), Di-n-octylphthalate (DNOP), Diisononylphthalate (DINP), Diisodecylphthalate (DIDP) and Dimethylphthalate (DMP). A selection of phthalate monoesters have also been included in this study to evaluate human exposure to phthalates. Research into the human health effects of phthalates is far from complete, and while phthalates including DBP, BBP, and DEHP have been banned or limited in manufacturing (in particular for items such as children's toys), new research is emerging which indicates that substitute plasticizers have similar deleterious health effects. This research is timely as the extent of phthalate contamination within Ireland, and the impacts on human health, are unknown.

MO300

Poly- and perfluoroalkyl substances (PFASs) in the sewage system of the Bordeaux city: high contribution of unidentified precursors of perfluoroalkyl acids

C. Simonnet-Laprade, University of Bordeaux UMR EPOC; P. Labadie, UMR CNRS EPOC Universite Bordeaux / UMR 5805 EPOC; M. Capdeville, LyRE Centre de Recherche et Développement SUEZ; P. Pardon, UMR CNRS EPOC Universite Bordeaux / EPOC UMR 5805; H. Budzinski, University of Bordeaux

This study proposes to identify the origin of 30 poly- and perfluoroalkyl substances (PFASs) to the sewage system of a French city (Bordeaux Metropolis). For this purpose, 16 samples of domestic wastewaters, 10 samples of wastewaters impacted both by industrial and commercial activities were collected within the sewage network upstream typical and representative treatment plants; in addition 4 samples of runoff waters were also targeted in order to explore the input of this type of potential source. PFASs were also analyzed in the influents, the effluents, and the sludges of the 4 main wastewater treatment plants (WWTP) of Bordeaux Metropolis to quantify global inputs to the natural aquatic environment. The results highlight distinct patterns and levels of contamination between different types of samples and potential sources. Overall, wastewaters impacted by industrial inputs have the highest levels (ΣPFAS = 4.6-501.7 ng.L^{-1}) with the predominance of PFOS, PFHxS, C_4-C_8 PFCA and 6:2 FTSA. High levels of 8:2 and 10:2 FTSA (> 100 ng.L^{-1}) are found in wastewaters from the harbor area and a major industrial area. Domestic wastewaters have the highest levels of 6:2 diPAP (median concentration of 4.5 ng.L^{-1}), probably related to its use in food packaging. Concerning runoff waters, the highest levels are found in the airport area with $\sum$PFAS of 227 ng.L^{-1}. The comparison of the profiles with those found for WWTP influents using a principal component analysis made it possible to highlight the importance of the industrial and commercial discharges on the global contribution to WWTPs. It has been possible to quantifly global fluxes of PFASs to the four WWTPs and they were estimated at about 14 g.d^{-1} for the sum of targeted PFASs; concerning removal in WWTPs, only the C_{10}-C_{14} PFCAs, the PFOS and their precursors with more than 8 perfluorinated carbons were shown to be partially or totally removed by the treatment processes. In addition to the targeted analysis and in order to estimate the proportion of unidentified perfluoroalkyl acid precursors, the total oxidizable precursors (TOP) assay developed by Houtz and Sedlack (2012) was applied to each of the matrices. This method, which was applied for the first time to domestic wastewaters and to WWTP sludges, revealed the presence of unidentified precursors of C_4-C_{12} PFCA in all the matrices representing 32 % (in runoff water) to more than 90 % (in domestic wastewaters) of the total PFAS molar concentrations.

MO301

Antibiotics and endocrine disrupting compounds in wastewater treatment plants and in receiving water bodies around the city of Rome (Italy)

J. Rauseo, National Research Council / Institute of Water Research IRSA-CNR; F. Spataro, N. Ademollo, T. Pescatore, National Research Council of Italy / Water Research Institute; L. Patrolecco, Water Research Institute-National Research Council / Water Research Institute

Several studies highlighted the occurrence of organic micropollutants such as pharmaceuticals residuals, ingredients of personal care products and endocrine disrupting compounds (EDCs) in wastewaters and surface waters with concentrations ranging from ng l^{-1} to few mg l^{-1}. Among the emerging compounds, antibiotics are of peculiar interests due to their potential adverse effects on aquatic ecosystems diversity and function and because they can act as a potential driver for dissemination of resistance genes. Previous studies show that classes of synthetic antibiotics, such as quinolones, sulfonamides, tetracyclines, betalactams and macrolides, widely used in human and veterinary medicine, are rather resistant to microbial degradation, providing an indication as to why these compounds might persist within wastewater treatment plant (WWTP). Nonylphenol ethoxylates (NPEOs), commonly used as detergents, wetting agents, emulsifiers and dispersants, are precursor of nonylphenols (NPs) by the loss of the ethoxy groups. NPs are estrogenic because they can bind to estrogen receptors and can block or alter endogenous endocrine functions in various reproductive and developmental stages. In fact, NPs have been included in the European list of priority hazardous substances for surface waters in the Water Framework Directive. The aim of the present study was to investigate the occurrence and fate of selected classes of emerging and ED compounds in four WWTPs serving the city of Rome and in the receiving Tiber and Aniene rivers, in order to assess the possible risk for the aquatic ecosystem associated to their presence. Two sampling periods were planned to consider the different hydrological regimes of the receiving waters that affect contaminant dilution. The extraction-clean-up from aqueous samples was performed by solid-phase extraction (SPE) followed by an analytical determination with HPLC-MS/MS. The results showed the presence of selected antibiotics and NPs in influent-effluent from WWTPs around the city of Rome and in contaminated sites along the urban stretch of Tiber and Aniene rivers. The results confirmed that WWTPs were the main source of river contamination. Although the effluent wastewater input into receiving water should produce a dilution of contamination, the continuous release of these xenobiotics into the aquatic environment may lead to chronic exposure of organisms at all levels of the food chain.

MO302

Mass flows of antimicrobial compounds in Swedish sewage treatment plants

M. Östman, J. Fick, M. Tysklind, Umea University / Department of Chemistry

Antimicrobial biocides are used as disinfectants, antiseptics and preservatives to prevent unwanted microorganisms. In the same manner as antibiotics, they are entering our sewage system and passing on to the sewage treatment plants. Sewage treatment plants has been suggested as a possible high-risk environment when it comes to development of antibiotic resistant bacteria. Concerns has been raised that biocides might promote antibiotic resistance via co- and cross-resistance mechanisms. It is therefore important to quantify loads and understand the fate of these compounds within the sewage treatment plants to be able to design and construct more efficient future plants, as well as an aid in risk assessment of these chemicals. The aim of this study was therefore to determine detailed mass flows of eleven antimicrobial compounds within three Swedish sewage treatment plants. In total, the three plants were sampled for nine days for samples representing the major flows in respective plant i.e. incoming wastewater, treated effluent, water after the primary clarifier, primary sludge, surplus sludge, digested sludge. All samples were analysed by liquid chromatography tandem mass spectrometry (LC-MS/MS). The compounds included different quaternary ammonium compounds (QACs), such as 12-BAC, 14-BAC, DDMAC , CPC and CTAB as well as other compounds such as chlorhexidine, benzotriazole, ciprofloxacin and fluconazole. QACs and chlorhexidine were efficiently removed from the water phase (~99% reduction) but the majority remained in the digested sludge. The total yearly loads in the treated effluent in three studied plants was 29.4 kg and 2900 kg in the digested sludge. For more polar compounds such as trimethoprim and fluconazole, about half of the amount found in the incoming sewage water was found in the treated effluent. To our knowledge this is the first detailed mass balance study for CPC and chlorhexidine reported. The study provides not only new scientific understanding but also important knowledge to e.g. sewage treatment plant operators and law- and policy makers.

MO303

Herbicides and fungicides in watersheds of agricultural regions of Ontario

T. Sultana, Trent University / Environmental and Resource Studies; P.A. Helm, Ontario Ministry of Environment and Climate Change / Environmental Monitoring and Reporting Branch; C.D. Metcalfe, Trent University / Water Quality Centre

Herbicides and fungicides are widely used in agriculture to control weeds and fungal diseases that can reduce crop yields. There is potential for these compounds to be transported from treated fields into surface waters via agricultural runoff. The objective of this project was to evaluate the distribution of selected current-use fungicides and herbicides in 5 major rivers and 13 smaller streams within regions of intense agriculture in southern Ontario, Canada. The Polar Organic Chemical Integrative Sampler (POCIS) was selected as a principal monitoring technique, although grab samples of surface waters were also collected throughout the POCIS deployment periods. The sampling rates (Rs_{cal}) for each target compound were determined in the laboratory with synthetic water over 14 days at 15^{0}C. The sampling rates were adjusted for the influence of environmental factors (e.g. temperature, flow) by measuring the loss of Performance Reference Compounds (PRCs) spiked into POCIS deployed in the field. Extracts from POCIS and grab samples were analyzed by liquid chromatography with tandem mass spectrometry (LC-MS/MS) using an AB Sciex QTrap 5500 instrument with electrospray ionization coupled with an Agilent 1100 HPLC. Among the six herbicide target compounds, the highest maximum concentrations were observed for atrazine (1,070 ng/L), dicamba (845 ng/L) and 2,4-D (691 ng/L). The highest maximum concentrations of fungicides were for azoxystrobin (959 ng/L), myclobutanil (86 ng/L) and boscalid (74 ng/L). The rest of the fungicides and herbicides were detected at concentrations below 60 ng/L. There was no correlation between the watersheds that had the highest levels of fungicides vs the highest levels of herbicides. This may reflect differences in crops grown across the region, or differences in the timing of application of the pesticides. Overall, this study indicated that selected current-use fungicides and herbicides are widely distributed at ng/L concentrations in agricultural watersheds in Ontario, Canada.

MO305

A Study on the Distribution and Behavior of Nonylphenol in the Suyeong River, Korea

D. Kim, Pukyong National University / Department of Ecological Engineering; S. Kim, National Institute of Fisheries Science; K. Roh, Pukyong National University / Department of Environmental Engineering; Y. Kim, Pukyong National University / Department of Food science and Technology; Y. Chung, Pukyong National University / Department of Ecological Engineering

Nonylphenol is known, as one of Endocrine Disrupting Chemicals is the degradation product of Nonylphenol ethoxylates being used as nonionic surfactant. Nonylphenol is classified as an endocrine disrupter capable of interfering with the hormonal system of numerous organisms. In order to understand the current contamination and behavioral characteristics of Nonylphenol by measuring the concentration of Nonylphenol in the surface water in the downstream of Suyeong

River of Korea, and based on that, estimating the material balance. During the survey period, the range of Nonylphenol concentration in estuary of Suyeong River had the range of 142.0 ~ 569.6 ng/L and the average of 271.0 ng/L. The target area was divided into 3 regions to estimate the material balance of Nonylphenol in the downstream of Suyeong River. The dissolved Nonylphenol of 282.3 g/day occurred in region 1 of Suyeong River, and Nonylphenol influx load occurred in particulate suspended solids of 1,582.8 g/day. The dissolved Nonylphenol outflow discharge toward the region 2 was 192.5 g/day, while the adsorption to the particulate suspended solids was 89.8 g/day. In the case of Nonylphenol within the particulate suspended solids, the outflow to the region 2 was 1,250.0 g/day and the amount of settling toward the sediment was estimated to be 422.7 g/day. The adsorption from the dissolved Nonylphenol to the particulate suspended solids in the region 1 and region 2 was 31.8% and 54.9%, respectively. In the region 3, the desorption rate was 8.8%.

MO306

Drugs of abuse distribuiton in Turia River based on geographic information and ecotoxicological assessment

M. Andrés Costa, Universitat de Valencia / Environmental and Food Safety Research Group; J. Pascual-Aguilar, V. Andreu, CIDE CSIC UV GV; Y. Pico, University of Valencia / Medicine Preventive

The aim of this study establishes the influence and the effects of the human pressure in a typical Mediterranean River Basin to determine accurately the point sources of contamination through an environmental forensics methodology. A total of 42 drugs of abuse and metabolites were analysed in surface water samples collected from 22 sampling sites in 2012 and 31 in 2013 distributed along the river. Analysis of the target illicit drugs was performed using solid phase extraction and liquid chromatography coupled mass spectrometry (SPE-LC-MS/MS). To determine spatial incidence of drugs of abuse, analytical results of target compounds were georeferenced and integrated into a geographical information systems (GIS). Ecotoxicological risk of drugs of abuse detected in the Turia River was evaluated by calculating risk quotient (RQ). In 2012, 3,4-methylenedioxymethamphetamine (MDMA) and 4-methoxyphencyclidine (4-MeO-PCP) were detected in one sampling point at a concentration of 22.8 and 37.6 ng/L, respectively. In 2013, 4-MeO-PCP was detected in a different sampling point of 2012 at a concentration of 7.55 ng/L and ecgonine methyl ester (ECME) was detected at a concentration of 15.03 ng/L. Bufotenine (BUF), methadone (MET) and p-methoxyamphetamine (PMA) were found out in 3 or 4 sampling points at concentrations < 70 ng/L in 2012. Ephedrine (EPH) and codeine (COD) were detected in 3 sampling points at average concentrations of 11.6 ng/L for EPH and 91.3 ng/L for COD in 2013. The compound detected more frequently along the river was benzoylecgonine (BECG), a cocaine metabolite, with an average concentration of 25.4 (2.91–76.8) ng/L in 2012. In 2013, MDMA was detected in 5 sampling points (mean of 4.67 ng/L, ranged from 2.34 to 7.21 ng/L) and BECG and MET were detected in a total of 8 and 7 sampling points, respectively, each one at a mean concentration of 14.02 (1.83–12.7) ng/L for BECG and 11.4 (2.29–40.1) ng/L for MET. GIS provided the spatial incidence of drugs of abuse along the Turia River Basin. The occurrence of these drugs is higher near of the cities with highest population densities according to the descriptive model of territorial presence. Compounds used as drugs of abuse and prescribed as pharmaceuticals (MET, COD and EPH) were mostly detected in Valencia city and its metropolitan area where most hospitals are located. Although risk assessment showed low ecotoxicological hazard, further studies are also needed in order to assess long term toxicity.

MO308

Occurrence, fate and environmental risk assessment of benzophenone-type UV filters in a tropical urban watershed

K. Gin, National University of Singapore / Civil & Environmental Engineering; F. Mao, National University of Singapore / Civil and Environmental Engineering; L. You, National University of Singapore; M. Reinhard, Stanford University; Y. He, Shanghai Jiao Tong University

A variety of benzophenone compounds (BPs) have been used as ultraviolet (UV) light absorbers in personal care products and synthetic products that are exposed to sunlight. Following use, BPs can enter ambient environments directly via recreational activities and indirectly from sewage discharges. Numerous studies have been conducted to detecte the occurrence of BPs in the aquatic environments. However, the occurrence and fate of BPs in tropical waters remains poorly understand. In this study, occurrence and fate of seven BP-type UV filters (i.e., 2,4-dihydroxybenzophenone (BP-1), 2,2',4,4'-tetrahydroxybenzophenone (BP-2), 2-hydroxy-4-methoxybenzophenone (BP-3), 2,2'-Dihydroxy-4,4'-dimethoxybenzophenone (BP-6), 2,2'-dihydroxy-4-methoxy-benzophenone (BP-8), 4-hydroxybenzophenone (4OH-BP) and 4,4'-dihyroxybenzophenone (4DHB)) were investigated in a tropical urban watershed consisting of five major tributaries that discharge into a well-managed water body. The BPs concentrations were measured in four compartments, i.e., bulk water, suspended solids, pore water and sediments. Results showed that benzophenone concentrations varied from widely < LOQ to 122.6 ng L^{-1} in dissolved phase and < LOQ to 2774 ng L^{-1} in solid phases. Suspended solids in the water column contained significantly higher amount of BPs than sediments, while the concentration difference between bulk water and pore water was insignificant. Further study will evaluate the vertical concentration profile in the aqueous phases and in the solid phases. The concentration ratio of BP-1 to BP-3 will also be addressed, aiming at assessing the degradation of BP-3 in the field. This will be followed by a preliminary risk assessment.

MO309

Formation of disinfection byproducts throughout various drinking water treatment processes

C. Postigo, IDAEA, CID-CSIC / Environmental Chemistry; P. Emiliano, ATLL Concessionària de la Generalitat de Catalunya, SA; D. Barcelo, IIQAB-CSIC / Department of Environmental Chemistry; F. Valero, ATLL Concessionària de la Generalitat de Catalunya, SA

This work investigates the formation of disinfection byproducts (DBPs) throughout the treatment processes operating in the various drinking water treatment and desalination plants and distribution system that supply drinking water to more than 4.5 M inhabitants living in the city of Barcelona and its metropolitan area. For this, DBP formation potential tests with chlorine were performed with the water entering each plant and produced after the individual processes carried out in each plant to treat the water. DBP mixtures were generated for each investigated matrix at two different temperatures (15ºC and 25ºC) and reaction times (0, 24 h, and 48 h, and also 72h in the plant effluent) so that different scenarios in the drinking water distribution network were simulated. Six different DBP classes in total were investigated in the DBP mixtures generated by means of gas chromatography coupled to mass spectrometry detection. The list included the regulated trihalomethanes (4THMs), and the non-regulated (at European level) iodinated trihalomethanes (I-THMs), trihalogenated haloacetaldehydes (THALs), halogenated acetonitriles (HANs), halogenated acetamides (HACMs) and haloacetic acids (HAAs). Overall it could be concluded that the potential of the water entering the plants to form all investigated DBPs decreased throughout the treatment process, due to the removal of DBP precursors in the different treatment steps. The work performed contributed to evaluate the risk associated with changes in the water treatment process and prevent population exposure to DBPs in the event of scenarios that may alter the good performance of the whole process.
Acknowledgments: C.P. acknowledges support from the Secretary for Universities and Research of the Ministry of Economy and Knowledge of the Government of Catalonia and the COFUND Programme of the Marie Curie Actions of the EU's FP7 (2014 BP_B00064). This work has been financially supported by the Government of Catalonia (Consolidated Research Groups 2014 SGR 418-Water and Soil Quality Unit and 2014 SGR 291-ICRA).

MO310

Formation of N-nitrosodimethylamine during water treatment for potable use: an update

B.G. Slencu, University of Medicine and Pharmacy Grigore T. Popa Iasi / School of Pharmacy; L. Avasîlcăi, I.D. Morariu, Grigore T Popa University of Medicine and Pharmacy of Iasi / School of Pharmacy

Nitrosamines can form in water in specific conditions. N-nitrosodimethylamine (NDMA) could be carcinogenic at levels as low as 0.7 ng/L of drinking water. NDMA is the most widely detected N-nitrosamine in drinking water which was subjected to chloramination. NDMA is also found in water subjected to disinfection with chlorine, hypochlorite or other chlorine-generating compounds, but in lower amounts. NDMA can also form through ozonization or nitrosation. Multiple mechanisms can be involved in NDMA formation: a) reaction between monochloramine or dichloramine and organic amine precursors; b) ozonization of typical secondary amine precursors; c) chlorination of nitrite in the presence of nitrosamine precursors; d) catalytic formation on activated carbon, from secondary amines; e) UV or sunlight photolysis of nitrite, in the presence of secondary amines. Many organic nitrogenous substances can be NDMA precursors (pharmaceuticals, substances used in cosmetics, pesticides, chelating agents, amine-based polymers, etc), but not all can be present in significant amounts in the source water. In the context of water treatment for potable use, amine-containing coagulation polymers and some anion exchange resins constitute the main source of NDMA precursors. Although NDMA is the most prevalent nitrosamine detected, according to the surveys conducted until now, it may account for only a minor fraction of all nitrosamines formed during chloramination. More research is required in order to establish ways to avoid NDMA and other nitrosamines formation.

MO311

Presence and environmental hazard of psychoactive pharmaceutical compounds in coastal waters and biota from North-Western Spain.

J. Frenandez, Rey Juan Carlos University; J. Rodriguez Gil, University of Ottawa / Department of Biology; N. Mastroianni, Institute of Environmental Assessment and Water Research IDAEA-CSIC / Department of Environmental Chemistry; C. Postigo, IDAEA, CID-CSIC / Environmental Chemistry; M. López de Alda, Institute of Environmental Assessment and Water Research IDAEA-CSIC / Department of Environmental Chemistry; B. Huerta, Catalan Institute for Water Research ICRA / Water Quality; S. Rodriguez-Mozaz, Institute for Water Research (ICRA) / Water Quality; M.E. Valdés, Catalan Institute for Water Research ICRA / Water Quality; D. Barcelo, IIQAB-CSIC / Department of Environmental Chemistry; Y. Valcárcel, Research and Teaching Group in Environmental

Toxicology and Risk Assessment TAyER / Rey Juan Carlos University
Historically, coastal and transition waters have preferentially been used for human settlement, leading to a continuous input of wastewater effluents into these habitats. Water quality monitoring of these systems, with the goal of improving their protection under the water framework directive is highly needed. The number of studies characterizing the presence of psychoactive pharmaceuticals (PAs) in coastal waters and marine biota in Spain is particularly limited. Our work represents the first attempt at monitoring these compounds in the Rias Baixas area (North Western Spain). This area was chosen as the location for this study due to its economic and ecological importance as a national and global leader in shellfish (especially mussels) production. In this study, the presence of 16 PAs (benzodiazepines and anxiolytics) was studied in samples of sea water and tissue from 7 economically valuable and highly consumed marine species. Samples were collected from 22 sites along the three main Rias Baixas. Fourteen out of the 16 assessed substances were detected in the water samples with venlafaxine (64%) and citalopram (41%) showing the highest detection frequencies. The highest concentrations in water were also measured for venlafaxine (291 ng/L), followed by lorazepam (95.90 ng/L) and citalopram (92.50 ng/L). Only 3 PAs (alprazolam, citalopram and venlafaxine) were present in the collected biota samples (razor clam, clam and octopus) in concentrations up to 14 ng/g d.w. (citalopram), 3 ng/g d.w. (venlafaxine) and 0.31 ng /g d.w. (alprazolam). Hazard quotients (HQ), calculated from the measured concentrations in water and available chronic aquatic toxicity data resulted in values higher than 1 (indicating elevated hazard and possible risk) for venlafaxine, citalopram, and sertraline. The venlafaxine concentrations resulted in the largest HQ values (up to 128). A human health risk assessment based on the measured tissue concentrations is underway. Our results confirm the presence of PAs in coastal waters of the Rias Baixas area in concentrations potentially able to cause chronic effects in exposed organisms, as well as the presence of these compounds in biota inhabiting the area. Based on the results obtained further monitoring of venlafaxine and citalopram in coastal waters is recommended. This work was possible thanks to the Spanish Ministry of Economy and Competitiveness FIS (PI14/00516) and the European Regional Development Fund (ERDF).

MO312

Determination of glyphosate and AMPA in fish bile from the Marne River, France

H. Blanchoud, EPHE UMR 7619; T. Ferreux, F. Alliot, EPHE / UMR Metis; A. Goutte, UMR METIS EPHE

Glyphosate is a widely used herbicide. In France, it is used during intercultural operations and to control weeds in non-agricultural areas. Glyphosate and AMPA (its main degradation product) can reach high concentrations in rivers, especially in the Seine basin including the Paris suburb (France). Although it is not bioaccumulative, the associated use of surfactants in pesticide formulations allows better assimilation in biological tissues. The aim of our study was first to develop a method to assess glyphosate and AMPA levels in biological tissues and then to determine the contamination of an freshwater fish, the European chub (*Squalius cephalus*) by glyphosate and AMPA. Fish were fished in the Marne River (a tributary of the Seine River situated in the East part of Paris) at 4 sites characteristic of agricultural and urban areas. Water was also sampled for analysis to compare sites contamination. Bile is an ideal material to identify metabolites of pollutants. This biological fluid was taken directly from the gallbladder with a syringe on freshly euthanized chub and frozen for further analysis. Then, 100µL of bile was taken and 500µL of internal standard (13C-Glyphosate and 13C-AMPA) were added before extraction with milliQ water (5mL) and ultrasonic method for 30 minutes. The extract was then derivatizated (FMOC-Cl) and concentrated on SPE OASIS HLB cartridge (60cc) before LC MS MS analysis. Preliminary tests were performed to establish and validate the protocol and to find the lowest limit of quantification and the best reproducibility. Results showed that glyphosate is detected in a fish sample coming from the most contaminated site by AMPA. This suggests that glyphosate is assimilated in fish and is still detectable after glyphosate has been degraded to AMPA in the water river. Glyphosate content in fish could be an indicator of environmental contamination. Further developments are needed to validate the protocol and complete the study with other organs than bile.

MO313

From source to food: following emerging pollutants

A. Garduno, The University of Nottingham; S. Pathasarathy, The University of Nottingham / Faculty of Engineering; J. Duran-Alvarez, Universidad Nacional Autonoma de Mexico / CCADET; C. Ortori, D. Barrett, The University of Nottingham / Faculty of Science; T.P. Dodsworth, The University of Nottingham / Biosciences; R.L. Gomes, The University of Nottingham / Faculty of Engineering

The current global population growth is putting an increasing strain on the world's natural resources. Water is no excpetion; the current situation brings a surge in the demand of water for drinking, sanitation, municipal, industrual and agriultural uses. At the same time the amount of wastewater generated is unprecedented. Given these facts, it makes sense to look for ways in which to adequatley resue wastewater therby reducing freshwater demand. One such solution is the reuse of wastewater for agricultural irrigation. Benefits of this practice include the reduction in fertilizer use due to the high nutrient content of wastewater, the enviornmental benefits of reusing an unwanted resouce and the economic advantage for farmers who have to pay little or nothing to use the resource . Furthermore it has been shown that wastewater pollutant load can be reduced as it goes through the environment through processes such as photolysis, biodegradation and adsorption . Using these natural processes to our advantage can reduce the costs of treating wastewater. However it has been shown that treated and untreated wastewater contain emerging pollutants (e.g. pharmaceuticals, personal care products, antibiotics, hormones, etc). When reusing wastewater for irrigation we are creating a pathway for these pollutants to enter the environment and possibly the human and animal food chains. Therefore to adequatley assess this practice it is necessary to have a clear understanding of the presence, fate and prevalence of emerging pollutants from source (irrigation water), through soil and finally in plant tissue. Therefore this paper presents a robust method to analyse the relevant environmental matrices (i.e. water, soil and plants). This method will help determine the risk posed to humans, animals and the environment when resusing wastewater for irrigation. Furthermore it will aid in evaluating the natural attenuation of emerging pollutants in the agricultural environment and therefore inform on the level of treatment necessary to undertake this practice sustainbly.

MO314

Psycoactive compounds in mussels: analytical method development and occurrence assessment

E.L. Garcia, IDAEA-CSIC / Department for Environmental Chemistry; C. Postigo, IDAEA, CID-CSIC / Environmental Chemistry; M. López de Alda, Institute of Environmental Assessment and Water Research IDAEA-CSIC / Department of Environmental Chemistry

It is well stablished that wastewater treatment plant effluents release pharmaceutical compounds to the aquatic environment impairig water quality. The environmental presence of these compounds may cause negative effects in the exposed aquatic organisms. In order to evaluate the ecological risk that they may pose, it is relevant not only to evaluate the concentrations at which organisms are usually expose but also their bioaccumulation potential. In this context, the objective of this work was to investigate the occurrence of different classes of psycoactive substances and metabolites in mussel tissues, and to assess the bioaccumulation potential of these compounds in these organisms. To this end, an analytical method based on a "quick, easy, cheap, effective, rugged, and safe" (QuEChERS) extraction and subsequent determination by means of liquid chromatography–electrospray–tandem mass spectrometry (LC–ESI-MS/MS) was developed and validated for analysis of over 40 psychoactive compounds and metabolites, including various illicit drugs (opioids, amphetamine-type stimulants, cocainics, cannabinoids, and hallucinogens) and therapeutic drugs (anxiolytics, antidepressants, sedative/antihistaminics and stimulants) in mussels. This relatively fast and simple methodology allowed the quantification of most of the target analytes at the low ng/mL level. Poor analyte absolute recoveries, which could be attributed to ionization suppresion effects by matrix components, were obtained especially for cannabinoids. However, analyte losses and matrix effects are satisfactorily compensated by the use of deuterated analogues as surrogate standards in the analytical process. The application of the method to different specimens coming from diverse areas purchased in local markets has shown little accumulation of these substances in the mussels analysed. This work was possible thanks to the Government of Catalonia (2014 SGR 418), the Spanish Ministry of Economy and Competitiveness FIS (PI14/00516) and the European Regional Development Fund (ERDF), and Bekolut and Merck for their gift of QUeCHERS and LC columns, respectively.

New Horizons in Particulate Polymer Analysis: Micro- and Nanoplastics and Tire Rubber Detection, Characterisation and Impacts in the Environment (P)

MO315

MPHunter: a dedicated software for µFTIR-Imaging Microplastic data analysis. First development steps and future perspectives

A. Vianello, Aalborg University / Civil Engineering Department - Section of Water and Environment; S. Primpke, G. Gerdts, Alfred Wegener Institute / Shelf Sea System Ecology; M. Simon, N. van Alst, K.B. Olesen, F. Liu, J. Vollertsen, Aalborg University / Civil Engineering Department

Imaging-Micro-Fourier Transform Infrared Spectroscopy (Imaging-µFTIR) applying a Focal Plane Array (FPA) is the most promising analytical tool for direct and rapid analysis of microplastics (MP) deposited on IR windows or collected on suitable membrane filters. The amount of data produced during an imaging analysis (many GB), combined with the lack of specific features addressed to MP identification and quantification in commercial FTIR software, makes the analysis of the IR map extremely time consuming and partially operator dependent. Although a novel automatic analysis pipeline has already been developed by Primpke et al. (2017), the spectral identification is still performed using a commercial FTIR software, limiting the use of the pipeline to the FTIR software's owners. Here we present a dedicated software (MPHunter) for MP analysis which can export, convert and manage datasets from the two FPA-µFTIR Imaging suppliers. The software, which can manage several million single spectra and many thousands of reference spectra in one run, is written in RAD Studio (Embarcadero Delphi IDE), an object-oriented programming environment which allows easy construction of user-friendly interfaces. As an example, an imaging dataset of 196 tiles from a 128x128 pixel FPA detector (totaling 3.2 million individual spectra) can be easily managed using the software's features. The software calculates the Pearson's correlation coefficient between the unknown spectra and a reference spectral library for the raw spectrum and its first and second derivatives. The reference spectra can be easily created and uploaded to the software as a .csv file. The calculation time for comparing 3.2 million spectra to a library of 150 spectra is around 6 hours on a standard laptop. Software features include conversion from %Transmittance to Absorbance and vice versa, selection of multiple customizable spectral ranges/whole spectral range for correlation and filters for residual noise removal. The correlation results can be further refined to define particles boundaries. Potential MP can then be marked, measured (main axes, area) and saved. MPHunter is open source freeware. It allows a semi-automated MP identification and quantification, decreasing the time demand for the interpretation of FTIR-imaging data and increasing the data accuracy. Further improvements are ongoing to interface MPHunter to the Analysis Pipeline from Primpke et al. (2017) and use it as the searching engine.

MO316

From alpine regions to dense populated areas: A comparison of microplastic contamination between 15 rivers across Germany

M. Loeder, I. Schrank, H. Imhof, University of Bayreuth; M. Hess, LANUV NRW / Water management, water protection; P. Diehl, State Environment Agency Rhineland-Palatinate; J. Mayer, Hessian Agency for Nature Conservation, Environment and Geology; H. Rahm, North Rhine-Westphalia State Agency for Nature, Environment and Consumer Protection; W. Reifenhäuser, Bavarian Environment Agency, Wielenbach; J. Stark, State Institute for Environment, Measurements and Nature Conservation Baden-Württemberg; J. Schwaiger, Bavarian Environment Agency / Aquatic Toxicology and Pathology; C. Laforsch, University of Bayreuth

Among marine litter, plastic waste is of growing concern, as nowadays it has become ubiquitous in the oceans. A large portion of the plastic waste is produced onshore and then enters the marine environment via water discharge to the river systems. Although, the oceans are considered as the main sink of plastic debris, recent studies also reported on the contamination of freshwater ecosystems with microplastics. Therefore, freshwater ecosystems do not only act as a source of plastic particles for the oceans, they also act, at least temporarily, as a sink. This may come along with all the associated harmful consequences that have been reported previously for marine ecosystems. Nevertheless, there is a considerable gap of knowledge about the impact and contamination of freshwater ecosystems with plastic particles. The lack of harmonized methods for microplastics sampling and detection hamper the comparability of data on concentrations and the composition of synthetic polymers in the freshwater environment. We compared microplastic contamination down to 20µm between 15 rivers across Germany, by the use of a harmonized sampling, sample processing and sample analysis (FTIR) currently verified by the JPI Oceans project BASEMAN. Results show a high variability between rivers and due to the lower size fraction measured relatively high concentrations compared to the few studies conducted in surface waters of freshwater ecosystems so far. Further, our data may shed light on major pathways and sources of microplastics in freshwater ecosystems. (M. Loeder, I. Schrank and H. Imhof contributed equally to the work as first co-authors).

MO317

Analytical approach for the identification and quantification of microplastic particles in environment samples by particle analysis in combination with FTIR and Raman microscopy

D. Fischer, Leibniz-Institut f. Polymerforschung Dresden / Analytics; A. Kaeppler, J. Muche, K. Eichhorn, Leibniz Institute of Polymer Research Dresden; S. Oberbeckmann, Leibniz Institute of Baltic Sea Research Warnemuende; M. Labrenz, Leibniz Institute of Baltic Research Warnemuende

The detection of microplastic particles in an environment sample in the wide range from 1 µm to 5 mm nearly quantitatively in a reasonable time is a challenging mission. This task should start with well-defined sampling procedures and sampling locations. Next task is the sample preparation procedure to remove organic and inorganic parts in such a manner, that the microplastic particles will not be destroyed. A further important point is the subsequent loss free transport to an analytical lab and the use of blind samples from the sampling location through all steps until the lab. Starting from this point our poster describes the following procedures to identify and quantify the microplastic particles in environment samples. All operations and analyses are performed in dust-free rooms in flow boxes and all equipment is plastic-free. First procedure is a three-step vacuum filtration to divide the particles in four fractions, above 500 µm, from 500 to 50 µm, from 50 to 10 µm and below 10 µm. After filtration the particles of the different fractions are on silicon filters made from wafers which are IR transparent. Now ptical particle identification programs determine all particles in shape and dimension and store there coordinates for the subsequent measurements with FTIR and Raman microscopy. Both methods identify the microplastic particles on the filter by their chemical structure using spectral databases. This spectral search runs automatically. The databases include polymers, copolymers, fillers, inorganic and organic substances, paints and lacquers. The IR libraries use transmission, ATR and reflection spectra and the Raman libraries use spectra measured with 532, 633 and 785 nm lasers. Every particle is assigned about the spectrum to a substance. This can be a polymer or in case of a mixed spectrum a polymer with paint/filler or also not a polymer. The fractions above 500 µm and from 500 to 50 µm are measured mainly by FTIR and the fraction from 50 to 10 µm and some selected samples below 10 µm mainly by Raman. However, several samples of the fractions above 10 µm are measured also with both methods since a combination of both delivers complete microplastic particles analysis. Further parameters, as the integration time, the number of accumulations, the magnification of the objectives and the lateral resolution, which influence the results concerning measurement time and locating and identification of mainly smaller particles will be discussed.

MO318

Using pyrolysis GC-MS in combination with multivariate tools to identify and differentiate polymer type and weathering of microplastics

T. Størseth, L. Sørensen, K. Almaas, SINTEF Ocean / Environmental Technology; M.O. Hepsø, Norwegian University of Science and Technology; O. Brakstad, A. Booth, SINTEF Ocean / Environmental Technology

Pyrolysis gas chromatography coupled to mass spectrometry (pyGC-MS) is a promising tool for identifying and quantifying trace amounts of microplastic (MP) in environmental samples. For pristine plastic samples, it has been demonstrated that polymer type and additive chemicals can be elucidated from the obtained pyrograms and their underlying mass spectra. However, the approach requires manual interpretation of the data, which requires a high level of competence and is time-consuming. Pyrograms obtained from environmental samples are typically complicated by the presence of naturally occurring organic compounds and the presence of multiple polymer types. Furthermore, weathering processes such as oxidation and biodegradation may alter the chemical composition of the polymers, especially at the surface. In the current study, an automated method for MP classification was developed. Pyrograms with associated mass spectra (m/z range 50-600) were obtained for a range of the most common polymer types, as well as for polyethylene and polystyrene microplastic samples subjected to different types of simulated environmental weathering (UV, additive leaching, abrasion, biodegradation) in the laboratory. An untargeted analysis approach was first used to classify pristine and environmental MP samples. Multivariate tools were then applied to classify the samples based on the global pyGC-MS derived composition of the polymers, and to compare pristine materials with samples from the environment. The technique shows promise where manual techniques fail or have difficulty due to the lack of visual resolution of chromatographic peaks with important diagnostic mass spectral features.

MO319

Marine Microplastic: Production and characterisation of realistic test materials for studying ecosystem impacts

S. Kuehn, Wageningen Marine Research / Institute for Marine Resources Ecosystem Studies Wageningen UR; A. van Oyen, Plastic Partner GmbH; A. Meijboom, Wageningen Marine Research; J.A. Van Franeker, Wageningen Marine Research / Institute for Marine Resources Ecosystem Studies Wageningen UR; A. Booth, SINTEF Ocean / Environmental Technology

Reported studies investigating the possible effects of plastic litter on marine biota have almost exclusively utilised pristine plastic materials that are homogeneous in polymer type, size, shape and chemical composition. This is particularly the case for microplastics (marine litter < 5mm), as collecting samples of such material from the marine environment in quantities sufficient for use in laboratory impacts studies is simply not feasible. Crucially, weathered plastics collected from the marine environment show considerable physical and chemical differences to pristine and post-production consumer plastics. In the current study, we describe the preparation and characterisation of a more environmentally realistic marine litter-derived microplastic reference material (≤3 mm) for use in fate and effects studies. Weathered marine plastic litter (351 items) was collected from the coast of the island of Texel (The Netherlands) and carefully identified and categorised (fibre-based, packaging, foam, plastic boxes and jerry cans, bottles, gloves and miscellaneous plastic materials). Ropes, nets and sheeting comprised ~70% of the collected material, which contained 9 different polymer types. The macroplastic material was sub-sampled and subjected to a cryo-milling and sieving process to create the microplastic reference material. To confirm that the original macroplastic polymer distribution was mirrored in the generated microplastic sample, it was subjected to ATR-FTIR and differential scanning calorimetry analysis. Particle size distribution (PSD) of the microplastic, measured using laser diffraction and sieving, showed that 68% (by mass) of the particles were in the range between 0.5 and 2.0 mm. Particle number increased with decreasing particle size fraction. Scanning electron microscopy revealed a wide range of particle sizes and shapes reflecting the properties of the different polymers. ICP-MS and ICP-OES analyses revealed the presence of a broad range of metals and other elements (e.g. Al, Cr, Fe, Mg, Pb, S and Zn) associated with the final sample. Many of these represent common inorganic plastic additives used as colourants, fillers and stabilisers. The additive organic chemical profile of the microplastic mixture was also determined by GC-MS analysis following extraction by ethyl acetate and ultrasonication. A broad range of plasticisers, stabilisers, antioxidants and flame retardants were identified.

MO320

Optimization of the preparation of standards of high density polyethylene microplastics and quantification techniques by stereoscopic and confocal microscopy.

j. sanchez nieva, University; J. Perales, CACYTMAR University of Cadiz / Department of Environmental Technologies; E. Rojo-Nieto, Helmholtz centre for environmental research - UFZ / Department of Cell Toxicology; J. Gonzalez-Leal, University of Cadiz

A non-complex procedure has been developed for preparing HDPE microparticles as standard for microplastic determination in sediments. Always keeping environmentally relevant materials in mind, different bottle caps from several brands were studied in order to identify those that could provide a clear spectrum for HDPE using Raman spectroscopy (considering that Raman spectroscopy is sensitive to those additive and pigment chemicals in microplastics that interfere with the identification of polymer types). Red caps from a popular brand of mineral water were selected as the raw material as their spectrum was easily comparable with those provided in the literature for HDPE. The large pieces of plastics were converted into microplastics by using a conventional machining process, i.e., a drill with a sandpaper implement (Dremmel 300, 13 mm-60 grain size sandpaper). For this purpose, several sequential sessions of the machining processes were carried out in order to avoid changes in the physical properties of the plastics that would result from temperature increase due to due to prolonged friction. After the machining, the obtained HPDE particles were sieved using ethanol (96%) through two different mesh sizes until a final standard ranging from 0.1 to 0.850 mm size was achieved. This range was selected as it is relevant in biota ingestion risk as well as is appropriate for visual counting using microscopes. The standard HPDE microplastics used in all the extraction experiments were the same. It was thoroughly mixed previously to spiking in order to avoid differences in the distribution of sizes. To prepare standard dispersions, HPDE microplastics were suspended in ethanol 96% and shook using a magnetic stirrer. An effective method for determining the particle distribution of microplastics is microscopy. In this work, two types of quantification using microscopy were used and compared: optical microscopy by visual sorting (Leica ICC50 HD, 4x lens, using a mesh for counting with sections of 3x3 mm prepared for this study) and, confocal microscopy (Zeta Instruments, model Zeta 300). The last one, included object detection algorithms (Mathematica 10) which not only allows quantification of plastic particles but also their classification into size groups.

MO321

First Report of Microplastics in Pacific-side Arctic Ocean

H. Lee, S. Kim, Incheon National University / Department of Marine Science; S. Kang, Korea Polar Research Institute / Division of Polar Ocean Science

The Arctic is one of the pristine areas that are sensitive to global environmental changes and have a relatively low environmental pollution. However, Arctic is already affected by floating microplastics (MPs) according to previous studies (38-234 pieces/m^3 in sea-ice and 0.34 pieces/m^3 in the Atlantic arctic polar water). Previous research on the Arctic has concentrated on the waters associated with the Atlantic Ocean (for example, the Barent Sea), while the Arctic Sea (e.g., the Chukchi sea, East Siberian sea, etc.) linked to the Bering strait has never been studied. This area can be particularly important because it links Asian marginal seas and the Pacific Ocean, which is regarded as a global hot-pot of MPs input to the ocean. We conduct annual surveillance in every summer since 2016 using a Korean icebreaker (R/V ARAON) to identify the presence, distribution, fate and effect of MPs in the Arctic Ocean connected with the Bering Sea. This is first result for the Pacific ocean-side polar region investigated in 2016 (Aug./05-21/2016) & 2017 (Aug./06-25/2017) Araon Expeditions. Here, we present the results observed in some media including seawater (surface and subsurface water), sea-ice core, and snow. Seawater samples were collected by manta-trawl net (200 mm mesh, n=12) for surface water, bongo net (330 mm; n=16) for subsurface water, sea ice (n=27) by ice-corer, and snow (n=6). MPs were detected in all samples with average concentrations of 0.41 n/m^3 (surface water in 2016), 0.55 n/m^3 (subsurface water in 2016), and 12.90 n/L (in sea-ice core). We are progressing the analysis for sea-ice core and snow samples collected in 2017. And then the data will be added later. It is generally known that plastics are light and float, therefore they could be enriched on the water surface layer. However, MPs abundance observed in the bongo net (subsurface water) was similar to that of the manta nets (surface water), which can be a strong evidence of the possible sinking of MPs into the deep water of the Arctic Ocean. On the other hand, the sea ice’s contamination level was observed to be several tens of thousands higher than seawater. This indicates the necessity of further study on the trapping mechanism in the freezing process and the effect on the environmental change. The results of this study can be applied to further study on their major origins & mass balance of MPs in the Arctic Ocean, and contribution of MPs to environmental changes in the Arctic Ocean.

MO322

Analysing microplastics in samples of terrestrial systems

A. Mueller, E. Dümichen, Bundesanstalt für Materialforschung und -prüfung / 5.3 Mechanics of Polymers; P. Eisentraut, Bundesanstalt für Materialforschung und -prüfung; C. Bannick, Umweltbundesamt; U. Braun, BAM- Federal Institute Materials Research and Testing / 5.3 Mechanics of Polymers

The occurrence of microplastics (MP), i.e. solid synthetic polymer particles between few micrometres up to five millimetres, in marine and limnic water systems, is already manifold documented. On the contrary, less is known about the occurrence and the fate of MP in terrestrial systems. In the ongoing discussion about a general monitoring of plastic pathways in the environment this is a gap, because MP in terrestrial environment could influence the quality of soil, but might be also relevant for the final transport of plastics into the aquatic environment, e.g. via erosion. In this regard, one critical point is the lack of harmonized or standardized protocols. The matrix of solids is usually more complex than the matrix of aqueous samples. For a first assessment of a potential exposure situation, the determination of the total content of plastics is sufficient. The goal of this work is the development of a systematic protocol for sampling, sampling pre-treatment and analysis of MP in terrestrial samples, which ends up in a fast, quantitative method. Up to now, various studies about the analysis of MP at beaches, in marine sediment and along rivers are available. In these studies, as sample pre-treatment density separation techniques were used to reduce the inorganic matrix. In some cases, a chemical oxidation step to minimize the organic matrix is additionally carried out. FTIR or Raman spectroscopy were used to analyse the sample with enriched concentrations of MP. Using these techniques, only small proportions of inserted samples were analysed. Similar investigations techniques were used for terrestrial samples. The few existing studies investigated sewage sludge, compost fertilization or areas of intensive utilization of plastics foils in agriculture or from industries. In the present work, we present primary results of MP detection using TED-GC-MS in various worst-case or rather polluted hot-spots (i.a. leaf-compost, soil along frequently used roads). A special emphasis is given to easy and fast working steps and techniques for representative sample amounts. A quantitative assessment of highly occurring MP from littering (standard thermoplastics) as well as tire abrasion (synthetic elastomers) is intended.

MO323

Microplastics in Expanded Global Table Salt Product Samples and its implication

J. Kim, Incheon National University / Department of Marine Science; C. Kim, Greenpeace East Asia; S. Kim, Incheon National University / Department of Marine Science

\n Microplastic pollution is becoming a global issue in marine environment pollution. Among the various environmental media, some recent studies have identified microplastic contamination that remains in salt (sea salt, lake salt, rock salt). As salt is an essential human/animal food-item, microplastic contamination in salt means that salt intake is a pathway of microplastic exposure. Additionally, sea salt, which is produced through the evaporation of seawater, can represent the degree of contamination of microplastic remaining in seawaters. This indicates that sea-salt may be a monitoring media for global seawater contamination of microplatic. The purposes of this study are 1) to identify the contamination of microplastic in commercial table salt products sold worldwide, 2) to elucidate any relationship of microplastic contamination between sea-salt and seawater, and 3) to calculate the human exposure of microplastics resulting from the consumption of commercially available salt products. To do this, we purchased and analyzed the salt samples sold in 17 countries (8 countries in Asia, 7 in Europe, 1 in Africa and 1 in North America) in four continents. Each salt sample was selected in consideration of the salt production area, production method, and salt consumption in each country. Total 37 salt samples were analyzed, including sea salt, lake salt, and rock salt. Each sample was duplicated (n = 2) and two blank samples were analyzed for each batch to check contamination during the analysis. Size, color, polymer, and shape of each microplastic was determined under microscopic and spectroscopic analysis (FTIR). Thousands of microplastics were detected per 1 kg of the salt samples of this study, and the predominant forms were fragment and fiber, which were frequently detected in the order of PP > PE > PET. Significant correlation was observed between microplastic discharge rate via the rivers near the sea-salt production and microplastic contamination in the sea-salt. After further analysis, human exposure, characteristics of microplastic distribution, and application of sea-salt as an alternative monitoring medium will be announced.

MO324

Biodegradability of pristine and weathered car tire rubber using different inocula

F. Polesel, Technical University of Denmark (DTU) / DTU Environment; T. Ahonen, Technical University of Denmark DTU / Department of Environmental Engineering; N.B. Hartmann, Technical University of Denmark (DTU) / DTU Environment

Car tire wear is estimated to represent a major fraction of microplastic pollution in the environment. Rubber particles are transported by wind transport and road runoff, thereby reaching soils and wastewater treatment systems. Information on their occurrence, fate and degradability in environmental and engineered systems is limited yet crucial to determine their persistence and potential risks associated to their exposure. In this study, we assessed the biodegradability of car tire rubber (90-125 μm) under controlled laboratory-scale conditions. Standardized batch tests (OECD 301 and ISO 14851) were used to determine complete mineralization under aerobic conditions of pristine and UV-weathered tire rubber in the presence of three different microbial inocula, i.e. activated sludge, soil particles and soil supernatant. Acetate and poly(D,L-lactide-co-glycolide) (PLGA) were used as positive controls in terms of readily degradable substrate and degradable plastic material, respectively. Pristine and weathered rubber exhibited low but measurable biodegradation levels in the presence of activated sludge (3.8-------7.6% ThOD) and soil supernatant (0.8%-2.3%), while no degradation was observed when using soil particles as inoculum. Accelerated UV weathering and higher biomass concentration was found to increase the biodegradability by activated sludge. Interestingly, increased response in terms of oxygen consumption was observed with acetate as co-substrate, indicating improved rubber degradability in the presence of a readily degradable carbon source. PLGA exhibited limited biodegradability (< 17.4%). Scanning electron microscopy analysis revealed increased porosity and roughness on rubber surfaces over the course of the experiments, seemingly indicating degradation via surface colonization. Overall, ready biodegradability tests proved suitable to obtain information on degradation of car tire rubber. This study provides first evidence of their degradability, especially for weathered rubber in the presence of a co-substrate, which should be considered for future studies and fate assessment. No inhibitory effect of rubber on microbial activity was observed. PLGA as positive control in short-term tests is not encouraged due to its limited biodegradability. Future work should consider longer test durations, which may be necessary to provide sufficient biofilm colonization of rubber particles, and assess other (abiotic) degradation mechanisms.

MO325

Evaluating sorption properties of tire materials using poly-parameter linear free-energy relationships (ppLFER)

M. Wehrhahn, University of Vienna / Environmental Geosciences; T. Hüffer, T. Hofmann, University of Vienna / Department of Environmental Geosciences

Tire materials are common representatives of microplastics found in the environment. They are introduced on the one hand as tire wear, an abrasion product, which reaches the environment via road runoff.[1] On the other hand recycled and shredded tire crumb rubber (TCR) is applied as filler material for example on turf fields.[2] It was recently shown that tire materials are a substantial share (~ 66 %) on waste that is introduced into the environment as microplastic particles.[3] Tires generally consist of a mixture of polymers (40-60 %), mostly styrene butadiene rubber (SBR). These elastomers are compounded with carbon black or silica as reinforcing agent (20-35 %), oils (15-20 %) as softeners and extenders as well as vulcanization chemicals (e.g., zinc oxide and sulphur (1-2 %)).[4] Although tire materials are known to pose the risk of leaching toxic substances, they are one of the most popular construction materials and are widely distributed for example in the application of artificial reefs[5], or recycled as passive sampler to remove organic pollutants from water.[6] The precise characterization of molecular interactions between tire materials and (organic) compounds is therefore important to evaluate and predict the behaviour of tire materials in aqueous systems. Poly-parameter linear free-energy relationships (ppLFERs) provide the opportunity to describe the contributions of individual molecular interactions to overall sorption processes taking into account both the physico-chemical properties of the sorbate as well as the sorbent.[7] They have been successfully used to describe and predict sorption of organic compounds to various sorbents.[8] This work hence intends to investigate sorption properties of tire crumb rubber using poly-parameter linear-free energy relationships. [1] B. Liebmann, Mikroplastik in der Umwelt, **2015**. [2] B. Bocca, G. Forte, F. Petrucci, S. Costantini, P. Izzo, Sci. Total Environ. **2009**, 407, 2183. [3] C. Lassen, Microplastics: Occurrence, Effects and Sources of Releases. **2015**. [4] Y. R. Lin and H. Teng, Microporous Mesoporous Mater. **2002,** 54, 167. [5] R. B. Stone, L. C. Coston, D. E. Hoss, F. Cross, Mar. Fish. Rev. **1975,** 37, 18. [6] L. Alamo-Nole, O. Perales-Perez, F. R. Roman, Desalin. Water Treat. **2012,** 49, 296. [7] M. Abraham, A. Ibrahim, A. Zissimos, J Chromatogr A. **2004**, 1037, 29. [8] S. Endo, P. Grathwohl, S. Haderlein, T. Schmidt, Environ Sci Technol. **2009,** 43, 3094.

MO326

Particle toxicity in the daggerblade grass shrimp (Palaemonetes pugio): micronized tire wear particles and microplastics

J.E. Weinstein, The Citadel / Department of Biology; R. Leads, College of Charleston / Biology; S. Kell, College of Charleston / Graduate Program in Marine Biology; A.D. Gray, University of North Carolina at Greensboro / Biology

Recent surveys of Charleston Harbor, SC (USA) have demonstrated that >75% of total microplastics at some locations are tire wear particles (TWP). The aim of the present study was to investigate the toxicity of wet prepared TWP in adult grass shrimp (*Palaemonetes pugio*) and compare it to that of other microplastic particles. For our TWP assays, we conducted a 96-hour acute toxicity test and an immune challenge. Acute mortality was not observed at concentrations up to 100 g/L (1.9x10?7 particles/L). In our immune challenge, grass shrimp were exposed to TWP, polypropylene fragments, polyethylene spheres, polyester fibers, or sediment for 96-hours. Grass shrimp were then injected with either HEPES-buffered saline or *Vibrio campbellii* (5x10^3 CFU/shrimp). After 48 hours, no significant decrease in immune function was observed in exposed shrimp (p=0.8). We also conducted assays examining the size and shape dependent effects of microplastic particles (spheres, fibers and fragments), including TWP, on grass shrimp. Grass shrimp were initially exposed to various size fractions of plastic spheres (30, 35, 59, 75, 83, 116, and 165 µm), fragments (34 and 93 µm), fibers (34 and 93 µm), and TWP (50, 106, and 302 µm) at a concentration of 50,000 particles/L for three hours. Following exposure, grass shrimp were placed in particle-free water and monitored for survival, ingested and ventilated particles, and residence time in the gills and gut. Grass shrimp readily ingested and ventilated all tested particles. The time for microplastics to be cleared from the gut ranged from 27-75 hours with an average at 43.0±13.8 hours. Gut clearance for the TWP was 25.2±3.0 hours. Within the gill chambers the time for microplastics to be removed ranged from 27-45 hours with an average of 36.9±5.4 hours. Gill clearance for TWP was significantly longer at 52.1±21.2 hours. Mortality in these assays ranged from 0-55%, with microplastic spheres and fragments under 50 µm not acutely toxicity. All sizes of TWP were not acutely toxic. Fibers were acutely toxic at both size fractions tested (34 and 93 µm) with mortalities of 55 and 35%, respectively. Results from the present study suggest that wet prepared TWP are less acutely toxic than that of other synthetic particles, especially fibers.

MO327

Acute and chronic toxicity of micronized tyre rubber to Hyalella azteca

F. Khan, L.L. Halle, Roskilde University / Science and Environment; A. Palmqvist, Roskilde University / Department of Science and Environment

An average car tire lasts for 40000 km and during its life time 30% of the tire tread will emitted into the aquatic environment. Inevitably it interacts with aquatic biota. In comparison to the wealth of research on the impacts of microplastics (MPs), there is little on micronized tire rubber (microrubber, MR). Recent reports suggested that tire rubber contributes a significant proportion of ocean's plastic and when rubber is found in the environment it is often classed with MPs, but MR is fundamentally different from MPs in terms of structural and chemical properties, and perhaps should be considered as a distinct pollutant. MR contains a suite of toxic substances; trace metals (notably Zn, Cd), polycyclic aromatic hydrocarbons (PAHS, such a pyrene) and assorted volatile organics used in vulcanization and as antioxidants (e.g. aniline) which have been shown to leach into the aqueous environment. Currently, little is known about the ecotoxicological impacts of MR. The present study was to conceived to determine the toxicty of tire rubber particles to *Hyallela azteca*, an established freshwater model organism, over acute and chronic exposures, and to delinieate particle effects from those of the leachate. The acute toxicity (48 h) of MR particles compared to the MR leachate show similar LC50s (3426±172 particles/mL for MR and 3628±672 particles/mL for leachate), but significant differences are found at LC10 and LC90, suggesting that at low particle concentration the leachate is more important, but at high concentrations the particle may act to deliver chemicals *in vivo* following ingestion. The results of the 21 day study showed that mortality, reproductive output (neonate production) and net growth were significantly impacted at the higher exposure concentrations of MR. MR is an emergent contaminat of concern that is similar but disctint to microplastics in many aspects. Very little is known about the toxicity of MR, but our results show that MR exposure has short-term and longer-term toxicty on a key freshwater species.

MO328

Acute and chronic effects on Hyalella azteca and chemical analysis of rubber particles and leachate - comparison of pristine micronized car tire to previous data on worn car tire particles

L.L. Halle, Roskilde University / Science and Environment; A. Palmqvist, Roskilde University / Department of Science and Environment; K. Kampmann, A. Jensen, Danish Environmental Analysis; F. Khan, Roskilde University / Science and Environment

Microrubber (MR) from car tires constitutes a significant contribution to particulate contamination of the aquatic environment. MR is produced from driving, in the interface between tires and asphalt, and may eventually be lead out to the surface waters, possibly together with leached granulated rubber particles used in artificial turf. Although rubber particles have been detected in the aquatic environment the potential environmental impacts of this contamination are largely unknown. *Hyalella azteca* is an ecologically relevant freshwater amphipod that is also a well-established model organism in ecotoxicology. This study aims to investigate the acute and chronic effects of MR on *H. azteca* by comparing the biological effects and chemical characteristics of a pristine tire with previous data from a worn tire of same make and model. Effects are assessed as changes in survival, growth and reproduction and both the effects of rubber particles and rubber leachate is investigated with the aim of determining whether there are particle effects and/or if the mode up uptake of chemicals leached from tire influences effects observed in the acute phase. The chemical characteristics of both pristine and worn tire particles are quantified by GCMS. The preliminary results indicate that, surprisingly, pristine tire both as particles and leachate is much more toxic than worn tire in acute tests. Although the main source of MR undoubtedly is worn tire, these results points toward further ecotoxicological testing of tire coatings used during manufacturing. Results from this ongoing study will be presented and discussed in relation to the microparticle debate.

MO329

Applying nuclear techniques to study the biokinetics and toxicodynamics of

microplastics and co-contaminants in marine biota

C. Lanctot, International Atomic Energy Agency / Radioecology Lab; M. AL SID CHEIKH, University of Plymouth / Marine sciences and engineering; A.I. Catarino, Heriot-Watt University / Institute of Life and Earth Sciences; T. Cresswell, ANSTO Environmental Research / Institute for Environmental Research; B. Danis, Université Libre de Bruxelles; T. Mincer, Woods Hole Oceanographic Institution; F. Oberhaensli, P. Swarzenski, International Atomic Energy Agency / Radioecology Lab; I. Tolosa, International Atomic Energy Agency; H.K. Karapanagioti, University of Patras / Chemistry Departament; M. Metian, IAEA-EL / Radioecology Lab

Despite recent efforts in understanding the risks associated with marine plastic pollution, there remains a great deal of uncertainty regarding the potential impacts of microplastics and nanoplastics on wildlife and humans. This largely relates to the methodological and analytical limitations associated with studying relatively low and environmental concentrations of these plastics. The IAEA Radioecology Laboratory, in collaboration with a team of external experts, is tackling these challenges by applying nuclear and isotopic techniques to address important outstanding questions on the risks of microplastics to marine organisms. Novel approaches using radiolabeled plastic particles and associated organic and inorganic contaminants are being developed to very precisely quantify their movement, fate and impacts on a range of aquatic biota, under controlled laboratory conditions. Nuclear techniques are uniquely suited for this research given their sensitivity and capacity to measure biokinetic and toxicodynamic parameters overtime. As such, these tools will allow us to address important knowledge gaps, including the evaluation of (1) the biokinetics, biodistribution and potential biological impacts of realistic concentrations of small plastic particles (< 100 µm) in marine biota; (2) the sorption kinetics of trace pollutants to microplastics; and (3) the influence of microplastics on the bioaccumulation of co-contaminants. Importantly, this research will allow us to test if microplastics can truly be bioaccumulated (i.e., cross epithelial membranes/tissues), and if they can act as a vector for contaminant transfer in the marine environment under low level exposures. This poster will provide an overview of the techniques used to address these questions, as well as preliminary outcomes and future directions.

MO330

Aggregation kinetics of plastic nanoparticles in fresh and marine phytoplankton culture media

z. venel, EPOC, University of Bordeaux / UMR EPOC 5805; J. Gigault, University of Rennes 1 / Laboratoire Geosciences Rennes; M. Baudrimont, Université Bordeaux1 / UMR EPOC 5805

Release of plastics debris in the environment has been catching more and more concerns in recent years, especially in aquatic environment. It has been proved recently, that plastics break down to produce nanoparticles by photochemical degradation in marine waters. However, there is a lack of suitable analytical tools, and the environmental fate and transport mechanisms of nanoplastics have not yet been investigated. Indeed, several ecotoxicology studies investigate the impact of nanoplastics on aquatic organisms without addressing their aggregation state in aqueous medium, whereas fate and toxicity of nanoplastics depends on their aggregation behavior. This lack is clearly due to the complexity of environmental matrix and the extremely low concentration of nanoplastics, which push towards beyond the classical detection limits of analytical instruments dedicated to their physical characterization. The aim of our work, is to develop unprecedented methods to measure ultra-trace concentration of engineered nanoplastics in environmental like water. In our study, we investigated aggregation kinetics of plastic nanoparticles in culture media for fresh water (Dauta) and marine (F/2) phytoplankton. Polystyrene nanospheres (20 and 100 nm) and crushed nanoscale plastics particles were added in culture medium at environmental concentrations (10^9-10^{12} particle/cm^3). Immediately after, evolution of size distribution, fractal dimension and stability were investigated using *in situ* dynamic light scattering, static and multiangle light scattering, laser induced breakdown detection, asymmetrical flow field-flow fractionation and zeta-meter. Results show a rapid aggregation of nanoparticles during first hours, followed by a slow aggregation until reaching a maximum value after 24 to 48 hours. The different analytical tools used in this study, allowed us to monitor aggregation state and exhibit different aggregation kinetics of PS nanoparticles, depending on nanoparticle size, form, and salt concentration in order to better characterize the conditions of exposure of phytoplankton by these PS nanoparticles.

MO332

Mytilus spp. as sentinel species for water borne microplastic ingestion; a case study from the Norwegian coast

I. Nerland Bråte, Norwegian Institute for Water Research NIVA; A.L. Lusher, K. Iversen, NIVA   Norwegian Institute for Water Research; N.W. Green, J. Beyer, NIVA   Norwegian Institute for Water Research / marine pollution; K.V. Thomas, QAEHS   The University of Queensland; M. Olsen, NIVA   Norwegian Institute for Water Research / Queensland Alliance for Environmental Health Sciences (QAEHS)

Mytilus spp. is widely used as a sentinel species for coastal pollution monitoring, and ICES have *suggested M. edulis* and *M. galloprovincialis* as suitable to also monitor waterborne microplastic pollution in the marine environment. Due to several methodological differences between studies (e.g. sample collection, samples processing identificon of microplastics such as visual ID, detection limit and chemical verification of polymers) comparability can be challenging. When looking at spatial trends it is necessary to use comparable methods. Several studies have found microplastics in *Mytilus spp.* and a recent study found accumulating levels of microplastics in mussels compared to the surrounding waters. In this study, a total of 252 mussels were investigated from 13 different sites along the Norwegian coast by using KOH digestion followed by visual ID and µFTIR. Occurrence of plastics were found in a total of 76.6% of the analysed mussels from the Norwegian environment, with the overall average plastic load being 1.85 particles gram^{-1} w.w (ranging from 0 – 24.45) and with the highest values found in mussels from the Barents Sea. Microplastics consisted of fibers (85 %), fragments (11 %) and films and foams (4 %) and most particles were < 1mm, with the most dominant polymer group being semi-synthetic materials (rayon/viscose). Based on litterature and this current study, *Mytilus spp.* seems to be a promising sentile species to monitore pollution of the smallest waterbourne microplastics in the marine envrionment. However, for the method to be fully quantitative, imporvements/furhter investigations are needed e.g. same sized mussels from each site, same possition in the water column as well as understand inter-site variation within populations. Furthermore, the presence of semi-synthetic materials in the marine environment needs to be further investigation as well as their potential effects on biota.

Mercury Biogeosciences - Fate, Effects and Policy (P)

MO333

Influence of biofilm composition on mercury bioaccumulation

P. Dranguet, University of Montreal / Département de sciences biologiques; S. Bouchet, IPREM CNRS UMR   LCABIE / Institut des Sciences Analytiques et de Physico-chimie pour Lenvironnement et les Materiaux; M. Leclerc, Universite de Montreal / Département de sciences biologiques; D. Amouroux, IPREM CNRS UMR   LCABIE / Institut des Sciences Analytiques et de Physico-chimie pour Lenvironnement et les Materiaux; M. Amyot, Universite de Montreal / Département de sciences biologiques; V. Slaveykova, University of Geneva / Département F.-A. Forel des sciences de lenvironnement et de leau; S. Le Faucheur, Institute F.-A. Forel, University of Geneva / Département F.-A. Forel des sciences de lenvironnement et de leau

In aquatic systems, the dominant lifestyle of microorganisms (bacteria and microalgae) is to live together interlocked in exopolymeric substances (EPS), rather than as single cells. Besides their involvement in several biogeochemical processes, biofilms are sites of accumulation and transformations of mercury (Hg). Their natural assemblage of heterotroph and autotroph microorganisms makes them an important entry of Hg into aquatic food webs. The objective of the present study is thus to better understand the mechanistic processes that control Hg accumulation in biofilms and we focused on the elucidation of the role of biofilm composition on the kinetics of Hg uptake. For that end, two couples of biofilms were exposed to IHg (~ 100 pM, precisely measured) using microcosms. The first couple of biofilms was obtained using different times of colonisation e.g. 85 and 127 days whereas the second couple (bottom/surface biofilms) was obtained owed to different colonisation depth in the Versoix River (CH). Prior Hg exposure, biofilm biomass and microbial composition (chlorophyll content and abundance of *16S rRNA* gene) were determined as well as the colloidal and capsular EPS thiol composition. The main water quality parameters (pH, concentrations of dissolved organic carbon, Hg, anion and cation) of the exposure media were also analysed. Accumulation of total Hg and non-extractable IHg (determined after a cysteine washing step) in biofilms were measured at different step times (t < 24 h) to model non-extractable Hg uptake kinetics using a non-linear pseudo first order one-compartment model. In each biofilm, non-extractable IHg accumulation was very rapid, within minutes of exposure. The uptake rate constant of the younger biofilm was measured to be 10 times higher than that of the older biofilm. That same ratio was also obtained between the bottom and the surface biofilms. Except for the older biofilm, Hg accumulation reached a plateau at ~6 h Hg exposure. A decrease in the EPS thiol concentration was observed in the bottom biofilm upon Hg exposure, suggesting a change in Hg bioavailability in the microorganism environment living in that biofilm. Our study demonstrated that biofilm microbial and EPS composition as well as thickness influence Hg uptake by microorganisms living in biofilms.

MO334

Gaseous elemental mercury concentration and diurnal evasional fluxes from the water-air interface in coastal environments of the northern Adriatic Sea

S. Covelli, Dipartimento di Matematica e Geoscienze / Dept. of Mathematics and Geosciences; A. Acquavita, ARPA FVG; F. Floreani, E. Petranich, E. Pavoni, University of Trieste

Among pollutants widespread in the environment, mercury (Hg) is well recognised for its toxicity, mobility and bioaccumulation potential. In coastal areas the presence of this element generates conflicts with important resources of profitable value such as fisheries and aquaculture. The Marano and Grado Lagoon (Adriatic Sea) experienced a double Hg impact. The first is due to the mining activity conducted at Idrija (western Slovenia) for approximately 500 years, whereas the second is the result of discharge of a chlor-alkali plant effluents. Fish farming is a

historical activity covering 14% of the total lagoon area. Recently, one fish farm was long-term monitored in order to understand the role of the sediment-water interface in recycling Hg and to estimate benthic fluxes and Hg mobility in the water column. An important further step toward a better comprehension of the Hg biogeochemical cycling in the lagoon environment, is represented by the estimate of its evasional fluxes, as gaseous elemental mercury (GEM), at the water-air interface. A dynamic flux chamber coupled with a real-time atomic adsorption spectrometer (Lumex-RA 915+) has been used to measure GEM and to estimate the diurnal evasion flux at the water-air interface during three seasonal campaigns in four selected sites: two in a lagoon fish farm, one in the open lagoon environment highly impacted by Hg mining activities and the last one in an uncontaminated area of the Gulf of Trieste, the Bay of Piran (Slovenia). Accomplished to these measurements the regional background level of atmospheric GEM was determined together with the main chemico-physical parameters influencing Hg behaviour. This new insights will be of help for future estimates of Hg mass balance in one of the most contaminated areas in the Adriatic Sea. *Keywords*: atmospheric mercury; mercury fluxes; fish-farm; Grado Lagoon

MO335

Atmospheric mercury assessment: a contribution to global monitoring and effectiveness evaluation within the Minamata Convention

A. Fino, Italian National Research Council - Institute of Atmospheric Pollution Research (CNR-IIA); F. Sprovieri, A. Macagnano, E. Zampetti, P. Papa, G. Esposito, CNR Institute of Atmospheric Pollution Research Italy; P. Nicola, Institute of Atmospheric Pollution Research

In 2013, the Minamata Convention on Mercury was adopted by governments recognizing mercury as a pollutant of global concern for both human health and the environment. After reaching the 50th ratification the convention entered into force on 16 August 2017. According to the Article 22, the Conference of the Parties should establish of arrangements for providing itself with comparable monitoring data on the presence and movement of mercury and mercury compounds in the environment as well as trends in levels of mercury and mercury compounds observed in biotic media and vulnerable populations on the basis of available scientific, environmental, technical, financial and economic information. UN Environment in close collaboration with Italian National Research Council - Institute of Atmospheric Pollution Research (CNR-IIA) and WHO implemented a UN Environment - Global Environmental Facility (GEF) project entitled *"Develop a plan for global monitoring of Human exposure to and environmental concentration of Mercury",* from late 2014. The project contributes to scientific knowledge for development of effectiveness evaluation mechanism. The main aim of the project was to harmonize approaches for mercury monitoring and to strengthen the capacity for mercury analyses in humans and in the environment. The experience made within the project suggests that there is a urgent need to coordinate the global efforts in atmospheric mercury monitoring by integrating existing monitoring programs worldwide in cooperation with other on-going programs such as GEO (Group on Earth Observation, www.earthobservations.org) and specifically with the GEO Flagship *"Global Observation System for Mercury (GOS^{4}M)*" which is aimed to support the policy process in relation to the Minamata Convention implementation. CNR-IIA proposed a selection of monitoring sites, mostly background sites but also including those highly impacted, to undertake passive sampling and analysis of Hg in ambient air in order to strengthen capacity to provide globally comparable data. The poster gives an overview of preliminary results coming from the pilot survey campaigns carried out with mercury novel passive sampling.

MO336

Assessment of Hg impacts on mountain river ecosystems

S. Le Faucheur, Institute F.-A. Forel, University of Geneva / Département F.-A. Forel des sciences de lenvironnement et de leau; F. Pierron, Université de Bordeaux / UMR EPOC CNRS 5805; C. Moinecourt, Université de Genève; G. Daffe, University of Bordeaux / UMR EPOC CNRS 5805; A. Boullemant, RioTinto

Mountain rivers are high-flow systems which can experience, even daily, high water height variations due to the presence of dams along their courses. These conditions limit the use of water and sediment analyses to identify pollution point source. That observation is especially true when contaminants are metals, notably Hg, which concentrations in natural waters are very low (few ng L^{-1}). The present study thus focused on the use of bioindicators to evaluate the potential releases of Hg from waste landfill and subsequent impacts. The two studied rivers are located in the Alps, both of which having mineral processing landfills along their courses. Several sites were sampled, upstream and downstream of the landfills, four times during one year to get seasonal variations of the water quality. For both rivers, water, sediments, bryophytes, biofilms and fish (*Salmo trutta fario*) were collected. Water was analysed for its physico-chemical parameters (pH, concentrations of dissolved organic carbon (DOC), anions, cations, metals, including Hg). Sediments were also analysed for their metal concentrations as well as for their composition. Finally, metal concentrations were also measured in both biofilms and bryophytes (total and intracellular) as well as in fish (in gills, muscle, gonads and liver). Results indicate those two streams are moderately hard in summer to very hard water in winter and are characterized by a very low DOC concentrations (~ 0.2 mg L^{-1}). Sediments are mainly composed of coarse sand (~ 95%) with very low organic matter (0,5 – 2%). In summer, when the river flow was at its highest, Hg concentrations in sediment and bryophytes are low, with no observable differences between up- and downstream sites. However, fish gills, liver, and muscle demonstrated more elevated Hg concentrations downstream of sites compare to upstream ones. In fall as well as in early-spring, when the flow decreased by more than half as compare as that of summer, Hg concentration in the water column is very low (close to 0.2 ng L^{-1}) but sediment, bryophyte, and biofilm Hg concentrations are in accordance with fish analyses and demonstrated the Hg increase downstream the landfills. However, no oxidative stress and impairments are observed in fish. The present study confirms the need to address all compartments to properly assess the water quality of an aquatic system and therefore to understand potential impact of landfills and industrial sites on freshwater ecosystems.

MO337

Mercury Photo-reduction and Total Photoreducible Mercury Dynamics in the Lakes of Kejimkujik National Park, Nova Scotia

N.J. O'Driscoll, Acadia University / Department of Earth and Enviromental Science; T. Christensen, Acadia University; E. Vost, Nova Scotia Department of Natural Resources; S. Klapstein, E. Mann, Acadia University

Photo-reduction and photo-oxidation are fundamental mechanisms controlling mercury volatilization and accumulation in freshwaters. In all surface waters dissolved gaseous mercury (DGM) is produced as a net result of the reduction of reducible mercury, which is believed to be primarily divalent mercury (Hg(II)) bound to specific carbon-based ligands, and the oxidation of elemental mercury (Hg(0)). These two processes control the amount of DGM available for evasion across the water-air interface; however, determination of the fundamental rate constants and mechanisms of these reactions in freshwaters are still areas that require more research. In particular, the total amount of photoreducible mercury is emerging as a key variable that requires more exploration. Here, we review the progress our group has made in this field over the past 10 years; we present rate constants as well as temporal dynamics in total reducible mercury derived from two recent projects that examined water samples from a series of freshwater lakes in Kejimkujik National Park, Nova Scotia, Canada. We examined the hypothesis that gross photoreduction and photooxidation rates would be significantly different in lake water. Another hypotheses was that the amount of mercury available for reaction with solar radiation (i.e. reduction of Hg(II) to gaseous Hg(0)) in surface waters would significantly change over a summer. A Luzchem photo-reactor was used to irradiate 200 mL water samples in quartz beakers continuously exposed to ultraviolet radiation for 24 h with concurrent Hg(0) analysis to derive pseudo-first order gross reduction rate constants and batch experiments were used to derive net reduction rates (and gross photooxidation by difference). Results showed that the net photo-oxidation rates for freshwaters were low, with mercury reduction and oxidation reactions very close to being in balance. We also found that the amount of total reducible Hg(II) changed significantly in three of the lakes over several sampling months. Dissolved organic carbon concentration was a key factor positively correlated with these results. This research provides the first quantitative measurements of gross photooxidation and photoreduction rates as well as total photo-reducible mercury over a season in surface freshwater lakes.

MO338

Influence of Avian Biovectors on Mercury Speciation in a Wetland

J. Kickbush, Acadia University / Biology; M.L. Mallory, Acadia University / Biology department; J. Murimboh, Acadia University / Chemistry; J. Rand, Acadia University / Engineering; S. Klapstein, Acadia University / Earth & Environmental Science; A. Loder, Acadia University; N.M. Hill, Fern Hill; N.J. O'Driscoll, Acadia University / Department of Earth and Enviromental Science

Mercury is a persistent and bioaccumulative chemical that is present in many remote environments due to its ability to be transported long distances in the atmosphere, and to be deposited far from the original source (Sunderland and Chmura 2000). Wetland ecosystems are important "hot spots" for mercury in eastern Canada, providing anoxic environmental conditions that promote the bacterial methylation of mercury. Methyl mercury is the most biologically available form of mercury and the form which biomagnifies in food webs (Gochfeld 2003). Seabird guano is a well-documented biovector for metals – including mercury – and nutrients, which may indirectly affect metal speciation (Choy et al. 2010). The site for this study, Big Meadow Bog (Brier Island, Nova Scotia, Canada) has a history of ditching in the 1950s, which changed hydrology significantly, resulting in colonization by 3000 pairs of herring gulls (*Larus argentatus*) in the 1980s. To quantify changes in mercury mobilization and speciation in response to this biovector, groundwater samples were collected from this site as well as a reference bog with similar geological and hydrological characteristics. The filtered samples were analyzed for total mercury, methyl mercury, and water chemistry (pH, conductivity, anions, cations, and dissolved organic and inorganic carbon). Results show significantly higher nutrients (nitrate, phosphate, and sulfate), total mercury, and methyl mercury concentration when compared to the reference bog that is minimally impacted by avian biovectors. This elevated availability of methyl mercury could potentially pose a threat to the local ecosystem and wildlife population due to methyl mercury's toxicity to living organisms (Akearok et al. 2010, Singh et al. 2011). Citations: Akearok J et al. 2010. Science of the Total

Environment 408(4):836-840. Gochfeld M. 2003. Ecotoxicology and Environmental Safety 56(1):174-179. Choy E et al. 2010. Science of the Total Environment 408:1858-1867. Singh R et al. 2011. Indian Journal of Pharmacology 43(3):246-253. Sunderland E, Chmura G. 2000. Science of the Total Environment 256(1):39-57.

MO339

Organohalogen and mercury residues in fish from the Western Mediterranean Sea: concentrations, bioaccumulation and dietary exposure

E. Junqué, Institute of Environmental Assessment and Water Research (IDAEA-CSIC); M. Garí, IDAEA-CSIC / Environmental Chemistry; R. Llull, General Direction of Public Health and Consumption; J. Grimalt, Institute of Environmental Assessment and Water Research IDAEA CSIC / Department of Environmental Chemistry

Organochlorine compounds (OCs) and mercury (Hg) have diverse deleterious health effects and are persistent in the environment. They tend to bioaccumulate and biomagnify along the food chain. Diet is the major source for the incorporation of these pollutants into humans, especially trough consumption of fish. This research focuses on the presence of OCs, total mercury (THg) and methylmercury (MeHg) in lean fish from the Western Mediterranean Sea. Determinant factors of these concentrations such as trophic level and weight of fish have been studied. The estimated weekly intake (EWI) from the Spanish population of these pollutants has been assessed. Samples were collected between March 2015 and August 2016. Most of them were from Balearic Islands (Majorca n=67, Menorca n=17, and Ibiza n=18) and the rest of the samples were from Tunisia (n=2) and Egypt (n=1). Additional fish samples were collected from the Atlantic Ocean, in front of Senegal (n=4) and Mauritania (n=10) coasts for comparison. The OCs levels found in fish were similar or lower than in other previous studies. In contrast, 15% of the most frequently fish species consumed by the Spanish population had Hg concentrations above the maximum level set forth by the European Union MRL for human consumption. The concentrations of OCs and Hg between trophic levels have been compared. Except for HeCB, the values were higher in the upper trophic level with statistically significant differences for ΣDDTs and ΣPCBs ($p< 0.05$). The relationship between fish weight and pollutant concentrations were also studied. A positive relation between Hg concentrations and weight was found ($R2=0.58$; p-value< 0.001).This trend was not observed for any other OC. The concentrations found in dusky grouper from the Mediterranean Sea and Atlantic Ocean were also compared. The former group presented higher levels for ΣDDTs, ΣPCBs and Hg ($p< 0.05$). The estimated weekly intake of OCs were well below the reported Tolerable Intakes. However, for Spanish population that only consume Mediterranean fish, the estimated weekly intake for Hg (4.42 µg/kg bw) exceeded the THg established by EFSA in 2012, 4 µg/kg bw. The equivalent estimations for MeHg, involving provisional tolerable weekly intakes of 1.3 µg/kg bw were six and three times higher than these provisional tolerable weekly intakes in adults and children (7-12 years of age), respectively.

MO340

Mercury, Comercial Fish & Risk assessment: a Review study (1994-2015)

H. Coelho Vieira, University of Aveiro; J. Von Osten, Autonomous University of Campeche / Instituto EPOMEX; A.M. Soares, F. Morgado, University of Aveiro / department of Biology & CESAM; S. Abreu, University of Aveiro / Dep. Biology & CESAM

Fish consumption is linked to the prevention of some human diseases, especially regarding cardiac and circulatory disorders, due to the content of high-quality protein, vitamins and n-3 fatty acids. At the same time, fish consumption is considered a major pathway of mercury (Hg) exposure in human. More than 90% of this Hg present in fish tissue is found essentially in its organic form (methylmercury (MeHg)), which is the most toxic form of Hg. Due to the potential adverse human health effects, international agencies have established Reference doses (RfD) as recommendations to Hg intake. Some studies have been associating the fish consumption with the Hg bioaccumulation, in areas along the Mid-Atlantic Ridge (MAR) exposed to active hydrothermal fields. The Azores archipelago is located in the North Atlantic Ocean close to the MAR. The last fishery statistics for fish consumption per capita in the Azores archipelago shows that, each Azorean consumes about 80 kg of fish per year being the region with the highest consumption of fishery products in Portugal. This study is the result of a review of all published articles indexed in Web of Science that presented Hg concentration in the muscle for fish species captured in the Azorean Exclusive Economic Zone, and additional new data from fish obtained by recreational fish. The selection of species was based on the fish landing reports (1994-2015) of Azores Fisheries Statistics (SREA) for the entire Azorean ports. At the Azorean ports, in average, about 10000 tonnes per year of these commercial fish species are discharged. Despite low Hg levels in fish, every year the population of this area is exposed to more than 1500g of Hg via fish consumption. However, the species with the highest concentration of Hg are not always those that contribute to a higher human exposure. The fish species *Mora moro* exhibit higher values than the permitted for fish consumption and carnivores fish species generally exhibit higher concentration of Hg than omnivores fish species. On the other hand, demersal fish species demonstrated higher Hg concentration than pelagic fish species. Finally, the target hazard quotient (THQ) is < 1 for all fish species, meaning that the level of exposure is lower than the reference dose, and indicating that the daily exposure is not likely to cause any negative health effects during a lifetime in the human population.

MO341

Mercury concentrations in black bream from the Gippsland Lakes, Victoria, Australia.

L. Metzeling, EPA Victoria / EPA Victoria; S. Balshaw, Department of Health and Human Services; R. Goudey, EPA Victoria

The Gippsland Lakes are a coastal lakes system in eastern Victoria, Australia. They represent a unique aquatic ecosystem of significant ecological significance (Ramsar classified) as well as supporting significant tourism, recreational and commercial fishing industries. The Lakes area has been subject to several contaminant monitoring investigations over the last 45 years that have identified sediment and soil samples with detectable mercury concentrations, often exceeding screening levels set to protect the environment, aquatic plants and animals. The sources of mercury to the Lakes have been identified as having anthropogenic origins. Historic sources are gold mining and wastewater discharges from a paper mill. Ongoing sources of mercury include emissions from coal-fired power plants in the Latrobe Valley to the east of the Lakes and wood smoke from bushfires and planned burns. This study, conducted in 2015, aimed to determine if the concentrations of mercury in black bream had increased over time, and whether or not existing dietary advice issued nationally by Food Standards Australia New Zealand for the protection of consumer health against the effects of mercury in seafood, was appropriate for fish sourced from the Lakes. Three previous studies investigated the concentrations of mercury in fish from the Gippsland Lakes, the earliest being in 1980. Comparisons by others between the first two studies (1980 and 1998) had suggested an increasing trend of mercury in fish. Assessment of the mercury concentrations in fish caught in 2015 against those reported in the previous studies found that the concentrations have remained relatively stable from 1980 to 2015, regardless of the location from which fish were collected. There was no indication of increasing concentrations of mercury over time with the recent results on average lower than the 1998 results. While some variation was observed in the levels of mercury in fish between studies, this variation appeared to be due to differences in the size and age of fish between studies, rather than a result of increasing availability of mercury to fish.

MO342

Mercury health risks due to the substitution of fish meat with shark meat.

P. Ramirez Romero, U.A.M. Iztapalapa / Hidrobiologia; L. Elizalde Ramirez, Universidad Autonoma Metropolitana Iztapalapa / Hydrobiology; H. Barrera Villa Zevallos, UAM Iztapalapa / Hidrobiologia

A previous three years study of mercury content in a variety of edible marine fish from Mexico showed that shark meat was being sold as fish meat when people bought processed fish meat. However, the magnitude of this practice was not known, therefore the objective of this study was to quantify the substitution of fish meat with shark meat in Mexico City's fish market (Central de Abasto) to evaluate the risk due to non-intentional shark meat ingestion. Samples were bought in the fish market as fish for ceviche, quesadillas or soup, economical fillet, battered or breaded fish from sea bass, catfish, tilapia, red snapper and other popular species. Chondrichthyes universal oligonucleotides in PCR were used to analyze the samples. 777 surveys were applied to obtain information regarding fish consumption habits, portion sizes and other characteristics of the population of the Mexico City metropolitan area. Shark mercury content was taken from the previous study. Hg average daily dose, lifetime average daily dose and total dose were calculated for two concentrations (lowest = 0.3 mg Hg/Kg and average = 2.7 mg Hg/Kg). Health risk was calculated using USEPA equations. Of the 52 "fish samples" analyzed 61.53% were identified as sharks of the following species: Leopard (*Galeocerdo cuvier*), Common sawshark (*Pristiophorus cirratus*), Goblin (Mitsukurina owstoni), nurse (*Ginglymostoma cirratum*), whale shark (Rhincodon typus), scalloped hamerhear (*Sphyrna lewini*), daggernose (*Isogomphodon oxyrhynchus*), silky (*Carcharhinus falcimormis*). With regards to the health risk, when considering the lowest Hg concentration, children may eat only one 188 g portion/month, while fertile women and older people, two 190 g portions/month and men up to five 260 g portions/month. When considering the average Hg concentration, the number of portions/month is drastically reduced to less than one/month. If the amount of portions described before are respected, the calculated risk for babies was 0.83 but significantly reduced later in life if the lowest Hg concentration is considered. However, when calculating the risk with the average Hg concentration the risk quotient was always above 1 and up to 7.2 for babies. In conclusion there is a health risk when eating fish that cannot be identified, so people must buy whole fish to secure their identity and authorities should implement a monitoring program to penalize the sale of shark meat as fish meat.

MO343

Mercury in trophic webs of estuaries in South-Southeastern Brazil.

T.H. Trevizani, Universidade de Sao Paulo / Oceanografia Química; M.C. Vedolin, Insituto Oceanográfico da Universidade de São Paulo / Oceanografia Química; R.C. Figueira, Instituto Oceanográfico da Universidade de São Paulo / Instituto Oceanográfico; C. Domit, Universidade Federal do Paraná / Centro de Estudos do Mar

The estuarine regions of Brazil are susceptible to anthropic pressures due to urban,

industrial, agricultural and harbor activities, which impact the environment through the release of contaminants such as metals. Among these metals, mercury (Hg) is highlighted due to its toxicity and capacity of biomagnification. In the Paranaguá estuary, in the state of Paraná, and in the Cananéia estuary, state of São Paulo, these potentially polluting activities are present in different levels. However, these regions comprise the largest remnants of the Atlantic Rainforest ecosystem, fact that gives them the titles of World Heritage site and biodiversity hot spot (UNESCO), therefore a environmental monitoring and conservation acts of the areas are necessary. Thus, this study investigated the concentrations of Hg and nitrogen isotope ratio ($\delta^{15}N$) in the trophic web composed by benthic invertebrates, benthivores fish (*Stellifer rastrifer, Paralonchurus brasiliensis and Isopisthus parvipinnis*) and marine mammals (*Sotalia guianensis and Pontoporia blainvillei*), to understand the trophic dynamics of Hg and compare these estuaries. Samples were taken in the summer of 2015; analyses of $\delta^{15}N$ were carried out by Elemental Analysis - Isotope Ratio Mass Spectrometry (EA-IRMS), and mercury analyses were carried out by Optical Emission Spectrometry, Inductively Coupled Plasma with Vapor Generator Accessory (OES-ICP-VGA), in the muscular tissue of the organisms. The results of $\delta^{15}N$ varied from 6.4 to 13.8 ‰ in Paranaguá and from 7.1 to 14.3 ‰ in Cananéia, with a continuous enrichment among the trophic levels. Concentrations of Hg were significantly higher in Paranaguá (0.02 to 5.8 mg kg-1) than in Cananéia (0.02 to 0.9 mg kg-1), with maximum values in marine mammals, followed by invertebrates benthic and fish. Through linear regressions between Hg and $\delta^{15}N$, positive correlations were observed only in Paranaguá, but they were not significant, which indicates a trend of biomagnification of this element. Such a behavior is expected since this estuary suffers greater pressure from anthropic activities than Cananéia, and presented similar values to highly degraded Brazilian estuaries, such as Santos Bay (SP) and Guanabara Bay (RJ). The results showed a current panorama of the trophic distribution of Hg in these estuaries being a useful tool in environmental monitoring and coastal management in regions of great ecological importance.

MO344

Biological and Geochemical Drivers of Mercury Toxicity in Yellowknife, NWT, Canada

M. Azdajic, E. Yumvihoze, A.J. Poulain, J.M. Blais, University of Ottawa / Biology

Mercury (Hg) is a global pollutant that bioaccumulates in aquatic and terrestrial foodwebs as monomethylmercury (MMHg). Microbial activity is the main driver of MMHg production, with sulfate reducing bacteria being a major contributor. The roasting of arsenopyrite at Giant Mine in Yellowknife, NWT, has created strong environmental gradients of sulfate in lakes in the surrounding area with distance from the mine. Whereas total Hg levels remain constant with increasing distance from the mine, the ratio of MMHg relative to total Hg increases with proximity to the stack. We hypothesized that high sulfate in lakes near the mine may be responsible for elevated MMHg concentrations in those same areas. To test our hypothesis, we sampled water and sediments from lakes spanning a range of distances from the Giant Mine. We determine simultaneous methylation and demethylation rates using stable isotope analysis and characterized the microbial community using high throughput sequencing of 16S rRNA genes. By analyzing methylmercury production and microbial community composition, we have identified sulphate as being the main driver of both final concentrations of methylmercury and microbial community structure.

MO345

Use of green tea to reduce mercury and methylmercury bioaccessibility in raw and cooked fish

V.F. Barbosa, Portuguese Institute of Sea and Atmosphere IPMA / Division of Aquaculture and Seafood Upgrading; P. Anacleto, Instituto Português do Mar e da Atmosfera (IPMA) / Division of Aquaculture and Seafood Upgrading; R. Alves, King Abdullah University of Science and Technology (KAUST); A. Maulvault, Instituto Português do Mar e da Atmosfera / Division of Aquaculture and Seafood Upgrading.; F. Fogaça, Embrapa; T. Langerholc, Faculty of Agriculture and Life Sciences, University of Maribor; A. Marques, Portuguese Institute of Sea and Atmosphere IPMA / Division of Aquaculture and Upgrading

Human exposure to mercury (Hg) and methylmercury (MeHg) increases in diets rich with high trophic level marine species, raising human health-related concerns through seafood consumption. Nevertheless, the overall concentration of Hg and MeHg detected in seafood does not always reflect the amount that will be available for absorption by human intestinal epithelium after the digestion process. On the other hand, several studies have highlighted the health benefits attributed to green tea consumption and their potential effects on reduction of bioaccessibilty of contaminants. In this context, the aim of the present study was to assess the effect of green tea in Hg and MeHg bioaccessibility in raw and cooked marine fish species. Results demonstraded that total Hg/MeHg concentration in seafood does not reflect the bioaccessible fraction. Hg bioaccessibility in raw samples ranged between 60% (yellowfin tuna and black scabbardfish) and 37 % (European conger), with most species presenting a bioaccessibility below 50%. Moreover, after grilling, Hg/MeHg bioaccessibility significantly decreased in all species, ranged between 31% (yellowfin tuna) and 8% (Atlantic wreckfish). The bioaccessibility of Hg and MeHg was also affected by the presence of green tea, decreasing the amount of these contaminants. In raw samples with tea, Hg bioaccessibility ranged between 47% (black scabbardfish) and 26% (swordfish); and between 18% (yellowfin tuna) and 7% (swordfish) after grilling. Green tea significantly decreased Hg/MeHg bioaccessibility in raw samples of yellowfin tuna, common smooth-hound and swordfish, as well as in grilled yellowfin tuna, common smooth-hound, atlantic wreckfish and blue shark. Bioaccessibility variability may be explained by changes in the chemical composition of species during grilling and green tea catechins bioavailability, once they are relatively unstable. This work clearly reveals that green tea is able to reduce Hg and MeHg bioaccessibility, leading to lower the risks associated with seafood consumption. Nevertheless, a better understanding of green tea bioaccessibility is needed, and how its protective effect affects other contaminants. Such information is certainly useful to help consumers to wisely select their food, and to enable food safety authorities to integrate this information in risk assessment and communication activities.

MO346

Importance of a tidal flat-saltmarsh system as a source-sink of mercury in a contaminated coastal lagoon environment (northern Adriatic Sea)

E. Petranich, University of Trieste / Dept. of Mathematics & Geosciences; L. Terribili, University of Trieste; S. Covelli, Dipartimento di Matematica e Geoscienze / Dept. of Mathematics and Geosciences; A. Acquavita, ARPA FVG; E. Pavoni, University of Trieste

Saltmarshes are important constituents of marine-coastal transitional environments that provide several services to the ecosystem. Due to anthropogenic activities, several contaminants are increasingly introduced into these environments where are disperse and can accumulate in the saltmarsh sediments. The Marano and Grado Lagoon (northern Adriatic Sea) is affected by Hg contamination, which mainly coming from historical regional mining activities (Idrija, western Slovenia) and, subordinately, from more recent industrial input due to a chlor-alkali plant. Several studies have also demonstrated the Hg contamination of saltmarsh sediments and halophyte vegetation that cover them. Moreover, saltmarshes not only trap contaminated sediments, but can act as a secondary source of contamination. This work aims at determining the potential role of the tidal flat-saltmarsh (TF-S) system as a sink or secondary source of Hg in this coastal lagoon in relation to the chemical-physical processes leading their remobilisation. The main objective was to understand the role played by periodic flow of tide in a TF-S pilot site in terms of transport, accumulation and release of Hg. Tidal flows and water chemistry were measured at the mouth of a principal tidal creek which collect the waters of a dense channel network draining a 5.5-ha tidal flat-saltmarsh system. Tidal fluxes were estimated by combining discrete hourly tidal flow measurements with weighted measurements of particulate (PHg) and dissolved (DHg) mercury obtained by water samples. The highest values of DHg and PHg were recorded during ebb tide and the quantitative fluxes estimated for all parameters highlighted a tendency for metal to be exported from the TF-S system due to the tidal flows in ebb tide conditions. The results obtained for the PHg fluxes, in particular, are in agreement with those observed on a macro-scale at one of the lagoon tidal inlets considering an annual mass-balance of PHg performed via several water column sampling campaigns. A simple estimation provides a negative sedimentary budget for the TF-S system, which loses PHg towards the main lagoon channel during a tidal semi-cycle thus confirming other evidence of serious morphological deterioration of this critical coastal environment. *Keywords*: tidal flat-saltmarsh system, mercury, tidal fluxes, sedimentary budget.

MO347

Main sources of mercury releases in Armenia

A. Aleksandryan, Hazardous Substances & Waste Policy Division / Head of Division; A. Saghatelyan, Center for Ecological-Noosphere Studies NAS RA; G. Tepanosyan, Center for Ecological-Noosphere Studies NAS RA / Environmental geochemistry department

National mercury releases inventory was done with the use of UNEP's "Toolkit for identification and quantification of mercury releases (January 2013)". The following main sources of mercury releases in the Republic of Armenia were identified: - Coal combustion and other coal use - Combustion of other types natural fuels (petrol, kerosene, diesel, liquid petroleum gas) - Natural gas - Zinc concentrate production - Copper concentrate production - Black copper converter production - Pig iron and steel production - Cement production - Consumer goods and other intentional use (luminescent/fluorescent lamps, thermometers, manometers and gauges, etc.) - Use and disposal of other products - Production of recycled metals - Waste incineration and open waste burning The key mercury releases here are releases to air (the atmosphere), to water (freshwater bodies, including via waste water systems), to land, to general waste, and to sectors specific waste treatment. An additional output pathway is “by-products and impurities” which designate mercury flows back into the market with by-products and products where mercury does not play an intentional role. In 2016-2017 studies were carried out in Vanadzor City of Armenia: at the territory of Chemical Combine and at the adjacent area. The highest content of mercury (3.3 mg / kg) has been recorded in dust of air sampled from the industrial area of the Combine. In air samples from adjacent urban area mercury content made 0.027-3.3 mg / kg.

MO348

Spatial and temporal variation of mercury accumulation in Thelypteris

hispidula in the upper Felidia river basin, Colombia

W. Correa Barragán, G. Duque, Universidad Nacional de Colombia / Facultad de Ingeniería y Administración, Departamento de Ingeniería; P. Cogua, Universidad Santiago de Cali; S. Cuero Salazar, Centro de Diseño Tecnológico Industrial - SENA Regional Valle

The upper basin of the Felidia River, located in the Farallones National Park of Cali, Colombia, is subject to different anthropogenic stressors, such as mercury, the product of illegal mining. Using a direct quantification method (EPA 7473), it was studied the variation of total mercury (HgT) in specimens of the riparian fern *Thelypteris hispidula*, sediments and water in three streams: El Socorro, El Roble and El Pato, during the dry season, dry-rain transition and rainy season. Using non-parametric statistics (Kruskal Wallis), significant differences were found in the HgT concentration at a spatial level in each studied matrix ($p < 0.05$), accumulating mainly in El Socorro. The HgT in the root of the plants presented differences in distinct sampling times ($p = 0.005$), increasing in the rainy season. The Spearman's bivariate correlations showed that the dynamics of HgT accumulation in the root, is directly related to the concentration of HgT in the stem (rho = 0.918, $p = 0.000$) and leaves (rho = 0.900, $p = 0.000$). It was also evidenced that the accumulation of HgT in the root, is influenced by the concentration of HgT in the sediments (rho = 0.764, $p = 0.001$). These results demonstrate the environmental effects caused by mining activities in protected areas in Colombia.

MO349

Temporal integration of diurnal variations of metals and mercury concentrations by passive sampling method in a highly polluted site on the Deûle River, northern France

M. Bretier, Irstea   Centre de Lyon - Villeurbanne; a. dabrin, Irstea / Unité de Recherche Milieux Aquatiques, Ecologie et Pollutions (MAEP); G. Billon, L. Lesven, Université de Lille1, Sciences et Technologies / Laboratoire de Spectrochimie Infrarouge et Raman; G. Grisot, Irstea   Centre de Lyon - Villeurbanne / Unité de Recherche Milieux Aquatiques, Ecologie et Pollutions (MAEP); P. Superville, Université de Lille1, Sciences et Technologies / Laboratoire de Spectrochimie Infrarouge et Raman; L. Dherret, M. Coquery, Irstea Centre de Lyon - Villeurbanne / Unité de Recherche Milieux Aquatiques, Ecologie et Pollutions (MAEP)

Due to several metallurgical plants along the river, the Deûle River is a highly polluted site in the northern France. Previous studies showed that sediments are polluted with Cd, Pb, Zn and Hg (Vdovic et al., 2006). Moreover, intense fluvial traffic generates regular resuspension of sediments leading to diurnal variations of metals concentrations in the dissolved and particulate phases. This was previously highlighted by in situ voltammetry approaches allowing to measure Pb, Zn and Cd with a high frequency data acquisition (Superville et al., 2014). However, these measurements could not be performed for Hg with such probes, while previous field deployment of DGT (Diffusive gradient in Thin films) passive sampler suggested such variations. Indeed, time weighed average concentrations were 20 times higher (20 ng/L) than those measured in grab samples (≈ 1 ng/L). Thus, a field campaign was conducted during 15 days to assess diurnal variations of metals and Hg concentrations (dissolved, particulate and labile phases) related to fluvial traffic in the Deûle River, in the city of Auby, downstream a metallurgical plant (Umicore). During the first week, DGT were exposed and grab samples were collected with a high sampling frequency (night and day). Then, a second set of DGT were exposed during the second week. The resuspension of particles and anoxic sediment caused by fluvial traffic was highlighted by the simultaneous increase of ammonium (< 0.02 µg/L to 0.38 µg/L) and SPM concentrations (3 to 34 mg/L). The analysis of metals and Hg in SPM showed increases of Pb, Zn, Hg and Cd concentrations in the particulate phase. The analysis of particulate Hg after two different filtrations at two cut-off points (0.45 and 0.70 µm) showed that particulate Hg re-suspended by fluvial traffic was mostly in the coarse fraction of SPM. Furthermore, the increase of SPM concentrations was related with an increase in dissolved Hg concentrations. These results suggest that when the anoxic sediment is remobilized by barge traffic, particles could be oxidized inducing a release of Hg. Moreover, since changes in redox conditions could also induce modifications in Hg speciation, further analysis will be carried out to measure Hg^{2+} and CH_3Hg^+. Finally, the interpretation of DGT measurements will show how well DGT integrate variations of inorganic contaminants concentrations during the exposure period.

MO350

The effect of activated carbon amendment on mercury methylation in contaminated sediment

E. Sormo, G. Cornelissen, L. Silvani, E. Eek, Norwegian Geotechnical Institute; H. Veiteberg Braaten, NIVA   Norwegian Institute for Water Research; N.W. Johnson, University of Minnesota Duluth / Civil Engineering

The conversion of elemental and inorganic mercury (Hg) to monomethyl mercury (MeHg) has been widely studied due to MeHg being more bioavailable, bioaccumulative and toxic to humans than the inorganic and elemental species. The net production of MeHg is controlled by both mercury methylation and demethylation, and a range of factors affects both processes. Sediments are known sites for MeHg production, as they are sinks for Hg, they have suitable red-ox conditions, a presence of methylating bacteria and more. Activated carbon has been much explored as a remediation tool for contaminated sediments: The carbon can immobilize contaminants, inhibiting the release to the water column and uptake in biota. Studies have shown that activated carbon also sorb Hg-species, but the mechanisms of how a carbon amendment affects the processes of Hg-methylation in contaminated sediments is not well understood. A lab trial was set up with sediment from two sites in Norway: The Gunneklev fjord (GF) and Bergen Harbour (BH) – two heavily contaminated locations following long histories of industrial activity. Bulk concentrations in the sediment samples were 25.5 and 9.5 mg/kg total mercury (THg) for GF and BH respectively. Two treatments were investigated: Activated anthracite char (AC) and activated biochar (BC). Treatments were set up for time series of 0, 1, 3 and 6 months in sealed glass jars, stored dark at room temperature. At each time series sediment and pore water was sampled. Additionally, THg and MeHg in pore water was measured using DGTs with an aragose diffusion gel and a spheron-thiol resin gel. Pore water data show a net production of MeHg in the GF control, from an initial 8.7 to 393 ng/l within the first month, but it then drops off to 147 and 18.4 ng/l after 3 and 6 months respectively. Compared to the control, an initial reduction of 86% MeHg in pore water is seen for the AC treatment, that increases to >95% for the 1, 3 and 6 month time series. The BC treatment cause an initial 55% reduction of MeHg, but after 1, 3 and 6 months the reduction is >99% compared to the control. In the BH sample, there was no increase of the initial 2.1 ng/l MeHg in the pore water of the control during the 6 months of the experiment. Both AC and BC treatments however, reduced MeHg in the pore water by >50%. Pore water MeHg-concentrations measured by DGT were similar to concentrations in extracted pore water, indicating that pore water MeHg is available for uptake.

MO351

Bayesian Human Health Risk Assessment of Almadén Mining Area

M.F. Ortega, D. Bolonio, C. Rodríguez, M. García-Martínez, Universidad Politecnica de Madrid / Energy and Fuels; J. Esbrí, Universidad de Castilla la Mancha / Geology and Mining Engineering; J.F. Llamas, Universidad Politecnica de Madrid / Energy and Fuels; P. Higueras, Universidad de Castilla la Mancha / Geology and Mining Engineering; L. Canoira, Universidad Politecnica de Madrid / Energy and Fuels

Almaden, with the largest and richest known mercury deposit, is located in the southwest of Ciudad Real (Spain) with a population of 5,657 inhabitants (2016). This area can be considered one of the most affected by mercury in the world, both by its natural origin and by anthropogenic pollution since there are indications that the cinnabar mines of this region have been mined without interruption since before the fourth century BC until 2002. A probabilistic human health risk assessment has been carried out in order to establish whether mercury contamination of Almaden endangers human health, taking into account exposure pathways related to soil, air, vegetable and fish ingestion and an adult resident receptor. The difficulty of the probabilistic analysis is that it is not always easy to obtain distribution functions for the different parameters of a given population, being frequent to use the literature to be able to complete the necessary information. To address this problem, Bayesian statistics have been used. Thanks to that, a combination between stablished density functions (*a priori* distributions) and data collected at the study site can be carried out. In this way, the exposure variables are better defined by *a posteriori*-determined distributions that allow a better estimation of the risk. The results show that the human health risks obtained by ingestion of fish bought in local markets and vegetables grown in the area (lettuce, beans, tomato, onion, pepper, potato, cucumber and zucchini were analyzed) are not acceptable. The Minamata Convention on Mercury entered into force on 16 August 2017. As Minamata Convenction includes a ban on new mercury mines, and the phase-out of existing ones, this methodology could be used to establish if mercury contamination after mercury mines closure around the world endanger human health. E-mail contact: david.bolonio@upm.es, https://orcid.org/0000-0002-9166-1861

MO352

Concentrations of mercury in two offshore skates: sandy ray and shagreen ray

E.E. Nicolaus, Cefas Lowestoft Laboratory / Environment and Ecosystems

Mercury concentrations in muscle and liver tissues from two offshore species of skate were examined. Concentrations of mercury in muscle of *Leucoraja circularis* (n = 20; 23–110.5 cm total length, 157–490 m water depth) and *L. fullonica* (n = 24; 28.5–100 cm total length, 130–426 m water depth) were 0.02–1.8 and 0.04–0.61 mg kg^{-1}, respectively. Concentrations of Hg increased with total length. Only the largest specimen had a concentration of Hg in muscle >1.0 mg kg^{-1}. Data were limited for specimens >90 cm long, and further studies on contaminants in larger-bodied skates could usefully be undertaken.

MO353

EMPIR project "MercOx - Metrology for oxidised mercury"

I. Fettig, Federal Environment Agency (Umweltbundesamt); M. Horvat, Jozef Stefan Institute; I. de Krom, VSL; D. Douglas, LGC; T. Rajamaki, VTT

Mercury (Hg) is one of the most toxic metals, and is regulated by the Industrial Emissions Directive (IED) 2010/75/EU, the Air Quality Directive 2004/107/EC, the Waste Incineration Directive 2000/76/EC and the Minamata Convention adopted in 2013; which is a global treaty to protect human health and the environment from the adverse effects of Hg. In addition to its elemental form Hg also exists in oxidised forms (i.e. Hg(II)) that are reactive and can be transformed into organic Hg species such as methylmercury (MeHg), the most toxic Hg species and the one most prone to bioaccumulation in aquatic systems. Half of atmospheric Hg emissions are of natural origin whilst the rest are of anthropogenic sources, primarily from fossil fuel burning and other high temperature industrial processes (cement clinker production, waste incineration, ore roasting, steel production). Knowledge of Hg speciation both in air and in stack gas emissions is critical when validating models for predicting Hg emissions, transport, deposition and fate at the European level as well as on a global scale. Therefore, atmospheric Hg isotopic signatures that can be used to trace the origin and fate of atmospheric Hg also need metrological support and development. The overall goal of the EMPIR – MercOx project (Oct 17 – Sept 20) is to develop SI traceable measurements, for monitoring and control of mercury and its different species in gas emission sources and in the atmosphere. The project will achieve significant improvements in the measurement comparability and uncertainty of Hg measurement results. Currently, traceable calibration methods only exist for elemental mercury, but such measurements are also needed for oxidised Hg species in order to meet the requirements of EU regulation and the implementation of the Minamata Convention. The development of reliable and direct Hg(II) measurement techniques and reliable and traceable Hg(II) standards is needed to solve the traceability problem that currently exists in the measurement of total mercury (Hg^{tot}) and oxidised Hg concentrations originating from different Hg sources. Furthermore, methods for measuring oxidised Hg and for accurately comparing the Hg^{tot} concentration in generated elemental and oxidised Hg reference gas standards are required, as well as improved sampling methods, traceable reference standards, validated methods for the on-line measurement of Hg under field conditions and a comparison of Hg species inter-conversion.

MO354
PBTK/TD assessment of mercury (Hg(II)) accumulation in freshwater tilapia species

C. Liao, National Taiwan University / Department of Bioenvironmental Systems Engineering; W. Chen, Kaohsiung Medical University / Department of Biomedical Science and Environmental Biology; Y. Yang, National Taiwan University / Bioenvironmental Systems Engineering

Waterborne and dietborne exposures of freshwater fish to inorganic mercury (Hg(II)) affect their growth and reproduction. However, a mechanistic model to predict the impact of Hg(II) on freshwater fish is lacking. The purpose of this study is to develop a physiologically-based toxicokinetic/toxicodynamic (PBTK/TD) model to assess bioaccumulation of Hg(II) in freshwater tilapia. A PBTK model consisted of six interested compartments can be constructed including blood, gill, liver, kidney, intestine, and muscle. The essential physiological and physicochemical parameters can be estimated from published tilapia-related studies. The partition coefficients were estimated for each tissue or organ based on the experimental data by dividing Hg burden in tissues of that in blood at specific days after Hg(II) exposure. A series of experimental data were analyzed to reconstruct the dose-response profiles describing the relationships between tissue/organ-specific burden of Hg(II) and mortality. A four-parameter Hill model was used to describe the dose-response relationships. Here we showed that tissue/organ burdens would reach equilibrium before 180 days of exposure in all six rivers. Among all exposed tissues/organs, kidney had the highest internal exposure doses of Hg(II) ranging from 0.0208 – 0.1348 $\mu g\ g^{-1}$ ww. In contrast, muscle had the lowest internal exposure doses of 0.0001 – 0.0003 $\mu g\ g^{-1}$ ww Hg(II), indicating that Hg levels in muscle might be well below levels considered at risk for human consumption based on regulation from Taiwan FDA. The highest accumulative internal dose of Hg(II) was in gill of 0.0111 (95% CI: 0.0007 – 0.1907). The effective Hg(II) burden in tissue/organ at 50% mortality for liver, gill, and muscle were 10.410 ± 1.047, 6.307 ± 0.756, and 2.839 ± 0.575 $\mu g\ g^{-1}$ ww, respectively. A fair quantitative agreement between model predictions and experimental data was also reached. Sensitivity analysis indicated that the amount of Hg accumulated in tilapia whole body was most influenced by sediment uptake rate, indicating that sedimentborne Hg exposure was the most influential factor on accumulation of tilapia that is bottom-feeding fish. We suggest that more dose-response data of sublethal and chronic effects are required to improve future risk assessment in a more realistic and practical way. In a broader way, our model can be applied to predict continuously chronic Hg accumulation in fish that are deemed safe for human consumption.

MO355
Mercury in fish, fish intake and fish consumption recommendation

H. Coelho Vieira, University of Aveiro; A.M. Soares, F. Morgado, University of Aveiro / department of Biology & CESAM; S. Abreu, University of Aveiro / Dep. Biology & CESAM

Fish consumption is recognized as an important component in the human diet, due to the high-quality protein content and n-3 fatty acids, however, it is also considered the main route of mercury (Hg) exposure from ingestion of contaminated seafood. Hg is released into the environment by natural and anthropogenic sources and recognized as a pollutant of high importance, due its high degree of toxicity, persistence and bioaccumulative properties. Since exposure to mercury poses human health at risk, the Food and Agriculture Organization (FAO)/World Health Organization (WHO), Joint Expert Committee on Food Additives (JECFA) and also by the United States Environmental Protection Agency (USEPA) have been established reference doses (RfD) or "Provisional Tolerable Weekly Intake" (PTWI), in order to minimize that risks. JECFA established a PTWI for MeHg of 1.6 $\mu g\ kg\ bw^{-1}\ week^{-1}$, whereas USEPA pointed a lower value of MeHg intake, setting the RfD at 0.1$\mu g\ kg\ bw^{-1}\ day^{-1}$ (equivalent to 0.7$\mu g\ kg\ bw^{-1}\ week^{-1}$). Recently (2012), PTWI suggested by JECFA for MeHg was revised by the European Food Safety Authority (EFSA) to 1.3 μg MeHg $kg\ bw^{-1}\ week^{-1}$. This study, presents a brief review of the Hg concentration present in the muscle of commercially valuable fish species caught near the mid-Atlantic ridge (Azores Archipelago) where fish consumption e relatively hight (…..) and compares these Hg concentration with the maximum levels of Hg for certain contaminants in foodstuffs established by the European community, evaluates the human exposure to Hg, using the Hg concentration quantified in scalp hair and related with fish consumption using a food frequency questionnaire and establishes isocurves pointing the maximum number of fishmeal per week without exceeding the MeHg RfD (USEPA RfD), by combining number of meals (per week), amount of fish ingested (by meal) and levels of MeHg in fish. The Hg concentration found in the hair indicates that individuals with higher fish consumption per week generally have higher concentrations of Hg and in order to meet the USA dietary guidelines, which recommend a consumption of 227g; only fish with MeHg concentrations below 0.34 $\mu g\ g^{-1}$ could be selected to be consumed so that the PTWI established by JECFA would not be exceeded, despite the concentration of 0.5 $\mu g\ g^{-1}$ (for most of the fish species) or the concentration of 1.0 $\mu g\ g^{-1}$ ("exception list") is allowed for fish consumption.

Mechanistic effect modelling for risk assessment: applications, use in a regulatory context and future directions (P)

MO356
Ring-test of different implementations of the General Unified Threshold model of Survival (GUTS)

R. Ashauer, University of York / Environment; T. Jager, DEBtox Research / Dept of Theoretical Biology

The additional information and insight gained through the application of toxicokinetic-toxicodynamic modelling can strengthen the environmental risk assessment of chemicals in consumer products or plant protection products. For the endpoint survival the most suitable and powerful tool is currently the General Unified Threshold model of Survival (GUTS), which consists of two complimentary models: GUTS-SD (stochastic death) and GUTS-IT (individual tolerance). In order to ease the use of GUTS and increase trust and acceptability we recently carried out a ring-test of eleven different implementations of GUTS. The frequency of erroneous results due to programming errors and less appropriate settings for numerical solvers or parameter search and convergence algorithms indicated that user training and experience is key. However after correction of user errors all software implementations resulted in comparable and similar results. Estimated parameter values generally agreed well and the implementations returned similar results in scenarios mimicking pesticide risk assessment. We suggest that new users and new GUTS implementations should be trained using this ring-test and refer to these results as benchmark. Any new user should run the ring test exercises and improve their modelling techniques until they achieve comparable results. Standardisation of typical use cases could also help to reduce sources of error as well as corresponding, user-friendly, robust GUTS software. This software could reduce sources of error by restricting user-choice to only those options suitable and relevant for the regulatory risk assessment under consideration.

MO357
Feeding impairment in fish explained by a TK-TD model

S. Augustine, Akvaplan-niva; A. Gergs, Bayer AG - Crop Science Division / Department of Environmental, Social and Spatial Change; K. Ladermann, Research Institute gaiac; E. Zimmer, IBACON GmbH; T. Preuss, Bayer Ag / Environmental Safety; V. Ducrot, Bayer Ag / Environmental Safety Ecotoxicology

In current environmental risk assessment (ERA), plant protection products (PPPs) are tested on a diversity of standard test species for harmful effects. Recent developments in mechanistic effect modelling provide the possibility to extrapolate results from standard studies to untested species and untested ecological scenarios or exposure situations, which will improve the quality of ERA as well as saving time and resources. Toxicokinetic-toxicodynamic (TKTD) models for lethal effects have already proven the ability to identify patterns in effects across compounds and species. The Dynamic Energy Budget (DEB) theory may have the potential to provide a general modelling framework for sublethal effects. Models based on DEB theory have been used in ecotoxicology for decades, and these models are currently under discussion as standard approach for risk refinement at the level of tier-2. Models based on DEB theory allow for mechanistic interpretation of effects on feeding, energy expenditure, growth and reproduction. The same modelling framework can be used for all organisms, which is crucial for across - species extrapolation of effects. Many compounds, especially those that target the nervous system, act on the ability of the organisms to feed or assimilate energy. Thus, predicting effects on feeding and assimilation is a crucial characteristic for a TKTD model to predict sublethal effects in the context of ERA. We present here the results of the i-ERA project (integrated ERA) on the responses to low food conditions / feeding impairment in four fish species (rainbow trout, fathead minnow, zebrafish and medaka). We tested the DEB model for predicting organism level responses of juveniles (rainbow trout) and adults (all others) under low food conditions. We find that under low food conditions, fish do not change their metabolism compared to the standard DEB model. This indicates that the model can be used in ERA for the four fish species analyzed here to predict effects of compounds that act on feeding inhibition without any adaptation. The differences in the organism-level response to low food conditions / feeding impairment between the four species can be explained by differences in their model parameters. The standard DEB model can be extended to account for effects to prolonged low food conditions. We suggest the model adaptations needed in such case, and discuss how the model can be used in risk assessments for weight-of-evidence in tier-1 and tier-2 as suggested by EFSA.

MO358

TK-TD modelling as additional line of evidence in the risk assessment for aquatic macrophytes: chlorotoluron as a case study

J. Klein, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Ecotoxicology; U. Hommen, Fraunhofer IME; G. Weyman, ADAMA

To assess effects of the use of plant protection products based on chlorotoluron as active substance, various designs of laboratory tests with *Lemna* sp. and other species and also mesocosm studies including different macrophyte species are available. Since it is not possible to experimentally study every possible exposure scenario, TK-TD modelling was used as an additional approach to address the potential effects of short-term exposure as predicted for some FOCUS surface water scenarios. The *Lemna* TK-TD model developed by Schmitt et al. (2013) was used to simulate laboratory tests assuming exponential growth as observed in the experimental controls. Growth under field conditions was modelled as dependent on time variable temperature and light conditions as well as density dependence. The substance-specific TK-TD parameters were calibrated using the results of a growth inhibition test with 7 days of exposure followed by 7 days of recovery in fresh medium without test item. The so calibrated model was verified by comparing its predictions with results of three other tests with different exposure patterns, some of which were designed with this purpose in mind. Modelling efficiencies were close to or above 0.9 for all four tests and, thus, the model was considered suitable for simulating effects of different exposure patterns on the growth of *Lemna*. We simulated laboratory refined exposure tests with PEC profiles of the 7 days worst-case time window of the FOCUS step 3 scenarios as well as field populations using the full FOCUS profiles as inputs. For the exposure profiles characterized by short-term pulses, margins of safety were above 10 to reach a 50 % inhibition of the growth rate over 7 days, the endpoint used in Tier 1. For the simulated field tests, maximum deviation of biomass under control and exposure conditions was used as assessment endpoint. If up to 25 % deviation of biomass of an exposed population from a control population is considered a negligible effect, the Margins of Safety was above 20 all analysed scenarios. The experimental results and the additional line of evidence provided by the modelling indicate that the exposure profiles considered here will, with a high probability, not lead to unacceptable effects on macrophytes. This project demonstrates the usefulness of modelling as additional tool in risk assessment of plant protection products, particularly for extrapolation between scenarios which cannot all be tested experimentally.

MO359

TK/TD modelling as a tiered approach to reveal interspecies variability of toxicity in fish

F. Gabsi, Rifcon GmbH; T. Preuss, Bayer Ag / Environmental Safety

Species sensitivity distribution (SSD) analysis can be used in higher tier risk assessment to describe the variation in sensitivity of a group of species to a certain contaminant. Contrary to the standard procedure in which toxicity endpoints are derived by considering only effects at the end of a constant exposure experiment, this method has the potential to additionally make use of time-variable exposure and organism response over time. Here, changes in SSD (and the corresponding HC_5 values) with the exposure scenario were investigated in a group of fish species using the toxicokinetic/toxicodynamic (TK/TD) model for survival GUTS. The GUTS model was parametrized based on standard acute tests for each fish species and both stochastic death (GUTS-SD) and individual tolerance (GUTS-IT) assumptions were tested. Then, the 58-day LC_{50}s for constant exposure and for two different pulse exposure scenarios (single and double pulses) were derived. The LC_{50} values were subsequently used as input data for the SSD calculations. The SSDs were derived by fitting probability distributions to the LC_{50} data, and the corresponding HC_5s were determined. The analysis was performed separately for two compounds. Results with both toxicants revealed that the sensitivity ranking for the fish species and consequently the HC_5 values were not the same among the tested exposure scenarios. Predictions with either GUTS-SD or GUTS-IT models also had an impact on the SSDs and did not yield the same results for the same exposure profile. Additionally, longer exposure durations did not always result in lower HC_5s. These findings infer that SSD strongly depends on the exposure scenario, and reveal the interference of substance toxicokinetics and organism responses to toxicity in determining the sensitivity ranking of the species. Therefore, it is essential for a reliable environmental risk assessment not only to consider realistic exposure scenarios, but also the TK/TD processes related to the substance and the organism. With a set of standard data, the GUTS model can help to achieve this goal for untested exposure patterns.

MO360

RIFCON EasyGUTS: Ready-to-use and freely available software for TK/TD modelling of survival

D. Nickisch, O. Jakobi, A. Mediancev, Rifcon GmbH

GUTS (**G**eneral **U**nified **T**hreshold model of **S**urvival) is one of the most commonly used models used for toxicokinetic and toxicodynamic evaluations of aquatic experiments in the context of the European registration of plant protection products at lower tiers in the ecotoxicological risk assessment. One user friendly implementation of this model is the Windows based program EasyGUTS. This implementation and its functionalities were recently tested and verified using published data. Results obtained with EasyGUTS are in good agreement with results obtained from various other publications and model implementations. However, one limitation of the program during this verification process was that it was only possible to select the log-normal distribution for the individual tolerance model rather than giving the possibility to also use other functions like a log-logistic distribution. This was the case since the GUTS R package to which EasyGUTS is linked, is restricted to only this possibility. Since a TK/TD draft guideline is expected earliest next year and no preference for a distribution is given in recent publications, we extended the distribution selection of the R GUTS package and consequently also the selection possibilities in EasyGUTS. The new implementation of EasyGUTS was tested using the aforementioned data and the model implementation was verified using additional data given in recent publications, in line with the EFSA 'Scientific Opinion on Good Modeling Practice'. Moreover, EasyGUTS as a functional tool was tested in internal and external modelling workshops. Our experience is that the usability of the software and the robustness of the calibration algorithm was fitting well, so that even all users could reproduce results and decisions. Since EasyGUTS is finally verified and harmonised with the R GUTS package, it is ready to use under free license agreement and can be downloaded from the RIFCON homepage beginning of 2018. This poster presents the verification process of EasyGUTS and gives insight on the sensitivity of the model to initial parameter values and the influence of different distributions used for the individual tolerance model.

MO361

A new test design to inform TKTD models on species sensitivity

E. Bruns, Bayer AG, Division Bayer CropScience / Ecotoxicology; K. Kuhl, Bayer AG - Crop Science Division; J. Hager, Bayer Ag; T. Preuss, Bayer Ag / Environmental Safety

Recently, several TK/TD population modelling approaches have been developed and are applied in different risk assessment areas. The European Food Safety Authority (EFSA) guidance on tiered risk assessment for plant protection products for aquatic organisms in edge-of-field surface waters, states TK/TD modelling as an approach for the aquatic risk assessment for the evaluation of time variable exposure. For aquatic invertebrates two key questions exists for the ecological threshold option. 1) What is the species sensitivity distribution 2) What are the effects from short-term / peak exposure patterns on long-term survival and reproduction. Currently TK/TD models are parameterised on the standard Tier 1 or Tier 2 datasets. Particularly in chronic studies, test organisms are continuously exposed over long time periods (week to months), which makes these experiments costly, time consuming and which limits the number of non-standard species to be investigated, as these species - in the absence of appropriate husbandry and test methods and due to complex biologies - are particularly challenging to test reproducibly in chronic set-up's. Here we will present an approach using the GUTS model (TK/TD model for survival) informed by specifically designed peak-exposure experiments to answer both questions. We will employ short-term experiments, lasting 48h with two short peaks of 4h duration, at 3 different treatment levels of an insecticide, in combination with several observation time points for 5 aquatic insect species and 3 crustaceans. The outcome of these experiments will inform the GUTS model and will allow for a suitable calibration, after which it will then be possible to construct pattern-specific species sensitivity distributions to be used in acute effect assessments for time variable exposure patterns like FOCUS scenarios. Moreover, the approach may also provide further insights whether peak exposure experiments in an acute study design can be used to investigate sensitivity differences also on a longer-term time scale, by concurrently preventing the pitfalls and potential artefacts arising from not yet developed, adequate long-term husbandry- and test protocols for non-standard species.

MO362

Impact of temperature on species sensitivity distribution in aquatic invertebrates

K. Ladermann, S. Classen, Research Institute gaiac; T. Strauss, M. Hammers-Wirtz, Research Institute gaiac / gaiac - Research Institute for Ecosystem Analysis and Assessment; A. Gergs, Bayer AG - Crop Science Division / Department of Environmental, Social and Spatial Change

Aquatic effect assessment uses results from laboratory experiments at constant environmental conditions. However, for these tests organisms are kept under optimal temperatures which might differ across species. If done for multiple species, results from these toxicity test are use to statistically derive community level endpoints, such as the HC5, from species sensitivity distributions (SSDs). Therefore, data from acute toxicity tests are ranked using cumulative distribution. Apparent toxicity outcomes, such as the LC50s, have been reported to depend on ambient temperature particulary in aquatic invertebrates and other ectotherms and if comparing species sensitivity the results might be biased by the experimental conditions. It has been demonstrated that changes in physiological rates with different temperature regimes can be described by the Arrhenius function. Part of this study is to examine if the Arrhenius function is also able to predict TKTD model rates, such as GUTS, for different temperatures. We use GUTS to extrapolate toxic effects across temperatures and investigate the impact of temperature on a species sensitivity distribution (SSD) with aquatic invertebrates regarding Chlorpyrifos.

MO363

Lemna toxicokinetic and toxicodynamic (TK/TD) modelling - Impact of the ecological scenario on the risk assessment

S. Heine, Bayer Ag / Effect modelling; E. Bruns, Bayer AG, Division Bayer CropScience / Ecotoxicology; T. Preuss, Bayer Ag / Environmental Safety

Recently, several TK/TD population modelling approaches have been developed and are applied in different risk assessment areas. The European Food Safety Authority (EFSA) guidance on tiered risk assessment for plant protection products for aquatic organisms in edge-of-field surface waters, states TK/TD population modelling as an approach for the aquatic risk assessment. Although, the EFSA aquatic guidance states TK/TD population modelling as a method for the risk assessment, there is a lack of guidance's and practical experiences for this new technique – especially to what extent the environmental scenario in which a TK/TD population model is applied influences the outcome of the risk assessment. Unfortunately, it is not obvious which environmental scenario is a conservative one, e.g. a high or a low level of nutrient or temperature. In this contribution, we analysed the sensitivity of a *Lemna* model (Schmitt et al., 2013) to changes in environmental conditions in a risk assessment case study. For this case study we considered exposure to a toxicant and conducted several simulations with the *Lemna* model. While the exposure situation was kept equal in all simulations, the environmental conditions were changed. Results demonstrate that population dynamics are altered the most in cases where the exposure occurred in phases with strong growth of *Lemna*. This analysis can be the basis to set a conservative ecological scenario for environmental risk assessment for *Lemna* TK/TD modelling approaches.

MO364

Defining ecological lake scenarios for population modelling as part of the Ecological Risk Assessment of chemicals

T. Strauss, Research Institute gaiac / gaiac - Research Institute for Ecosystem Analysis and Assessment; A. Gergs, Bayer AG - Crop Science Division / Department of Environmental, Social and Spatial Change; K. Ladermann, Research Institute gaiac; M. Hammers-Wirtz, Research Institute gaiac / gaiac - Research Institute for Ecosystem Analysis and Assessment

The ecological risk assessment of chemicals (ERA) aims to minimize adverse ecological effects on populations and ecosystems. This assessment strongly depends on the selection of the underlying ecological scenarios and the species sensitivity to anthropogenic stressors. This also applies to the populations of planktonic species and fish in standing waters (lentic systems), many of them being focal species in ERA. For use in population modelling, we suggest a classification of ecological scenarios of lentic systems based on the EU Water Framework Directive (WFD). As a result of the European intercalibration process, a list of general lake types has been defined which includes many of the aspects that are important for lake modelling. Besides abiotic characteristics, the German lake classification system for the national implementation of the WFD additionally makes use of biocoenotic and trophic descriptors, and provides short characterizations of typical characteristics for relevant lake types. For the German lake types, data on e.g. phytoplankton biomass and nutrient concentrations are available from natural reference lakes which can serve for model validation. As case studies, we have chosen three lake types from this list of general lake types, which differ in relevant lake properties such as morphometry, trophic state, water depth, stratification regime during summer, and food web structure of the pelagic food web. We additionally considered common anthropogenic lakes and ponds e (i.e. artificial eutrophic and very shallow lakes and ponds) as further scenarios which are relevant for the ERA of chemicals in Europe. For the simulations of these ecological scenarios, the biogeochemical lake model StoLaM was used, in which several phytoplankton and zooplankton groups as well as fish are implemented. Additionally, the one-dimensional vertical structured hydrodynamic model HyLaM as part of the StoLaM allows high resolution of the lake internal physical environment which is required for simulating the nutrient and plankton dynamics in detail. Based on scenario analyses, simulations of typical planktonic dynamics in lake systems will be presented and discussed.

MO365

The use of population models in copper risk assessment: a case study with Acipenser transmontanus

K. Vlaeminck, Arche consulting / GhEnToxLab; K. Viaene, Ghent University / GhEnToxLab; P. Van Sprang, ARCHE; K. De Schamphelaere, Ghent University (UGent) / Applied Ecology and Environmental Biology

Current metal risk assessment consists of assessing single-species data on metal toxicity and constructing a species sensitivity distribution (SSD) for the derivation of safe thresholds. Despite their usefulness, SSDs have been criticized over the last decades for being ecologically unrealistic, and for typically only accounting for individual-level endpoints. Population models as an alternative are becoming more popular in ecotoxicology as they translate a pollutant's effects on individuals (e.g. survival) to the population level (e.g. growth rate). Additionally, ecological models are less expensive and time-consuming to develop and perform research with compared to population experiments. In this study, we aimed at adapting an existing white sturgeon (*Acipenser transmontanus*) population model to predict population level effects of copper toxicity. The white sturgeon is a fish species particularly sensitive to copper during early developmental life stages. An individual-based model (IBM) was implemented using the software platform NetLogo. Copper effects were integrated by adjusting the mortality rate for the sensitive life stage (age-0 individuals) For different scenarios (i.e. environmental configurations, exposure profiles, etc.) population-level effects were assessed as a function of the copper concentration. As expected, population equilibrium density decreased with increasing copper concentrations. Effect concentrations (EC_x values) for population equilibrium density were situated in the same range as (traditional) lethal concentrations (LC_x values) at the individual level. Nonetheless, the magnitude of the population's response to copper depends on several environmental factors such as habitat fragmentation and distribution of the pollution in the river system (random, heterogeneous, or homogeneous). Population EC_x values were derived with the IBM by extrapolating observed (conventional) LC_x values from literature. However, the adapted population model for *A. transmontanus* contains some inherent assumptions which need further fine-tuning. By investigating the mortality profile (i.e. mortality over time) in depth, the mortality sub-model could be improved further, increasing predictability of the model. Additionally, investigating population density-dependent effects on the survival of age-0 individuals could increase accuracy as well. This study shows that population models could be used as more ecologically-relevant tools in metal risk assessment.

MO366

Comparison of toxic effects on Daphnia magna between a metal, a pesticide, and a PAH, in a toxicokinetic-toxicodynamic framework

K. Vlaeminck, Arche consulting / GhEnToxLab; K. Viaene, Ghent University / GhEnToxLab; P. Van Sprang, ARCHE; K. De Schamphelaere, Ghent University (UGent) / Applied Ecology and Environmental Biology

Modelling techniques are becoming more prominent in the risk assessment of chemicals. Mechanistic models, such as individual-based models (IBM) with a foundation in the dynamic energy budget (DEB) theory, are increasingly promoted as alternative tools in ecological risk assessment. In this context, toxicokinetic-toxicodynamic (TKTD) models are often used to describe (sub-)lethal effects on the life cycle of the modelled organism. Inherently, the mode of action will differ between compounds (i.e. compounds will affect different physiological processes). The current study compares TKTD parameters of three different compounds, and examines their influence on the dynamic energy budget (DEB) of *Daphnia magna*. A comparison is made between three model substances: a heavy metal (Cu), a pesticide (endosulfan), and a poly-aromatic hydrocarbon (pyrene). The TKTD model was calibrated for each compound based on life cycle experiments with *Daphnia magna* effects of the three compounds. During life cycle experiments (21-days), growth, reproduction and survival were monitored at different concentrations for each of the compounds. Using all three endpoints, the modes of actions and the TKTD parameters were estimated for copper, endosulfan, and pyrene. Combining the TKTD model with DEB-IBM, effects on physiological processes can be translated to the organism level.

MO367

Deriving predicted no-effect concentrations for perfluoroalkyl acids in the Po river ecosystem through a novel methodology based on the AQUATOX ecosystem model

A. Gredelj, A. Barausse, L. Grechi, L. Palmeri, University of Padua / Department of Industrial Engineering

Health and environmental risks posed by perfluoroalkyl acids (PFAAs) have been rising concerns through the past decades, especially in the most contaminated world areas. One of those is the Northern Italy, because of its high industrialization and population. Nevertheless, the real risk connected to PFAAs as emerging contaminants, both for ecosystems and for human health, is still somewhat unexplored. Linking external exposure to the effective dose of the chemical is one of the main tasks of Environmental Risk Assessment procedures, through the establishment of safe ecological thresholds such as Predicted No-Effect Concentration (PNEC), based on procedures incorporated in the REACH regulation and Water Framework Directive and in related guidelines. These policies offer three methodologies for deriving PNEC: use of assessment factors (AF), species sensitivity distribution (SSD), and results from model ecosystems and field studies whose task is to extrapolate single-species data to ecosystem-level responses. Although AF and SSD methods are described by strict guidelines making them commonly applied, they do not consider the effects of ecological interactions between species on the assessed risk level, which is potentially not-negligible since population dynamics in polluted environment are not only driven by direct toxicity of chemicals on single species. One cost-effective alternative for assessing ecological risk of chemicals considering also indirect ecological effects is the use of mechanistic ecosystem models, simulating the multiple interactions between biotic and abiotic ecosystem compartments. However, there is lack of official guidance for models choice, development and use, resulting in scarce implementation of ecological models for regulatory purposes. Accordingly, two main goals of this work were to develop a methodology for deriving PNEC by the use of the US-EPA AQUATOX ecosystem model, and to evaluate the risk posed by PFAAs (represented by two long-chained and two short-chained compounds) in the ecosystem of the Po, the greatest river in the Northern Italy. Through AQUATOX, water concentrations of PFAAs resulting in a non-negligible biomass loss for each modelled population of the ecosystem were assessed, thus connecting biomass density (a model output) to a “safe” concentration (PNEC). The resulting PNECs were compared to PNECs derived with conventionally used AF and SSD methods to assess the performance of the proposed novel methodology.

MO368

Incorporating spatially explicit metapopulation models as the endpoint of an Adverse Outcome Pathway-based Bayesian Network-Relative Risk Model

J.D. Stark, Washington State University / Dept of Entomology; C. Mitchell, Washington State University / School of the Environment; V. Chu, Western Washington University / Environmental Science; M.E. Harris, Western Washington Unibersity; L. Wallis, Western Washington University / Institute of Environmental Toxicology; G. Young, Advisian WorleyParsons Group / Aquatic Sciences; W.G. Landis, Western Washington University / Institute of Environmental Toxicology; K. von Stackelberg, NEK Associates LTD / Department of Environmental Health

Population viability analysis is useful tool for assessing the environmental risk of toxicants because it produces endpoints relevant to managers and can be manipulated to compare the potential outcomes of conservation actions. In general, many Environmental Risk Assessments (ERAs) lack utility and realism because they fail to incorporate the combined effects of lethal and multiple sub-lethal impacts, environmental stressors, and chemical mixtures into a relevant endpoint for managers. To improve the utility of regional scale risk assessment, we are developing a Bayesian Network-Relative Risk Model that incorporates the combined effects of toxicants and environmental stressors into an Adverse Outcome Pathway (AOP) framework linking environmental conditions to spatially explicit metapopulation models. As a primary case study for this new model, we are examining the impacts of organophosphate (OP) insecticides on ESA-listed chinook (*Oncorhynchus tshawytscha*)and coho (*Oncorhynchus kisutch*) salmon populations using site specific data from the Lower Skagit, Nooksack, Cedar, and Yakima River watersheds in Washington State. The AOP within the BN-RRM links concentrations of OPs to % Acetylcholinesterase (AChE) inhibition which is then linked to sublethal impacts that are incorporated into matrix metapopulation models through age-specific reductions in survival and reproduction. The outcome of this effort will be an adaptable management tool that uses existing, disparate data to link realistic toxicant concentrations to probabilistic population outcomes. The preliminary results of this model development suggest that environmentally realistic concentrations of OPs may slow the growth of salmon populations, undermining the success of current restoration efforts.

MO369

Modeling and monitoring the effects on the central nervous system of a chronic exposure to low dose of pollutants: an innovative strategy with first results

T. Claudepierre, URAFPA-INRA / URAFPA INRA; F. Desor, C. Cakir Kieffer, Université de Lorraine UL / URAFPA INRA; M. DELANNOY, URAFPA-INRA / URAFPA INRA; A. El Hajj, T. Oster, C. Malaplate, Université de Lorraine UL / URAFPA INRA; N. Tran, Université de Lorraine UL / Ecole de chirurgie, Faculté de Medecine de Nancy; F. Yen-Potin, C. Feidt, Université de Lorraine UL / URAFPA INRA

Chronic low dose exposure and possible cumulative effects of various pollutants could affect consumer health and may contribute to the development of neurodegenerative pathologies. Due to the highly complex and long exposure, a clear link between diet contaminants and neurodegenerative diseases has often been suspected, but rarely proven. An additional drawback is the high diversity of *in vitro* models (cancer cell lines, stem cells, primary embryonic cells), bringing additional complexity in the deciphering of the observed effects of pollutants. To properly assess the risks and to reevaluate the maximal acceptable dose of specific pollutants in the food chain, there is a need for efficient modeling of pollutant effects on the central nervous system (CNS). To address this need, we are developing a new approach to evaluate the consequences on neuronal health of long time exposure to pollutants. We are actually re-evaluating the neurotoxic effects of chlordecone (CLD) as proof-of-concept of our strategy. Several concentrations of CLD were used to treat a variety of mouse primary neurons isolated from different postnatal CNS areas. We then assessed neuronal functions using specific markers for neuronal death, neurite development and synapse formation. In parallel, we produced cerebrospinal fluid (CSF) from pigs exposed to CLD via contaminated food. This CSF containing CLD and its by-products that are able to cross the blood brain barrier could then be used on the same cultures to compare its effect with that obtained following direct treatment with the molecule. We were able to identify a direct neurotoxic effect (10 µM) on specific purified neuronal cultures together with more subtle damages at lower concentrations including neurite arborization defects (0.1 to 1µM depending on the respective neuronal cultures). Additional effects on pure glia cell cultures at higher concentrations (100 µM) suggest a reactive gliosis in the whole animal. These observations were confirmed by CSF treatment using CLD-contaminated CSF but not with CSF from control pigs. In addition, the dose with no observable effects is at least 10 times lower using primary postnatal neuronal cultures compared to embryonic cultures. Our experimental model is therefore much more sensitive and may reflect more precisely the consequences of chronic CLD exposure. Our strategy could help to re-evaluate the CNS effects of this remnant pollutant present in West Indies soils.

MO370

A new classification method for mechanisms of toxic action

F. Bauer, KREATiS; P.C. Thomas, CEHTRA SAS / Ecotoxicology and Risk Assessment

A knowledge of the mechanism of action (MechoA) of substances is a crucial first step in risk assessment approaches, especially when using in silico models to predict (eco)toxicity. Mechanisms of Action are similar to Molecular initiating Events which govern molecular interactions between xenobiotics and biological material. Using the accumulated knowledge of MechoAs covering hundreds of molecules, we developed a set of structural alerts associated with specific MechoAs. Consequently, a new method to predict MechoAs with high accuracy and with simple rules was developed, using a MechoA classification with 6 general MechoAs including 23 detailed MechoAs. The MechoAs are given mainly for mammals and fish but information on other species was also included. We used a training set of 301 molecules, and validation set of 491 molecules. Our method was built as a linear decision tree composed of 62 decision rules. This method achieved 92.0% correct classifications for the training set and 92.3% for the validation set. 6% of the predicted classifications were slightly different from the literature MechoAs for the training set (3.4% for the validation set) and 1% of the training set was misclassified (4.3% in the validation set). Finally, only 1% was out of the applicability domain for the training set while no molecules from the validation set were unclassified. This model is both simpler and performs better than the previous model we developed (Bauer et al 2018). We compared this method with Verhaar (as updated in 2008) (Verhaar *et al.*, 1992, 2000, Enoch *et al.*, 2008) and Russom (MOA classification by OASIS implemented in OECD QSAR Toolbox) (Russom *et al.*, 1997) methods, and our decision tree showed the best statistics. This method is currently being implemented into a software, and it will be made freely available and we consider it as a useful support in risk assessment. This model will be continuously enhanced with the addition of new rules and minor corrections as needed.

Biocides and Veterinary Medicines: latest developments in regulatory risk assessment, research and monitoring (P)

MO371

Biocide leaching from building facades: Pseudo-persistence in soil due to reoccurring emissions

U. Bollmann, Aarhus University / Environmental Science; D. Fernández-Calviño, K.K. Brandt, University of Copenhagen / Department of Plant and Environmental Sciences; M.S. Storgaard, Aarhus University, Department of Environmental Science / Department of Environmental Science; H. Sanderson, K. Bester, Aarhus University / Environmental Science

Facade paints and render are commonly protected against biological deterioration using biocides. Mixtures of in-can as well as film preserving bactericides, algeacides and fungicides are added to the materials. Nevertheless, active ingredients leach from the treated facades, if contacted with wind-driven rain. Especially in suburban residential areas a large fraction drains directly to soil, e.g., flowerbeds, gravel strips or the lawns surrounding the houses. Consequently, the soil in areas with biocide-treated buildings is exposed to rain runoff water highly polluted with biocides. In the present study, the degradation rates of eleven biocides in soil were determined in laboratory microcosms. Degradation half-lives ranged from rapidly degrading ($T_{1/2}$ < 10 d) to compounds with higher persistence ($T_{1/2}$ >> 120 d). For two selected biocides (terbutryn and octylisothiazolinone) a set of transformation products were quantified in the microcosms as well. This showed that the mass balance for terbutryn could be closed with nine analysed transformation products for the entire incubation period (120 d), revealing that relative persistent metabolites are formed. In contrary, the mass balance including seven transformation products of octylisothiazolinone was not closed, as transformation products were degraded as well. However, Microtox tests revealed reduced toxicity of transformation products towards *Aliivibrio fischeri* than the respective parent compounds. Nevertheless, for most biocides the degradation half-live is longer than time intervals between rain events in Northern Europe. Hence, though many of the used biocides are degrading relatively rapidly in soil most of the compounds residues may accumulate in soil surrounding biocide treated buildings, due to repeated input with every driving-rain event. Consequently, most biocides can be considered as “pseudo-persistent”-contaminants in this context. This was verified within the present study by (sub)urban soil screening, where concentrations of up to 0.1 µg g^{-1} were detected for parent compounds as well as terbutryn degradation products in soils below biocide treated facades.

MO372

Biocides in facade coatings: Influence of pigments on the phototransformation of biocides

M.M. Urbanczyk, Aarhus University (AU) / Department of Environmental Science (ENVS); U. Bollmann, Aarhus University / Environmental Science; N. Borho, Dr. Robert-Murjahn-Institut; U. Schoknecht, BAM Federal Institute for Materials Research and Testing; K. Bester, Aarhus University / Environmental Science

Biocides are common additives in façade coatings to protect the materials against biological deterioration. In-can as well as film preservatives are used for this purpose. Nevertheless, these biocides leach to the environment when the façade is getting in contact with driving-rain. Long-term exposure tests in natural weather showed large gaps in the mass balances, indicating towards other loss mechanisms. The present study focused on phototransformation as a major pathway for active ingredient loss. In laboratory experiments in UV-weather chambers the formation and fate of phototransformation products were studied. As pigments intensively interact with the spectrum of the incoming light, the effect of pigments on phototransformation rates and pathways of the biocide transformation was studied using red, black, white paints and a pigment-free formulation in comparison. It could be shown, that pigments have a huge influence on phototransformation of biocides. First, pigments are shielding the biocides from phototransformation. Biocides are much faster transformed in the pigment-free formulation, while similar transformation rates can be determined for the red, black and white paint. Second, pigments interact with the biocide´s phototransformation, leading to different transformation pattern with different pigments. The loss rate of the parent for the red and the black paint were nearly indistinguishable, while small differences concerning formation of transformation products were determined for the white paint.

MO373

New Developments in Environmental Emission Scenarios of Biocides - Rodenticides

E. Petersohn, German Environment Agency (UBA) / Section Biocides; K. Wege, A. Friesen, German Environment Agency UBA; M. Anthe, S. Hardt, DR. KNOELL CONSULT GmbH

Rodenticides as biocidal products are regulated according to Regulation (EU) No 528/2012 (BPR). In both frames - evaluation of active substances as well as authorisation of biocidal products – a risk assessment needs to be carried out for human health and environment. The latter is based, inter alia, on Emission Scenario Documents (ESD) providing methods for release estimation of active substances from biocidal products to the environment. In case of rodenticides (product type 14 of BPR), the current available ESD for Rodenticides (2003) has been reviewed to take account of realistic biocidal product applications as well as worst-case environmental exposure assessment. The German Environment Agency (UBA) has commissioned Dr. Knoell Consult GmbH for drafting a revised ESD for PT 14 (rodenticides) on the basis of European Competent Authorities experiences gained during active substance approval and product authorisation, experiences from a workshop on risk mitigation measures for anticoagulant rodenticides, knowledge and common practice of trained pest operators, rodenticides associations, experiences from awarding public and private authorities and furthermore. New scenarios or sub-scenarios have been developed in case of application of rodenticides in sewer systems (with reference to the different types of pipe systems) and of application in and around buildings (distinction between direct applications on paved and unpaved soil; integration of an indoor baiting scenario). A new scenario for bank slopes of water courses has been established, whereas the waste dump/landfill scenario and the open area scenario from the original ESD for PT14 have undergone minor adaptations. When exposure of the terrestrial compartment is considered the transport of biocidal active substances to aquifers and groundwater has to be allowed for. In case of rodenticide application an appropriate approach for estimation of local concentrations in groundwater is newly included in the revised ESD for PT14. The risk assessment for primary and secondary poisoning of non-target organisms was revised in order to provide a more generic approach, i.e. identifying focal non-target organisms. Furthermore, guidance already provided for plant protection products has been considered. The presentation aims at providing an overview of current developments in environmental emission and exposure estimation of rodenticides as biocidal products.

MO374

New Developments in Environmental Emission Scenarios of Biocides - Preservatives for products during storage

K. Michaelis, German Environment Agency (UBA); M. Schwander, German Environment Agency Umweltbundesamt; M. Galler, M. Schweitzer, SCC GmbH

Preservatives for products during storage are also known as in-can preservatives and are regulated according to Regulation (EU) No 528/2012 (BPR). These preservatives are biocidal products which are used in many different end-products (e.g. detergents, paints, glues etc.) to prolong their service life. The environmental emission of these end-products is evaluated with an emission scenario document (ESD), whereas the existing ESD for in-can preservatives does not contain calculations for the variety of all end-products. Consequently, the German Environment Agency (UBA) initiated a research and development project for the further development of the evaluation method of in-can preservatives. The draft for the revised ESD has been prepared by SCC GmbH on behalf of the German UBA. Due to the variety of different applications of in-can preservatives, a differentiation in 6 sub-categories was defined. Additionally, for a complete environmental emission estimation different life cycle steps of the biocidal end-product have to be assessed. Consequently, the incorporation of the in-can preservative into the end-product (formulation) as well as the uses of the end-product (application and service life) within a subcategory have to be considered. To reduce the workload and harmonize the emission estimation it was decided to define emission scenarios which describe a realistic worst-case situation for the environment refer to application amount, emission days and release fractions. On the basis of expert knowledge, draft competent authority reports of in-can preservatives and a survey between stakeholders, industry and other EU member states worst-case scenarios were identified and discussed at EU level. Finally, the revised ESD suggest one or a few worst-case emission scenarios for each subcategory. In addition to the worst-case scenarios, calculation sheets for the estimation of the emission from other uses are provided as Appendices, so that the emission from other end-products (non-worst-case scenarios) can be calculated as well, by using this ESD.

MO375

Monitoring of Biocides in German Sewage Treatment Plant Effluents - First Results

C. Meier, German Environment Agency (UBA) / Biocides; K. Pohl, German Environment Agency (UBA) / Section Biocides; M. Ahting, I. Noeh, German Environment Agency UBA / Biocides; A. Thoma, F. Sacher, DVGW Water Technology Center; M. Kaiser, S. Fuchs, Karlsruhe Institute of Technology KIT / IWG

Due to a widespread use, biocidal active substances and their transformation products are expected to be found in the environment. Projections show that there will be an increase of biocide entries in the environment, mainly in urban areas due to an increased use of e.g. disinfectants and especially masonry preservatives. Biocidal substances enter the environment through numerous entry pathways. One main entry path is through sewage treatment plants (STP). Therefore, the German Environment Agency (UBA) initiated a project where the effluent of 29 public STPs from all over Germany will be investigated over a period of one year, starting in November 2017. Additionally, selected samples from influents as well as from sewage sludges will be in the focus. Using a prioritisation concept for biocides a list, ranking substances that enter the environment through the STP-pathway, was generated. The list was judged by experts and finally, for this project 23 biocidal active substances or transformation products were chosen for analysis. First results show that several substances can be detected at measurable concentrations in the effluents. This ongoing project will provide better knowledge about the fate and behavior of biocides entering the environment through public STPs. It will give us a time dependent picture of the environmental pollution by biocides in Germany through urban STPs and will also show possible fields of action for regulatory purposes.

MO376

The 'risk envelope approach' applied to environmental risk assessments for disinfectants - a strategy to reduce workload for biocidal product families

A. Vanden Bosch, ARCHE; L. Jansen, Arche consulting; S. Navis, Arche consulting / Laboratory of Aquatic Ecology, Evolution and Conservation; E. Van Ael, A. Ghekiere, Arche consulting; F. Verdonck, ARCHE; T. De Wilde, Arche consulting

Under the Biocidal Products Regulation, applicants can apply for authorisation of biocidal product families (BPFs), which consist of products with similar uses, the same active substances, similar compositions within specified variations and similar levels of risk and efficacy. Especially when consortia are formed and products from multiple companies are grouped into a single dossier, building a dossier to demonstrate safe use for all products may become burdensome. Hence, there is a strong need to reduce the amount of risk assessments required to support the BPFs, in the interest of the applicants as well as the competent authorities. BPFs are typically subdivided into subfamilies called ‘*meta* SPCs’. The subgrouping in *meta* SPCs considers a.o. the composition, formulation type, product type (PT), risk mitigation measures (RMMs), classification and labelling (C&L) and shelf-life of the individual products. The parameters that drive the human health and environmental risk assessments, however, most often do not coincide with the factors that determine the *meta* SPC structure. Instead, other grouping strategies are more fit for purpose. The risk envelope approach is a strategy routinely applied in plant protection product dossiers. It entails that - for each area of risk assessment - the key parameters driving that risk assessment are identified. Subsequently, the uses are grouped and ranked according to these key parameters. As such, one or more worst case or 'critical' uses can be identified. If it can be demonstrated that there is no undue risk to men or environment for the critical use, all other uses are considered to be covered as well. A case study will be presented whereby the concept of the risk envelope is applied to the environmental risk assessment for a BPF of disinfectants (PT 1-5). Risk assessments can be grouped (a) for different products/uses within a *meta* SPC, and (b) for different products/uses across *meta* SPCs. Overall, applying the risk envelope approach may lead to a great reduction in workload, whilst allowing for easy addition of products/uses to the BPF at a later stage. The benefits and potential difficulties of this approach will be discussed in detail.

MO377

Are biocide emissions into the environment already at alarming levels? Recommendations of the German Environment Agency (UBA) for an approach to study the impact of biocides on the environment

K. Pohl, German Environment Agency (UBA) / Section Biocides; C. Meier, German Environment Agency (UBA) / Biocides; M. Ahting, I. Noeh, German Environment Agency   UBA / Biocides

More than 40,000 biocidal products were registered on the German market, including disinfectants, preservatives, pest control and antifouling products. All biocides act as intended on living organisms and the use of these biocides can result in alarming impacts on the environment. This has already been confirmed by individual findings of only a few substances, particularly in surface water. However, a comprehensive picture of the actual pollution of the environment with biocides – one that goes beyond such individual findings – is not available, since there is no biocide-oriented, systematic environmental monitoring in Germany to date. To tackle this problem, the German Environment Agency (UBA) has developed recommendations for an environmental measuring programme for biocides based on the results of a research project and two international workshops. These recommendations contain a prioritization concept for biocidal substances as well as a proposal for a systematic monitoring programme. At first, we established a database containing information relevant for the environmental risk assessment according to the Guidance on Biocidal Products Regulation (BPR) for all biocidal substances currently available on the market. A multi-criteria prioritization approach was applied to prioritize substances based on their 1) emission relevance, 2) environmental effect data, and 3) environmental persistence. Thereby creating lists of high-prioritised biocidal substances and relevant transformation products that are of particular concern for the environment. Instead of monitoring individual environmental compartments, our approach aims at monitoring the entry pathway of relevant biocidal substances. Therefore, we developed different entry path scenarios (work packages), which represent the different use pattern and entry paths of particular biocidal products. Based on the obtained prioritised substances and the different entry paths a systematic monitoring strategy is suggested for a German wide inventory of biocides in the environment. This will provide on one hand better knowledge about the fate and behavior of biocidal substances of concern and their impact on the environment. On the other hand, these monitoring data could help to support a more comprehensive risk assessment of biocides by providing a basis for risk mitigation measures or for the exclusion and substitution of environmentally hazardous active substances.

MO378

A case study on exposure assessment of biocides in PPCP using exposure assessment models

M. Kim, H. Kwon, KIST Europe / Environmental Safety Group; Y. Seol, University of Science and Technology; J. Kim, KIST Europe; E. Choe, Korea Institute of Industrial Technology; S. Kim, KIST Europe / Environmental Safety Group

Several accidents caused by the use of chemical products created a need for risk assessment of chemicals used in Pharmaceuticals and Personal Care Product (PPCP) in Korea. CMIT/MIT which is a mixture of 5-chloro-2-methyl-2H-isothiazol-3-one and 2-methyl-2H-isothiazol-3-one is used in PPCPs as preservative and disinfectant. Despite of its inhalation toxicity, this mixture has been used as humidifier disinfectant from the 1990s without considering its exposure route and caused a lot of victims to suffer from its adverse effect. In contrast, CMIT/MIT was detected in toothpaste and the products containing the mixture were recalled in 2016 even though its toxicity via oral route is not known. The aim of study is assessing the exposures of CMIT/MIT in PPCP and comparing different level of exposure models to discuss the applicability of European exposure model in Korea. In this study, the exposure of CMIT/MIT is assessed using European exposure models. New exposure scenarios were developed based on the real use conditions, considering products which contained CMIT/MIT in Korea. Both inhalation and oral route were especially considered in this research under the assumption that the mixture is contained in spray type cleaners and toothpastes and assessed with two different levels of consumer exposure tools. ECETOC TRA.3 was used as tier 1 model which is basic and simple but conservative calculations and ConsExpo was used as tier 2 as it can be more precisely redefined and covers more specific estimations. The gap of estimated exposure values which have been derived from these two different model was identified. It is concluded that difference between exposure calculation equations and input values affects the results. And each level of model has its strengths and weaknesses. Several improvements are needed to apply European models assessing in reflection of Korean exposure scenarios.

MO379

Hazard evaluation of biocides and its metabolites for the aquatic compartment

D. Hernandez-Moreno, INIA / Environment; M. Blazquez, INKOA SISTEMAS / RTD; O. Andreu-Sánchez, Xenobiotics; A. Bermejo-Nogales, M. Fernandez-Cruz, INIA - National Institute for Agricultural and Food Research and Technology / Environment

The LIFE-COMBASE project main target is to promote the sustainable use of biocidal active substances by developing prediction models. As a first step, a database compiling aquatic toxicity data to the aquatic compartment for biocides and derived metabolites has been implemented. The aim of the present study is to make a critical review of this information in order to have a wide view about their potential risk for the aquatic compartment. All the biocides approved and under review were compiled (277 active substances; Regulation (EU) 528/2012). A selection of 192 biocides with possibilities to be modeled and a search using several official and scientific databases, looking for any possible metabolite derived from their release in the aquatic compartment were done. Data was collected in an excel file, including identification of the biocide or metabolite (EC NR, CAS NR, SMILES), classification data (main group, product type, regulatory status), LogP, half-life, degradation reactions and acute and chronic toxicity data for fish, invertebrates, algae and WWTP microorganisms. The EU Regulation (EC) No 1272/2008 on classification and labeling was considered to group these compounds in four toxicity categories taking into account the values of NOEC or $L(E)C_{50}$ as: 1 ($\leq$ 1 mg/L), 2 (>1 to $\leq$ 10 mg/L), 3 (>10 to $\leq$ 100 mg/L) and 4 (>100 mg/L). Most of the found data was related to toxicity in fish, followed by invertebrates and algae, microorganisms being the least studied. There was not reported data for around 80 of the 185 metabolites found, probably due to, in some cases, their commercial unavailability. Another identified problem was that some data were developed with formulated products or with active substances for which purity was not reported. Data already analyzed for the acute toxicity indicated that, 62% of the biocides were located in category 1 for invertebrates, 54 % for fish and 52 % for algae. Only 2 biocides belong to this category for the microorganisms group. Metabolites are mainly less toxic than the parent biocides, however many of them present the same toxicity and very few (< 7%) are more toxic. The ongoing work indicates that biocides and a considerable percentage of their metabolites present a high toxicity for the aquatic species. It also identifies data gaps related to the ecotoxicological potential for metabolites. **Acknowledgement**s: LIFE-COMBASE project (LIFE15 ENV/ES/000416)

MO380

Synchronous decreasing levels of imposex and tributyltin (TBT) in dogwhelk (Nucella lapillus) from Norway, 1991-2015

M. Schøyen, Norwegian Institute for Water Research (NIVA) / Centre for Coastal and Marine Research; L.A. Tveiten, Norwegian Institute for Water Research NIVA / Marine Biology; B.A. Beylich, S. Øxnevad, Norwegian Institute for Water Research NIVA / marine pollution; D. Hjermann, Norwegian Institute for Water Research NIVA / Oceanography; J. Beyer, N.W. Green, NIVA   Norwegian Institute for Water Research / marine pollution

Imposex is TBT-induced development of male sex-characters in female dogwhelk (*Nucella lapillus*). This biological effect is quantified by the *Vas Deferens Sequence Index* (VDSI). Levels of imposex and TBT in *N. lapillus* have been monitored annually in Norway since 1991. Populations around the North Sea were critical reduced in the 1990s, due to increased use of TBT-based antifouling paints. Before the global TBT-ban in 2008, increased TBT-levels coincided with increased imposex prevalence at many monitoring sites located close to high maritime activity. After 2008, decreasing TBT-trends at former impacted sites, lead to population recovery of *N. lapillus*. The observations in *N. lapillus* further corroborated by monitoring data showing decreased TBT levels in blue mussel (*Mytilus* spp.). This monitoring data confirm the rationale of implementing strict international regulations on industrial chemicals when these can be linked to ecological perturbations in coastal ecosystems. The TBT/imposex monitoring was conducted at eight coastal stations representing the Norwegian coast from the Oslofjord to the Varangerfjord, following the guidelines given by OSPAR and ICES. Subsequently, 50 specimens from each station was analysed individually for imposex/VDSI and pooled (only females) for TBT and other organotin like triphenyltin (TPTIN). The levels of imposex (VDSI< 0.828) and TBT (< 6.3 µg/kg w.w.) were low in *N. lapillus* at eight stations in 2015. At most stations, VDSI was 0 or close to 0 and below the OSPARs Background Assessment Criteria (BAC=0.3). The highest level (VDSI= 0.828) was found at the shipping channel Karmsundet, which were above BAC but below the OSPARs Ecotoxicological Assessment Criteria (EAC=2). There were significant downward long-term (whole period 1991-2015) and short-term (recent 10 years 2006-2015) trends for both imposex/VDSI and TBT based on time trend analysis. These results show that the Norwegian legislation banning use of TBT on boats less than 25 m in 1990, on larger ships internationally from 2003, and the total ban in 2008 have been effective in reducing imposex in *N. lapillus* and have re-established some of the populations. Low levels or significant downward long-term and short-term trends for TBT in common periwinkle (*Littorina littorea*) and blue mussel (*Mytilus* spp.) substantiate this.

MO381
Risk assessment issues for algaecides under BPR
C. Durou, M. Darriet, J. Rivera, CEHTRA SAS
A prospective and comprehensive environmental risk assessment (ERA) must be performed on the active substance for the market authorisation of biocidal products. The technical guidelines are made publicly available by ECHA. The data required for the ERA include the determination of a set of properties (physical-chemical, fate properties, short-term and long-term ecotoxicity), an effective in-use dose or concentration, frequency of application of the biocidal product etc… . In coming years, a prospective risk assessment has to be prepared in order to address the risk associated with the use of biocidal containing these active substances and also any substance of concern. The initial approach for a prospective environmental risk assessment will proceed with a Tier 1 calculation, which assumes 100% of the applied chemical will be released and ignores the formation of degradation by products neither biodegradation of the active substance, neither consumption of active substance due to its biocide activity. These initial assumptions may lead to an overestimation of the environmental exposure and risks to the active substance. The initial environmental risk assessment can be refined with supporting data e.g. on the degradation and/or dissipation of the active substance and also by consideration of risk management measures to lower emission of the active substance. In the case biocidal products applied in swimming pools to disinfect or to control algae growth in water, several actives substances are under evaluation or are recently approved in the EU which includes halogenated compounds, inorganic compounds, quaternary ammonium compounds. The present work focuses on the application of the Biocide risk assessment methodology to algaecide applied in swimming pools. The poster will focus on following key aspects: to determine an effective in-use concentration which is an input parameter for assessing the risks associated with the active substance to define the ecotoxicological dataset which is needed in order to determine accurate PNEC values for characterising the risks posed by the active substance to discuss possible options to refine the exposure of environment including new studies and risk management measures

MO382
Could a spatially distributed modelling approach enhance post approval considerations for veterinary medicines?
C. McMillan, G. Hughes, J. Carnall, Cambridge Environmental Assessments
The environmental risk assessment framework adopted in Europe for assessing the surface and groundwater risk from veterinary medicines used to treat livestock follows a tiered approach. The initial exposure assessment is a simplistic approach, with the FOCUS suite of models (FOrum for Co-ordination of pesticide fate models and their USe) often subsequently required for higher tier surface and groundwater refinement. Standard FOCUS scenarios defined within the guidance are intended to represent realistic worst case scenarios for assessing leaching behaviour and surface water risk. However, this approach does not help inform post approval considerations such as identifying the regions most vulnerable to groundwater contamination or surface water risk. In this presentation we consider how this could be achieved for groundwater assessments by using the current Okehampton scenario and comparing it to a spatially distributed version of PEARL at the EU scale, modified for grassland. The spatially distributed version of PEARL was underpinned by an environmental database at 1 km resolution describing (i) land cover using the CORINE 2012 dataset (ii) soils using the newly created 3D soil hydraulic database of Europe (Tóth *et al.*, 2017) and (iii) MARS daily weather. Arable soil parameterisations were modified to represent permanent grassland soils. The spatially distributed PECgw produced were analysed at a range of spatial scales including NUTS2, Member State (NUTS1), pesticide registration zones defined by directive 1107/2009/EC and climatic zones. The results indicate that the current approach used in the registration process for veterinary medicines masks a wide variation in the risk as predicted by a more detailed, spatially distributed approach. This presentation illustrates how a more spatial approach to the environmental risk assessment of veterinary medicines could help provide more clarity on the environmental risk posed by authorised products in regions within Europe, particularly in situations where environmental risks are identified but the veterinary medicine product is approved due to other considerations (e.g. animal welfare). This type of approach could help inform decisions on risk management and facilitate a more targeted approach to ecopharmacovigilance and drinking water protection. Tóth, B., Weynants, M., Pásztor, L., Hengl, T. 2017. 3D soil hydraulic database of Europe at 250 m resolution. Hydrological Processes 31: 2662-2666 \n

MO383
Are currently-adopted European guidelines on veterinary medicine product and feed additive risk assessment sufficiently cautionary?
A. Di Guardo, Universita degli Studi di Milano-Bicocca / Department of Earth and Environmental Sciences; B. Kolar, National Laboratory of Health, Environment and Food; A. Finizio, University Milano - Bicocca / Department of Earth and Environmental Sciences
Veterinary medicine products (VMPs) are used in livestock production to preserve animal health or to promote growth in certain categories of animal; feed additives (FAs) are products aimed at improving the quality of feed and the quality of food from animal origin, or to improve the animals' performance and health. These substances may not be put on the market unless authorisation has been given following a scientific evaluation demonstrating that they have no harmful effects, on human and animal health and on the environment. In particular, according to European Framework Directive 2001/82/EC, the environmental risk assessment (ERA) procedures for VMPs are based on technical guidance documents which propose a tiered approach to calculate PECsoil and PECgw of VMPs from livestock manure spread on the field. On the same way, the ERA procedure for feed additives is reported in a technical guidance document from EFSA which describe a two-tiered approach to calculate PECsoil and PECgw from spread manure. Calculation of PECsoil proposed by the two ERAs in the first tier is directly related to the "annual nitrogen (N) immission standard" which is the amount of nitrogen per Hectare spread on or into the field. Both ERAs propose a default value of 170 kgN Ha^{-1} which is the maximum allowed annual amount of nitrogen originating from animal manure on a farm within nitrate vulnerable zones (NVZ). On the other side, in Europe, NVZs are a minor part of the agriculture areas, hence in several zones higher thresholds of N immission standard are allowed. Both ERAs procedures could therefore underestimate the PECsoil with a potential environmental toxicity for non-target terrestrial organisms. This study is aimed to evaluate if PECsoil, calculated using standard models currently used in the authorization procedures of VMPs and FAs, are sufficiently adequate to protect soil non-target organisms when compared to those obtained using more realistic scenarios of manure applications on soil. As a case study, we considered Lombardia Region in Italy which shows one of highest livestock manure loads in Europe. Information on manure use at field scale were gathered from ValorE system, which is currently used in Lombardia Region to improve the manure management.

MO384
Quick scan to monitoring data of veterinary pharmaceuticals in the Netherlands
S. Kools, T. ter Laak, KWR Watercycle Research Institute
On the Dutch market, approx. 260 active substances are used in different veterinary medicines. In a quick scan, we investigated the potential contribution from farm animals to environmental occurrences of veterinary medicines in Dutch waters to feed possible policy measures. For this, we gathered information from detected compounds in both groundwater and surface water, as these sources are related to drinking water production. From the bulk of the compounds on the market no measurement data are known (84%). Of the 260 compounds used in veterinary medicines, only 42 are measured, and in total 23 substances actually detected. These detected involved 15 antibiotics, four anti-parasitic resources, three anthelmintics and one painkiller. Our quick-scan confirms that a good insight into the presence of veterinary medicines in the water cycle is still lacking. Not only measurement data is limited, it became clear that data on local and regional surface waters is missing or fragmented. We propose inclusion of monitoring data in national and international databases, so data becomes available for other purposes, including prioritization. We also see that current monitoring schemes is not specifically aimed at veterinary medicines, and due to the fact that emissions, temporal and spatial trends, emission routes and concentrations at relevant locations are little available, no clear overview of risks is yet existing. We further noted that the origin of a detected compound cannot always be properly traced back to veterinary use only. We noted that several compounds are also used in human medicines or as pesticides. Admission to the market, based on active substances, is therefore sometimes regulated in different rulemaking and also, usage data is scattered. This makes priority setting difficult when performed in a segregated view on the universe of chemicals

MO385
Comparing methods for estimating environmental emissions
A. Kowalczyk, SC Johnson EurAFNE Limited / Global Safety Assessment & Regulatory Affairs; S.D. Walker, S.C. Johnson & Son, Inc. / GSARA
The environmental risk assessments consist of information on exposure and hazards of chemicals to environmental compartments. Environmental emissions for biocidal products are estimated according to Emission Scenario Documents (e.g. OECD). In some ESDs, the emission is calculated based on the use/consumption of the product as specified on the label (e.g. PT18), with a number of default assumptions applied. However, other ESDs include the facility to take account of tonnage information as well as average consumption values in product specific calculation models (e.g. PT2). Both approaches have strengths and weaknesses. However, the importance of establishing realistic and reliable methods of estimating environmental emissions cannot be understated, especially in light of the intention to develop guidance on aggregate assessment. Accordingly, this poster will illustrate on two approaches for estimating emissions: regional tonnage (top down) or consumption (bottom up). Opportunities and limitations of the applicability of the data and their implications for use in EU environmental exposure assessments will be evaluated.

MO386

SETAC Dung Organism Toxicity Testing Interest Group
J. Roembke, ECT Oekotoxikologie GmbH

Interpretation and uncertainty - overcoming challenges of translating LCA results into reliable information (P)

MO387
Recommendation on Steam Cracker allocation for the sake of comparability of petrochemicals products datasets used in LCA studies
G. Castelan, PlasticsEurope / LCA; P. Saling, BASF SE / Sustainability Strategy
The steam cracker process turns fossil hydrocarbon feedstocks into several different main products, like ethylene and propylene, benzene, toluene, xylene, etc. They are all basic building blocks of many chemicals and polymers used in nearly all products and sectors. Thus LCA data of steam cracker products directly influence a huge amount of further downstream products. It is therefore important that LCA data for steam cracker products are modelled consistently, enabling a reduction of uncertainty and a better interpretation by LCA experts, particularly in perspective of comparability, in LCA studies of these downstream products. Basing on ISO 14044 and on the abundant existing literature on this topic the Life Cycle Thinking and Sustainability working group of PlasticsEurope, composed of experts from its member companies, plus some experts of the Chemical Sector of the World Business Council for Sustainable Development, and some LCA consultants have issued a recommendation built through a consensual 5 years long process. The presentation will elaborate on the discussions and on the recommendation finally issued, considered as the best compromise between comparability and specific representativeness. For multi-output processes, such as a steam cracker, ISO 14040 and 14044 standards define a hierarchy of several options. Due to the nature of steam cracker processes allocation is considered as the preferred option. The concept of defining a main "products" fixed list in combination with a mass-based allocation for steam crackers has led to a consistent LCA approach, independent from market prices, technological changes or market driven adaptations of steam cracker outputs. It gives practitioners a clear guidance for the allocation process. This results in less differing environmental data for steam cracker products and will lead to a higher comparability. The results are much more stable, although the same product will have slightly different LCI results depending on different amounts of products derived from the steam cracker. Such a collaborative work towards streamlining should be engaged for all chemicals, and should be applied within all database both to background and foreground parts, like for example in the European EF compliant database.

MO388
Actual versus default inventory uncertainty in ecoinvent database
F. Belizario, L.A. Oliveira, Institute for Technological Research IPT; M.R. Saade, V. Gomes, University of Campinas UNICAMP; M.G. Silva, Federal University of Espirito Santo; G. Moraga, Universidade Federal do Rio Grande / NORIE; A.B. Passuello, Federal University of Rio Grande do Sul; V.M. John, University of São Paulo USP; O.S. Yoshida, Institute for Technological Research IPT
Variability of national life cycle inventory flows is a relevant uncertainty source and should be properly informed in public databases. Within the scope of the Sustainable Recycling Industries project, life cycle inventories for Brazilian construction products were developed and submitted to ecoinvent following its guidelines, including the preferred use of the lognormal distribution for uncertainty modelling, which requires converting sample average and variation into the geometric mean and the unbiased variance of the underlying normal distribution. However, dataset reviewers inform that most data providers do not perform these conversions and simply use the sample average for flow amounts, frequently associated to default basic uncertainty factors suggested by ecoinvent. This work discusses the implications of three different uncertainty modelling approaches: 1) using both converted mean and variance, 2) using the sample average with the converted variance; 3) using the sample average and default basic uncertainty variance (probably the most common approach). Primary data collected in 25 concrete block factories were used in the analyses. Influence on life cycle impact assessment results was assessed using Monte Carlo simulation with 10.000 iterations, CML 1-A method and ecoinvent v.3.2 "Rest of the World" datasets for upstream processes. Results show that the sample weighted average and the geometric mean differed significantly. Therefore, using the sample weighted average as a proxy for the lognormal geometric mean may overestimate impacts, in our case by aproximately 10%, considering only the effects of the concrete block production process flows. Since existing datasets may have followed this approach, the overall effect is possibly higher. Furthermore, basic uncertainty values are significant lower than measured variations across manufacturing sites, which is inconsistent with a conservative estimation approach. Thus, uncertainty information provided by ecoinvent might contain inconsistencies and lead to errors in uncertainty assessment, such as impact overestimation. Uncertainty modelling can be improved in the database by allowing the input of different amount parameters, performing automatic conversions in the submission software or simplifying the provision of uncertainty data using simpler probability distributions.

MO389
Life cycle assessment of battery systems with harmonized life cycle inventories considering different storage applications
X. Zhang, Paul Scherrer Institute / Laboratory of energy systems analysis; C. Bauer, Paul Scherrer Institute / Laboratory for Energy Systems Analysis; T. Terlouw, Utrecht University / Copernicus Institute of Sustainable Development; M. Beuse, ETH Zurich / Energy Politics Group, Department of Humanities, Social and Political Sciences
The penetration of renewable electricity has greatly increased in the past decade. Battery is a key storage technology to balance supply and demand and to facilitate the worlds's transition towards a sustainable energy system. However, having a comprehensive overview of batteries's life cycle environmental performance still remains a challenge, because battery technologies are of various kinds and the applications of batteries vary. These applications are different from each other in terms of required power and energy size as well as number of cycles. Due to these different requirements by applications, the same battery technology needs to be operated differently and sized accordingly. Numerous studies in the past investigated the life cycle environmental performance of batteries; however, most of them are focused on the application of batteries in electric vehicles, considering a limited number of lithium-ion battery technologies, while the stationary applications of batteries were less explored in limited studies. In addition, these studies are mostly conducted based on diversed sources of life cycle inventory data, without harmonizing the assumptions that are not necessarily different. Peters et al. have recently harmonized the inventory data for several types of batteries, but they are compared without considering the applications. Another study by Baumann et al. considers the applications of battery in the assessment, without addressing the country of application, which results in partial understanding of contributions in the life cycle emissions. This study therefore addresses these challenges, by considering six battery technologies for five storage applications in three representative application countries in Europe. On the basis of previous studies, the harmonization of inventory data is carried out to a greater extent. We also extend the scope of the system, which is often limited to battery pack, to include the complete balance of systems, which ensures the operation required by the applications.

MO390
LCA of nano-adsorbents - Interpretation of laboratory results
A. Kazemi, Tarbiat Modares University / Department of Environmental Science, Faculty of Natural Resources; S.I. Olsen, Technical University of Denmark / DTU Management Engineering Division for Quantitative Sustainability Assessment; N. Bahramifar, Tarbiat Modares University / Department of Environmental Science, Faculty of Natural Resources; A. Heydari, Tarbiat Modares University
Nano-adsorbents as an emerging product and a special application of nanomaterials can increasingly play an important role in the control and removal of environmental pollutants. An example of this is the use of nano-iron to remediate contaminated groundwater. However, even though particularly this example has been successfully applied in real scale, the application of nanomaterials as adsorbents is still an emerging technology at the early stages of development. Hence, this study enables an environmental assessment of nano-adsorbents as an emerging product/technology based on the results from the laboratory. Two nano-adsorbents with graphene-based (MGO-NH-SH) and Fe_3O_4-based (Fe_3O_4@SiO-NH-SH) composites, which function with a similar thiol group for Hg(II) removal are compared at different stages of the production. Removal of mercury is important due to its historic cases of fatal contamination and its continued use. Although mercury must be removed from the contaminated sites it is still very relevant to make an LCA in order to ensure a balance between the impacts of producing the nanoadsorbent versus the avoided impact of the mercury that is being removed. The environmental impacts of synthesised adsorbents including energy use, climate change, water use, human toxicity, and ecotoxicity are investigated by a stepwise procedure during their synthesis processes, regarding their potential to remove mercury from polluted water (functional unit is removal of 1 kg of Hg(II)). Accordingly, characterization results showed that although the process of the functionalization of nanoadsorbents leads to the increase of the adsorption capacity of nanoadsorbents, it is also paired with a significant enhancement of negative environmental impacts. A "what-if" perspective was applied to assess the uncertainties of using lab-scale data for parameters including amounts of acid (HCl + H_2SO_4), ammonia, ethanol, methanol, DCC (N,N'-dicyclohexylcarbodiimide), NHS (N-Hydroxysuccinimide), water recovery, and electricity. The results of t-test comparing the impacts between MGO-NH-SH and Fe_3O_4@SiO-NH-SH estimated approximately 37, 34, 40, 31, and 26% more climate change, water use, energy use, human toxicity, and ecotoxicity, respectively for the latter. Sensitivity analysis were employed to determine the uncertainties for scale-up production and it is shown that especially potential reductions of electricity use, ethanol and DCC can reduce the impacts significantly.

MO391
Quantifying the influence of consumer behaviour on water, energy and greenhouse gas footprints of showering
S. Shahmohammadi, Radboud University / Environmental Science; Z. Steinmann,

Radboud University Nijmegen; H. King, Unilever; H. Hendrickx, Unilever RD Colworth; R. University, Radboud University Nijmegen / Department of Environmental Science
Life Cycle Assessment (LCA) has been used as a tool for environmental footprinting of a wide range of household cleaning activities. Even though differences in the way household activities are performed by consumers may alter the outcome of LCAs, the variability in consumer behaviour is generally ignored in LCAs, which use the average behaviour as basis for quantifying the environmental impacts. The goal of our study was to demonstrate how the data on consumers' reasoned choices, consumers' habits, climatic parameters, manufacturing of products and infrastructure of countries can be combined to quantify the variability in the energy use, greenhouse gas emissions and water footprints related to the life cycle of showering. The impacts of showering were modelled in 4 countries namely Australia, Switzerland, the United Kingdom and the United States using various data sources to quantify the associated variability. Results showed that both inter-country behavioural, climatic and infrastructural differences as well as intra-country variation in consumer behaviour are crucial for determining the variability in the life cycle environmental impacts. Inter-country variability - the ratio between the highest median footprint and the lowest median footprint over the four countries- in the 4 main output variables of the model i.e. energy use, GHG emissions, water withdrawal, water consumption and water scarcity was a factor of 1.5, 2.2, 1.4 and 5.8 respectively. Intra-country variability - the ratio between the 95th percentile and the 5th percentile of the distribution- was typically higher than inter-country variability and ranged between factors of 5 and a factor of 20 depending on the country and indicator considered. Sensitivity analysis showed that consumers' reasoned choices - particularly heater type and shower flow rate- and their habitual behaviours -particularly shower duration-, are the dominant sources of variabilities. Reductions in the water and energy related impacts of showering through changing of reasoned choices are achievable by one-off decisions such as buying an energy efficient water heater. However, reducing the impacts through changing of consumers' habits could be challenging and needs more systematic approaches as consumers tend to keep their old habits.

MO392

Recommendations on the Creation, Management and Use of Data Quality Information for Life Cycle Assessment

A. Edelen, ORISE; W. Ingwersen, US EPA

The average life cycle assessment (LCA) model can combine hundreds or even thousands of data points in order to describe a product system. LCA practitioners and generators are very familiar with the labour and time intensity, which accompanies data collection and processing. While the amount of life cycle inventory data is growing, and there are efforts to improve access to LCA data, questions of 'best fit' data and the appropriate use of results in supporting decision making still plague the LCA community. Ultimately, these are questions of data quality. Updates to the Weidema 2013 pedigree matrix include flow and process level data quality indicators. Five indicators resembling those of Weidema et al. (Weidema et al. 2013) matrix are described at the flow level. Two new indicators are provided at the process level. In previous matrices, all the data quality indicators were not necessarily orthogonal, in that the indicators were capturing overlapping information. In the updated table, all indicators are independent. The adaptation of a framework that contextual data quality continuously changes and must be re-applied situationally, it is important that individual practitioners and data collectors be offered training on the application of a data quality system to ensure representational consistency of data quality results. A needs and capabilities assessment highlighted lack of guidance on documentation and storage ofDQC. Although datasets are documented, the DQC of the original data is either missing or partially stored in the background documentation. The lack of clear documentation of the DQC by generators is a hindrance to the interoperability of the data, since users must search through background documentation and or find original documentation of data in order to perform an evaluation on the contextual indicators. A method for data quality aggregation is proposed that extends earlier work (Rousseaux et al. 2001) to provide aggregate data quality scores for LCIA results. The use of data quality is recommended alongside, and not mixed with, quantitative uncertainty assessment. References [1] Weidema B, Bauer C, Hischier R, Mutel C, Nemecek T, Reinhard J, Wernet G (2013) Overview and methodology: Data quality guideline for the ecoinvent database version 3. The ecoinvent Centre, St. Gallen [2] Rousseaux P, Labouze E, Suh Y, Blanc I, Gaveglia V, Navarro A (2001) An overall assessment of life cycle inventory quality. INT J LIFE CYCLE ASS 6 (5):299-306

MO393

Site-specific N-emissions of rapeseed cultivation in Germany

H. Stichnothe, Thünen Institute / Agricultural Technology; J. Augustin, ZALF; R. Fuss, H. Flessa, Thuenen Institute of Climate-smart Agriculture; R. Ruser, University of Hohenheim; H. Kage, University of Kiel

Globally growing population increases the demand for food, which should be produced as efficiently but also environmentally friendly as possible. Simultaneously climate change requires the reduction of Greenhouse gases (GHG) in order to keep the global temperature increase below 2°C. Germany has defined ambitious goals to reduce its GHG-emissions, The reduction targets are 40% by 2020, 55% by 2030, 70% by 2040 and 80-95% by 2050. Within the last 15 years, GHG emissions from agriculture have not decreased. Simultaneously the European Water Framework Directive requires a good status of water bodies, which is in particular regions in Germany not achieved. Winter oilseed rape (Brassica napus L., WOSR) is the major oil crop cultivated in Germany.Nitrogen field emissions are usually estimated using IPCC-emission factors that are not specific for the crop and associated with strong uncertainty. N_2O field emissions are controlled by N fertilization and dominate the GHG balance of WOSR cropping due to the high global warming potential of N_2O. The same applies for nitrate emissions that dominate the Eutrophication potential or ammonia emissions for the Acidification potential of WOSP when organic nitrogen fertiliser is applied. To address these issues and support decision makers, our project aims to educe specific emissions factors for ammonia and nitrous oxide that can be included in regional-specific life cycle assessment studies for WOSR cultivation.Thus, field experiments were conducted to increase the data basis and subsequently derive WOSR-specific emission factors. Furthermore, the project strives to develop robust but also generalisable statements about nitrous oxide emissions and ammonia volatilisation due to returning digestate from biogas plants to the field using state-of-the-art application methods. The results of the joint research project lead to an improved understanding of trade-offs in the environmental assessment of WOSR production including additional aspects such as fertiliser use efficiency.

The environment as a reactor determining fate and toxicity of nanomaterials (P)

MO394

Ecotoxicity and fate of Ag and CeO2 nanomaterials in outdoor lysimeter experiments

K. Schlich, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology; M. Hoppe, Federal Institute for Geosciences and Natural Resources; M. Kraas, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Ecotoxicology; D. Rückamp, Federal Institute for Geosciences and Natural Resources; K. Hund-Rinke, Fraunhofer IME / Department of Ecotoxicology

Nanomaterials (NM) will enter the environment via diverse pathways. Sewage sludge for example is repeatedly applied as fertilizer on farmland due to its high nutrient content. This may lead to a significant increase of NMs in soil over years. However, there are other scenarios like the exposure of the terrestrial environment via runoff. Therefore, our aim was to investigate the ecotoxicity and fate of CeO_2-NM and Ag-NM under environmentally relevant conditions in outdoor lysimeters over around 2 years (CeO_2-NM) and 3 years (Ag-NMs). Nanomaterials of the OECD Sponsorship Programme, namely NM-212 (CeO_2) and NM-300K (Ag), were used for the experiments. Two concentrations for each CeO_2-NM and Ag-NM were applied via sewage sludge into the top 20 cm of lysimeter soil. In addition, CeO_2-NM were applied via simulated rainfall over four weeks on the surface of the lysimeter soil and afterwards mixed into the top 20 cm to simulate ploughing. Subsamples of the soil were incubated under laboratory conditions for 180 days to study the comparability of outdoor and laboratory results regarding ecotoxicity. The results from our long-term lysimeter experiments showed no detectable horizontal displacement in combination with very low remobilization for both tested NM over 2 to 3 years. Thus, indicate that the sludge applied NM and the NM applied via simulated rainfall remained nearly immobile in the pathway between soils and leachate. However, Ag uptake in the roots of wheat, canola and barley indicates that the chemical conditions in the rhizosphere induce Ag-NM remobilization from the incorporated sewage sludge even after three harvesting cycles. The CeO_2-NM did not induce any adverse effect on the investigated soil microorganisms and the plant growth. At the higher Ag-NM concentration, a constant inhibition of the soil microflora (ammonium oxidizing bacteria and substrate-induced respiration) was observed over about 3 years in the lysimeter study, while there was no effect at the lower Ag-NM concentration. The ecotoxicological results of the laboratory experiment over 180 days reflect the findings of the lysimeter study. For Ag-NM and CeO_2-NM the results indicate that a hazard assessment based on data from laboratory tests is acceptable.

MO395

Long term effects of three different silver sulfide nanomaterials, silver nitrate and bulk silver sulfide on soil microorganisms and plants

K. Schlich, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology; M. Hoppe, Federal Institute for Geosciences and Natural Resources; M. Kraas, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Ecotoxicology; J. Schubert, Leibniz Institute for Polymer Materials; M. Chanana, Institute for Building Materials (IfB); K. Hund-Rinke, Fraunhofer IME / Department of Ecotoxicology

Silver nanomaterials (AgNMs) are subjected to various transformations along their way into the sewage treatment plant (STP). Hereby the AgNMs are mainly transformed to silver sulfides (Ag_2S) (Kaegi et al, 2011). Sparingly soluble Ag_2S is considered as none toxic to soil organisms. In the STP the AgNMs adsorb to sewage sludge (Schlich et al., 2013) and the arising biosolids will be applied in large quantities on agricultural land within the European Union. The main goal of the present study was to determine, if different types of sulfidized AgNMs evoke a difference in the toxicity of the AgNMs. A realistic exposure scenario was chosen. The five test materials NM-300K, previously sulfidized NM-300K, a nanoparticulate Ag_2S, and bulk Ag_2S were added with an influent concentration of 1 mg/L and $AgNO_3$ with an influent concentration of 0.5 mg/L into the denitrification of a simulated STPs continuously for 10 days. The sewage sludge of each treatment was dewatered and the biosolids were mixed with soil. After 0, 60, 90, 140 and 180 days the effects on ammonium oxidizing bacteria (AOB, ISO 15685) and the substrate induced respiration (SIR, OECD 217) were observed. In addition, after 60 days of aging of the AgNM in the test soil a sub-sample was taken from each treatment and a chronic plant test was carried out with oat (*Avena sativa*) and both the roots and the shoots were examined for an uptake of the Ag. We found an increasing inhibition of the ammonia oxidizing bacteria (AOB) from day 60 until day 140/180 in both tests. The inhibition due to the different nanosized AgNMs was mainly comparable throughout the test. In the first test the bulk Ag_2S had no effect on the activity of the AOB. Surprisingly, in the second test we found an effect of the bulk Ag_2S on the AOB, whereas all other results were in good agreement with the first test. The substrate induced respiration (OECD 217) occurred to be a less sensitive test system to determine the effect of the different test materials on the soil microorganisms. Effects were found only after 180 days of the test due to the silver nitrate (70% inhibition) and the nanosized Ag_2S (30% inhibition). There were no effects on the emergence or plant growth of *Avena sativa* over 8 weeks in the chronic plant test. An uptake of a low Ag concentration into the roots of the plants was observed.

MO396

Influence of soil type on the toxicokinetics of Ag and Ag2S nanoparticles and ionic Ag in soil invertebrates

C. van Gestel, Vrije Universiteit Amsterdam / Ecological Science

The rise of nanotechnology and the increased use of nanomaterials in consumer products may lead to an increased emission of nanoparticles (NPs) to the environment. Since NPs may leach from products during use, waste water treatment plants (WWTPs) may be an important sink but also an important source of NP emission to the environment. The use of sewage sludge in agriculture may, for instance, lead to NP exposure in soils. NPs may undergo transformation when passing WWTPs, with sulphidation being an important process. Silver nanomaterials are among the most used, suggesting that Ag-based NPs also will be among the NPs most likely ending up in soils. And considering the transformation processes taking place in the WWTP, Ag_2S may be a form in which the NPs likely will reach the soil. In soil, sorption, aggregation and dissolution processes will determine the availabilty of the NPs or released ions for uptake by organisms. Bioavailabilty will also depend on soil properties that play an important role in governing these processes. This study aimed at assessing the influence of soil type on the bioavailability of Ag and Ag_2S NPs to enchytraeids (*Enchytraeus crypticus*) and springtails (*Folsomia candida*). Four soils with different pH (4-7), organic matter (2-17%) and clay contents (3-13%) were used. An uptake and elimination kinetics approach was taking, in which the animals were exposed to a single concentrations (nominal 10 mg Ag/kg dry soil) for 14 days (uptake phase), after which they were transferred to clean soil for a 14-day elimination period. A first-order one-compartment model was used to calculate uptake and elimination rate constants. Results for the enchytraeids showed k1 values for the uptake of Ag ranging between 0.009 and 0.057 g soil/g animal/day for Ag_2S NPs and of 0.107-0.671 g soil/g animal/day for $AgNO_3$. These data suggest a lower availabiliity of the Ag from the Ag_2S NPs than from the ionic Ag. The k1 values for the uptake of Ag_2S NPs did show a different trend with soil properties than those for $AgNO_3$. Where lowest availabilibity was expected in the soil with the highest cation exchange capacity (CEC), this indeed was the case for $AgNO_3$, but not for for Ag_2S. Elimination rate constant values (k2) ranged between 0.057 and 0.565 per day, and were not dependent on soil type or Ag form. Tests on the springtails are still running.

MO397

Terrestrial isopods as models to assess the biotransformation of nanoparticles inside the organisms: an example with silver and gold nanoparticles

A. Jemec Kokalj, University of Ljubljana, Biotechnical Fac. / Department of Biology; M. van der Zande, RIKILTWageningen University & Research; I. Lynch, University of Birmingham / Geography Earth   Environmental Science; B. Fryer, University of Birmingham / School of Geography, Earth and Environmental Sciences; S. Novak, University of Ljubljana / Biotechnical; D. Drobne, University of Ljubljana / Department of Biology

Physico-chemical properties of nanomaterials, such as their size, shape and dissolution, depend on their environment. Most commonly anticipated alteration of metal based nanoparticles is their dissolution and alteration in size, which are interrelated. Our previous *in vivo* studies with crustacean isopod *Porcellio scaber* have shown that the dissolution of some metal nanoparticles (NPs), such as copper oxide and silver NPs, drastically increase inside the animals. These *in vivo* studies were typically the 14 days feeding experiments and afterwards the total metal content (both NPs and metal ions) was analysed in digestive glands of the animals. With the advancement of analytical techniques, such as single particle (sp)-ICP MS, it is now possible to analyse only the NPs content in the digestive gland and distinguish the signal from metal ions. This also enables to proof whether NPs are formed secondary in the organisms after ingestion of metal salt solution. We present a study where terrestrial isopods were fed silver and gold NPs and their respective metal salt controls via feeding on leaves, and afterwards the NPs and metal content in the digestive gland and rest of the body was measured. In parallel, we also performed an experiment where we exposed the same NPs in simulated *in vitro* invertebrate digestive juice and assessed the dissolution rate using (sp-) ICP MS. Our preliminary data show that there are mostly ions present in the digestive glands, but NPs were also detected when the animals were exposed only to metal solution. This points to the formation of secondary NPs inside the organism. *In vitro* digestive juice model does not entirely represent the expected dissolution rate of NPs that was concluded from *in vivo* exposure. The usefulness of terrestrial isopods as models to assess the transformation of NPs will be discussed. This work was funded under NanoFase project (grant agreement No 646002).

MO398

Energy reserves and respiration rate in the earthworm Eisenia andrei after exposure to zinc in nanoparticle or ionic forms

Z.M. Swiatek, Institute of Environmental Sciences, Jagiellonian Univeristy / Institute of Environmental Sciences; A. Bednarska, Polish Academy of Sciences / Institute of Nature Conservation

The energy budget is an indicator of the organisms' overall condition and the changes in the energy reserves and/or energy consumption rate have been used as biomarkers of toxic stress. To understand better the effect of different forms and concentrations of Zn and possible costs connected with the effective Zn regulation by the earthworm *Eisenia andrei*, we designed a TKTD experiment in which individuals were sampled over time for the available energy reserves (total lipid, sugar and protein contents), energy consumption (measured at both the cellular level and as the whole animal respiration rate) and internal Zn concentration measurements. The earthworms were exposed to $ZnCl_2$ or zinc nanoparicles (ZnO-NPs) in Lufa 2.2 soil for 21 days (uptake phase), followed by 14 day elimination in clean soil (elimination phase). Two concentrations were tested for both $ZnCl_2$ (250 and 500 μg Zn g^{-1} dry soil) and ZnO-NPs (500 and 1000 μg Zn g^{-1} dry soil), corresponding to EC_{25} and EC_{50} for reproduction, plus control without added Zn. The results suggest that the earthworms are able to regulate internal Zn concentrations efficiently, regardless of its form and concentration, without any serious impact on their energy reserves. Sugar content was the only energy reserve component which was significantly lower in 1000 ZnO-NPs than control (p=0.03) in the uptake phase. The total available energy reserves (*Ea*) and protein contents did not differ significantly between treatments but significant effect of day of exposure was found ($p \leq 0.0003$). Neither treatment nor the exposure day affected the lipid content in the uptake phase. In the elimination phase, no treatment or time effect was found on *Ea* or any of its components. The whole-organism respiration rate (measured as oxygen consumption) was not affected by Zn treatments in any of the two toxicokinetic phases. The results for the whole organism respiration rate will be additionally compared with those for the respiration rate measured at the cellular level as an electron transport activity, which is probably more prone to rapid temporal changes in conditions - as is the case for most biochemical biomarkers. The relationships between biomarkers linked to metabolism (i.e. respiration rates measured at different levels) or energy budget (i.e. all energy reserve components) and internal Zn concentrations will be discussed. This study was supported by the National Science Centre, Poland (2015/17/N/NZ8/01576).

MO399

Evaluating the Cellular & Humoral Immune Responses of the Terrestrial Isopod, Porcellio scaber, to Gold Nanoparticles

C.A. Mayall, University of Ljubljana, Biotechnical Fac. / Biotechnical Faculty; D. Drobne, University of Ljubljana / Department of Biology

Gold nanoparticles are popular due to their stability, the ease with which they can be synthesised and the myriads of potential uses they could have, which includes drug delivery systems, cancer therapy and biosensors. It is inevitable that these nanoparticles (NPs) will find their way into the environment and therefore the possible effects they could have need to be evaluated. In particular, it is anticipated that organisms may recognise NPs as "foreign" and respond by modulating their immune system. To date, only a few studies have dealt with this issue. The aim of this study was to investigate whether ingestion or direct intravenous injection of gold NPs alters the immune response of the terrestrial isopod, *Porcellio scaber*. These organisms are well-studied and have previously been used as models for environmental toxicity. As the immune system is an early responder to foreign matter, studying it in conjunction with traditionally used parameters of toxicology can give more information into the possible effects these particles may have. This experiment used two types of gold NPs: one with a sodium citrate coating and another with a PVP coating, both of which were approximately 26nm. For the ingestion route, animals were fed gold NPs for 14 days. During this time the feeding, defecation and survival rates of the animals was recorded. After 14 days, hemolymph was removed and the number, viability and proportion of hemocytes were counted. Along with the cellular tests, the humoral side of the immune system was investigated by measuring the activity of the enzyme phenoloxidase, which is associated with melanisation and wound healing, in the hemolymph. The levels of immune markers, glutathione S-transferase and soluble acetylcholinesterase, were also assayed. As the gut is thought to impede the NPs' ability to journey into the hemocoel, isopods were injected with gold NPs and then left for 48 hours to recover from the injection; in previous experiments 48 hours was shown to be enough time for hemocyte numbers to return to pre-injection levels. The total number of cells, viability and the proportion of hemocyte types were counted. These counts were then compared to the animals which had been fed NPs and to others that had been injected with a non-lethal dose of LPS. Preliminary data shows that the isopods cellular immune response is altered upon direct injection of NPs, but no such effect was found after their ingestion. The study is still ongoing.

MO400

Determining the comparative ecotoxicity of Cd/Te quantum dots with three different functional groups in three species of soil dwelling organisms

T. Botha, North-West University / School of Biological Science; S. Bosch, D. van Rooyen, G. Bouwer, North-West University / Unit for Environmental Science and Management; M. Maboeta, North-West University / Unit for Environmental Sciences and Management; V. Wepener, North-West University - School of Biological Sciences / School of Biological Sciences

Soil is a natural resource that is important for a number of ecological reasons relating to ecosystem and biosphere processes. These processes include plant production, nutrient cycling of organic matter, storage of water and carbon, and richness of pathogens in agricultural crops. Pore water is the interstitial water found between sediment and soil. Soil acts as a biological habitat and gene reserve for a variety of species which are involved in all the soil ecosystem health. Therefore contaminants released into soil can affect the organisms which dwell in them directly affecting soil richness. As nanomaterials are being released into the environment they are able to form complex structures with organic material and soil particles. In order to address the fate and behaviour of Cd/Te QDs three different functional groups (COOH, PEG, NH_3) were used for soil ecotoxicity studies. The earthworm *Eisenia andrei*, pot worm *Enchytraeus albidus and* soil nematode *Caenorhabditis elegans* were used following OECD and ISO protocols to determine comparative ecotoxicity of nanomaterials in soil. The nanomaterial distribution is soil was determined by using a flow through system combined with microwave digestion and ICP-MS where nanomaterials were poured onto soil as well as homogenously mixed and eluted using ultrapure water. It was found that a predominant amount of metals were found within the eluted interstitial water and that NH_3 functional groups had a higher binding affinity to the soil. There was no mortality seen for both earthworms and pot worms exposed up to 500 mg/L over 21 and 28 days respectively. Significant stimulation in reproduction was seen at 5 mg/L for NH_3 and 5 and 30 mg/L in the COOH for earthworms. Pot worms showed an insignificant bimodal response but a significant decrease in reproduction was seen at 5 mg/L in the NH_3 group only. The nematodes showed a significant decrease in reproduction across all exposure concentrations tested (1 – 100 mg/L) within all functional groups. A dose dependent nanomaterial uptake was seen within the tissue of both the pot worms and nematodes but was only observed in the PEG group of the earthworm group. As nanomaterials are released in to the soil environment they exhibit a high mobility within pore water, this mobility is dependent on the functional groups of the nanomaterials release. Soil nematodes show the highest ecotoxicological response compared to earthworms and pot worms and should be used as an indicator species for nanomaterial release.

MO401

Assessment of the differential effects of transformation on the toxicity of nanomaterials with different size and coating propertiesto to soil bacteria and the nematode Caenorhabditis elegans

C. Schultz, Centre for Ecology and Hydrology; J. Adams, NERC Centre for Ecology & Hydrology; S. Lofts, NERC Centre for Ecology & Hydrology / Shore Section; C. Svendsen, CEH, Wallingford / Pollution and Ecotoxicology; D. Spurgeon, Centre for Ecology & Hydrology

Much of the work conducted to-date in nanoecotoxicology has focussed on understanding the toxicity of as-produced nanomaterials. However, environmental fate studies have shown that nanomaterials are frequently transformed in waste streams and natural systems, and that such transformation can modify toxicity. The aim of this study was to understand changes in absolute and relative toxicity of nanomaterials with different starting characteristics; In particular how environmental transformation processes relevant to waste stream and environmental conditions lead to a convergence of nanomaterial characteristics and observed toxicities as compared to those of pristine forms. In order to establish the toxic effect of these materials three bacterial species (*Arthrobacter globiformis*, *Janthobacterium lividium* and *Pseudomonas putida*) were exposed to a range of concentration of nanoparticles with different size and surface properties and their growth inhibition determined. The reproductive toxicity of the selected nanomaterials on the nematode *Caenorhabditis elegans* was also assessed. Investigated were 4 types of silver (25 and 50 nm, uncoated and PVP), 5 types of polystyrene (50 nm unfunctionalised, amine (+ charge), carboxyl coated (- charge) and 100nm, 300nm unfunctionalised), 4 types of TiO_2 nanoparticles (uncoated, PVP, F127, Pleuronic coatings, under dark and light conditions). Initial tests identified effects of particle properties for each core material. Size was found to have the greatest impact on Ag nanoparticle toxicity, whereas surface charge altered polystyrene toxicity the most. In TiO2 nanoparticle exposures uncoated and F127-coated nanoparticles showed the greatest differences in toxicity under dark and light conditions. Thus differences in the toxic effects of the pristine materials were established, although their ranking was not conserved between the species. Studies with chemically transformed or environmentally aged nanomaterials are currently under way to assess whether these differences persist after the silver nanoparticles are sulfidised and the polystyrene and TiO_2 nanoparticles are aged in sewage treatment plant effluent.

MO402

Toxic Effects of Silver Nanoparticles and Its Transformation Product in Soil Applied with Biosolid

E. Topuz, I. Koyuncu, Istanbul Technical University / Environmental Engineering

Biosolids, which are produced as a result of biological wastewater treatment, need to be managed as a separate waste category. Land spread of biosolids to agricultural land, as a resource of nutrients and organic matter, is encouraged under the "*Resource Efficiency Roadmap of Europe"* [1]. However, the presence of contaminats in biosolids such as engineered nanoparticles can cause concerns. Total Ag concentrations in biosolids can be up to 195 mg Ag/kg dry soil in biosolids according to Johnson et al. [2] which is close to observed EC50 concentrations of Ag nanoparticles (AgNPs) [3]. Moreover, AgNPs are mostly transformed to Ag sulphide nanoparticles (Ag_2SNPs) due to the reducing conditions present in the wastewater treatment plant (WWTP) [4]. Recent studies suggest the possibility of AgNP residuals because of the partial sulfidation of AgNPs [5]. Land spread of biosolids might lead to the transfer of AgNPs and Ag_2SNPs to the soil which could pose harm to soil organisms. Hence, this study aims to investigate the toxic effect of AgNPs and its transformation product, Ag_2SNP, on *Enchytraeus crypticus*, which is suitable species for soil ecotoxcity testing. The results are expected to adress the possible threats of AgNPs and its transformation product, Ag_2SNP, on soil ecosystem in the case of their spread via biosolid application. Polyvinylpyrrolidone coated AgNP (AgNP-PVP) and Ag_2SNP are tested in order to investigate their possible different effects on the survival and reproduction of *Enchytraeus crypticus*. $AgNO_3$ is tested as an ionic control for AgNP. Then, AgNP-PVP and Ag_2SNP are mixed at different ratio (1:5, 2:5, 3:5, 4:5) in order to understand the toxic effects of AgNP-PVP in the case of its different transformation rates at WWTPs. Lufa 2.2 soil is mixed with biosolid (500:3) to be used as exposure medium. Biosolid is sampled from a WWTP where anaerobic digestion and sludge drying are used for wastewater sludge management. AgNP-PVP, Ag_2SNP and their different mixtures are spiked to the exposure medium. Survival and reproductive toxicity are determined with the standard toxicity test explained by Castro-Ferreira et al. [6]. Animals and exposure media are analyzed for total Ag concentrations. Therefore, lethal and/or reproductive toxicity are evaluated by considering the Ag concentrations in the exposure media (soil and porewater) and in the animals. Moreover, bioaccumulation potentials of AgNP-PVP and Ag_2SNP in soil organisms are determined. References [1] Ex-post evaluation of certain waste stream Directives Final report European Commission – DG Environment 18 April 2014 retrieved from http://ec.europa.eu/environment/waste/pdf/target_review/Final%20Report%20Ex-Post.pdf [2] Johnson, A.C., et al. (2014). *Chemosphere, 112*, 49-55. [3] Topuz, E. and van Gestel, CAM. (2017). Water Research, 47, 3866-3877. [4] Hennebert, P., et al. (2013). *Waste Manag ,33*, 1870–1881. [5] Kent et al. (2014). *Environ Sci Technol, 48, 8564−8572.* [6] Castro-Ferreira, et al. (2012). Chemosphere, 87,1222–1227. Acknowledgement This study is funded by Horizon 2020 Marie Sklodowska-Curie Actions Individual FellowshipsProject "TESinSAB" (Project Number: 704803) and Istanbul Technical University, Scientific Research Projects Fund (Project # 39961).

MO403

Short-term induced molecular stress responses in coelomocytes of Eisenia fetida earthworms in vivo exposed to silver nanoparticles

s. curieses, CONICET PRIET UNLU; N. Garcia Velasco, E. Urionabarrenetxea, University of the Basque Country UPV/EHU; M. Saenz, PRIET CONICET, National University of Luján; M. Soto, University of the Basque Country UPV/EHU; W.D. Di Marzio, CONICET-PRIET / PRIET

In recent years the production of a great variety of products containing nanoparticles (NPs) has increased massively. The subsequent release of NPs into the environment has created a need to assess the potential ecological risk in soil, water and air. Silver nanoparticles (Ag-NPs) have the highest degree of commercialization due to their high thermal and electric conductivity, high catalytic activity, and powerful antimicrobial properties. *Eisenia fetida* is a model specie in soil toxicity studies and has been broadly used due to its sensitivity to different toxicants at different levels of biological organization. The main aim of the present investigation was to understand the effects produced by AgNPs (5.08±2 nm sized and PVP-PEI coated) in comparison with the soluble form of the metal ($AgNO_3$) at molecular level in coelomocytes of *E. fetida* at different exposure times. *E. fetida* were *in vivo* exposed to different concentrations of Ag-NPs and $AgNO_3$ (0.05 and 50 mg Ag/kg soil) through OECD artificial soil for 1, 3 and 14 d. Then, the transcription levels of selected genes associated to oxidative stress (Catalase) and metal detoxification (MTs-metallothioneins) were determined in coelomocytes extruded from exposed earthworms. In addition, the enzymatic activity (Catalase) and protein content (MTs) were quantified. The responses varied significantly among days, exposure concentration and Ag form. Exposure to Ag-NPs led to a significant induction of CAT at day 1, followed by an increase in its transcription levels after 3 and 14 d of exposure. Similarly, exposure to $AgNO_3$ induced the transcription of CAT at day 1 but at day 14 a downregulation was observed. The CAT activity increased at both treatment and exposure times (1 and 3 d). After 14 d of exposure, CAT activity was inhibited at the highest concentration tested. The highest increase of MTs at protein level was observed after 3 d of exposure. Our results indicate that short-term exposures to Ag-NPs induced early molecular stress responses (MT induction and oxidative stress) in coelomocytes that precede other responses at higher levels of biological organization. The responses in translational level in *E. fetida* tissues were according. The study indicates the importance of using integrative biomarkers for the evaluation of the potential risk of Ag-NPs in soils.

MO404

Effects of Cerium Nanoparticles with deferent surface-charge in coelomocytes of Eisenia fetida

s. curieses, priet conicet; O. Tsyusko, University of Kentucky, Department of Plant and Soil Sciences; J. Li, University of Kentucky, Department of Toxicology and Cancer Biology; M. Saenz, PRIET CONICET, National University of Luján; W.D. Di Marzio, CONICET-PRIET / PRIET; j. unrine, University of Kentucky, Department of Toxicology and Cancer Biology

With the rapid development of nanotechnology and its broad applications, a wide variety of engineered nanoparticles are used in commodities, pharmaceutics, cosmetics, biomedical products and industries. Cerium oxide nanoparticles (CeO_2-NPs) are used in diesel fuels as a combustion catalyst, and as chemical-mechanical planarization agents in production of silicon wafers. This study investigated the toxicity of CeO_2-NPs with polymer coatings in of different charge in coelomocytes of *Eisenia fetida* earthworms. The CeO_2-NPs (2-5 nm primary particle diameter) were coated with dextran to confer a neutral charge (DEX-CeO_2 (0)), diethylaminoethyl dextran to confer a positive charge (DEAE-CeO_2 (+)) and carboxymethyl dextran to confer an negative charge (CM-CeO_2 (-)). The range of exposure concentrations were 0.02-1562.5 mg Ce /L. The coelomocytes were exposed *ex situ* for 1 h for each treatment. Then, the transcriptions levels of genes associated with stress (catalase and heat shock protein 70) were determined by q-RT-PCR. In addition, cytotoxicity and genotoxicity were determined by using tripan blue assay and comet assay respectively. The responses varied significantly among exposure concentration and charge of polymer coatings. The results showed that positively charged DEAE-CeO2 (+) were more toxic that negative and neutral CeO_2-NPs. The results show that initial surface chemistry had a profound impact on the toxicity of CeO_2-NPs to coelomocytes.

MO405

The uptake of pristine and aged silver nanoparticles by wheat, Triticum aestivum, in a soil exposure

A. Green Etxabe, CEH Wallingford; C. Schultz, Centre for Ecology and Hydrology; D. Tarnovska, M. Matzke, NERC Centre for Ecology and Hydrology; D. Spurgeon, Centre for Ecology & Hydrology; C. Svendsen, CEH, Wallingford / Pollution and Ecotoxicology; E. Lahive, NERC Centre for Ecology and Hydrology

It is expected that most nanoparticles (NPs) which reach the soil will not be in their pristine form but instead will have been transformed by the environment (e.g. sulfidisation of Ag in waste water treatment processes). This will greatly influence the form of NP to which soil organisms are exposed and their ultimate bioavailability. The bioaccumulation of NPs inside the organisms can govern their fate and transformation in the environment; and uptake studies can give insight into how organisms act as sources and sinks for NPs in food webs. Most data currently available are for pristine Ag
NPs, and consequently the difference in the bioavailability of the aged forms, predominantly Ag2S, is uncertain. The aim of this study is to compare the uptake kinetics of Ag NPs, both pristine (PVP coated Ag NPs, 20 and 50 nm) and aged (Ag2S, 20 nm), in the crop species, wheat, *Triticum aestivum*. Wheat plants were exposed from seed to each of the NPs at two nominal concentrations of Ag, 3 and 10 mg Ag/kg, in the soil Lufa 2.2. Samples were collected at five time points over the 42 day post-emergence exposure period. The growth rate, Ag accumulation and the translocation from root to shoots were determined. The toxicokinetic parameters of the Ag uptake in the roots and shoots were calculated using total soil concentration and soil pore water concentrations as metrics of exposure. Pore water was collected at all sampling points and at the end of the exposure period pore water was ultra-filtered as a measure of the dissolved Ag in the pore water. The accumulation of all silver forms was greater in the roots, with only a small fraction transported to the shoots. The uptake of Ag2S was lower compared to pristine Ag particles but there was no difference between the uptakes of the two pristine Ag particles. This study shows that environmentally relevant forms of Ag NPs are bioavailable to plants and show different uptake kinetics than the pristine forms.

MO406

In vitro effects on Dendrobaena veneta coelomocytes of Ag and TiO2 nanoparticles before and after wastewater treatment processes

A. Georgantzopoulou, Norwegian Institute for Water Research NIVA; C. Coutris, Norwegian Institute of Bioeconomy Research NIBIO / Dpt for Soil Quality and Climate; K. Ndungu, Norwegian Institute for Water Research; P.A. Carvalho, SINTEF Materials and Chemistry; A. Almeida, Norwegian Institute for Water Research NIVA; A. Macken, NIVA / marine pollution

The majority of nanomaterials (NMs) used in commercial applications are likely to enter the wastewater stream and reach wastewater treatment plants. In many countries, wastewater effluent and sewage sludge are discharged in aquatic environments or applied on agricultural land, however, the transformation of the particles and the potential hazard they pose in these compartments are poorly understood. Recent studies have shown high association of NMs with sewage sludge, therefore soils can be a sink for NM pollution making terrestrial organisms vulnerable. The main aim of the study is to understand the transformation of NMs during wastewater treatment processes and to evaluate the potential environmental hazard of aged particles compared to pristine ones. In this study, coelomocytes (primary immune cells) isolated from the epigeic earthworm *Dendrobaena veneta* are used as a model to assess the effects of Ag and TiO_2 NPs. Initial investigations focus on Ag (PVP coated, 25 nm, nanoComposix) and TiO_2 particles (uncoated anatase, nominal primary size of 5 nm, NM-101, JRC) and their mixture, to better understand their uptake, interaction with coelomocytes and subsequent cellular effects. Moreover, a lab-scale wastewater treatment system is used to study the transformation of Ag and TiO_2 NPs through biological wastewater treatment processes, and the potential effects of the aged particles through biosolids application is evaluated. Extensive characterization of the particles in exposure media is performed with dynamic light scattering (DLS), single particle-Inductively Coupled Plasma Mass Spectrometry (sp-ICP-MS) and transmission electron microscopy (TEM), while sequential filtration/ICP-MS, sp-ICP-MS and TEM are performed on the sludge containing Ag and TiO_2 NPs. The effects of the pristine and aged particles on the metabolic activity, lysosomal integrity, reactive oxygen species formation, immune response and coelomocyte population are assessed. Moreover, nanoparticle uptake and intracellular localisation are evaluated with TEM and sp-ICP-MS.

MO407

Differential biomarker responses of Daphnia magna to pristine and wastewater borne silver nanoparticles

V. Galhano, Department of Biology & CESAM - University of Aveiro / Biology (dBio); M. Monteiro, Aveiro University / Biology; S. Loureiro, Universidade de Aveiro / Biology; S. Hartmann, University of Siegen, Institute of Biology / Department of Chemistry and Biology; K. Witte, University of Siegen / Department of Chemistry and Biology; A.M. Soares, University of Aveiro / department of Biology & CESAM; I. Lopes, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro

The increasing use of silver nanoparticles (AgNPs) as antibacterial agents in a variety of products have raised the need to assess their environmental impact. The ever-growing application of AgNPs leads to their introduction into wastewater treatment plants (WWTPs) via sewer systems. During treatment, AgNPs are mainly retained in sewage sludge but part of transformed AgNPs is released into the environment. This study aims at investigating the effect of pristine and wastewater borne AgNPs on biochemical markers of neurotransmission, oxidative stress and anaerobic metabolism in *Daphnia magna*. Organisms (14-d old) were exposed to 25-125 µg/L of NM-300K for 96-h in a WWTP effluent or in ASTM medium. Daphnids were analised for changes in acetylcholinesterase (AChE), glutathione *S*-transferase (GST), catalase (CAT), lactate dehydrogenase (LDH) activities, and lipid peroxidation (LPO). Results showed a significant increase of CAT activity in effluent-control comparatively to ASTM-control, thus suggesting induction of oxidative stress by effluent. The dispersant used in ASTM (4% w/w of each Tagat® TO and Tween® 20) showed both significant decreases (AChE, GST, CAT) and increase (LDH) of enzymatic activities in dispersant-control relatively to negative-control, suggesting deleterious effects of dispersant to daphnids. Biomarker responses to NM-300K were more marked when added to effluent comparatively to ASTM, especially for higher concentrations. There was a significant decrease of AChE activity in effluent (25 and 75 µg/L) and ASTM (125 µg/L) media, which implies impairment of control and modulation of neural transmission signal in these experimental conditions. The significant increase of GST and CAT activities at 100 and 125 µg/L in effluent, respectively, suggest oxidative stress. The significant increase of LDH activity at 50, 100 and 125 µg/L in effluent suggests an increase in anaerobic metabolism and higher stress for daphnids. Unexpectedly, there was a significant decrease on LPO at 125 µg/L in ASTM, which could be explained by a decrease of synthesis of total lipids. This study shows that (i) the response of biomarkers to used dispersing agent highlights the need for further study on its effects in organisms prior to its application, in order to understand the AgNPs behaviour in standardized test media, and (ii) there is a distinct biomarker response-pattern in daphnids exposed to WWTP effluent containing NM-300K and ASTM supplemented with pristine NM-300K. \n

MO408

Outlining the behaviour and ecotoxicology of biomedical nanoparticles in natural waters

G. Grassi, M. Cuperlo, University of Siena / Department of Physical, Earth and Environmental Sciences; D.R. Hristov, University College Dublin / School of Veterinary Medicine; K.A. Dawson, University College Dublin / Centre for

BioNano Interactions; I. Corsi, University of Siena / Physical, Earth and Environmental Sciences
Engineered nanoparticles (NPs), given the multitude of uses, can be released in aquatic environments, both intentionally and accidentally. To date there is no shortage of studies concerning the environmental fate and ecotoxicity of widely used NPs, such as titanium and silver oxides. However, much less is known about NPs employed in novel cutting-edge applications as nanomedicine. Here, we studied five biomedical NPs, namely aminated polystyrene ($PSNH_2$), europium doped-cerium oxide (CeO_2@Eu), carbon dot-doped silica (Si@C), bare and polyethylene glycol-functionalized silica (SiO_2B and SiO_2PEG, respectively), and we assessed their behaviour and biological impacts in natural river- (NRW) and seawater (NSW). Hydrodynamic sizes were monitored for 30 days by dynamic light scattering (DLS) and showed remarkable differences in NRW compared to NSW of both bare and PEGylated SiO_2 NPs. In fact, SiO_2 NPs dispersions were found to be stable in NRW, while an immediate instability was observed in NSW. PS-NH_2, CeO_2@Eu and Si@C NPs did not show such a clear distinction between the two natural media, reaching micrometric sizes after 24 h. In order to address sedimentation phenomena, normalized derived count rates (DCR) were used to estimate the abundance of suspended NPs in the both media. SiO_2B and SiO_2PEG NPs remained suspended in NRW until 10 days, while in NSW the sedimentation regime was steeper and hardly any signal was collected form suspensions after 24 h. On the contrary, no such difference was observed for $PSNH_2$, CeO_2@Eu and Si@C NPs, which completely settled within 24 h. NPs structural integrity was monitored as well over 30 days by means of spectrofluorometric assays. SiO_2-based NPs underwent disintegration processes in both media, which was confirmed by transmission electron microscopy (TEM) imaging, while $PSNH_2$ maintained an intact structure in NRW and NSW. Finally, algal growth inhibition tests were performed using freshwater and marine microalgae (OECD, 1994). $PSNH_2$ and CeO_2@Eu were toxic in the OECD synthetic freshwater media only, while the remaining NP types did not show any sign of toxicity. A significant ($p < 0.05$) reduction in $PSNH_2$ and CeO_2@Eu NPs toxicity was observed repeating the tests in NRW, while again no toxicity was confirmed in NSW. Altogether, our results provide a realistic insight in the fate and toxicity of diverse NPs, also highlighting the importance of testing complex natural matrices for a more realistic risk assessment.

MO409

Development of a method for the analysis of nanoparticles in the freshwater clam Corbicula fluminea

B. Meisterjahn, Fraunhofer Institute for Molecular Biology and Applied Ecology IME / Ecological chemistry; B. Knopf, Fraunhofer Institute for Molecular Biology and Applied Ecology IME / Environmental Specimen Bank and Elemental Analysis; N. Schröder, Fraunhofer Institute for Molecular Biology and Applied Ecology IME / Ecological chemistry; S. Kühr, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Bioaccumulation and Animal Metabolism; D. Volker, German Environment Agency; C. Schlechtriem, Fraunhofer IME / Department Bioaccumulation and Animal Metabolism
Bioaccumulation studies with manufactured nanomaterials (MNMs) were carried out with the freshwater clam *Corbicula fluminea.* Silver nanoparticles (AgNPs) and TiO_2-NPs were chosen as model particles. The aim of this project was to detect and characterize MNMs in the tissue of animals collected at the end of the exposure period. For the analysis and characterization of nanoparticles in the tissue samples, two promising analytical methods were applied: (i) single particle inductively coupled plasma mass spectrometry (spICP-QQQ-MS) as well as (ii) Flow-Field-Flow-Fractionation (Flow-FFF) coupled to ICP-QQQ-MS. The spICP-QQQ-MS technique uses short acquisition times in the 0.1-10 ms range for the detection of individual particle events during transient analysis and enables the measurement of number-based particle size distributions. Flow-FFF is a separation technique based on the diffusion coefficients of particles and separates particles according to their hydrodynamic diameter without using a stationary phases. Both techniques can deliver complementary information about size and concentration of particles present in the samples. Furthermore to the analytical method a tissue extraction procedure had to be developed to transfer the particles into stable suspension which is a prerequisite of both analytical techniques. Traditional digestion using strong acids likely leads to the dissolution of most MNMs. Therefore, special emphasis was placed on the development of a mild method for the isolation of MNMs from the environmental matrices. Two procedures were applied (i): enzymatic digestion with Proteinase K and (ii): additional alkaline hydrolysis with TMAH. The samples were examined with the former mentioned analytical method. Complete tissue digestion of the mussel samples were achieved with both extraction procedures. For evaluation of the methods, nanoparticles were measured in original samples as well as in spiked samples. The results obtained for the selected model particles are compared.

MO410

The aquatic ecotoxicity of a marketed nanosilver product - a direct comparison with ionic silver

K. Arijs, ARCHE; D. Leverett, wca; K. Oorts, ARCHE; J. Mertens, Precious Metals and Rhenium Consortium c/o EPMF; K. Schlich, M. Brüggemann, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology
As part of the REACH Substance Evaluation for silver, new data was required to be generated to further justify read-across from ionic silver to silver nanoforms. Therefore, the aquatic ecotoxicity and fate and behaviour of ionic silver and the smallest silver nanoform with the highest specific surface area registered under REACH were tested. An ecotoxicity testing programme was undertaken comparing the effects of this silver nanoform with silver nitrate using the following internationally standardised and accepted aquatic ecotoxicity tests: Toxicity to the alga, *Pseudokirchneriella subcapitata* (OECD Test Guideline No. 201). Long-term toxicity to *Daphnia magna* (OECD Test Guideline No. 211). The silver nanoform was fully characterised and was an aqueous suspension containing approximately 37% nanoparticles with spheroidal-like shape (mean primary particle size 9.4 nm). Total silver, 'conventional' dissolved silver (0.45 µm membrane filtered) and 'truly' dissolved silver (3 kDa centrifuge filtered) were measured (ICP-MS) in samples taken from test vessels. Membrane filters (0.45 µm) and centrifuge filters were conditioned before use with the test solution/dispersion to be filtered. Particle size & Zeta Potential were determined (DLS) in vessels without test organisms. Elemental particle size distribution was analysed in separately prepared samples of the test item in test medium by means of asymmetric Flow-Field-Flow-Fractionation (4F) coupled to ICP-MS (for the silver nanoform only). Based on measured silver concentrations, silver nitrate was more toxic than nanosilver to both algae growth and *Daphnia* reproduction, for all silver fractions. Size and Zeta Potential measurements are inconclusive for all tests and it appears that the test concentrations were too low / particles too few to resolve from control / background level (using the Zetasizer Nano equipment). In addition, the dissolution rate of the tested silver nanoform was determined for the specific test media used in the ecotoxicity tests over a period of 28 days (following OECD Test Guideline No. 29), with measurements of the same three silver fractions. This showed a different dissolution behaviour in both test media, which agreed with observations in the ecotoxicity tests.

MO411

Investigations on the uptake pathway and accumulation of silver from manufactured silver nanoparticles in the freshwater amphipod Hyalella azteca

S. Kühr, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Bioaccumulation and Animal Metabolism; J. Klehm, Fraunhofer Institute for Microstructure of Materials and Systems IMWS / Business Unit Biological and Macromolecular Materials; C. Schlechtriem, Fraunhofer IME / Department Bioaccumulation and Animal Metabolism
Testing nanomaterials (NMs) under environmentally relevant conditions is an important aspect regarding the risk assessment of nanomaterials that enter the water cycle. Due to the fact, that sewage treatment plants (STPs) are the main pathway of NMs into the aquatic environment we developed a coupled test system using the effluents of model STPs in a chronic exposure test with the epibenthic amphipod *Hyalella azteca,* which is commonly used for ecotoxicity studies. Previous studies with this test system showed that silver (Ag) from silver NMs is accumulated by *H. azteca* exposed to model STP effluents. However, the pathway of Ag accumulation, via ingestion of particulate Ag and/or bioconcentration of dissolved ionic silver, is still unknown. To further elucidate the uptake pathway of silver from model STP effluent, two groups of *H. azteca* with five animals each were placed in a single test vessel. The two groups were separated by a stainless-steel strainer. One group was fed contaminated sludge from model STPs, loaded on glass fibre filters. The second group, located in the stainless-steel strainer, was fed uncontaminated control STP sludge and had no direct contact to the test sludge containing Ag NMs. The study was carried out with five replicated test vials with two groups of amphipods each. Water samples were taken within the strainers to measure the silver content in the media and to prove that the animals fed control sludge were not in contact with Ag NMs potentially released from the contaminated sludge. After an exposure period of 7 (21) days Ag content of the water and animal samples collected at the end of the exposure period was measured by ICP-MS or ICP-OES to determine the accumulation of Ag in both groups. The presence of NMs in the animals was examined by high-resolution transmission electron microscopy (TEM) and methods of correlative microscopy. The derived accumulation factors and the results of the TEM investigations allow to evaluate the contribution of particulate and dissolved ionic Ag to the accumulation of Ag from STP effluent.

MO412

Ecotoxicity of silica and silver nanoparticles (ENPs) on hyporheic copepods as a function of their bioavailability by dissolved organic matter (DOM) and water hardness of environmental samples

W.D. Di Marzio, CONICET-PRIET / PRIET; M. Saenz, PRIET CONICET, National University of Luján; D. Galassi, Universita dellAquila; J. Alberdi, National University of Lujan; T. Di Lorenzo, Institute of Ecosystem Study CNR
The development and production of engineered nanoparticles has increased the potential for interactions of these nanomaterials with aquatic and terrestrial environments. ENPs are one of the commonly used particles in nanotechnology-based products. They are used in a wide spread application such as chemical industry, cosmetics, medicine and agriculture. The ENPs predicted concentrations using market study production estimates based on life cycle release models and measurements, in biosolids, was 5 - 123 and 0.02 – 2.01 mg/kg and, in

liquid effluents 0.03 - 6.74 and 0.003 – 0.26 µg/L, for Si and Ag NPs respectively. Environmental exposure models have shown that soils and sediments can provide important reservoirs of these nanomaterials, especially in the presence of high concentrations of DOM. These such as the humic substances found in water, sediment, and soil, are ones of the substances capable of interacting with ENPs. To understand and assess the effects of NPs on the environment, should be well established quantitatively the concentration-response relationships. Also, to know what environmental variables can regulate their bioavailability and, thus their toxicity. Designed studies are therefore required in order to understand the fate, transport, stability, and toxicity of nanoparticles. By the other hand, there are not many studies about the effect of ENPs on hyporheic copepod species and less related with DOM concentrations. The hyporheic zone is a region underneath streambed that integrates surface and groundwater. Its location is central to biogeochemical linkages between the riparian zone, dissolved nutrients, and benthic biota. Even if in this DOM sources are relatively constant, biogeochemical processing within the hyporheic zone resulted a DOM pool that is temporally dynamic regarding its composition and concentration. In this study we evaluated how DOM concentrations and water hardness are related with the acute ecotoxicity of Si and Ag NPs on the survival of *Metacyclops gracilis* a widespread hyporheic species. Toxicity of AgNps was related with DOM concentrations and showed a non-significative *Beta* for water hardness. On the contrary, for SiNps, DOM and water hardness quantitative relationships were negatively correlated with ecotoxicity on this freshwater invertebrate.

MO413

Long-term exposure of ZnO nanoparticles to freshwater microalgae cultivated in batch and semi-continuous mode

A.F. Aravantinou, F. Andreou, I. Manariotis, University of Patras / Civil Engineering

Nanoparticles (NPs) have always existed in the physical environment. The rapid development of commercial applications involving the use of a large variety of synthetic nanoparticles has resulted in the introduction of higher amounts of nanoparticles in the environment. As the use of NPs increases, their effect to the coastal food chain and ecosystems is crucial. The aim of this work was to investigate the toxic effect of zinc oxide (ZnO) NPs on freshwater microalgae in batch and semi-continuous feeding mode for longer period than the time used in typical toxicity tests. *Scenedesmus rubescens* was selected as model microorganism since it is a common freshwater microalgae. *S. rubescens* exposed to ZnO NPs concentrations varying from 0.081 to 810 mg/L for 28 days in batch mode conditions, while in semi-continuous mode it was exposed to 0.081mg/L of ZnO NPs. The cultures were grown in modified Blue-Green 11 medium (BG-11). The effect of ZnO NPs on microalgae was assessed by the determination of growth rate, nutrient removal and lipid production. The toxic effect of ZnO NPs was estimated by the half maximum inhibitory concentration (IC_{50}) and the growth inhibition rate (I%) according to OECD 201 guideline. The experimental results in the batch mode conditions revealed that microalgae growth was significantly affected by the exposure time and the NPs concentrations. Specifically, the results showed that after a period of time the microalgae were adapted in the presence of ZnO NPs and were more resistant. In the semi-continuous mode the growth of *S. rubescens* was greater in the presence of ZnO NPs, and the lipid content was higher.

MO414

Effects of sunscreen-derived TiO2 nanoparticles on freshwater and marine organisms

s. schiavo, ENEA CR; M. Oliviero, University Parthenope; A. Philippe, Universität Koblenz-Landau / Institute for Environmental Sciences Group of Environmental and Soil Chemistry; s. manzo, ENEA / SSPT-PROTER-BES

Sunscreens represent one of the main source of engineeredTiO_2nanoparticles (TNPs) source in coastal ecosystems, especially during summer period.Their adverse effects were generally investigated using bare model TNPs and only few studies were based on the NPs extracted fromcommercial products.Therefore, this study aims to evaluate the effect of TNPs extracted from threedifferent commercial sunscreens upon freshwater and marine organisms: microalgae (*Pseudokirchneriella subcapitata; Dunaliella tertiolecta)* and crustaceans (*Daphnia magna*; *Artemiasalina*). Microalgae are suitable indicator for marine water pollution and because of they are at the base of the aquatic food web, any modification of their growth could affect higher trophic levels such as zooplancton. In order to understand the effective role of the other sunscreen components in the toxic action of the TNPs, TNPs directly purchased from the industry (ITNPs), TNPs extracted from sunscreens and the whole sunscreens werealso tested Preliminarily results showed thatfor microalgae, bothfreshwater and marine ones, the toxic effect exerted by the whole sunscreen was lower in respect to the toxicity of extracted $TiO_2$and of TiO_2 industrial nanopowder. In *D. magna* instead the LD_{50} of particles extracted from one sunscreen was lower than values previously determined for ITNPs. No significative differences between tested substances were highlighted with the acute test upon *A. salina*. These findings suggest that the product formulation may mitigate the toxic effects of TNPs either by direct modification of TNP properties (reactivity, bioavailability) or by providing organic and inorganic nutrients promoting phytoplankton and hence microalgae growth. Moreover the organic UV filters could limit the amount of light that the particle receive and thus limit the photocatalytic activity. Our preliminary results then showed a different TNPs toxicity in the two ecosystems: in particular,the TNPs different physico-chemical behaviour and reactivity depending on testing environmental media and even the specific interaction with organisms should be taken into account in designing the experimental assessment. Further studies are needed to better understand the real availability of the NPs for organisms also taking into account the UV radiation.

MO415

Silver nanoparticles affect the early development of Tisbe battagliai: pristine vs aged particles

A. Georgantzopoulou, Norwegian Institute for Water Research NIVA; J. Farkas, SINTEF Ocean / Environmental Technology; K. Ndungu, Norwegian Institute for Water Research; P.A. Carvalho, SINTEF Materials and Chemistry; A. Booth, SINTEF Ocean / Environmental Technology; A. Macken, NIVA / marine pollution

Silver and titanium nanoparticles are used in numerous consumer products and applications and they are likely to enter wastewater streams, reach wastewater treatment plants and aquatic systems through wastewater and effluent discharge. Nanomaterials undergo transformations in different matrices altering their fate, behaviour, bioavailability and toxic potential that could differ greatly from the pristine counterparts. There are challenges on the detection and quantification of nanomaterials at environmentally relevant concentrations in complex media and matrices such as whole organisms. Morevoer, studies on uptake and effects of transformed particles on marine species is largely lacking. The aim of the present study is to better understant the transformation of Ag and TiO_2 NPs in wastewater and assess the uptake, bioacumulation and subsequent developmental effects of pristine and transformed particles on the marine organism *Tisbe battagliai*. In this study the harpacticoid copepod *Tisbe battagliai* was selected as a relevant marine species and the effects on the naupliiar development was assessed over an exposure period of 6 days. The mortality and the developmental stage of the organisms was assessed daily. Initial investigations focus on the study of Ag (PVP coated, 25 nm, nanoComposix) and TiO_2 particles (nominal primary size of 5 nm, NM-101, JRC) and their mixture. Moreover, the particles were aged in synthetic wastewater (for 4 hours under stirring conditions) and the effects of "aged" particles on the uptake, bioacumulation and naupliar development was assessed. Extensive characterization of the particles in synthetic wastewater, seawater and exposure media was performed with dynamic light scattering (DLS), ultraviolet–visible spectroscopy (UV-VIS), single particle-Inductively Coupled Plasma Mass Spectrometry (sp-ICP-MS) and transmission electron microscopy (TEM). Moreover, nanoparticle uptake, bioacummulation and intracellular localisation is evaluated with TEM and sp-ICP-MS. Results showed that the both particles types were stable in the synthetic wastewater during the 4 h aging time. In contrast, particle concentrations decreased over time in seawater, which was likely due to both dissolution and aggregation for Ag, and aggregation in TiO_2. TiO_2 particles were found to be more stable in seawater after the aging process, which can affect their impacts on exposed organisms.

MO416

Silver concentration in the haemolymph of a tropical marine amphipod fed with silver nanoparticles and silver chloride

M. Vannuci-Silva, UNICAMP / Institute of Biology; S. Cadore, University of Campinas; G. Umbuzeiro, School of Technology, UNICAMP / LAEG

The relatively recent development of engineered Ag nanoparticles has expanded silver uses considerably. Silver nanoparticles (AgNP) tend to agglomerate in the aqueous phase and settle to sediment surfaces exposing deposit feeding organisms. Amphipods, like *Parhyale hawaiensis*, are deposit-feeding species ecologically relevant, becoming an excellent model for ecotoxicology studies. In addition to the traditional toxicity studies, internal doses determination, for example in the haemolymph, can provide information on the level of exposure to toxic metals. The aim of this study was to investigate Ag concentration in the haemolymph of the marine amphipod *Parhyale hawaiensis* exposed to food containing AgNP and AgCl. We hypothesized the actual AgNP could be absorbed by the gut leading to a higher amount of Ag in the haemolymph when compared to food containing AgCl. Silver nanoparticles < 100nm (Sigma Aldrich) or elemental Ag (from AgCl, Sigma Aldrich) were incorporated into formulated fish feed pellets in a concentration of approximately 200 mg kg^{-1}. *P. hawaiensis* organisms (8 months) were placed individually into a plastic container (100 mL of reconstituted saline water) and fed on alternate days with control, AgNP, or AgCl amended feed pellets. After 1 hour of feeding, each organism was washed and placed into a new plastic container with clean salt water to ensure that the exposure was only via food. The amphipods were exposed during 7, 14 and 28 days. After exposure, the haemolymph was collected using a thin glass capillary, weighted and analysed. Three pooled samples of 4 organisms (2 females and 2 males) were tested per exposure concentration. The silver determination in haemolymph was carried out by a Graphite Furnace Atomic Absorption Spectrometer (GFAAS). A higher amount of silver in the haemolymph was absorbed from AgNP feed, reaching 8.4 ± 0.7 ng mg^{-1}, in comparison to 3.7 ± 1.0 ng mg^{-1} for AgCl, at the longest exposure time. The increase of silver concentration was related to the exposure duration. Therefore, it appeared that ingested silver is more bioavailable to *P. hawaiensis* as AgNP than when it is in its salt form. Data strongly suggest that nanoparticles were uptaken by the gut and distributed in the haemolymph causing this increase in Ag content. More studies are required to verify the Ag form present in the haemolymph and how it will induce damage in the exposed organisms.

MO417

Toxic effects of multi-walled carbon nanotubes on bivalves: comparison between functionalized and nonfunctionalized nanoparticles

L. De Marchi, University of Aveiro, Department of Biology & CESAM / Departamento De Biologia & CESAM; V. Neto, Department of Mechanical Engineering & Centre for Mechanical Technology and Automation (TEMA), University of Aveiro 3810-193, Portugal; C. Pretti, Department of Veterinary Sciences, University of Pisa, San Piero a Grado; E. Figueira, University of Aveiro / Biology CESAM; F. Chiellini, Department of Chemistry and Industrial Chemistry, University of Pisa; A. Morelli, Department of Chemistry and Industrial Chemistry, University of Pisa, Udr INSTM Pisa, Pisa 56126,; A.M. Soares, University of Aveiro / department of Biology & CESAM; R. Freitas, University of Aveiro / Departamento de Biologia CESAM

The use of carbon nanomaterials (CNMs) has increased rapidly in the last years, namely due to their important properties such as electromagnetic, optical, catalytic, mechanical, thermal, and pharmokinetics. Currently, carbon nanotubes (CNTs) are one of the most important and commercially used CNMs. CNTs are hollow graphene cylinders that are microns to millimeters in length and can be divided in single-walled (SWCNTs) with a diameter of 0.7 to 3 nm, and multi-walled (MWCNTs) with a diameter of 10 to 25 nm. CNTs are engineered with a wide variety of core structures and surface functionalizations that change their chemical and physical properties to enhance their suitability for different industrial applications. However, despite of the large array of available CNT configurations, their impacts on aquatic organisms, especially on invertebrate species, are still limitedly known. To our knowledge, no information is available on how surface chemistry alteration (functionalization) of CNTs may impact the toxicity of these CNMs to bivalve species. For this reason, the impacts induced by chronic exposure (28 days) to unfunctionalized MWCNTs (Nf-MWCNTs) in comparison with functionalized MWCNTs (f-MWCNTs), by introducing polar groups such as carboxyl groups (-COOH) in order to achieve better dispersibility in water, were evaluated in the Manila clams *Ruditapes philippinarum*. one of the most dominant bivalve of the estuarine and coastal lagoons environments. Alterations induced in clams' oxidative status, neurotoxicity and metabolic capacity were performed. The results obtained clearly showing that both Nf-MWCNTs and f-MWCNTs were able to generate oxidative stress in the exposed clams and were also responsible for changes in organisms' metabolism (expressed in alteration of energy reserves) and neurotoxicity induction in *R. philippinarum*, however greater impacts were caused by f-MWCNTs, namely in terms of metabolic activities (GLY and ETS), oxidative stress biomarkers responses (LPO) and antioxidant enzymes activities (SOD and GPx) compared to Nf-MWCNTs. In the present study, it was clearly demonstrated that nanomaterial toxicity can be attributed to core structure and surface functionalization, which have been shown to alter the level of toxicity.

MO418

Assessment of genotoxic and proiflammatory effects of different Silica and Titania Nanoparticles on human bronchial cells

D. Cavallo, Italian Workers Compensation Authorithy INAIL / Occupational and Environmental Medicine, Epidemiology and Hygiene; C.L. Ursini, R. Maiello, A.M. Fresegna, A. Ciervo, S. Iavicoli, Italian Workers Compensation Authority-INAIL / Occupational and Environmental Medicine, Epidemiology and Hygiene

The widespread production and use of titanium dioxide (TiO_2) and silica (SiO_2) nanoparticles (NPs) in consumer products and in different industrial and medical applications raises concerns about their possible toxicity. We studied potential genotoxic-oxidative and inflammatory effects of two amorphous silica NPs (precipitated NM200 and pyrogenic NM203) and two anatase TiO_2NPs (NM100 size 50-100 nm and NM101 size 5-8 nm) furnished by JRC. NM characterization was performed by TEM and DLS. Human bronchial (BEAS-2B) cells were exposed for 24h to 0.1-100 µg/ml of selected NMs to evaluate: cytotoxicity (trypan blue assay), direct/oxidative DNA damage (Fpg-comet assay) and inflammatory effects (IL-6, IL-8 and TNFα release by ELISA). In culture medium NM200 was better dispersed than NM203, NM100 resulted better dispersed than NM101 at 100 µg/ml and both titania showed similar agglomerate sizes at 10 µg/ml. We found lack of cytotoxicity for all NPs. Slight direct DNA damage at 10 and 100µg/ml and slight oxidative DNA damage at the lowest concentration were induced by NM200. NM203 induced dose-dependent direct DNA damage statistically significant at 100 µg/ml and oxidative DNA damage at low concentrations. NM100 induced dose-dependent direct DNA damage and oxidative DNA damage at 1 and 10 µg/ml. Direct DNA damage, statistically significant at 10 and 100 µg/ml, and induction of oxidative DNA damage at 100 µg/ml were found for NM101. Both silica NPs induced slight IL-8 release at 100 µg/ml, NM203 induced also IL-6 release at 10 and particularly at 100 µg/ml (262.2 fold of control). Both TiO_2NPs induced slight IL-8 release at 100 µg/ml but only NM101 induced significant IL-6 induction at 100 µg/ml. The findings show higher genotoxic/oxidative and inflammatory effects for NM203 in respect to NM200, probably due to its higher surface reactivity determining a strong interaction with the proteins in the medium and higher protein-mediated cell interaction. The findings also show DNA damage for both TiO_2NPs and oxidative DNA damage for NM101, correlated with the proinflammatory IL-6 cytokine induction, probably due to its smaller size, higher agglomeration tendency and capacity to induce ROS. This study is partially financed by FP7-NANoREG project, Grant n. 310584.

MO419

Transformations of engineered nanomaterials during wastewater treatment: the role of engineered surface coatings and the impact on environmental fate

M. Surette, Oregon State University / School of Chemical, Biological, and Environmental Engineering; J.A. Nason, Oregon State University / Chemical Biological and Environmental Engineering; R. Kaegi, Eawag - Swiss federal Institute of Aquatic Science and Technology

Previous research has highlighted the importance of particle-particle interactions in controlling the environmental fate of engineered nanomaterials (ENMs). Yet our ability to accurately predict the outcome of these interactions within environmental systems is still limited. One obstacle is the inherent complexity of these interactions which depend on several factors, including the properties of the ENM. However, the properties of the ENM are dynamic and can be altered via myriad transformation processes (*e.g*., over-coating via natural macromolecules, surface coating displacement, etc.). When considering the pathways by which ENMs may be released to the environment, wastewater treatment plants (WWTPs) not only act as gateways controlling the release of ENMs but they may also serve as reactors adjusting the properties of the ENMs. Therefore, to improve our understanding of ENM interactions within environmental systems we must first understand the extent to which ENM properties are altered within WWTPs. \n The objective of this research is to develop a protocol that simulates the transformations or 'aging' ENMs experience within a WWTP. The initial focus is on the effect of the dissolved components within the wastewater medium and whether ENMs with initially dissimilar properties will have similar properties after aging. To accomplish this, 12-15 nm gold nanoparticles (AuNPs) with different engineered surface coatings were selected as model ENMs. A series of batch reactors, each containing a sample from a different stage in a WWTP, were used to assess the impact of each stage on the ENMs. Each wastewater sample was first filtered to remove suspended solids and then dosed with a single type of model ENMs. Each reactor was mixed and aliquots were collected over time. The aliquots were then analyzed by a variety of techniques to investigate the effect of the media on the properties of the ENMs, including size, surface charge, stability/aggregate structure, and hydrophobicity. \n Future research will investigate the impact of the suspended solids and the overall effect of the transformations on the aggregation behavior of the ENMs upon their introduction to different environmental mediums (*e.g.*, surface water). In simulating the discharge of the aged ENMs to the environment, the effect of the transformations induced by a WWTP on the aggregation behavior of the ENMs will be evaluated. Ultimately, this will help refine our understanding of ENM environmental fate.

MO420

Freshwater sediments as an environmental reactor: defining biologically relevant fate parameters to provide context for nanomaterial bioaccumulation

R. Cross, University of Exeter; C. Tyler, University of Exeter / Biosciences College of Life and Environmental Sciences; T.S. Galloway, University of Exeter / Biosciences

As the field of nanotoxicology matures there is a call for the research focus to progress from hazard identification to more ecologically relevant assessments of the risk that engineered nanomaterials (ENM) pose as they undergo a range of transformations in the environment. This will require test designs prioritising environments most at risk of contamination, and which not only measure ecologically relevant endpoints, but also characterise the fate, transformations and behaviours of particles within the test system, providing the context for differences observed between treatments. Freshwater sediments present an ecosystem in need of further research, as these are predicted to be major sinks of ENMs entering the aquatic environment though waste water treatment and terrestrial pathways during material production, use and disposal. Whist freshwater sediments have been identified as an ecological compartment at risk of contamination, very little is known about the fate of ENMs entering these sediments. We present a simple separation method to isolate the colloidal (< 200 nm) and dissolved (< 1kDa) fractions of the sediment pore water, which can be run alongside biological exposures. This provides the context for how these biologically accessible fractions of ENMs in the sediments may relate to intrinsic particle properties such as size, core composition and coatings. Using cerium oxide (CeO_2NP) and silver nanoparticles (AgNP) we investigate the routes of bioaccumulation of these materials in the freshwater sediment dwelling worm *Lumbriculus variegatus*. By following the fate of these particles in the solid bound, colloidal and dissolved fractions of the sediment, we provide context to explain differences in both the route and extent of uptake of these materials by the worm. This poster presents the successful application of this method to investigate the implications different stabilisation mechanisms (electrostatically stabilised citrate and sterically stabilised PEG coatings) have upon the route of uptake of CeO_2 and AgNPs and transformations they undergo during sediment exposures. Accumulation of CeO_2 through dietary uptake is linked to their strong associations to the solid fraction of

the sediment and lack of dissolution (< 1% of spiked cerium was extractable with water). Transdermal uptake of AgNP was attributed to dissolved silver in the pore waters and uptake of soluble silver, potentially through localised dissolution of particles at the worms' surface.

MO421

Examining the role of TiO2 nanoparticle surface transformations on transport and toxicity

A.R. Deline, Oregon State University / Environmental Engineering; J.A. Nason, Oregon State University / Chemical Biological and Environmental Engineering

Titanium dioxide nanoparticles (TiO_2 NPs) have great potential for use in a variety of commercial and environmental applications, including the photocatalytic treatment of contaminants. While processes like microbial inactivation and the generation of reactive oxygen species (ROS) have been studied under a variety of irradiation and water chemistry conditions, there exists limited mechanistic insight as to how these processes are related to the molecular-level surface transformations that may occur under different environmental conditions. This study is using surface-sensitive characterization techniques, including x-ray photoelectron spectroscopy (XPS), to examine the impact of UV irradiation, temperature, and water chemistry exposures on the metal oxide surfaces of both pure anatase and mixed anatase-rutile TiO_2 NPs. Initial XPS studies of the impact of simulated solar irradiation revealed that solar irradiation resulted in a decrease in the aliphatic carbon present on the TiO_2 surface and an increase in the oxygen-bonded carbon, with no observable effect on the oxidative properties of the metal oxide. Additional studies will examine the surface transformations that result from the use of a Fresnel lens to increase the irradiation intensity and solution temperature. The relationships between these molecular-level surface properties and the extrinsic properties of the TiO_2 NPs are being further explored using a suite of functional assays. Assays that have been optimized for the characterization of TiO_2 NPs in this study include methylene blue dye degradation (photocatalytic activity), rose bengal dye adsorption (hydrophobicity), and fluorescein dye conversion (ROS generation). Ultimately, changes in the properties of the TiO_2 NPs will be compared to larger scale environmental behavior, allowing for a better understanding of the specific role that surface structure plays in nanoparticle transport and toxicity.

MO422

Influence of organic compounds on the sulfidation kinetics of copper oxide nanoparticles

A. Gogos, Eawag, Swiss Federal Institute of Aquatic Science and Technology / Process Engineering, Particle Lab; A. Voegelin, R. Kaegi, Eawag - Swiss federal Institute of Aquatic Science and Technology

Once released to the aquatic environment, engineered nanomaterials (ENM) will inevitably come into contact with different types of dissolved organic matter (DOM). It has been shown in a large number of studies that DOM influences the colloidal properties of ENM, which in turn impact subsequent transport and transformation processes. Sulfidation, as an important environmental transformation process, has significant implications for the fate and the ecological effects of ENM that are based on chalcophile elements. However, to date, our knowledge on the influence of DOM on the kinetics and mechanisms of this transformation reaction are very scarce. For copper oxide nanoparticles (CuO NP), the sulfidation reactionand its kinetics have been described in detail. However, the influence of DOM on this reaction has not been investigated, yet. Due to their high content of bisulfide (HS^-), wastewater systems represent major sulfidizing compartments, where the DOM mainly consists of proteins, polysaccharides and humic substances. In this study, we therefore selected three organic model compounds (Bovine serum albumin (BSA, model protein), Alginate (model polysaccharide) and Polyacrylic acid (natural organic matter analogue)) and investigated their influence on the sulfidation of CuO NP. All experiments were conducted in solutions buffered to pH 8 at concentrations of 1.3 mM CuO and 4 mM HS^-. Variable amounts of the organic compounds were added to reach final concentrations of 10, 100 and 1000 mg L^{-1}. Reacted CuO NP were collected at selected time points and characterized using Cu K-edge X-ray Absorption Spectroscopy (XAS). In addition, selected samples were characterized using analytical electron microscopy. XAS analyses revealed that at a concentration of 10 mg L^{-1}, none of the selected organic compounds affected the sulfidation rate and observed reaction products. However, at BSA concentrations ≥100 mg L^{-1} a reduction of the reaction rate was observed. In addition, at these high concentrations, BSA hampered the recrystallization of amorphous Cu_xS to covellite. Electron microscopy also showed that in the presence of BSA, amorphous Cu_xS was the dominating particle type. Our results show that at high concentrations, proteins such as BSA influence both the reaction kinetics and the reaction pathway of the CuO sulfidation. In real municipal wastwater, however, lower protein concentrations and thus a complete sulfidation of the CuO NP can be expected.

MO423

Evaluating spICP-TOF-MS for Exploring Environmental Nano-scale Processes

M.D. Montaño, University of Vienna / Environmental Geosciences; B. Gerstmann, A. Laycock, N. Tepe, T. Hofmann, F. von der Kammer, University of Vienna / Department of Environmental Geosciences

The advent of single particle ICP-MS (spICP-MS) has helped advance the field of nanometrology, specifically at concentrations and in matrices that are environmentally relevant. However, the concentration of naturally occuring nanoparticles (NNPs) and nanominerals far outweigh the expected released concentrations of engineered nanoparticles (ENPs), making their detection by single element spICP-MS and their subsequent risk assessment a challenge. The introduction of ICP-time-of-flight-MS (ICP-TOF-MS) has the potential to overcome these challenges, as elements are detected *quasi*-simultaneously at dwell times of 46µsec, covering nearly the entire atomic mass range (7-250 m/z). By examining differences in the chemcial composition on a particle-by-particle basis, NNPs and ENPs can be differentiated, and geochemical processes occuring at the nano-scale can be explored on an individual particle basis. In order to establish this technique, and its utility for environmental analyses, several multi-element and multi-isotope nanoparticles were analyzed using traditional spICP-MS (with quadrupole mass filtering) and with spICP-TOF-MS. The precision and accuracy for particle sizing and counting were evaluated for each technique for a range of elements to explore the advantages and potential limitations of these techniques as the apply to environmentally and geochemically relevant systems. To illustrate the advances made in multi-element monitoring by time-of-flight, single particle analyses were performed on both a quadrupole ICP-MS and an ICP-TOF-MS, and using 3ms and 100µs dwell times on both instruments. Particles analyzed consisted of mixtures of well-defined AuAg core-shell NPs with Au and Ag only NPs, polydisperse ceramic NPs with well-defined chemical compositions, and environmentally relevant colloidal suspensions containing ENPs. These systems were analyzed for both the size and number concentration in order to establish the efficacy of spICP-TOF-MS as a characterization technique. spICP-TOF-MS demonstrates considerable advantages over traditional spICP-MS and has the potential to examine the geochemical realm on an individual particle basis. The further development of this technique may also lead to a better assessment of ENP exposure in test systems and nature, improving on environmental risk assessment and gaining a better understanding of ENP interactions with naturally occuring colloids.

MO424

Assessing potential risks of Nanodrugs and their delivery systems in fish using Light Sheet Microscopy

D.L. Windell, University of Exeter / College of Life and Environmental Sciences; J. Moger, The University of Exeter / College of Engineering, Mathematics and Physical Sciences; M.J. Winter, The University of Exeter / College of Life and Environmental Sciences; S. Owen, AstraZeneca / Safety Health Environment; C. Tyler, University of Exeter / Biosciences College of Life and Environmental Sciences

Nanotechnology applications have increased dramatically in recent years including in the pharmaceutical sector. The unique properties of nanoparticles have been exploited in medicine in both drug development and drug delivery. Their small size and capability for manipulation and functionalisation allows for great improvements in drug efficacy. Nanomedicines can cross a wide range of biological membranes and barriers (including the blood brain barrier) facilitating the diagnosis and treatment of life threatening diseases such as cancer. Although nanotechnology may help to reduce the toxicity and side effects of drugs, the actual carriers themselves may also have the potential for inducing toxic effects, depending on their composition. This raises the need for safety evaluations of these drug delivery systems both in patients, but also with respect to their potential for environmental impact. Very little is known regarding the potential impacts associated with the release of these nanodrugs or their carrier systems into the environment, although some studies have begun to investigate the potential toxic effects of various nanoparticle shapes and coatings in aquatic organisms. Using various sizes of gold nanoparticles (between 10 and 100nm in diameter) with a non-reactive methyl polymer and fluorophore coating, we have traced their uptake and tissue partitioning using a casper mutant zebrafish and light sheet microscopy. We have constructed a light sheet system based on the OpenSPIM platform, (SPIM - Selective Plane-Illumination Microscopy) which allows us to create 3D images and 4D videos in real-time. Using this rapid image acquisition technique we showed a size selective uptake of the nanoparticles into the kidney and minimal uptake in other organs. Depuration studies indicate a steady loss of the gold nanoparticles from the pronephric kidney over time. We also investigated for biological responses using specific zebrafish transgenic lines for oxidative stress and kidney function. We are now investigating the effect different coatings and functionalisations have on the uptake and distribution of gold nanoparticles in the larval zebrafish ultimately with the aim of beginning to define the potential for this important new group of medicines for having an environmental impact on fish.

MO425

SETAC Nanotechnology Interest Group

C. Svendsen, CEH, Wallingford / Pollution and Ecotoxicology

Hydrophobic Chemicals and Mixtures: Reliable Investigations on their Environmental Fate and Effects (P)

MO426

Effect of ageing on polycyclic aromatic hydrocarbon composition of biochar

G. Sigmund, Technische Universität München / Chair of Analytical Chemistry and Water Chemistry; T. Bucheli, Agroscope ART / Environmental Analytics; I. Hilber, Agroscope / Environmental Analytics; T. Hüffer, M. Kah, T. Hofmann, University of Vienna / Department of Environmental Geosciences

The influence of ageing on biochar properties has been investigated by comparing three fresh biochars with biochars artificially aged by either H_2O_2 thermal oxidation or horseradish peroxidase enzymatic oxidation. In addition, a field-aged counterpart for one of the biochars was recovered from an agricultural field site, four years after application. Biochar bulk properties showed only minor changes following both artificial and field ageing, indicating high biochar stability. Concentrations of the 16 US EPA PAHs were measured in all of the biochars and a contaminant trap was used to investigate the effect of ageing on their bioaccessibility. The concentrations of total and bioaccessible PAHs ranged from 4.4 to 22.6 mg/kg and 0.0 to 9.7 mg/kg, respectively. Concentrations of the 16 US EPA PAHs decreased following field ageing, but the proportion of low molecular weight PAHs increased. The observed changes in PAH composition with field ageing can partially be explained by uptake from the surrounding soil. In addition, size discriminatory intra-biochar transfer processes also contributed to the changes in PAH composition. To better understand changes in PAH composition with ageing, an additional broad range of alkylated PAHs was also analyzed in selected samples. Our results show that the tested artificial ageing protocols are unable to approximate the changes in PAH composition resulting from field ageing. Nevertheless, total and bioaccessible PAH concentrations decreased for both artificially and field-aged biochars, indicating that biochars release most PAHs when they are freshly produced and that the risk of PAH release decreases with ageing. Therefore, well-produced biochars that meet European Biochar Certificate (EBC) and International Biochar Initiative (IBI) quality thresholds for total PAH concentrations are unlikely to present a risk with regard to PAH release following field application. These results have recently been published (doi: 10.1039/C7EM00116A).

MO427

Field testing of a new calibration approach for silicone passive samplers: Comparison of the concentration ratio method using samplers of different thicknesses with the PRC approach.

H. Fuchte, Institut fur Umweltforschung / Institute for Environmental Research BioV; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics; K. Smith, RWTH Aachen University / Institute for Environmental Research

Silicone passive sampling is a common method for sampling bioavailable concentrations of waterborne hydrophobic pollutants in the environment. Often silicone samplers have to be used in kinetic mode and sampler calibration is unavoidable. Most commonly, exchange kinetics are derived from the release rates of performance reference compounds (PRCs) spiked into the sampler prior to usage. Unfortunately, PRCs can be expensive and are not always available for all compounds. Due to these challenges, a complementary calibration approach using passive samplers of different thicknesses has been developed and investigated in lab studies. This study describes the testing of the approach *in situ* out in the field. The sampling location was a storm water retention pond collecting storm water run-off from a motorway. The pond has two basins, one of which is equipped with a Floating Treatment Wetland (FTW) for cleaning the run-off. Two sets of duplicate samplers with 3 different thicknesses were installed in the inflow, after the FTW and in the reference basin without a FTW. One set of samplers was taken out after three, and the second after five weeks. These were extracted and analyzed for PAHs, with the concentration ratios for the different thicknesses used to calculate the field dissolved concentrations. All samplers had been additionally loaded with PRCs, with the decreases also used to calculate the field levels. These were compared to the results from the concentration ratio approach, underlining its suitability as a complementary calibration method and its application domain.

MO428

Use of biochar for hexaclorocyclohexane sorption: a mechanistic approach

L. Silvani, g. cornelissen, s.e. hale, Norwegian Geotechnical Institute

Hexaclorocyclohexanes (HCHs) are halogenated compounds composed of 4 main isomers: α-HCH, β-HCH, γ-HCH and δ-HCH, which differ for their tridimensional structure. Commercial HCH in technical grade is a mixture of these isomers. HCH has been extensively used as a pesticide despite the fact that only γ-HCH (lindane) has insecticide properties. HCHs' toxic, carcinogic, teratogen and neurotoxin effects have been reported in humans; and the HCHs have a clear tendency to accumulate in the environment. For these reasons, HCHs are contaminants of worldwide concern and an adequate remediation approach is therefore necessary. Various remediation techniques have been used to remove HCHs in aqueous solution, among these, adsorption is the most common used one. Biochar (BC) is a carbonaceous material that is a promising sorbent amendment material due to its high adsorption of organic and inorganic contaminants, and to its low cost. In this study three standard biochars, from digestate (BC_d), from greenhouse tomato waste (BC_{gtw}) and from durian shell (BC_{ds}), have been used as sorbent materials for the HCHs removal from water. The BCs used cover a wide range of surface area (5.4 - 328.6 $m^2 g^{-1}$), pore volume (5.1 - 186.6 $cm^3 g^{-1}$), pore dimension (1.05 - 5.85 Å), pyrolysis temperature (400 – 700 ºC) and surface properties (including iron content). Batch isotherm tests were carried out in deionized water with the single isomers and the mixture of α-, β-, γ- and δ-HCH. The HCH concentration was ranged between 1 and 500 µg L^{-1} in the monocomponent isotherms and between 5 and 2000 µg L^{-1} (total concentration) in the mixture isotherms. Polyethylene (PE, 26 µm thick, 0.30 ± 0.01 g) was used as a passive sampler for assessing the HCHs concentration in water. The sorption performance of the biochars is related to physicochemical properties. Preliminary results have shown the adsorption performances are correlated with the BC surface area and iron content, where a higher adsorption is observed as surface area and iron content increase. Clear differences in the behavior of the isomers were observed.

MO429

Development of a Method for Measurement Freely Dissolved Concentrations of Alkylated PAHs Using Solid Phase Microextraction with PDMS Fibers

M. Reininghaus, RWTH Aachen / Biogie 5; T. Parkerton, ExxonMobil Biomedical Sciences Inc. / Toxicology & Environmental Science; G. Witt, HAW Hamburg / Department of Environmental Engineering

Frequently, the total PAH sediment concentration reported for a sample has been based on 16 individual priority pollutants according to the U.S. EPA Method 8310. For pyrogenic sources of PAHs (e.g. incomplete burning processes), the parent PAHs are the predominant species. In contrast, PAHs from petrogenic sources (e.g. crude oil) are dominated by alkyl PAHs. Therefore, the U.S. EPA narcosis model requires the measurement of 18 parent and 16 groups of alkyl polycyclic aromatic hydrocarbons (PAHs) (so-called 34 PAHs) in sediments expressed as organic carbon normalized concentrations (C_{soc}) for evaluating risks to benthic organisms based on the calculated magnitude of PAH toxic units (TUs) (Hawthorne et al., 2006). However, due to the heterogeneous nature of organic carbon in field sediments, potential risks of adverse biological effects from sediment-associated contaminants are most directly related to concentrations of freely dissolved chemicals (C_{free}) in sediment pore water, not to C_{soc}. Recent advances in equilibrium passive sampling methods (EPSMs) offer a promising alternative to the measurement of C_{soc} and support improved risk-based decision making since bioavailability of sediment contaminants can be directly quantified via C_{free}. (Mayer et al., 2015, Burkhard et al., 2017). When using EPSMs, polymer to water partition coefficients are crucial for reliable calculation of C_{free}. To date partitioning coefficients are available for parent PAHs across different polymers (e.g. PDMS, POM) (Lydy et al., 2015). In this study, an equilibrium passive sampling method was developed for investigating alkylated PAHs in marine and limnic sediments and used for risk evaluations of both pyrogenic and petrogenic PAHs. The method is based on solid phase microextraction (SPME) with different silicone materials (PDMS coated glass fibers and hollow fibers). Partitioning coefficients for the PDMS coated fibers (K_{PDMS}) were calculated for selected target alkylated PAHs which have previously not been available. K_{PDMS} for additional alkylated PAHs of interest were then predicted based on the experimentally reported K_{PDMS} values. Finally, the new method was demonstrated by in-situ deployment at seven field stations of different pollution levels. Further insights between in-situ and ex-situ EPSM deployment were obtained by comparing the results of in-situ C_{free} measurements with corresponding laboratory derived measurements using sediments collected from the same stations.

MO430

Spatial Distribution of HOCs on the Palos Verdes Shelf Superfund Site

A.R. Taylor, University of California Riverside / Environmental Sciences; J. Wang, University of California Riverside; D. Schlenk, J. Gan, University of California, Riverside / Department of Environmental Sciences

Hydrophobic organic contaminants (HOCs), such as DDTs, PCBs, and currently used pesticides contaminate soils and sediments all over the world. As they are hydrophobic in nature, these compounds are resistant to both chemical and physical degradation while also having a strong affinity for soil or sediment particles and organic matter.[1] Often, this contamination is due to the historic or current use and manufacturing of these compounds, such as the widespread contamination of the Palos Verdes Shelf by DDTs and PCBs, which continue to pose health hazards to ocean organisms and humans that eat fish from this area.[2] Several current use pesticides, such as fipronils and pyrethroids, have also been detected in sediment from the shelf during preliminary experiments, indicating that these contaminants may have been deposited onto the shelf via urban waterways. In this study, we assessed the spatial distribution of current-use insecticides pyrethroids and fipronils in the top 2 cm of sediment on the Palos Verdes Shelf Superfund Site. Concentrations of total pyrethroids (?Pyrethroids= ? of bifenthrin, fenpropathrin, lambda-cyhalothrin, cyfluthrin, cypermethrin, and *cis*-permethrin) ranged from n.d. to 170.15 ng/g and total fipronils (?Fipronils= ? of fipronil desulfinyl, fipronil sulfide, fipronil, fipronil sulfone) ranged from n.d. to 5.59 ng/g. On-going research also aims to understand the spatial distribution of legacy HOCs (PBDEs, DDTs, PCBs) in the shelf area and assess their bioavailability in order to determine their risk to both organisms living on the shelf and possible routes of human exposure. These findings will be made available to the federal and state agencies for use in environmental risk assessment and designing management strategies. **References**

Delle Site, A. Factors affecting sorption of organic compounds in natural sorbent/water systems and sorption coefficients for selected pollutants. A review. *J. Phys. Chem. Ref. Data*. **2001**, *30* (1)187–439. U.S. Environmental Protection Agency. 2010. *EPA Signs Interim Record of Decision: Remedial Work Begins*; Region IX, United States Environmental Protection Agency: San Fransisco, CA.

MO431

PAHs in water and surface sediments from Douro River estuary and Porto Atlantic coast (Iberian Peninsula, North Portugal) — Risks for biota and human health?

M. João Rocha, ICBAS U.Porto, CIIMAR CIMAR LA; J.L. Dores-Sousa, VUB / Department of Chemical Engineering; C. Cruzeiro, CIIMAR CIMAR LA, Porto, CEF FCTUC U.Coimbra; E. Rocha, ICBAS U.Porto, CIIMAR CIMAR LA

This study shows the presence of 16 priority polycyclic aromatic compounds (PAHs) in surface waters from the Douro River estuary and nearby Atlantic seacoast. These areas bath Porto and Gaia cities, widely known by the production/export of the famous Porto wine. This area, besides being highly industrialized, also holds an oil refinery, an important harbour, intense maritime traffic, and recreational marinas. For this study, water samples were taken from four strategic sampling sites, at six different times of the year. These samples were extracted by ultrasound technique (suspended fraction) and solid-phase extraction (dissolved fraction), before their quantitative analysis by gas chromatography–mass spectrometry (GC-MS). Data showed the presence of all analysed PAHs in all samples, which global amounts (Σ16PAHs) were extremely high in both analysed matrices and at all sampling sites. In fact, average concentrations attained ≈ 52 μg/g dry weight (dw) in surface sediments and ≈ 55 ng/L in water. In view of the evaluated concentrations, the surveyed areas were classified as highly polluted by these organics, suggesting that both mutagenic and carcinogenic responses can occur in both humans and aquatic animals living in these areas. This statement is supported by the measurement of carcinogenic PAHs for humans (group 1) dissolved in water (≈ 5%) and in surface sediments (≈ 6%) in biologically significant amounts. These data are the first reported in this geographic area and can be used as a starting point for future control of the PAHs levels either locally either at the European scenario. **Acknowledgements:** European Regional Development Fund (ERDF) through COMPETE, Framework of the Structured Program of R&D&I INNOVMAR – Innovation and Sustainability in the Management and Exploitation of Marine Resources (NORTE-01-0145-FEDER-000035), Research Line ECOSERVICES, supported by the Northern Regional Operational Programme (NORTE2020), through the ERDF. ICBAS – U.Porto. **Keywords:** PAHs, carcinogenic, estuary/sea, monitoring

MO433

Occurrence and availability of PACs and total AhR agonists in contaminated soils - Combining in vitro reporter gene assay and chemical analysis with passive sampling and column leaching

M. Larsson, Orebro University / Man-Technology-Environment research centre (MTM); M. Lam, RWTH University Aachen; P. van Hees, Orebro University / MTM Research Center; J. Giesy, University of Saskatchewan / Department of Veterinary and Biomedical Sciences and Toxicology Centre; M. Engwall, Orebro University / Man-Technology-Environment research centre (MTM)

Polycyclic aromatic hydrocarbons (PAHs) are common contaminants at industrial sites, and occur as complex mixtures of thousands of PAHs and heterocyclic compounds (NSO-PACs) among others, collectively referred to as polycyclic aromatic compounds (PACs). The contaminant composition differ widely due to contamination sources and weathering processes in the environment. Despite the high complexity in PAC-contaminated areas, current risk assessment of PACs is commonly based on chemical analysis of the 16 US EPA PAHs. Consequently, many PACs are unknown. There is an urgent need of improved and applicable analytical methods to assess environmental levels and fate of potential toxic PACs to evaluate risk to human health and the environment. An important concern regarding sites contaminated with PACs is the risk of groundwater contamination by release of the compounds from soils. The aim of this study was to investigate the occurrence of 77 PACs including PAHs, alkyl-PAHs, oxy-PAHs and NSO-PACs among total aryl hydrocarbon receptor (AhR)-agonists in soils from historical contaminated sites and to assess the availability of the compounds in the soils. A novel approach combining chemical (GC/MS) and bioanalytical measures (H4IIE-luc) combined with characterization of availability by use of a column leaching test and passive sampling was used. This approach allowed screening of potentially toxic metabolites of PACs in soils during remediation. The results show that chemical analysis of 16 US EPA PAHs to determine the degree of contamination of PACs in soils greatly overlooks toxicologically relevant PACs. Available concentrations of PACs were considerably lesser than total initial concentrations of PACs in all soils, indicating low availability of the compounds in soils. The leachable fraction was generally greater for more hydrophilic PACs, such as more polar PACs composed of two or three rings. Contribution of the analyzed PACS to the overall AhR-mediated activities detected in soils, leachates and passive samplers was pretty small and confirms presence of several other AhR agonists in soils. Only a small fraction of AhR agonists were available in soils, indicating an overestimation of the risk, if only total initial concentrations in soils would be considered in risk assessment. However, the results show that analysis of available fractions based on only 16 US EPA PAHs have the potential to underestimate the risk of the soils.

MO434

Verification of read-across for aquatic hazard properties of Petroleum Substances in REACH registrations

Y. Verhaegen, CONCAWE; C. Hughes, Shell Health / Risk Science Team; L. Camenzuli, ExxonMobil Petroleum and Chemical; A.D. Redman, Exxon Mobil Biomedical Sciences / Toxicology and Environment Science Division; M. Comber, Mike Comber Consulting; S.A. Villalobos, BP / Global Product Stewardship; V. Ochoa, Cepsa; S. Linington, BP; E. Vaiopoulou, European Petroleum Refiners Association

Petroleum substances are examples of UVCBs (substances of Unknown or Variable composition, Complex reaction products or Biological materials), whose complex chemical composition will vary depending on, amongst other things, the source of crude oil, the refinery processing and climate conditions. These substances present additional challenges when conducting environmental hazard and risk assessments under regulatory schemes such as REACH, and Concawe has developed bespoke models for these purposes. However, for purposes of hazard classification and labelling there is still a need for experimental aquatic toxicity test data on petroleum substances. Concawe substances have been organised into categories, based on similarities in refinery processes and physicochemical properties, resulting in a clustering of comparable chemical compositions and related hazard profiles. Applying read-across within a category is an established concept to fill in data gaps and to reduce unnecessary testing, and has been applied to available aquatic toxicity data for Concawe substances using a worst-case approach. ECHA recently released its Read-Across Assessment Framework (RAAF) for environmental endpoints, however the RAAF for UVCBs is still under development due to their added complexity. One recommendation of the RAAF when applying a category approach is to present data in a matrix to demonstrate that properties are similar or follow a regular pattern. In this presentation a category data matrix will be presented for the Concawe category of vacuum hydrocracked gas oils (VHGO). Available historical experimental aquatic toxicity data will be presented alongside substance identity information, predicted EL50 and toxic unit (TU) values calculated using PETROTOX, and results from biomimetic extraction solid phase microextraction (BE-SPME) screening studies. The latter is a technique which measures bioavailable hydrocarbons, and has been demonstrated to correlate well with experimental and predicted aquatic toxicity data. The resulting, complimentary dataset forms a weight of evidence upon which to justify category approaches to the read-across of experimental toxicity data.

MO435

Automated Solid Phase Microextraction (SPME) for measuring freely dissolved concentrations of hydrophobic chemicals in soils, sediments and other solid matrices

C. Vitale, University of Insubria; K. Knudsmark Sjøholm, University of Copenhagen / Dept. of Plant and Enviornmental Sciences; A. Di Guardo, University of Insubria / Department of Science and High Technology; P. Mayer, Technical University of Denmark / Department of Environmental Engineering

In risk assessment of hydrophobic chemicals that are strongly associated to the soil/sediment organic matter, freely dissolved concentrations (C_{free}) are more representative than total concentrations (C_{total}) of their actual bioavailability, potential for bioaccumulation and toxicity. Such freely dissolved concentrations can be measured by Solid Phase Microextraction (SPME) if operated in the equilibrium and negligible depletion mode. Furthermore, in order to reduce the measurement variability, increase sample throughput and to produce high quality data, automated SPME methods are promising. The aim of this study was thus to investigate (1) how to operate automated SPME on solid samples, (2) how to achieve equilibrium sampling for hydrophobic organics within a practical time span and (3) how to calibrate the new approach. Polychlorinated biphenyls (PCBs) served as model compounds and matrices included soil, sediment and sludge.

MO436

New approaches for determining solubility of volatile liquid chemicals

H. Birch, DTU Environment / Department of Environmental Engineering; L.N. Trac, Technical University of Denmark / Environmental Engineering; P. Mayer, Technical University of Denmark / Department of Environmental Engineering

Water solubility is a fundamental parameter in environmental risk assessment of chemicals, and is theoretically a simple parameter to determine. For hydrophobic chemicals in the liquid state, the main challenge is to establish equilibrium between the pure liquid phase and the water phase within a reasonable time frame, while avoiding formation of dispersions or micro-droplets. A slow-stir method for solubility measurements has previously been developed for this purpose, however it is time consuming as it requires weeks to equilibrate. In this work, two new approaches were used for solubility determinations. Both methods were originally developed for toxicity testing at the saturation level. Both approaches avoid direct contact between the pure substance and the water, thus minimizing the risk of droplet formation. The first approach uses passive dosing from a saturated silicone polymer in order to saturate the water, while the second approach equilibrates the water with the pure phase liquid through the headspace. Equilibrium time in the range of minutes to hours is expected for the two methods. Four liquid hydrophobic

chemicals within the logKow range of 4.4-8.6 will be included in the study, and results from both methods will be compared.

MO437

Headspace passive dosing for dose-response testing of volatile hydrophobic organic chemicals

L.N. Trac, Technical University of Denmark / Environmental Engineering; S.N. Schmidt, Technical University of Denmark / Department of Environmental Engineering; M. Holmstrup, Aarhus University / Department of Bioscience; P. Mayer, Technical University of Denmark / Department of Environmental Engineering

Constant and well-defined exposure is crucial for the toxicity testing of liquid organic chemicals with high Henry's constants, which are prone to substantial evaporative losses. A simple and effective headspace passive dosing method was developed and then applied to control the exposure of the freshwater algae *Raphidocelis subcapitata* and the terrestrial springtail *Folsomia candida* to terpenes and alkanes in toxicity experiments. The headspace passive dosing method applies a liquid partitioning donor placed in the headspace of the closed test vial for controlling exposure while avoiding direct contact and introduction of pure phase micro-droplets. Passive dosing from the pure liquid compound was applied for toxicity testing exactly at the solubility limit, and a dilution series of test chemicals prepared in purified vegetable oil served as donor for dose-response testing. The terpenes S-(-)-Limonene and a-(+)-Pinene were tested in both the algal growth inhibition test and the springtail test. In addition, n-nonane, n-undecane and n-tridecane were tested on the algae, while iso-octane, iso-dodecane and n-dodecane were tested on the springtails. Our first results demonstrated that (1) the headspace passive dosing method is a simple yet effective way to control exposure to volatile hydrophobic organic chemicals and (2) that the method is straightforward to apply in algal growth inhibition and springtail toxicity tests. Further analyses of exposure parameters are in progress to better understand and quantify the resulting toxicity.

MO438

Application of biomimetic solid phase microextraction to characterize aquatic hazard of petroleum substances

L. Camenzuli, ExxonMobil Petroleum and Chemical; A.D. Redman, Exxon Mobil Biomedical Sciences / Toxicology and Environment Science Division; J. Butler, ExxonMobil Biomedical Sciences, Inc / Environmental Toxicology and Chemistry Laboratory; D. Letinski, ExxonMobil Biomedical Sciences Inc; E. Vaiopoulou, European Petroleum Refiners Association

Environmental hazards of petroleum substances differ in response to variable substance composition. In response CONCAWE has initiated a comprehensize analytical program to extend analytical characterization of petroleum substances to further support hazard classification within and across petroluem substance categories. As part of this work, SPME and toxicity data for newly characterized substances (*n*=139), across approximately 10 major categories, were compared to historical data. New compositional data were used as input to PETROTOX to predict acute toxicity in terms of lethal loadings (LL50). The predicted LL50s in the present work were shown to compare favorably with historical measured and predicted toxicity data. Further, experimental work was performed to estimate the bioavailable concentrations of hydrocarbons using biomimetic solid phase microextraction (BE) on water accommodated fractions (WAF) prepared with each substance at a nominal loading of 50 mg/L.This method simultaneously extracts and concentrates dissolved hydrocarbons onto a polydimethylsiloxane coated fiber which is then thermally desorbed unto a gas chromatography for quantification by flame ionization detection. The measured BE data provide an analytical surrogate that correlates to target lipid and hence WAF toxicity. New BE data showed similar agreement with earlier data collected on WAFs prepared with substances from the same categories. The BE method is a convenient predictive tool used to screen petroleum substances for testing. In summary, predicted toxicity and BE measurements for additional petroleum substances presented in this work strengthen the basis for aquatic hazard classification of petroluem substance categories.

MO439

Bioaccumulation factors of synthetic musks and other hydrophobic contaminants in mangrove molluscs.

S. Bayen, McGill University / Singapore-Delft Water Alliance; E. Segovia, H. Zhang, W. Lee, G. Juhel, National University of Singapore; F. Smedes, RECETOX / Environmental chemistry and modelling; B.C. Kelly, National University of Singapore / Civil & Environmental Engineering

The occurrence of a range of historical and emerging hydrophobic organic contaminants in mangrove ecosystems in Singapore. In particular, the levels of synthetic musk fragrance compounds, polychlorinated biphenyls, organochlorine pesticides and polycyclic aromatic hydrocarbons were measured in mangrove sediments, clams and caged mussels. In addition, the freely dissolved concentration of these organic chemicals in water was assessed with silicone rubber passive samplers. Results showed that polycyclic musks are present in mangrove ecosystems, and can accumulate in the tissues of molluscs. In the present study, bioaccumulation factors (BAF_{fw} wet weights) were calculated for all the samples/sites and log BAF_{fw} averaged 4.0±0.4, 4.4±0.3, 4.7±0.3, 3.9±0.7 and 4.3±0.4 for galaxolide, traseolide, phantolide, celestolide and tonalide respectively. Overall, the empirical models fit reasonably well the bioaccumulation of polycyclic musks in both caged and native molluscs in tropical mangroves. The study of the bioavailability of hydrophobic compounds in highly dynamic environments such as mangroves can be sometimes intricate, and the usefulness of passive samplers and sentinels species such as bivalves was confirmed in the present study.

MO440

Effect-based characterization of mixtures of environmental pollutants in sediments collected between the Arctic and Australia

A. Jahnke, Helmholtz Centre for Environmental Research - UFZ GmbH / Cell Toxicology; M. Landmann, Helmholtz Centre for Environmental Research - UFZ GmbH / Department of Cell Toxicology; M. Bergmann, Alfred Wegener Institute Helmholtz Centre for Polar and Marine Research / HGF-MPG Group for Deep-Sea Ecology and Technology; J. Bräunig, The University of Queensland / Queensland Alliance of Environmental Health Sciences (QAEHS); S. Schaefer, Federal Institute of Hydrology / Biochemistry and Ecotoxicology; A. Sobek, Stockholm University / Department of Environmental Science and Analytical Chemistry ACES; B. Escher, Helmholtz Centre for Environmental Research GmbH - UFZ / Cell Toxicology

There is a multitude of pollutants that combine persistent and hydrophobic properties. In aquatic environments, they are largely deposited in sediments. The amount and characteristics of the organic carbon determine how strongly they are bound or if they are readily available for partitioning to biota and biouptake. These pollutants can be accumulated by aquatic organisms and biomagnified to higher trophic levels. Hence, it is important to explore the composition, activity and effects of environmental mixtures of pollutants in sediments of different origin, characteristics and pollution history. Sediments from Sweden, the European Arctic (coastal Svalbard vs. open sea), Queensland (Australia) and a French-German river were collected. The freely dissolved concentrations (C_{free}) of the chemicals were determined using equilibration with thin coatings of silicone on the inner walls of glass jars with subsequent solvent extraction. Total sediment concentrations (C_{total}) were determined using accelerated solvent extraction. While there is a wide range of pollutants that have been detected in sediments world-wide, traditional chemical analysis cannot cover all compounds and their transformation products. Therefore, in this study, the extracts were dosed into seven cell-based bioassays covering cytotoxicity, activation of metabolic enzymes (binding to the arylhydrocarbon receptor, AhR), specific, receptor-mediated effects such as estrogenicity (ERa); and adaptive stress response (oxidative stress, AREc32). Cytotoxicity was assessed in all bioassays and occurred occasionally. Moreover, four of the seven bioassays were active in this study: AhR, AREc32, the peroxisome proliferator-activated receptor gamma (PPARg) and ERa. The activation of the AhR was by far most responsive and showed a distinct pattern across the sampling locations. The other three assays showed responses only at higher enrichment factors of the extracts, also revealing specific contamination patterns. A comparison between C_{free} vs. C_{total} will enable assessing the actual risk (C_{free}) vs. the potential hazard of those chemicals that might be released in future scenarios (C_{total}). The presented work calls for more detailed studies at specific sites and testing of additional endpoints with the aim of obtaining a complete picture of mixture effects caused by the freely dissolved and total concentrations of hydrophobic organic chemicals in sediments.

MO441

Bioaccumulation of hydrophobic organic compounds in aquatic biota: addressing current challenges for in tissue passive equilibrium sampling

E. Rojo-Nieto, Helmholtz centre for environmental research - UFZ / Department of Cell Toxicology; J. Koschorreck, Umweltbundesamt; m. muz, UFZ - Helmholtz Centre for Environmental Research / Cell Toxicology; A. Jahnke, Helmholtz Centre for Environmental Research - UFZ GmbH / Cell Toxicology

Organisms living in environments contaminated with Hydrophobic Organic Compounds (HOCs) can enrich these chemicals, a process known as bioaccumulation. Current bioaccumulation assessment, based on exhaustive extraction of HOCs followed by lipid-normalisation, cannot distinguish between cases when bioaccumulation is driven by bioconcentration (passive uptake) or by biomagnification (uptake via food). This shortcoming obstructs our understanding of HOCs transfer in aquatic food webs, across trophic levels and between environmental compartments (sediment, water, biota). The recently proposed approach based on using ratios in chemical activity as a metric for bioaccumulation assessment represents a major advance relative to the traditional ones, since it aims at expressing the data on a common basis to enable direct comparison among compartments. Passive Sampling Devices (PSDs) have opened a new analytical window for measuring chemical activity. PSDs have been explored to compare contamination of sediments and biota with high lipid content and offer great potential to assess contaminant transfer in aquatic food webs. The presented work is one subproject of the ERC-funded project "CHEMO-RISK" which aims, amongst others, to address the bioaccumulation of HOCs in aquatic biota on a thermodynamic basis. We will develop silicone-based PSDs in order to broaden the use of these devices to those media that are equilibrating slowly, as is the case in lean tissues. For this purpose, homogenated fish tissues from the German Environmental Specimen Bank, with different lipid contents (ranging from 1 to 5%

lipid), have been selected to optimise silicone-based PSDs for sampling in lean tissues. For this study, silicone is used as common reference phase, with sampler relocations across the homogenated samples along the sampling period, as has been proposed by Rusina et al. [1], in order to avoid the local depletion of the sample in direct contact with the silicone, and both, kinetic and equilibrium approaches have been considered. The 7 indicator PCBs (28, 52, 101, 118, 138, 153, 180) have been selected as target compounds, covering a log K_{OW} range from 5.66 to 7.15. Reference: [1] Rusina TP, Carlsson P, Vrana B, Smedes F. 2017. Equilibrium passive sampling of POP in lipid-rich and lean fish tissue: Quality control using performance reference compounds. Environ. Sci. Technol., DOI: 10.1021/acs.est.7b03113.

MO442

Widespread occurrence of 4-Nonylphenol, BHT, and 2,4-DTBP in blue crab, Callinectes sapidus, megalopae in the northern Gulf of Mexico

S. Chiasson, Loyola University / EEB; E.K. Grey, Governors State University / Division of science, mathematics, and technology; D.A. Grimm, Tulane University / Coordinated Instrumentation Facility; C.M. Taylor, Tulane University / Ecology & Evolutionary Biology

The blue crab, *Callinectes sapidus*, is an ecologically and economically important invertebrate species in the northern Gulf of Mexico (NGOM). The NGOM receives nearly 60% of drainage from the river systems in the continental United States. Blue crab megalopae collected over three years from multiple estuaries in the NGOM from Texas to Florida were tested for alkylphenol contamination using GC/MS. We found widespread contamination of blue crab megalopae with 4-nonylphenol (NP), butylated hydroxytoluene (BHT), and 2,4-di-tertbutylphenol (DTBP). NP is an alkylphenol known to impair endocrine function and concentrations detected in megalopae in 2010 and 2011 exceeded the lower limit of the No Observed Effect Concentration range for aquatic invertebrates set by the U.S. Environmental Protection Agency. BHT is a common preservative in food, pharmaceuticals, and cosmetics and is considered safe. DTBP is considered a marine pollutant, but exhibits low toxicity. All these compounds have high partition coefficients, which explains their presence in animal tissue. Concentrations of NP, BHT, and DTBP were highly correlated in megalopae over time at most study sites, implying a common source. The relatively high NP concentrations detected in juvenile blue crabs across the NGOM suggests that management strategies of alkylphenols in the environment should be re-evaluated to mitigate potential sub-lethal effects of exposure to blue crab populations.

MO443

Real-time visualization and quantification of perylene bioaccumulation at single cell level

x. guo, School of Environment, Beijing Normal University; X. Jin, Beijing Normal University; F. Bai, Peking University

Hydrophobic organic chemicals (HOCs) are of special ecotoxicological concern because they can be directly incorporated and bio-concentrated in living organisms. However, the effects of self-clustering of HOCs on their environmental behavior and toxicology have not yet received enough attention. For the first time, single-molecule fluorescence microscopy (SMFM) with a microfluidic flow chamber and temperature control has enabled us to record the dynamic process of perylene bioaccumulation in single bacterial cells and examine the cell-to-cell heterogeneity. Although with identical genomes, individual *E. coli* cells exhibited a high degree of heterogeneity in perylene accumulation dynamics, as shown by the high coefficient of variation (C.V=1.40). This remarkable heterogeneity was exhibited only in live *E. coli* cells. However, the bioaccumulation of perylene in live and dead *S. aureus* cells showed similar patterns with a low degree of heterogeneity (C.V=0.36). We found that the efflux systems associated with Tol C played an essential role in perylene bioaccumulation in *E. coli*, which caused a significantly lower accumulation and a high cell-to-cell heterogeneity. In comparison with *E. coli*, the Gram-positive bacteria *S. aureus* lacked an efficient efflux system against perylene. Therefore, perylene bioaccumulation in *S. aureus* was simply a passive diffusion process across the cell membrane. With the use of SMFM, the motion and distribution of perylene naon-clusters (PNCs) formed in water at very low concentration were visualized with high temporal and spatial resolution. More interestingly, the transport of PNCs across the cell membrane was also real-time captured, demonstrating that they entered macrophage cells by endocytosis. Supplementing the well-recognized routine of passive diffusion through membrane lipid bilayer, the uptake of HOCs in the form of nano-clusters by endocytosis was proposed to be an additional but important mechanism for their uptake into living cells. HOCs distributing in the environmental systems in the form of nano-clusters, as exemplified by PNCs in this study, may have significant implications for understanding their environmental fate and potential toxicological effects.

MO444

Impregnation factors of freshwater fish by organic micropollutants in the Marne Hydrographic network

N.F. Molbert, UPMC UMR METIS 7619 / Biogeochemistry; M. Chevreuil, EPHE / UMR METIS 7619; F. Alliot, EPHE / UMR Metis; R. Santos, HEPIA; J. Mouchel, UPMC UMR METIS CNRSUPMC; A. Goutte, EPHE / UMR METIS

Ecotoxicology faces the challenge of monitoring the levels of an increasing number of chemicals on biota. While persistent pollutants have been largely studied, several pollutants are metabolized, especially by vertebrates. Despite the higher toxic potential of metabolites compared to their parent compounds, little attention has been given to metabolites. Several persistent micropollutant families (Polychlorinated Biphenyls (PCB), OrganoChlorine Pesticides (OCP)) and metabolizable ones (Polycyclic Aromatic Hydrocarbons (PAH), phthalates, pyrethroid pesticides), as well as their metabolites were measured in a free-living freshwater fish, the European chub (*Squalius cephalus*, N = 113) caught by electrofishing operations in the Seine watersheds. The occurrence of pollutants were characterized in muscles and their metabolites in bile and liver using gas chromatography (GC-MS / MS) and high performance liquid chromatography (HPLC-MS / MS) coupled to a mass spectrometer. Body burdens of pollutants in chubs were then explained according to the environmental (surface water and sediments) contamination and individual parameters (age, body length, health status and parasitic load). Despite restrictive legislation, persistent pollutants (OCP, PCB) were found in all environmental matrices and fish tissues. Phthalates were the most abundant chemicals, with concentrations in fish muscles in the range 41.6-22000 $ng.g^{-1}$. Positive correlations were detected between the environmental pollution and the levels of persistent pollutants (PCB, OCP) in chubs, but not for the metabolizable chemicals, likely due to their rapid degradation and excretion. No correlation was found between micropollutant levels and health status of chubs, suggesting low ecotoxicological effects of these contaminants exposure in the Marne hydrographic network. Surprisingly, chubs infected by the acanthocephalan *Pomphorhynchus laevis* were less contaminated than the uninfected ones for OCP and phthalates. Further validations are needed to confirm the transfer of these pollutants from host to parasites and to investigate the potential benefits of this detoxification pathway for parasitized chubs.

MO445

Environmental occurrence and distribution of organic UV stabilizers in the sediment of the North and Baltic Seas

C. Apel, Helmholtz-Zentrum Geesthacht; R. Ebinghaus, Helmholtz-Zentrum Geesthacht / Department for Environmental Chemistry

Organic UV stabilizers are of emerging environmental concern due to their large production volumes and potential endocrine disrupting properties. UV stabilizers are widely used in plastic products, paints and coatings to improve the product stability against UV light. Furthermore, some UV stabilizers are approved as ingredients in personal care products like sunscreens to protect human skin against harmful effects of UV radiation. The pathways into the marine environment are either indirect by wastewater treatment plant discharges or direct by recreational activities like bathing and swimming. Four benzotriazole UV stabilizers are classified as SVHC (Substances of Very High Concern) under the EU legislation REACH. Numerous others are currently listed under the European community rolling action plan (CoRAP) to be (re-)evaluated in the next years. Due to their chemical properties, most UV stabilizers accumulate in sediment (logKow > 3) and have potential for persistence or pseudo-persistence. Environmental data for the coastal and marine environment are sparse. For this study 60 surface sediment samples of the North and Baltic Seas were analysed for 19 commonly used organic UV stabilizers. The sample pretreatment and analysis was carried out as following: First, the samples were homogenized with sodium sulphate. Afterwards, extraction and clean-up was performed using an accelerated solvent extraction (ASE-350, DIONEX, Germany) method. For this, 22 mL stainless steel ASE cells were filled with 3 g of 10% deactivated silica and approximately 5 g sediment that was spiked with appropriate isotopically labelled standards. The cells were extracted using dichloromethane for three 10 min-cycles at 100 °C. The extracts were solvent-changed to methanol and reduced in volume to 150 µL. The instrumental analysis was performed on a LC-MS/MS system (1290 Infinity coupled to 6490 triple quadrupole LC/MS, Agilent Technologies, Germany) equipped with an APPI-source and a C18 column (Eclipse Plus RRHD 1.8 µm, 2.1 x 150 mm, Agilent Technologies, Germany). Total Organic Carbon (TOC) analysis was carried out with aliquots of the freeze-dried samples using a LECO RC612 multiphase (Germany). This study shows levels of contamination and distribution of organic UV stabilizers in surface sediments of the North and Baltic Seas for the first time. Several substances have been identified in concentrations in the low ng/g dw range.

MO446

Is Lake Como a "uniform lake"? Information from its inhabitants (zooplankton and fish)

M. Mazzoni, University of Insubria (Como) / DiSTA; A. Buffo, F. Cappelli, Water Research Institute - Italian National Research Council IRSA-CNR; R. Perna, University of Insubria; S. Polesello, Water Research Institute- CNR / Water Research Institute; S. Valsecchi, Water Research Institute - Italian National Research Council IRSA-CNR; R. Bettinetti, University of Insubria / DiSTA

Lake Como, a subalpine lake (Northern Italy), is an oligomictic lake, with complete water mixing occurring after particularly windy and cold winters. It presents a typical shape of an upside “Y” where a western, eastern and northern basin can be identified. In more detail, the western branch is distinctly separated from the rest of the lake by an underwater ridge, where the highest depth is measured (425 m at

Argegno), and does not present an outlet, resulting in a longer real water renewal time. On the other side, waters of the eastern branch are encouraged to flow towards south directly through the Adda River, which is also the main inlet in the northern branch. Western and eastern branches also present different level of trophic status. In the present work, we investigate if these main morphological features can lead to differences a) in zooplankton density and biomass, b) in the interactions between zooplankton and fish and c) in levels of pollution between the pelagic areas of the two branches, evaluated along a seasonal sequence. Preliminary data tell that the taxa composition of the pelagic planktonic communities is the same in the basins but differences in density and biomass are highlighted. These differences are found in levels of contamination of legacy compounds (DDT, PCB) while there are not differences between the two branches in concentrations of perfluoroalkyl substances (PFAS).

MO448

Kinetic Sorption and Bioaccumulation of Hydrophobic Organic Chemicals in Marine Plankton Food Chain

F. Ko, National Museum of Marine Biology and Aquarium/ National Dong Hwa University / Institute of Marine Biology; C. Chu, National Dong-Hwa University / Institute of Marine Biology

Bioaccumulation and distribution of polycyclic aromatic hydrocarbons (PAHs) over different amounts of exposure time were investigated in the plankton food chain including phytoplankton and zooplankton. The simulated plankton food chain was using phytoplankton (*Tetraselmis chuii*), rotifers (*Brachionus* sp.), and copepods (*Apocyclops* sp.) cultured in a gas purging system with a steady supply of PAHs for 7 days in this study. The results show that PAH accumulation in plankton can be roughly divided into three sections: 0.2-1 hours, 1-24 hours, and 24-168 hours. The PAH concentrations in plankton varied greatly over the 0.2-1 and 1-24 hour time intervals, then approached study-state at 24-168 hours exposure. The low molecular weight PAHs (ACN, AC) were found at significantly higher levels in copepods than in rotifer and phytoplankton, but the high molecular weight PAHs (FA and PY) were found at significantly higher levels in phytoplankton, indicating that plankton might have selectivity towards PAHs. In principal component analysis (PCA), the plankton could be separated significantly into phytoplankton and zooplankton. Parts of the PAH accumulation found in rotifers and copepods were similar, demonstrating that PAH composition in plankton might be affected by trophic levels. All PAHs demonstrated significantly linear relationships between bioconcentration factor (BCF) and PAH hydrophobicity (K_{ow}) in plankton, however the different linear regression slopes of log BCF and log K_{ow} between phytoplankton, rotifer and copepod, suggested that the plankton have different pathways of PAH accumulation.

MO449

Do weathered multiwalled carbon nanotubes influence the distribution of the biocide triclocarban in a sediment-water system?

L. Benner, I. Politowski, M.P. Hennig, H. Hollert, RWTH Aachen University / Institute for Environmental Research; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics

Multiwalled carbon nanotubes (MWCNT) are widely used nanomaterials in a variety of different products and processes, well-known for their high sorption capacity. Due to increasing usage and production, exposure to the aquatic environment either accidentally or via disposal of CNT-containing products might increase likewise. Weathering processes like radiation can alter nanoparticle properties and lead to changes in their environmental behaviour. The duration of stay of MWCNT in the water phase is very short; due to agglomeration and aggregation processes they preferably settle down in sediments, which represent a potential sink for carbon-based nanomaterials. Nevertheless, during their stopover in the water phase they may interact with water dissolved xenobiotics, and thus alter the fate of these substances. Due to the lack of information on the influence of MWCNT on organic chemicals in aquatic ecosystems, proactive research is needed to estimate potential risks, especially for sediment-dwelling organisms as a part of the `Trojan Horse' effect. In the present study MWCNT were irradiated by simulated sunlight (300-400 nm) for 90 days. The weathered MWCNT (wMWCNT) were used to investigate their influence on fate and distribution of the biocide triclocarban (TCC) in a sediment-water system. This substance was chosen because of its hydrophobic properties and strong affinity to sorb on MWCNT. The concentration of wMWCNT has a significant impact on the distribution of TCC in natural water. 100 µg and 1000 µg wMWCNT/L in Milli Q water led to an adsorption (log k_{MWCNT} in OECD medium: 7.6 L/kg) of 10% and 65% ^{14}C-TCC respectively. We will report experiments on the distribution of TCC in water/sediment in presence of 1 mg wMWCNT/L. ^{14}C-TCC will be shaken for 2 h with 1 mg wMWCNT/L and subsequently incubated in a sediment-water system in the dark for 180 d. A scenario with ^{14}C-TCC only will serve as control. TCC is expected to sorb onto the wMWCNTs and accumulate in the natural sediment by fast sedimentation of wMWCNT-TCC complexes. Production and release of carbon based nanomaterials are predicted to further increase in the near future, thus the interactions of nanomaterials with organic pollutants will be of growing importance to assess environmental consequences. **Acknowledgements:** The work is supported by the European Project NANO-Transfer that receives funding from the Bundesministerium für Bildung und Forschung (BMBF) under agreement with the FP7 ERA-NET SIINN.

MO450

When technical limits triggers risk assessment for non-biodegradable insoluble pharmaceutical molecule

B. Journel, E. Beltran, CEHTRA SAS; P. Adrian, CEHTRA

Fate of substances in the environment are driven by numerous factors. Among them, substance's properties such as Henry's constant (*i.e.* water solubility and volatility) and hydrophobicity (in terms of K_{ow} and adsorption) have a non-negligible impact on how substance's behaviour is modelised in the different compartments (soil, water, sediment, air), to predict exposure levels in risk assessments. Pharmaceuticals represent a specific category of substances as they are often difficult to analyse, making experimental results more subject to imprecision. Due to analytical difficulties, parameters such as water solubility and K_{ow} are often expressed as "lower than" or "higher than" and have no defined value. Additionally, model softwares such as EUSES impose maximum value for K_{ow} and minimum value for water solubility as input parameters, whereas EU TGD spreadsheet allows to consider the experimental results. The decision on the parameter values and models to be considered needs then expert judgment. We will present the case of a pharmaceutical molecule currently studied, for which water solubility and then K_{ow} cannot be precisely measured experimentally. While staying regulatory compliant and reflecting the experimental results, the input values chosen for these parameters have a significant impact on calculated PECs for this insoluble molecule. Studies on fate and behaviour in soil and water/sediment systems are to be conducted, however considering the technical difficulties to analyse the molecule, and the route of environmental exposure, it may be necessary to define a category of molecules for which some of current regulatory requirements could be waived related to their chemical properties. A proposal for an appropriate risk assessment will be provided.

MO451

Effect of environmental characteristics on the bioavailability of hydrophobic organic compounds to fresh water organisms from natural aquatic systems

L. Teunen, University of Antwerp; C. Belpaire, Research Institute for Nature and Forest INBO; R. Blust, L. Bervoets, University of Antwerp / Department of Biology (SPHERE Research Group)

Many aquatic ecosystems are under persistent stress due to influxes of anthropogenic chemical pollutants. High concentrations can harm entire ecosystems and be potentially toxic to humans. The European Water Framework Directive (WFD) obliges member states to monitor chemical compounds in surface waters and to set quality standards that are protective for the ecological integrity. Generally, most of the target chemical compounds are able to be measured in environmental samples. However, in the case of highly hydrophobic compounds, their very low water solubility precludes direct measurement in water, and thus alternative monitoring strategies are needed. Accordingly, the WFD has formulated biota quality standards (BQS) which refer to concentrations of compounds that have to be monitored in fish and invertebrates. In the present study we are investigating the reliability and relevance of BQS by studying the relationships between concentrations of hydrophobic compounds in environmental compartments (mainly in sediment) and concentrations in biota. Our study encompasses 22 field locations at which we are monitoring the concentrations of a set of hydrophobic organic compounds and total Hg in both sediment and biota (fish and mussels). In addition, some sediment characteristics, i.e. organic carbon content (TOC) and clay content are measured and water characteristics are monitored, i.e. pH, oxygen level and conductivity. For each of the measured compounds multiple regressions are being constructed to establish the links between the concentration of compounds in biota and in sediment. The interpretation will take into account dissolved concentrations (where feasible) as well as general water and sediment characteristics. Identification of robust links between the extent of bioaccumulation and sediment and/or water concentrations would strengthen the basis for use of surrogate monitoring methods.

MO452

Personal care products (PCPs) in the southeastern coast of Brazil: implementation of the analytical method and environmental occurrence

T. Combi, Instituto Oceanográfico da Universidade de São Paulo / Instituto Oceanográfico; R.C. Montone, Universidade de Sao Paulo / Oceanographic Institute

The use of large amounts and a broad variety of chemicals that are potentially harmful to the environment (including persistent organic pollutants (POPs), personal care products (PCPs), flame retardants, and others) threatens water, sediment and biota. The existence of such a high number of compounds in the environment leads to interferences during chemical analysis, hindering the assessment of the occurrence and distribution of contaminants in environmental matrixes. Thus, the development and optimization of analytical methods that can detect multiple classes of compounds at very low levels and the contamination assessment in marine and coastal areas are among the most complex and current issues in environmental chemistry. Possible adverse effects related to some groups of PCPs, such as potential to accumulate through the trophic food chain and in human adipose tissues, endocrine disruption and hazard to coral reef conservation have been recently detected. Consequently, their study has become a priority among the main bodies responsible for protecting public health and the environment, such as the European Commission and USEPA. However, the current knowledge about the occurrence and fate of PCPs is still scarce, especially in less developed or developing countries as Brazil. Thus, this study aims to evaluate the occurrence of PCPs in surface sediments of selected areas along the southern and southeastern Brazilian coast through the optimization and implementation of a state of the art methodology. Preliminary results obtained for surface sediment samples from São Paulo coastal areas through microwave-assisted extraction (MAE) and triple-quadrupole mass spectrometer analyzes (GC-MS/MS) revealed the presence of UV-filters (especially octocrylene and EHMC) and fragrances (tonalide and galaxolide). The next steps of this work include testing additional extraction methodologies, extraction solvents and clean-up procedures to improve the detection and quantification of these compounds. The final results of this work will provide the first extensive dataset on the occurrence, levels and fate of PCPs in the Southern Atlantic which will not only contribute with new and rather scarce data but also with valuable information for regional and global inventories.

MO453

IFRA Environmental Standards and RIFM Safety Assessment Program Advances Update for 2018

A. Lapczynski, RIFM / Environmental Science; D.T. Salvito, Research Institute for Fragrance Materials (RIFM) / Dept of Environmental Science; C. Gonzalez, IFRA

To assure safety of fragrance ingredients in consumer products, International Fragrance Association expanded the fragrance industry's self-regulatory safety program with the development of IFRA Environmental Standards for both risk and hazard in 2008. Fragrance material risk assessments for these Standards are incorporated in the Research Institute for Fragrance Materials' (RIFM) testing program in coordination with its Expert Panel. To identify materials for risk assessment refinement, fragrance materials were screened using the RIFM Environmental framework and 2008 IFRA volume of use survey as reported for both Europe and North America. The Framework for this evaluation was published in Environment Toxicology and Chemistry (Salvito et al., 2002, 1301-1308). In addition, hazard assessment on these materials was also performed and reviewed. As a result nearly 3,000 materials were screened with preliminary risk quotients estimated to rank priority materials for risk assessment refinement. In an effort to provide greater transparency to the IFRA Environmental Standards, RIFM reports the most recent results of these additional tests (for both risk and hazard assessments) at both the annual SETAC NA and Europe meetings. These studies include persistence testing (ready biodegradation tests and die-away studies), bioaccumulation, and acute and chronic aquatic toxicity. Incorporating these new data in a second tier risk and hazard assessment for these materials will also be presented.

MO454

Comparison of different sampling techniques for the identification fire effluents from low-density polyethylene burning

A. Dhabbah, King Fahad Security College / Forensic Science Departmen; A.A. Stec, University of Central Lancashire / Centre for Fire Hazards and Science, School of Physical Sciences and Computing; Y. Badjah-Hadj-Ahmed, King Saud University / College of Science, Chemistry Department; R. Hull, University of Central Lancashire / Centre for Fire Hazards and Science, School of Physical Sciences and Computing,

Huge amounts of various polymers are being used in many fields with numerous benefits. However, their great ability to ignition and rapid flame spreading make these materials dangerous for human life and properties due to the release of highly toxic combustion products. The present work aims to investigate several methods of sampling and analysis of polycyclic aromatic hydrocarbons (PAHs) produced by controlled burning of low-density polyethylene (LDPE) using a toxicity tube furnace. Five different sampling methods were used: solid phase micro extraction (SPME), syringe, tedlar bags, sorption tubes, and gas-solution absorbers. The produced hydrocarbons were analysed by gas chromatography coupled to mass spectrometry with and without pyrolysis. The analysis of PAHs released from polyethylene combustion showed that emissions with a potentially negative impact on the human health and the environment are produced in significant concentrations. Among the tested techniques, the most convenient sampling method was that using syringe with a glass vessel which allowed detection of the highest amount of PAHs at both 800 and 600°C, then followed by SPME. On the other hand, the use of gas-solution absorber (midget impinger) showed poorer results. Regarding the use of tedlar bags and sorption tubes, they did not give satisfactory results. Several carcinogenic or possibly carcinogenic compounds were identified in the combustion products, such as benzene, naphthalene, anthracene and pyrene.

MO455

PbTk modelling of super-hydrophobic chemicals

W. Larisch, K. Goss, Helmholtz centre for environmental research - UFZ / Analytical Environmental Chemistry

It is a widespread opinion that super-hydrophobic chemicals are not taken up by fish [1–3]. But so far, we haven't seen sufficient evidence why they should not cross membranes or aqueous boundary layers, although these processes might be kinetically slow. Super-hydrophobic chemicals are very challenging for controlled exposure experiments. Here we use the few existing data points of a fish feeding study with Dechlorane (log Kow (estimated) = 11.6) with our recently published PbTk model, TK-fish, to shed more light on this issue. We first validated the oral up-take pathway in our model and found that facilitated transport via albumin and bile micelles through the aqueous boundary layers must accounted for, for hydrophobic chemicals such as HCB in order to get correct results. Subsequent simulations with the super-hydrophobic chemical Dechlorane revealed that for an oral uptake route the diffusive transport through aqueous boundary layers in the gastro-intestinal tract and in the blood is indeed the limiting process. Good agreement of the predicted model results with measured values indicates that there is no principal hindrance for the oral up-take of super-hydrophobic chemicals. The results also indicate that it would take roughly 2 years or more for a steady state to be established which is too long for an experimental exposure study. 1. Dyer SD, Bernhard MJ, Cowan-Ellsberry C, Perdu-Durand E, Demmerle S, Cravedi J-P. 2008. In vitro biotransformation of surfactants in fish. Part I: linear alkylbenzene sulfonate (C12-LAS) and alcohol ethoxylate (C13EO8). *Chemosphere*. 72:850—862. 2. Sakuratani Y, Noguchi Y, Kobayashi K, Yamada J, Nishihara T. 2008. Molecular size as a limiting characteristic for bioconcentration in fish. *J. Environ. Biol.* 29:89—92. 3. 2016. *Guidance on Information Requirements and Chemical Safety Assessment, Chapter R.11: PBT/vPvB assessment Draft Version 3.0,European Chemicals Agency, Helsinki*. https://echa.europa.eu/documents/10162/23047722/ir_csa_r11_pbt_peg_en.pdf/ddac9031-daa4-4995-8ecf-3738162ba4e8

Migratory bird species at risk - the role of pesticides and other chemicals (P)

MO456

Main scientific gaps in knowledge of risk from pesticides to [migratory] wildlife globally, and potential contribution of WTIG to CMS questions

C.A. Bishop, Environment and Climate Change Canada / Wildlife Research Division

MO457

Main scientific gaps in knowledge of risk from rodenticides to [migratory] wildlife globally, and potential contribution of WTIG to CMS questions

P. Berny, VETAGRO-SUP / Toxicology

MO458

Main scientific gaps in knowledge of risk from Pb ammunition and shot to [migratory] wildlife globally, and potential contribution of WTIG to CMS questions

R. Cromie, Wildfowl & Wetlands Trust

MO459

Main scientific gaps on knowledge of NSAIDs [migratory] wildlife globally, and potential contribution of WTIG to CMS questions

M. Taggart, University of the Highlands and Islands / Environmental Research Institute

MO460

Main scientific gaps on knowledge of deliberate poisoning to [migratory] wildlife globally

M. Odino, Independent Environmental Services Professional

Big data analysis in ecotoxicology: how to get new information out of existing data? (P)

TU001

Holistic evaluation of long-term field effect earthworm studies with the fungicide Boscalid

F. Staab, BASF SE; J. Roembke, S. Jaensch, ECT Oekotoxikologie GmbH; P. Kabouw, BASF SE; S. Braaker, BASF France S.A.S.

In order to place a plant protection product on the market, the product and its active substances need to demonstrate an acceptable risk to earthworm communities. The current European risk assessment scheme follows a tiered approach using worst case environmental concentrations and endpoints from earthworm reproduction laboratory studies in tier 1. For the active ingredient Boscalid no risk to earthworms has been identified based on the chronic laboratory studies provided by BASF to EU registration authorities. However, for one of the formulated products containing Boscalid the tier 1 assessment did not allow to exclude a potential long-term risk to earthworms in the field. Therefore, a comprehensive field study program was conducted in different crops and field sites in Germany between 2000 and 2010. The study program went beyond regulatory requirements and comprised 6 independent long-term field studies that ran up to five years and were accompanied by a comprehensive residue analysis program. The number of earthworm field data generated is - to our knowledge - one of the highest ever collected for one plant protection product. We evaluated the extensive data set under "holistic" considerations putting representativeness/comparability of examined earthworm communities, site- and soil properties of the different locations as criteria in the analysis. Based on these criteria a statistical assessments of representative and comparable earthworm communities in relation to the field exposure were conducted. The assessment revealed that – using data from representative and comparable study sites - there was no concentration related effect of a five-year use of the product regarding diversity and abundance of different earthworm communities.

TU002

Contextualising statistically significant differences observed in mesocosm studies using historical control data

F. Joyce, Cambridge Environmental Assessments; H.S. Schuster, Cambridge Environmental Assessments (CEA) / Aquatic Ecotoxicology

Mesocosms (which aim to replicate communities residing in edge-of-field waterbodies) are used as part of the higher tier aquatic risk assessment for plant protection products (PPPs) in the EU registration process. When setting up mesocosm studies, care should be taken to standardise communities present in each replicate to reduce variability and maximise statistical power; indicated by minimum detectable differences (MDDs). However, being dynamic and complex systems, variability can often still occur between replicates, which can affect the reliability and interpretation of the results. Statistically significant differences can occur due to natural variability rather than biologically relevant effects, but demonstrating this to regulatory authorities using the results of a single mesocosm study can be challenging. One option is to contextualise the experimental results from a single study using available historical control data; this is an approach often used for laboratory studies performed under standard conditions. It is, therefore, proposed that this approach can be extrapolated to mesocosm studies, given that they are also performed under standardised conditions as much as possible. Cambridge Environmental Assessments (CEA) have a wealth of control data from historical mesocosm studies, with samples collected in spring, summer and autumn, thus capturing the variability in population and community dynamics over multiple years and seasons. Here we present our review of this historical control data, and how this provides a baseline to aid interpretation of results from individual studies, allowing an assessment of biological relevance and thus the appropriateness of influencing the regulatory acceptable concentration (RAC). When integrated into the aquatic risk assessment, this will represent a *realistic* worst-case scenario.

TU003

Enhancing the utility of the ECOTOX knowledgebase via ontology-based semantics mapping.

K.A. Fay, CSRA, Inc.; C. Elonen, U.S. EPA/ORD/NHEERL; D.J. Hoff, U.S. EPA ORD / Mid Continent Ecology Division; C. LaLone, U.S. EPA / Mid Continent Ecology Division; A. Pilli, M. Skopinski, CSRA, INC; R. Wang, U.S. EPA / Exposure Methods and Measurements Division

The US Environmental Protection Agency's Ecological Toxicology (ECOTOX) knowledgebase contains more than 30 years of reported single chemical toxicity effects data on aquatic and terrestrial organisms. Approximately 900,000 test results covering more than 11,000 chemicals and 12,000 species are available in ECOTOX. While the database is currently used by many sectors for a variety of purposes, a future goal is to allow for computational modeling of the data to identify novel adverse outcome pathways and networks, and assist in predicting species sensitivity. To accomplish these goals, the initial steps entailed:1) validating the chemicals within ECOTOX 2) mapping species to NCBI taxids and 3) mapping all relevant ECOTOX codes to corresponding ontological terms so chemical effects can be turned into computable phenotypic ontology classes. To semi-automate the code mapping, a Java-based lookup tool was developed using the ontology browser BioPortal (https://bioportal.bioontology.org/) REST API to conduct batch code mapping. This tool was designed to make use of BioPortal's annotator and recommender functions so that all ontological class identifiers relevant to a particular ECOTOX term would be returned and specific ontologies recommended. Using this approach, the majority of the 2000+ ECOTOX codes were mapped to ontological class identifiers; some terms required multiple identifiers to properly describe them. Further, manual curation was necessary for a proportion of terms. The results of the automated code mapping approach were evaluated against a set of manually annotated phenotypes as induced by exposures to ten well studied chemicals (atrazine, bisphenol A, cadmium chloride, chlorpyrifos, copper sulfate, cypermethrin, dioxin, EE2, malathion, or Tris(1,3-dichloroisopropyl) phosphate) in six vertebrate species (carp, zebrafish, fathead minnow, mouse, rat, trout). *The content of this presentation neither constitute nor necessarily reflect US EPA policy.*

TU004

ECOTOX Knowledgebase: New tools for data visualization and database interoperability

C. Elonen, U.S. EPA/ORD/NHEERL; J. Olker, C. LaLone, U.S. EPA / Mid Continent Ecology Division; D.J. Hoff, U.S. EPA ORD / Mid Continent Ecology Division; S. Erickson, M. Skopinski, S. Casey, A. Pilli, K.A. Fay, CSRA, Inc.

The ECOTOXicology knowledgebase (ECOTOX) is a comprehensive, curated database that summarizes toxicology data from single chemical exposure studies to terrestrial and aquatic organisms. The ECOTOX Knowledgebase provides risk assessors and researchers consistent information on toxic effects of chemical substances for use in deriving benchmarks and establishing criteria. ECOTOX has the capability to refine and filter data searches by 16 parameters (e.g. Species, Chemical, Effect, Control, Year, etc.) and customize output selections from over 100 data fields. Study details such as species taxonomic hierarchy, chemical purity, routes of exposure, and all calculated or statistically derived endpoints provided by the authors is encoded in discrete data fields for each test result. During the past 10+ years, ECOTOX has aligned the coding of the aquatic and terrestrial references by the addition of data fields, adapted search terminology to better focus literature searches, and updated search screens. To meet the data needs of 21st century toxicological assessments, new tools have been integrated into ECOTOX to improve data mining capabilities for end users such that environmental regulatory, the regulated industry, and researchers can more effectively and efficiently search and use existing toxic effects data. New data visualization and filtering options have been added to aid in data exploration. Efforts to enhance interoperability with other EPA databases have been employed to assist in efficiently accessing necessary data. These additions will be available in ECOTOX Knowledgebase version 5.0, to be released in FY18.

TU005

Edaphostat - A web application for automated and interactive meta-analysis of environmental data from the Edaphobase data warehouse

J. Hausen, RWTH Aachen University; B. Scholz-Starke, M. Ross-Nickoll, RWTH Aachen University, Institute for Environmental Research / Institute for Environmental Research; R. Ottermanns, RWTH Aachen University / Institute for Environmental Research

A steadily increasing number of databases in ecotoxicology and ecology combine and merge data from different studies and research programs. Usage of these databases promises new insights in species occurrence and distribution, allowing e.g. the derivation of reference values and answering environmental questions on a larger scale. However, data from different studies are often very heterogeneous because the studies differ in scope, design, and parameters sampled. This makes meta-analysis a challenging task, as database users need to know how to select, combine and process this mixed origin-data. Automated analysis tools, which are customized for certain data warehouse applications, can be a solution to this problem. We present a web application called "Edaphostat", which is part of the Edaphobase data warehouse (https://portal.edaphobase.org/). Edaphobase combines spatially explicit information on quantities of soil organisms, environmental parameters, and vegetation. The data in the warehouse are coming from museum collections, field data, literature and unpublished results. Edaphostat performs several steps of data cleaning, formatting, and transformation to make datasets comparable. Preprocessed data are analyzed and the results are visualized as interactive plots and dashboards. The tool depicts species distribution alongside environmental gradients (for example pH and C/N) and habitat parameters (such as soil classes) and species settlement in ecological niches. Edaphostat makes use of the flexibility of the Edaphobase data warehouse allowing meta-analysis of data selected by e.g. area, time period or study design. It performs automated analysis of environmental data to assess species-specific autecological preferences and ecological niches.

TU006

Deriving USEtox aquatic freshwater toxicity Effect factors from the REACH database for thousands of chemicals using R-Studio program

E. Saouter, EU Commission JRC / Sustainable Assessment UNit; F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Resources Bioeconomy Unit; A. Gissi, European Chemicals Agency; P. KARAMERTZANIS, ECHA European Chemicals Agency; J. Provoost, European Chemical Agency ECHA; S. Proenca, EU Commission Joint Research; D. Versteeg, EcoStewardship LLC

Product Environmental Footprint (PEF) and Organisational Environmental Footprint (OEF) form a core part of the Commission Recommendation "on the use of common methods to measure and communicate the life cycle environmental performance of products and organisations" (2013/179/EU). The potential impact of chemicals emitted during the life cycle of a product is assessed via the USEtox multimedia fate model [3]. This model requires for each single chemical dozens of physico-chemical parameters as well as data of ecotoxicity to freshwater aquatic life and toxicity to human for cancer and no-cancer effects. For PEF/LCA, those data are required for thousands of chemicals using the most up-to-date information [4,5]. The EU commission Joint Research Centre has obtained from the chemical agency (ECHA) for more than eight thousand chemicals all the physico-chemical properties (166'926 test results), ecotoxicity (242'729 test results) and human toxicity data (41'381 test results) available in the IUCLID 5.5 database (as of March 2017). The database has been used to calculate unique value for chemical properties and toxicity indicators for thousands of chemicals to be used in the USEtox model. The poster presents the methodology applied for the selection of the aquatic toxicity data available in REACH, the set of criteria used to derive various level of quality data to meet the requirement to produced Effect Factors for as many chemical as possible, and the various calculation procedure to derive final chemical effect factors. Correlation between acute and chronic toxicity for thousands of tests, for each taxonomic groups has been established, as well as the feasibility to calculate effect values based on Species sensitivity distribution. The following final calculation have been performed for thousands of chemicals: - Acute and Chronic species geometric means with standard deviation and number of individual test available per species - Arithmetic average of all the log of the species geometric mean with standard deviation and count of species as well as count of SSD group for each chemical - Lowest Acute and Chronic species geometric means with standard deviation - Comparison of the chemical toxicity based on the above calculations and chemical ranking based on GHS and CLP.

TU007

Deriving physico-chemical input data for the USEtox model from the REACH database for thousands of chemicals using R-Studio program

F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Resources Bioeconomy Unit; S. Proenca, EU Commission Joint Research; A. Gissi, European Chemicals Agency; E. Saouter, EU Commission JRC / Sustainable Assessment UNit

Product Environmental Footprint (PEF) and Organisational Environmental Footprint (OEF) form a core part of the Commission Recommendation “on the use of common methods to measure and communicate the life cycle environmental performance of products and organisations” (2013/179/EU). The potential impact of chemicals emitted during the life cycle of a product is assessed *via* the USEtox multimedia fate model. This model requires for each single chemical dozens of physico-chemical parameters as well as data of ecotoxicity to freshwater aquatic life and toxicity to human for cancer and no-cancer effects. For PEF/LCA, those data are required for thousands of chemicals using the most up-to-date information. The EU commission Joint Research Centre has obtained from the chemical agency (ECHA) for more than eight thousand chemicals data regarding physico-chemical properties (166’926 test results, as of March 2017) available in the IUCLID 5.5 database. These data have been processed to automatically derive accurate values for six of the physico-chemical properties required by UseTox for fate modelling: Kow, Koc, vapour pressure, water solubility, Henry law constant and biodegradability; in addition, adsorption partition coefficient (Kd) to suspended matter, sediments and soil for inorganic compounds were determined. In order to provide high quality results, criteria were applied for selecting data on the basis of their reliability (assessed by Klimisch scores), purpose and study type. Moreover, other specific criteria were defined for each properties according to the method used, such as experimental condition (temperature and pH). Geometric mean and the coefficient of variation, for their reliability evaluation, of the consistent data selected was performed. A correlation analysis of the results with values previously included in USEtox and with values obtained with computational methods (QSAR/QSPR) was established to assess the quality of this proposed automated approach. A quality levels approach is proposed. This, paired with the abovementioned statistical index, represents a helpful support to the user for evaluating the reliability of each parameter used in the fate model. For chemicals with no available data or not satisfying the minimum quality requirements, physico-chemical properties were derived using in-silico tools (QSAR/QSPR). In particular, OECD QSAR toolbox and the EPIsuite estimation models played a fundamental role for this data gap filling purpose.

TU008

Toward a possible Toxicity Test Battery integrated Index for Nanomaterials

M. Oliviero, University Parthenope; s. schiavo, ENEA CR; s. manzo, ENEA / SSPT-PROTER-BES

Nanotechnology is a rapidly expanding field of research continuously producing novel materials with nanoscale properties (nanomaterials, NMs), as result, it is inevitable that NMs will enter the aquatic environment. Usuallythe ecotoxicological approach is generally based on a battery of bioassays with organisms belonging to different trophic levels, by choosing the appropriate endpoint for each species. While these endpoints, taken individually, can indicate the presence of a potentially deleterious effect, it is often difficult to combine these effects into an assessment of the overall status for the selected environment. Therefore is necessary to integrate all results to evaluate the risk for NMs. To synthesize the results obtained with a battery of ecotoxicological tests, different approaches were proposed.such as Toxicity test battery integrated index (TBI). This index has the advantage to put in evidence the differences between the samples. However there are still some open issues and TBI integration procedure needed to be modified according to testing sample or substance in order to represent the test sensitivity towards the matrices.In particular, when NMs are investigated also different physic-chemical behaviour and interaction with organisms should be taken into account.Therefore, the aim of this work is to study the suitability of TIB procedure for the NM to determine the needed modification for tailoring the data integration. In particular, we considered metal bearing nanoparticles (NPs) such as TiO_2, SiO_2 and ZnO and a battery of toxicity test with organisms of different biological complexity and representative of different trophic levels with the aim to establish a unique toxicity ranking. From the analysis of the results integration with TBI it could be highlighted that to define the hazard associated with NPs is necessary to tailor the index parameters on specific NMs physic-chemical characterization. Moreover, to make the results more reliable, together with a larger number of tests, a longer testing time for some organisms and other endpoints (genotoxic and cytotoxic parameters) should be utilized

TU009

Historical analysis of the use of plant protection products in apple orchards (1970-2014): Combining handwritten farmers records with electronic data

L. de Baan, Agroscope / Institute for Plant Production Sciences IPS; M. Mathis, J. Stocker, Agroscope; O. Daniel, Agroscope / Institute for Plant Production Sciences IPS

Plant protection products (PPPs) are used to protect crops against pests and diseases and ensure yields and quality of crops. Because they are biologically active, they can cause negative side effects on the environment or humans. Data-sets on the use of PPP for specific crops over a long time would allow to get a better knowledge on the potential emissions of PPP within and beyond agro-ecosystems. However, consistent long-term datasets are mostly lacking. In addition, historic farmers’ records are often only available in handwritten paper format. In Switzerland, data on PPP use in apple orchards has been voluntarily recorded by farmers since the 1950-ies, to evaluate farm economics. Up to the 1990-ies, data were only available in handwritten paper format, since 1997 they were collected electronically. In this study, we digitised the handwritten records and combined it with the electronic data. We first developed a concept, how the handwritten records can be entered into a database, which contains similar information as the electronic data. We collected data on farms (productivity), apple plantations (year of plantation, size, type, variety), and plant protection measures per plantation (product, dosage, date of application). \n We also developed procedures to handle missing data and to detect mistakes in the indicated dosage or field size. Finally, a dataset of spray sequences in apple orchards over the period 1970-2014 was analysed, regarding the number of treatments per PPP category (e.g. fungicides), the total amount of active ingredients applied per season and PPP category and the ranking of chemical groups per PPP category. While the average number of treatments and the average amount of active ingredients remained within a similar range, major changes were detected in the composition of applied active ingredients. For example, in the 1970-ies and 80-ies, more than 75% of all insecticide treatments were organophosphates. After 1986, organophosphates were quickly replaced by carbamates and benzoylureas, and today only contribute to about 10% of all insecticide treatments. This study illustrates, that for a historic analysis of pollutants it is sometimes unavoidable to first digitise handwritten data, because the digital revolution is a rather recent trend. This effort resulted in a unique 44 year time series of PPP use in apple orchards. In a next step, we will analyse the historic development of ecotoxicological risks of PPP usage in apple orchards.

TU010

Using long-term datasets to assess the impacts of neonicotinoids on farmland bird populations in the UK over the last 21 years

R. Lennon, The University of York / Environment; N. Isaac, NERC Centre for Ecology & Hydrology; R. Shore, Centre for Ecology & Hydrology (NERC); K. Arnold, University of York / Environment; W. Peach, Royal Society for the Protection of Birds; C. Brown, University of York / Environment Department

Thus far the majority of research regarding neonicotinoids (NNs) has been focused on pollinator species; however, little work has been done to investigate the potential long-term impacts of these pesticides on other taxa, such as farmland birds. Birds can be directly exposed to NNs via two main exposure routes: ingestion of NN-coated grain, or seedlings germinated from coated grain. With bird abundance data, NN usage records and UK cropping data, a poisson log-linear generalised mixed model was used to investigate whether there are any impacts of NN use on farmland bird populations over a period of 21 years. Specifically, the main objectives of this study were to 1) establish whether there is a relationship between species population growth and NN usage in the UK, 2) establish whether species traits, such as body weight and home range are correlated with any effect of NNs on species population growth, and 3) to ascertain whether hypothesised exposure risk (direct) was able to explain differences between the impacts of NNs on individual species population growth. A total of 54 bird species were modelled, for which the estimated effect of NNs on population growth were highly varied. Relationships between the estimated effects and species traits, including hypothesised risk to exposure will be reported.

TU011

Regression-based models reveal sources of pollutants in Norwegian marine sediments

G. Everaert, Flanders Marine Institute / Laboratory of Environmental Toxicology and Aquatic Ecology; A. Ruus, NIVA / NIVA; D. Hjermann, NIVA Norwegian Institute for Water Research; K. Borga, Department of Bioscences, University of Oslo / Department of Biosciences; N.W. Green, NIVA Norwegian Institute for Water Research / marine pollution; S. Boitsov, Institute of Marine Research; H. Jensen, Geological Survey of Norway; A. Poste, Norwegian Institute for Water

Research
We characterized spatial patterns of surface sediment concentrations of seven polychlorinated biphenyls (PCBs), seven polycyclic aromatic hydrocarbons (PAHs), three chlorinated pesticides and five metals in Norwegian waters and Skagerrak. In total, we analysed 5,036 concentrations of 22 chemical substances that were measured between 1986 and 2014 at 333 sampling sites by means of generalized additive models (GAMs). We found that GAMs with organic carbon content of the sediment and latitude and longitude as covariates explained ca. 75% of the variability of the contaminant sediment concentrations. For metals, a predominantly hotspot-driven spatial pattern was found, i.e. we identified historical pollution hotspots (e.g. Sørfjord in western Norway) for mercury, zinc, cadmium and lead. Highest concentrations of PAHs and PCBs were found close to densely populated and industrialized regions, i.e. in the North Sea and in the Kattegat and Skagerrak. The spatial pattern of the PCBs suggests the secondary and diffuse atmospheric nature of their sources. Atmospheric inputs are the main sources of pollution for most organic chemicals considered, but north of the Arctic circle, we found that concentrations of PAHs increased from south to north most likely related to a combination of coal-eroding bedrock and the biological pump. The knowledge acquired in the present research is essential for developing effective remediation strategies that are consistent with international conventions on pollution control.

TU012

Application of a 'weight-of-evidence' model for assessing sediment quality and associated hazard with offshore gas platforms discharging produced water

A. Tornambè, ISPRA / National Center for Laboratory Networking, Ecotoxicology Area; L. Manfra, R. Di Mento, G. Moltedo, B. Catalano, ISPRA Institute for Environmental Protection and Research; G. Martuccio, ISPRA Institute for Environmental Protection and Research / National Center for Laboratory Networking Ecotoxicology Area; C. Sebbio, G. Chiaretti, O. Faraponova, M. Amici, C. Maggi, G. Romanelli, G. Sesta, G. Granato, F. Venti, P. Lanera, S. Maltese, F. Onorati, ISPRA Institute for Environmental Protection and Research

Environmental quality assessments and monitoring plans are key tools to all activities related to potential contamination of ecosystems, including marine systems. Potential effects of oil/gas production activities in Adriatic Sea (Italy) are successfully investigated since 2000 by water and sediment chemical analyses, sediment grain size analysis and bioaccumulation on native mussels. In this study, a multidisciplinary approach including chemical analyses, grain size analysis and bioassays on marine sediment, together with bioaccumulation and biomarker investigations in polychaetes exposed to sediment, is applied to assess potential impact due to offshore platforms and produced water (PFW) discharge. PFW is a complex mixture of contaminants and is the main discharge of gas/oil platforms. Marine sediment around two gas platforms (Central Adriatic region) were physic-chemically characterized by sampling of twenty-four stations at increasing distance from the platform/discharge, and in particular four stations, located at 0, 25, 50 and 100 m along the main local current, also for ecotoxicity. Different inorganic and organic contaminants were analyzed in sediment. To assess the effects of pollutants at different levels of trophic web, results of a bioassays battery composed by three different species (*Vibrio fisheri, Dunaliella tertiolecta, Tigriopus fulvus*) were considered. Moreover a battery of biomarkers at different biological levels together with bioaccumulation of some organic and inorganic contaminants were analyzed in polychaetes (*Hediste diversicolor)* exposed to sediment under laboratory conditions. A multidisciplinary weight of evidence (WOE) study was carried out, integrating different lines of evidence (LOE) as sediment chemistry, bioaccumulation, bioassays and biomarkers of four stations for each platform. These LOEs were elaborated within a quantitative WOE model which provides a synthetic hazard index for a comprehensive assessment of hazard associated to potential contaminated sediments. The WOE elaboration allowed to better summarize complex dataset of results, providing a more realistic evaluation of hazard and risk for produced water discharges.

TU013

Utilising biomarkers in a multispecies approach to relate organochlorine exposure and biological effects

V. Wepener, North-West University - School of Biological Sciences / School of Biological Sciences; J.H. van Vuren, University of Johannesburg / Zoology; R. Gerber, North-West University / Unit for Environmental Sciences and Management; N. Smit, NorthWest University / Environmental Sciences and Management

Due to their persistence and global distribution the use of organochlorine pesticides (OCPs) have been banned in most countries around the world. However, in a number of countries the application of DDT as malaria vector control agent is still allowed. This practice is not without controversy and reports on ecological and human health effects are increasing. The Phongolo River floodplain in north-eastern South Africa is a high risk malaria area where DDT is used as vector control agent through indoor residual spraying (IRS). This region is also regarded as a biodiversity hotspot in southern Africa and concern has been raised regarding the risk posed as a result of the long term use of DDT. Over the past seven year's studies have been undertaken to determine the degree of DDT exposure in the aquatic ecosystem through analyzing DDT and other OCP bioaccumulation in a number of different aquatic species. Concomitant biomarker analyses were undertaken to determine the biological effect of the DDT exposure. In this poster we collated and integrated the exposure (DDT and HCH bioaccumulation) and effect (biomarker) data of the different studies to test the hypothesis that increased DDT exposure will elicit similar biological responses across species. Bioaccumulation of DDT (and its metabolites) and HCHs were measured before and after IRS application periods in two decapoda, six fish and two amphibian species. Biomarkers of exposure (cytochrome P450 and acetylcholine esterase) and effect (catalase, superoxidase dismutase, malondialdehyde, protein carbonyl, and cellular energy allocation) were analysed in the same organisms. Using principal component analysis and discriminant functional analysis the exposure and effect data were integrated to elucidate the responses of aquatic biota to OCP exposure. Although higher trophic level organisms (i.e. tigerfish - *Hydrocynus vittatus* and Müller's clawed frog - *Xenopus muelleri*) displayed the highest DDT bioaccumulation there were no distinct biomarker responses evident. When exposure data of another banned OCP, *γ*-Hexachlorocyclohexane, were included in the analysis, significant relationships with cytochrome P450 and lipid energy reserves were obtained. The result therefore indicated that biological responses were not related to DDT but rather to HCH exposure.

Microbial community ecotoxicology in environmental risk assessment and ecosystem monitoring (P)

TU014

Identifying bacterial indicator taxa along an urbanization gradient in stream ecosystems

M. Simonin, Duke University / Biology; K.A. Voss, Regis University; B.A. Hassett, J.D. Rocca, S. Wang, Duke University / Biology department; C.R. Violin, University of North Carolina at Chapel Hill / Biology department; E.S. Bernhardt, Duke University / Biology department

The advent of high throughput sequencing enabled microbial eco(toxico)logists to better characterize the impact of stressors and especially pollution on microbial community structure. However, the overwhelming amount of information generated by sequencing and the high diversity of microorganisms led us to focus our analyses mainly at the community or phylum levels, ignoring all the key ecological knowledge potentially gained at the population level. In this study, we tried to move beyond beta-diversity patterns and exploit the full potential of high-throughput sequencing data by characterizing the response of individual taxa (OTUs) to a multiple stressor gradient and identifying bacterial indicator taxa. Taking inspiration from classic gradient analyses used for macro-organisms in Ecology (Threshold Indicator Taxa Analysis - TITAN, logistic and quadratic regressions), we identified bacterial taxa that presented positive, negative, neutral or subsidy-stress responses to a well characterized urbanization gradient in 41 streams in the Raleigh-Durham area (North Carolina, USA). We used a combination of environmental variables (% development, % forested, sediment Zn concentration, biotic index) that were significantly correlated to bacterial community structure to identify reliable bacterial indicator taxa along this multiple stressor gradient. Using TITAN, we identified more bacterial indicator taxa negatively impacted by urbanization than positively impacted (138 and 56 OTUs, respectively). Using quadratic regressions, we found 140 OTUs presenting a subsidy-stress response to the gradient. We observed that two bacterial families were strongly and consistently decreased by urbanization: Acidobacteriaceae (Acidobacteria) with 50% of OTUs identified as pure and reliable indicator taxa and Xanthobacteraceae (Alpha-Proteobacteria) with 39% of indicator taxa. Positive responders were distributed all over the phylogenetic tree and the family Comamonadaceae (Beta-Proteobacteria) presented the highest number of indicator taxa (14%). We calculated with TITAN that the community-level threshold, indicating the peak along the gradient where the maximum decline in all negative responders happened, was at 12.1% development. This community-level threshold occurs at very low levels of urbanization, indicating a high sensitivity of microorganisms to urbanization and the potential of bacteria to be used in bioindication or monitoring along with more traditional indexes.

TU015

Diuron sorption in freshwater biofilms: determination of isotherms

B. CHAUMET, Irstea; N. Mazzella, S. Morin, Irstea Bordeaux / UR EABX

In 2000, the EU Water Framework Directive (directive 2000/06/EC) was implemented with the objective of reaching the good ecological status of rivers. 45 chemicals were indexed as priority including 19 pesticides. The biofilm is at the basis of the trophic chain in aquatic environments and considered as an excellent bioindicator for water quality assessment (Edwards and Kjellerup 2013) because of its ability to integrate contamination (Vercraene-Eairmal et al. 2010). In this study, we used a photosynthesis inhibitor herbicide: diuron, one of the priority substances to the EU Water Framework Directive. Previous experiments carried out in order to characterize diuron bioaccumulation in biofilms, with two different exposure concentrations, suggest that pesticide uptake by microorganisms was not proportional to contaminant concentration in the water. For this experiment, we supposed that diuron absorption isotherms are not linear, and in order to confirm this hypothesis, bioaccumulation and toxic impact were simultaneously assessed at the equilibrium. To that aim, mature biofilm previously grown on glass slides during one month was exposed in channels at 6 increasing concentrations of diuron: 0, 1, 5, 10, 25 and 50 µg.L^{-1} for two hours, with a flow velocity of 2 cm.s^{-1}. Then, Langmuir isotherm equation (Praus et al. 2007) was fitted to the bioaccumulation data. During the determination of the isotherm, a plateau was reached over 5 µg.L^{-1} of diuron in the water. This suggested that all absorption sites were saturated, and then diuron concentration in the biofilm became independent of diuron concentration in the water. The fitting of a Langmuir isotherm allowed to estimate a maximal diuron concentration in biofilm at 2073 µg.g^{-1}, and an equilibrium constant of 0.378. Photosynthesis inhibition was correlated (R^2=0.75) to diuron concentration in the water. The data did not clearly highlighted a relationship between bioaccumulation and photosynthesis inhibition. This study establishes that diuron bioaccumulation in biofilm is nonlinear, and allows to calculate the equilibrium constant and maximal capacity of the biofilm regarding diuron uptake. These two constants can be used to further prediction of diuron bioaccumulated in biofilm from concentration in the water. The innovative coupling of toxicokinetic and toxicodynamic approaches would provide original information about pesticide behaviour and impact in periphytic microorganisms.

TU016

New insights into the biotransformation of sulfluramid: role of ammonia oxidizing bacteria and community shifts

T. Yin, National University of Singapore / Civil and Environmental Engineering; Y. Yang, S. Te, National University of Singapore; K. Gin, National University of Singapore / Civil & Environmental Engineering

Emerging organic contaminants (EOCs), such as perfluoroalkyl and polyfluoroalkyl substances (PFASs), are ubiquitously detected in the environment and have raised increasing concerns due to their adverse effects on ecosystems and humans. N-ethyl perfluorooctane sulfonamide (N-EtFOSA), belonging to PFASs, is used as the active ingredient in the pesticide, Sulfluramid, which is particularly important in the control of leaf-cutting ants in some developing countries. Previous studies have investigated its degradation kinetics and pathways in activated sludge, marine sediments and soil. However, little information is available on the contributions of different microbes to the biotransformation of N-EtFOSA. This study used Allylthiourea (ATU), an inhibitor of ammonia monooxygenase (AMO), to investigate the relative contributions of ammonia oxidizing bacteria (AOB) and other members to N-EtFOSA biotransformation and find potential N-EtFOSA degraders by analysing the microbial community shifts. In the reactors with ATU addition, N-EtFOSA was degraded faster with an apparent half-life of 1.3 days, which indicated that ATU had actually enhanced the biotransformation of N-EtFOSA. This implied that AMO was probably not involved in the biotransformation of N-EtFOSA, and thus the inhibition of AMO by ATU had no adverse effect on its biotransformation. ATU-treated sample was more diverse with a Shannon index of 4.04 while that of the ATU-untreated sample was 2.43. The abundance of *Candidatus Protochlamydia* increased significantly in the ATU-treated sample, which suggested that this genera could be a potential degrader for N-EtFOSA. Future work needs to determine the genes involved in the biotransformation process using metagenomics and metatranscriptomics.

TU017

How can three herbicides impact the fatty acids of the freshwater diatom Gomphonema gracile ?

F. Demailly, IRSTEA Bordeaux / Ifremer Nantes / EPOC (LPTC) / UR EABX; M. LE GUEDARD, LEB AQUITAINE TRANSFERT-ADERA / LEB AQUITAINE TRANSFERT; M. Eon, B. Delest, Irstea Bordeaux / UR EABX; P. Gonzalez, University of Bordeaux / UMR EPOC CNRS 5805; N. Mazzella, S. Morin, Irstea Bordeaux / UR EABX

Fatty acids are essential elements for the structure of biological membranes and for the storage of metabolic energy. They are used as a source of energy by metabolism at each trophic level, making fatty acids biochemically and physiologically important compounds (Neves et al. 2015). In the trophic chain, many fatty acids are only synthesized by microalgae and bacteria before being transferred *via* herbivorous invertebrates to fish and ultimately to humans (Arts et al. 2001). For example, highly unsaturated fatty acids (HUFA) such as eicosapentaenoic acid (EPA; C20:5n3), can not be synthesized *de novo* or in insufficient proportions by animals (Saito and Aono 2014). That is why fatty acid analysis is commonly used to study trophic interactions in food chains. Generally, microalgae with a high proportion of EPA, such as diatoms, are an excellent source of food for animals but the concentrations of these different fatty acids can vary according to the stage of growth of the organism and according to different environmental parameters including pesticide exposure (Brett et al. 2006, Robert et al. 2007, Burns et al. 2011, Filimonova et al. 2016). Moreover, for several years, the intensive use of pesticides caused many problems to the environment, making pesticides major pollutants of aquatic ecosystems (Aydinalp and Porca 2004). The aim of this study is to investigate the impact of 3 pesticides on diatom's fatty acids. To address this issue, a model freshwater diatom (*Gomphonema gracile*) was exposed to three herbicides, with three different cellular targets, at environmentally relevant and higher concentrations (diuron and S-metolachlor, C1= 1 µg/L and C2= 10 µg/L; glyphosate, C1= 5 µg/L and C2= 50 µg/L). After a 1-week exposure, fatty acid compositions of diatoms were determined by gas chromatography. In comparison with control samples the percentage of 1) polyunsaturated fatty acids (PUFA) decreased with S-metolachlor contamination (C2); 2) saturated fatty acid (SFA) and monounsaturated (MUFA) decreased with diuron and glyphosate exposure (C2). The decrease of PUFA is a direct impact and can be explained by the mode of action of S-metolachlor which inhibits elongases. Concerning diuron and glyphosate, the decrease of SFA and MUFA can reflect an indirect effect, which can be explained by the mode of action of these two pesticides which respectively blocks electron transfer in photosynthesis, and inhibits the synthesis of aromatic aminoacids.

TU018

Effects of Nickel on cell cycle progression, growth and antioxidant enzymes of green algae C. reinhardtii

M. Saenz, PRIET CONICET, National University of Luján; k. Bisova, Laboratory of Cell Cycles of Algae Institute of Microbiology; W.D. Di Marzio, CONICET-PRIET / PRIET

Freshwater ecosystems received industrial and domestic sewage discharged and natural chemical compounds as a result of anthropogenic activities. Heavy metals released in the environment have increased over the last decades causing environmental and human health problems worldwide. The known biological adverse effect of metals include growth disorders, dysfunction of photosynthesis and pigments synthesis pathways, induction of oxidative stress, mutagenic effects, among others. Among aquatic organism, microalgae have an important role in aquatic system as they are a key component of food chains. So that, it is crucial to has early assessment tools to evaluate effects of metals at the cellular level. In the present study effects of Nickel was evaluated on cell cycle progression, growth and antioxidant enzymes kinetic of the green algae *C. reinhardtii*. Synchronized cultures of this multiple fission dividing algae were used for the study. Aliquot from growing cultures were taken hourly during 36 hours. The attainment of commitment points (CP) was evaluated by transferring hourly aliquot into aerated tubes at 30 °C in the dark. Analysis of cellular division, nuclear divison (DAPI stain) changes in cell size, were performed. The proportion of mother cells and daughter cells were assessed at the end of the cell cycle. Ecotoxicity of metal was assessed by algal growth inhibition test, estimating toxicity endpoints, growth rates, protein, antioxidant enzymes activities of catalase, guaiacol peroxidase, ascorbate peroxidase, glutathion reductase and concentration of chlorophyll a, chlorophyll b and carotenoids at the end of 96 hs of exposition. Nickel provoked a block of cell cycle at the highest concentration tested. At lower concentrations, cell cycle progression was observed with different pattern of attained CP, depending the exposure concentration. Antioxidant enzyme activities were inhibited at concentration above 0,05 and 1 mg/L. The effects of metal on pigment concentration was less evident than the effects on growth rates, indicating a lower sensitivity of these parameters. Nickel provoked severe damage on algal cell growth, cell cycle progression, photosynthetic pigments as well as modification of antioxidant enzymes activities. An integrated analysis is done discussing the consequences on population performance in natural environment affected by heavy metal discharged from different anthropogenic sources.

TU019

Use of BiologEcoPlateTM to evaluate the effects of ZnO nanoparticles on soil microbial communities

V. Romano, Parthenope University of Napoli / Science and tecnology; v. pasquale, University Parthenope; s. schiavo, ENEA CR; M. Oliviero, s. dumontet, University Parthenope; s. manzo, ENEA / SSPT-PROTER-BES

Nanoagrochemicals seem to be the new frontier in modern agriculture due to theirincreased efficacy, durability, and to the reduction ofthe amount of applied fertilizer. Zinc oxide NPs have been largely usedas nanofertilizersas their spreedingto crops seems to increase their growth and yield.This increasing use could lead to their introduction in the environment occuring also via unintentionally pathways. Actually, some studies reported ZnO NPs negatively affect soil microbial activities and consequently the biogeochemical cycles.These effects could be evidenced by assessing metabolic profiles of culturable, aerobic, heterotrophic microorganisms (Biolog). BiologEcoplate was successfully used to detect short and long-term changes of functional diversity of soil microbial communities. This method was based on the determination of the oxide profiles related to several different carbon sources.This study aims to investigate the changes in the metabolic activity of culturable soil aerobic heterotrophic microorganisms along decimal dilution in response to the exposure for one month to two different type of fertilizers (F1 and F2). The fertilizers were added with ZnO Bulk, ZnO NPs and ionic zinc ($ZnSO_4$) at 230 mg Zn/kg. Then, the fertilizers with Zn compounds were added to the farm soil. After 15 days of soil exposure to fertilizers with ZnO Bulk, NPS and $ZnSO_4$, the eluates were obtained. Different eluate dilutions were tested upon *Pseudokirchneriella subcapitata* (6.25%, 12.5% and 25%), *Lepidium sativum* (50%). At time 0, 15, 30 days fresh soil samples were assessed by using the BiologEcoplate approach. The occurrence of the microbial oxidation of each BiologEcoplate™ C source was calculated as probability'p' on a binomial set of data in order to identify the treatments able to preserve the highest possible oxidizing ability of C substrates and those negatively affecting it. ZnO-nanofertilizers were more toxic than fertilizers with ZnO in bulk form, for algal growth, instead for plants, the effects of "ZnO-nanofertilizers" depended on the fertilizer type: only F1 + ZnO NPs resulted more stimulating than F1 + ZnO Bulk. Preliminary Biolog results seemed to highlight that the microbial community was mainly affected by ZnO NPs.The integration of the classical ecotoxicology with BiologEcoplate approach could represent a good strategy to establish the environmental risk related to the use of nanofertilizers. Keywords: microbial community, ecotoxicology, nanofertilizers

TU020

Environmental factors-regulated disease dynamics of tilapia lake virus (TiLV) transmission in farmed tilapia ponds

T. Lu, Nation Taiwan University / Department of Bioenvironmental Systems Engineering; Y. Yang, National Taiwan University / Bioenvironmental Systems Engineering; H. Lin, National Taiwan University; C. CHEN, National Taiwan University / Bioenvironmental Systems Engineering; W. Chen, Kaohsiung Medical University / Dept Biomedical Science and Environmental Biology; C. Liao, National Taiwan University / Department of Bioenvironmental Systems Engineering

BACKGROUND: Outbreaks of tilapia lake virus (TiLV) have caused substantial mortalities of farmed tilapia, posing a significant threat to worldwide tilapia industry. Environmental factors controlling TiLV disease dynamics should be clearly elucidated to prevent the potential economic impacts on aquaculture. **OBJECTIVE:** The main objective of this study was to make the TiLV disease dynamics by constructing an epidemiological model to implicate aquaculture management among farmed tilapia ponds. **METHODS:** The mortality of Nile tilapia infected by intraperitoneal (I.P.) injection with different TiLV dosage were fitted by two-parameter Hill model to estimate median lethal dose (LD50). To explore TiLV-induced disease dynamics, an epidemiologic three-compartmental susceptible-infectious-mortality (SIM) model was applied to describe cumulative mortality data to estimate mortality rate (α), transmission rate (β), and basic reproductive number (R_0) for Nile tilapia posed by TiLV under treatment of cohabitation. **RESULTS:** In toxicity assessment, LD50 estimate of Nile tilapia infected by I.P. injection with different TiLV dosage was 57127.5 TCID50 mL^{-1}. Under cohabitation scenario, disease parameters such as α, β, and R_0 derived from SIM model were 0.46 day^{-1}, 1.13 day^{-1}, and 2.46, respectively. **CONCLUSIONS:** TiLV transmission could be affected by environmental factors such as temperature and aquaculture density. Results of toxicity assessment and disease epidemics could provide insights into aquaculture management of TiLV disease by controlling potential factors in tilapia ponds. *Keywords*: Tilapia lake virus; Toxicity assessment; Susceptible-infectious-mortality model; Aquaculture management

TU021

Natural organic matter alleviates TiO2 and CuO nanoparticle toxicity in four algal species

E. Joonas, V. Aruoja, National Institute of Chemical Physics and Biophysics / Laboratory of Environmental Toxicology; K. Olli, University of Tartu / Institute of Ecology and Earth Sciences; A. Kahru, National Institute of Chemical Physics and Biophysics / Laboratory of Environmental Toxicology

Although the knowledge concerning synthetic metal nanoparticle (NP) effects on aquatic organisms has improved during the last decade, most research has been conducted in highly artificial environmental conditions. Sacrificing some of the experimental reproducibility to obtain more environmentally relevant data, we studied the 72-hour effects of uncoated CuO ($CuSO_4$ as ionic control) and TiO_2 NPs on two endpoints, biomass production and photosynthetic maximum quantum yield (Fv/Fm), in nutrient-adjusted natural water (ANW) and the OECD201 standard medium, using four freshwater species from three major algal groups: green algae (*Raphidocelis subcapitata* and *Chlamydomonas reinhardtii*), diatoms (*Fistulifera pelliculosa*), and cyanobacteria (*Synechocystis sp*). Metal toxicity on both parameters at 72 h was reduced in ANW in all algal species except the cyanobacterium, presumably because of natural organic matter (NOM) binding to the NPs and solubilized ions. The biofilm-forming diatom was most resistant to NPs when incubated in ANW, whereas both the diatom and the cyanobacterium were not inhibited by TiO_2 at concentrations up to 100 mg/L throughout. TiO_2 significantly inhibited biomass production of both green algae in the standard medium (EC_{50} 14-31 mg/L), but only *R. subcapitata* was inhibited in ANW (EC_{50} 31 mg/L). TiO_2 NPs did not significantly inhibit Fv/Fm of any species in either medium up to 100 mg/L, indicating a lack of toxic effect on the photosynthetic apparatus. The sensitivity to CuO remained at a similar level in the standard OECD medium (biomass based EC_{50} 0.2-0.9 mg/L) for all species, but differed over orders of magnitude in ANW (EC_{50} 0.3-16 mg/L). The cyanobacterium, that has the smallest cell of the four tested species, was consistently most susceptible to Cu toxicity. While shedding of Cu ions from particles explained CuO toxicity, TiO_2 effects were at least in part due to observed cell-nanoparticle heteroagglomeration. Overall, Fv/Fm was a less sensitive toxicity endpoint than biomass, but the two parameters were strongly correlated (Spearman's ρ=0.6-0.9) when toxicity was evident, again proving Fv/Fm as a rapid method for toxicity detection. The observed discrepancies in toxicity indicate that using different model organisms, experimental endpoints and conditions could provide valuable information about the behavior of emerging contaminants in the environment, thus improving the quality of risk assessment. Research was funded by IUT23-5.

TU022

Chlorinated solvent contaminated groundwater: a glimpse inside the environmental microbial communities and their potential for bioremediation

P. Pretto, R. Ricci, Biosearch Ambiente; T. Lettieri, European. Commission - Joint Research Centre / Directorate D   Sustainable Resources Water and Marine Resources Unit

Chloroethenes are among the most frequent pollutants affecting groundwater in North Italy due primarily to their extensive industrial use in the past. As many contaminants released in the environment because of inadequate disposal, they accumulate and persist in the ecosystem posing a threat for human and environmental health. Degradation of such harmful xenobiotics can occur thanks to the activity of autochthonous microbial communities able to break down the more chlorinated compound to lesser chlorinated ethenes which need to be detoxified as well. Bacteria able to metabolize such toxic substances are indeed well known as well as many of their metabolic pathways, but still an efficient and complete detoxification process is hard to achieve. The understanding of the microbial activity underpinning the whole process is crucial especially during a bioremediation process where microbes are stimulated through the amendment of nutrients in order to obtain the complete detoxification. The huge impact of metagenomics, and other molecular biology techniques for the comprehension of microbial composition and activities in different environments, is helping to shed light for the comprehension of the critical apparatus behind the detoxification process but we are still at the beginning. During the present work, two microbial populations inhabiting a chlorinated solvent polluted groundwater, with and without nutrient amendment, have been analyzed after whole genomic DNA extraction and sequencing. The data analysis, together with the chemical ones, will help to enlight the differences between the two populations in terms of genes expression and potential of biochemical pathways for pollutants' biodegradation in relation to the chemical and geochemical parameter characterizing the specific site. Metagenomics of polluted sites is a powerful tool that could help in the future to define the best strategy to employ in order to obtain a complete environmental detoxification. This approach will be useful both for companies operating in soil and water recovery and for policy makers.

TU023

Impact of the antihistamine fexofenadine on structure and functioning of leaf-associated microbial communities

P.T. Baudy, University of Koblenz-Landau / Institute of Environmental Sciences; J.P. Zubrod, University of Koblenz-Landau / Institute for Environmental Sciences; J. Fahlman, T. Brodin, J. Klaminder, Umea University / Department of Ecology and Environmental Science; M. Bundschuh, Swedish University of Agricultural Sciences / Department of Aquatic Sciences and Assessment

Effects of pharmaceuticals and pesticides designed to control microorganisms (e.g., antibiotics and fungicides) on aquatic microbial decomposers and the functions they provide are rather well-documented, while knowledge about effects of other micropollutants is scarce. In a recent study the antihistamine fexofenadine was shown to impact the microbial decomposition of plant detritus, however, the mechanistic basis for this remains unexplored. We therefore conducted a microcosm experiment, where we microbially colonized two plant substrates (i.e., black alder leaves and hay). Both precolonized substrates were subsequently exposed towards fexofenadine at concentrations of 0, 2, and 200 µg/L. Replicates (*n*=10-15) were harvested after 15 and 30 days. Substrates were used to determine mass loss or preserved to estimate microbial communities' structural or functional composition including fungal biomass, sporulation of aquatic fungi, bacterial abundance, fungal and bacterial DNA, and enzyme activities. Furthermore, water samples were analyzed for dissolved organic carbon (DOC) quality or preserved to analyze total organic carbon. After 15 days of exposure, there was a tendency towards reduced decomposition of black alder leaves (~40%) in both fexofenadine teatments, while after 30 days, decomposition in the 200-µg/L treatment was increased by ~45% (but both not significantly different from the control). On the contrary, the decomposition of hay tended to be increased by fexofenadine exposure after 15 days. After 30 days no differences could be observed among fexofenadine teatments for hay, while generally more hay was decomposed than black alder. Accordingly, in water samples of the two substrates, substantial differences in the DOC quality were observed. Furthermore, fexofenadine exposure lead in hay treatments to an increased proportion of microbially-derived DOC. These observations suggest that the microbial communities' structure and/or functional composition differ between the two tested substrates as well as among the antihistamine treatments. Moreover, potential implications in carbon and nutrient fluxes in exposed systems are indicated as the detected alterations in DOC quality may affect planktonic decomposer communities that are involved in DOC's remineralization in surface waters. To gain an in-depth mechanistic understanding of the observed effects, we are currently analyzing variables related to microbial community structure and functioning.

TU024

Innovative tools and metagenomics for the monitoring of rivers and lakes : the European project INTCATCH

M.D. Scrimshaw, Brunel University / Institute for the Environment; S. Marcheggiani, Italian Institute of Health ISS / Environment Health; M. Carere, Italian Institute of Health ISS; O. Tcheremenskaia, Italian Institute of Health ISS /

Environment Health; L. Mancini, Italian Institute of Health ISS; C. Mancini, AECOM / Environment Health; M. Delledonne, M. Rossato, University of Verona; F. Fatone, Università Politecnica delle Marche; A. Farinelli, J. Blum, D. Bloisi, University of Verona; A. Cordioli, Azienda Gardesana Servizi S.P.A. Peschiera del Garda; P. Varotto, Azienda Gardesana S.p.A. Peschiera del Garda; A. Tittonel, Technital S.p.A. Milano; D. Calisi, Algorithmica S.r.l. Roma; F. Giannone, Algorithmica S.r.l.; R. Allabashi, Boku University; A. Parsons, L. Parsons, Downstreams L.t.d.; T. Runnalls, Brunel University / IFE; G.C. Brighty, Environmental Sustainability Associates limited; T. Licha, Gottingen University; S. Malamis, Athens Technical University; T. Knutz, Go-Sys; A. Merkoci, ICREA

The European Project Horizon 2020 INTCATCH (Development and application of Novel, Integrated Tools for monitoring and managing Catchments) has the main goal to recommend and deliver new innovative tools for the monitoring of surface waterbodies in Europe. The tools foreseen by INTCATCH include sensors for the detection of heavy metals, nutrients, pH, temperature, pesticides, *Escherichia coli*, some of them are mounted on aquatic drones. An innovative tool of Intcatch is the portable sequencing laboratory/tool for metagenomic analysis that is performed in different surface waterbodies (e.g. Garda lake) selected through an analysis of the pressures. Bacteria are collected after concentration of water samples that are filtered by an automatic system applied on the aquatic drones. Using portable devices, the genomic tool allows the DNA meta-barcoding *on-site*, where samples are collected, and deliver the full bacterial composition of the water analyzed, including pathogens. The validation of the metagenomic results is performed with the use of traditional microbiological analysis of raw water. The metagenomic data can support the investigative monitoring of the Water Framework Directive because changes of the bacterial community can reflect variations in water quality, and be linked for example to the presence of unknown chemical substances or mixtures. In addition, given its capability to detect pathogens, such portable genomic tool can provide a valuable rapid readout in case of emergencies, that are increasing in frequency due to the effects of climate changes, such as flooding. Similarly, the metagenomics data, linked to the informations of the other tools, can be also used for the identification of pollution sources because the proportions of the bacteria groups vary in relation to several stressors (agricultural, urban, living stock, etc.).

TU025

Tolerance of sediment-microbial communities to copper indicates lake contamination

A. Tlili, Eawag / Department of Environmental Toxicology; C. Bonnineau, Irstea Lyon; A. Dabrin, Irstea Lyon-Villeurbanne / UR MALY; E. Lyautey, Universite Savoie Mont Blanc; B. Ferrari, Centre Ecotox EAWAGEPFL; S. Pesce, Irstea Lyon-Villeurbanne / Microbial ecology of anthropised river systems

In lakes, and fresh waters in general, sediments play a crucial role in biogeochemical cycles, and are often at the basis of the food-web. Heavy metals such as copper, zinc or silver can accumulate in lake sediments, which represent a risk for benthic microorganisms, with potential negative effects on the functioning of the whole ecosystem. Therefore, the goal of this field survey was to examine the impacts of copper on bacterial communities in sediments, along a contamination gradient by heavy metals in lake Geneva. Sediments were sampled with an Ekman grab sampler at four sites, located around the lake and close to the shore. Samples were analysed for total metal concentrations, organic matter content, abundance of metal-resistance genes (e.g *cop*A and *cus*A), bacterial biomass, and bacterial community composition. Moreover, tolerance of microbial communities to copper was determined according to the pollution-induced community tolerance (PICT) concept by measuring the inhibition of bacterial secondary production and the extracellular enzyme β-glucosidase activity. Results showed a clear gradient of copper contamination, ranging from 30 to 350 $mg.kg^{-1}$ DM sediment. The results also demonstrated that chronic *in situ* exposure to copper induced a decrease of bacterial biomass and a structural shift in the community composition. Interestingly, tolerance measurements to copper were strongly and positively correlated to the copper concentrations in the sediments. Overall, our findings support the fact that microbial communities in sediments are a good indicator for metal contamination in aquatic ecosystems, and the suitability of microbial tolerance measurements to pinpoint specific effects of metals at the community level.

TU026

Current challenges and perspectives in aquatic and soil microbial community ecotoxicology

K.K. Brandt, University of Copenhagen / Department of Plant and Environmental Sciences; M. Schmitt-Jansen, UFZ - Helmholz Ctre Environm. Research / Department of Bioanalytical Ecotoxicology; A. Tlili, Eawag / Department of Environmental Toxicology

Assessing the impacts of chemicals still largely relies on approaches that are based on assays in simplified laboratory settings at the single-species or subspecies level. Yet, it becomes increasingly recognised that to draw conclusions on potential effects on ecosystems, it is essential to consider higher levels of ecological organisation, and more intricate risks on ecosystem structure and function. Microbial communities provide a large range of ecosystem services such as primary and secondary production, nutrient recycling, pollutant degradation, and are sources of biochemicals. Microorganisms are also primary targets for chemicals, which can lead to structural and functional alterations of microbial communities, with potential negative consequences for ecosystem functioning and environmental selection of antimicrobial resistance. Hence, a microbial community-level perspective in ecotoxicology is more important than ever. In this presentation we will first provide an overview on the current status of microbial community ecotoxicology research in aquatic and soil ecosystems, and to which extent this field is considered in environmental risk assessment schemes. Second we will describe the challanges within both aquatic and terrestrial microbial community ecotoxicology. Finally we will discuss future research directions in microbial community ecotoxicology to accurately assess and predict impacts of chemicals on ecosystems, and to develop specific response indicators of chemical exposure and effects.

TU027

Hydrodynamic conditions alter the tolerance of biofilm communities towards chemical stress

B.H. Polst, Helmholtz centre for environmental research - UFZ / Department of Bioanalytical Ecotoxicology; F. Larras, Helmholtz Center for Environmental Research - UFZ GmbH; S. Lips, Helmholtz Centre for Environmental Research UFZ / Department of Bioanalytical Ecotoxicology; C. Anlanger, U. Risse-Buhl, M. Weitere, Helmholtz Centre for Environmental Research UFZ / Department of River Ecology; M. Schmitt-Jansen, UFZ - Helmholz Ctre Environm. Research / Department of Bioanalytical Ecotoxicology

Biofilms in rivers are complex communities built of bacteria, fungi, algae and protozoa embedded in a matrix of extracellular polymeric substances (EPS). They are important hotspots for biogeochemical processes in aquatic systems. A variety of stressors can potentially affect the structure and function of biofilms. Therefore their tolerance to one stressor may be influenced by former exposures to another stressor. Community composition and physical structure is influenced by hydrodynamics. Even though biomass and thickness of biofilms were reported to decrease with higher mean flow velocity and turbulence, the cell-to-EPS ratio increased. As the EPS content of a biofilm may influence the bioavailability of toxicants, differences in community tolerance towards herbicides are expected for biofilms grown under variable flow conditions. Still, the interactive effects of hydrodynamic growth conditions and herbicide tolerance are lacking. Using an artificial flow-through channel and water from the River Selke (Elbe catchment, Germany), we created heterogeneous flow regimes and related biofilm community structure and function to different mean flow velocities and values of turbulent kinetic energy. Taking the biofilms grown under such controlled hydraulic conditions, herbicide tolerance towards prometryn was tested according to the PICT-approach. Focusing on the phototrophic part of the biofilm communities, we 1) investigated the algal structure, function and herbicide tolerance under different near-bed turbulences (diatom composition, photosynthesis) and 2) assessed the role of EPS in stressor interactions. The relevance of EPS content in combined stressor interactions was confirmed by using artificial EPS and algal cultures.

TU028

Does fungicide exposure alter interspecific relationships of aquatic fungi during leaf-decomposition? - A case study using species-specific qPCR assays

N. Roeder, University of Koblenz-Landau; P.T. Baudy, University of Koblenz-Landau / Institute of Environmental Sciences; J.P. Zubrod, D. Englert, University of Koblenz-Landau / Institute for Environmental Sciences; M. Konschak, University Koblenz-Landau / Institute for Environmental Sciences; C. Baschien, Leibniz Institute DSMZ - German Collection of Microorganisms and Cell Cultures; R. Schulz, University of Koblenz-Landau / Institute for Environmental Sciences; M. Bundschuh, Swedish University of Agricultural Sciences / Department of Aquatic Sciences and Assessment

Aquatic hyphomycetes, a polyphyletic group of freshwater fungi, are considered key players in leaf litter breakdown, an important ecosystem function in aquatic systems. Structural implications of anthropogenic stressors in aquatic hyphomycete communities have mainly been analyzed using spore morphology, which does not allow assessing direct inferences on species-specific abundance and performance under stress. Therefore, we performed a microcosm experiment in which we related the effects of a model fungicide mixture on aquatic hyphomycete communities' leaf decomposition to individual species' abundances quantified via species-specific quantitative real-time polymerase chain reaction (qPCR) assays. Using a factorial design, every possible single, binary and quaternary species combination of four different aquatic hyphomycete species (*Alatospora acuminata*, *Heliscella stellata*, *Neonectria lugdunensis* and *Tetracladium marchalianum*), was exposed to the model fungicide mixture composed of five substances with different modes of toxic action (four sum concentrations ranging from 5 to 2500 μg/L and a fungicide-free control; n=5, N=275). In monocultures, aquatic hyphomycetes exhibited different fungicide tolerance levels, with concentrations ranging from 500 to 2500 µg/L resulting in significantly reduced abundances. Interestingly, only the two tolerant species (i.e., *N. lugdunensis* and *T. marchalianum*) were capable of decomposing leaf material to a significant degree. Moreover, abundances of single species within the model communities as well as their functioning were governed by dominance interactions (e.g., one species outcompeting the other), probably as a result of competition for leaf substrate. Depending on the species composition, interactions resulted in an up to 99 % reduced abundance of the inferior species. Species interactions were largely unaffected by fungicide exposure as dominant species where generally those identified as tolerant towards fungicide exposure. However, qPCR results revealed that one of the two sensitive species exhibited a significantly increased DNA yield in presence of the other one at field-relevant fungicide concentrations (5 µg/L). Species-specific qPCR assays proved to be a valuable tool for assessing ecotoxicological effects on as well as ecological interactions within aquatic hyphomycete communities. In the future, this technique might become an asset in aquatic risk assessment and environmental stress monitoring.

TU029

Cyanobacterial Bloom in the Lake Varese: Characterisation of Microbial Communities by Metagenomics analysis

D. Conduto Antonio, European Commission Joint Research Centre / Directorate Space, Security and Migration; R. Loos, I. Sanseverino, European Commission Joint Research Centre; A. Lahm, Bioinformatic consultant; A. Beghi, F. Pandolfi, ARPA Agenzia Regionale per la Protezione dellAmbiente della Lombardia; P. Genoni, Lombardy Regional Environmental Protection Agency; D. Napierska, T. Lettieri, European. Commission - Joint Research Centre / Directorate D Sustainable Resources Water and Marine Resources Unit

The increasing anthropogenic eutrophication and climate changes are contributing to the intense proliferation of cyanobacteria in waterbodies so causing a phenomenon known as bloom which may compromise the quality of drinking and recreational water. The dynamics of bloom events are not yet fully understood, however it is scientifically accepted that external factors such as water temperature, nutrient concentrations and light intensity, may influence the potential of a bloom.Our study focuses on the relationship between environmental factors and the composition of the microbial community of the lake Varese (Italy) for a period of several weeks before and after the bloom event.Sampling campaigns were performed on a weekly basis. Water samples were collected from the depeest region of the lake at 3 different water column depths, 0.5 m from surface (EPI), 13 m (MESO) and 2.5 times the Secchi disk depth measured in situ on sampling day (2.5x SECCHI). The samples were characterised for their chlorophyll a content, nutrients, cyanotoxins and genomic DNA was extracted for metagenomics.Purified DNA samples were subjected to 16S sequencing (variable region V3-V4) and for shotgun analysis. All 16S samples were MiSeq sequenced as 2x250bp paired reads, the corresponding shotgun samples as 100bp paired reads. Shotgun analysis was performed for sample collected from 31/8/2016 unitl 5/10/2016 and only for EPI and 2.5x SECCHI. The results showed that a peak of cyanobacteria was observed around 14.9/21.9 in the EPI (E) samples consistent with the high observed concentration of chlorophyll a. The lowest abundance of the cyanobacteria was in the week 31/8. Some variation of the overall microbial composition was also observed for proteobacteria and actinobacteria. Our result suggests that the major differences in bacterial community composition during the bloom are concentrated in the SECCHI depth region while composition of the EPI zone is more or less constant. Cyanobacteria were found highly abundant in Lake Varese and are therefore likely responsible for the bloom.This hypothesis is also supported by the cyanotoxin data although complementary 18S metagenomics sequencing would be recomended in order to discard a possible contribution of phototrophic eukaryotes.

TU030

Following copper bioaccumulation and internalization during freshwater biofilm development using stable Cu isotope

a. dabrin, Irstea / Unité de Recherche Milieux Aquatiques, Ecologie et Pollutions (MAEP); J. Gahou, Irstea Lyon-Villeurbanne; m. masson, c. brosse, Irstea Lyon; B. Volat, Irstea Lyon-Villeurbanne; C. Bonnineau, Irstea Lyon; S. Pesce, Irstea Lyon-Villeurbanne / Microbial ecology of anthropised river systems; M. Coquery, Irstea Centre de Lyon - Villeurbanne / Unité de Recherche Milieux Aquatiques, Ecologie et Pollutions (MAEP)

In small streams, microbial communities form river biofilms attached to solid substrates by producing extracellular polymeric substances (EPS). This matrix may act as a protective layer by limiting cellular contact with surface water contaminants. Thus, several studies have suggested that during biofilm growth, biofilm and EPS matrix thickness could limit cellular bioaccumulation. To test this hypothesis, we conducted an experiment under controlled conditions to follow the bioaccumulation of two Cu isotopes in different biofilm fractions throughout biofilm growth and maturation. During the early stages of its development (0 to 20 days), biofilm was grown on glass slides in water spiked with natural dissolved copper. Then, biofilm was transferred to a mono-isotopic (^{65}Cu) copper-enriched medium for 20 additional days. During these two successive exposure periods, dissolved Cu concentrations and the corresponding $^{63}Cu/^{65}Cu$ isotopic ratios were monitored every two days. At the end of each of the 2 exposure periods, biofilm was sampled from the slides and freeze-dried. A sequential extraction was then applied to recover Cu from the colloidal and capsular EPS fractions. The resulting pellet was mineralized to determine Cu concentrations in the intracellular fraction. Copper concentrations and isotopic ratios were determined by ICP-MS in water collected at various times of the experiment and after 20 and 40 days in the different fractions of the biofilm. The results showed constant dissolved Cu concentrations during the two exposure periods (~7 µg/L); while isotopic ratios $^{63}Cu/^{65}Cu$ widely differed between the first (2.23) and the second phase (0.25) of exposure. Bioaccumulation levels of Cu in the young biofilm (20 days) were similar between the colloidal, capsular and cellular fractions. Finally, the isotopic approach showed that after 40 days of exposure, the isotopic ratios in the three fractions of the biofilm were similar to the ratio in water of the second phase of exposure (~0.25). These results suggest (i) an intense and rapid renewal of the biofilm and of the bioaccumulated Cu and (ii) that Cu concentrations in a mature biofilm at a given time reflect the last period of exposure. In addition, although the isotopic ratios were very low, a significant difference of isotopic ratios between the EPS fraction (0.25) and the cell fraction (0.35) confirms the potential protective function of the EPS matrix. *\n*

TU031

Zirconium impact on freshwater periphytic communities

C.N. Doose, INRS - Centre Eau Terre Environnement; S. Morin, Irstea Bordeaux / UR EABX; C. Fortin, Institut national de la recherche scientifique Centre - Eau Terre Environnement

The growing world demand for metals increases metallic element mobilization in aquatic systems. Although the effect of metals on freshwater ecosystems is well documented, studies on the impacts of tetravalent metals are very scarce. Zirconium (Zr) is a tetravalent non-radioactive element for which the global demand has been increasing in the last decades. Benthic microorganism communities (periphyton) have shown good potential as a biomonitoring tool to assess metal exposure of aquatic organisms. In this work, the effect of Zr on periphyton biodiversity and biochemistry was investigated to apply this tool to zirconium and other tetravalent metals contamination assessment and to better understand their potential impacts on aquatic ecosystems. Glass slides were immerged in a pond in Cestas (near Bordeaux, France) for one month to be colonised by periphyton. They were distributed in 3 aquaria containing a synthetic culture medium (Dauta, 1982) and effective concentrations of 0.2 ± 0.1 nM (C0), 0.5 ± 0.3 nM (C1) or 2.9 ± 0.3 nM (C2) of Zr (n=3). One slide per section was sampled after 1, 2 and 4 weeks of exposure for subsequent analyses: dry weight, chlorophyll pigments fluorescence, photosynthetic activity, microscopic microorganism identification, polysaccharide and protein contents. Biomass, proteins, polysaccharides and diatoms abundance increased significantly showing the growth of the biofilm during the experiment. No significant Zr exposure effects were observed on biomass, proteins, polysaccharides contents but their productions appeared to be slightly lower in C1 and C2 at t2 and t4. Diatoms growth rate in the C2 condition was significantly lower than in C0 and C1. Results obtained by pigments fluorescence measurements showed significant cyanobacteria decrease in the C2 condition over the exposure time as well as the brown algae between t2 and t4. Principal response curve (PRC) analysis showed significant changes over time of micromeiofauna composition between the reference (C0) and the C2 condition. Ciliates were less impacted by Zr exposure than flagellates which tended to disappear in the C2 condition. Biofilm microorganisms play a wide role in major ecosystem processes. Regarding these results, Zr exposure can impact the periphyton microorganisms composition which could disturb periphyton key functions. A better understanding of effects of metals on micromeiofauna would improve risk assessment of metallic exposure in aquatic ecosystems.

TU032

DNA metabarcoding demonstrates effects from copper at environmental concentrations on microbial diversity in marine periphyton biofilms

N. Corcoll, University of Gothenburg, Sweden / Biological and Environmental Sciences; J. Yang, Nanjing University / State Key Laboratory of Pollution Control and Resource Reuse, School of the Environment; H. Kronenberger, T. Backhaus, University of Gothenburg / Department of Biological and Environmental Sciences; X. Zhang, Wisconsin Department of Natural Resources / State Key Laboratory of Pollution Control and Resource Reuse, School of the Environment; M. Eriksson, Chalmers University of Technology / Department of Shipping and Marie Technology

Copper pollution is common in coastal areas. In particular, the use of copper-based antifouling paints on ships hulls elevates copper concentrations in these environments. This study assesses the effects of dissolved copper on community structure and function of marine periphyton biofilms. Microbilal diversity and community structure was studied by 16S and 18S amplicon sequencing, targeting prokaryotic and eukaryotic organisms, respectively. Community function was studied as impacts on algal biomass, photosynthetic pigment profiles and primary production. Additionally, we studied Pollution-Induced Community Tolerance (PICT) using photosynthesis as the endpoint. Periphyton was exposed for 18 days to five copper concentrations, between 0.01 and 10 uM, in a semi-static test. The amplicon sequencing yielded 7.1 and 5.7 million high quality 16S and 18S reads, and the average numbers of 16S and 18S Operational Taxonomic Units among the samples were 9405 and 1242, respectively. Analysis of Unifrac distances showed that copper significantly changed the eukaryothic community structure at concentrations as low as 0.01 uM. The prokaryotic community structure was changed at slightly higher concentrations (0.06 uM). A total of 23 taxa, including species within the *Proteobacteria*, *Bacteroidetes*,_ *Stramenopiles* and *Hacrobia* classes, were identified as particularly sensitive to copper. Algal biomass, photosynthetic pigment profiles and primary production, were reduced at Cu concentrations of 0.06 uM and higher. PICT measurements confirmed that copper induced community tolerance in exposed communities. Taken toghether, these findings indicate that negative impacts from copper might be common in coastal ecosystems.

TU033

A Time-series Study of Soil Microbial Community Compositional and Functional Shift in Biodiesel vs. Petrodiesel Contaminated Soils

D.L. Carr, Texas Tech University / Biological Sciences; M. Dong, Texas Tech University / Biological Science

The spill of petrodiesel on land can irreversibly damage the soil ecosystem, and there are limited studies comparing petrodiesel and biodiesel impacts on soil microbial communities. Biodiesel has been considered as a viable substitute for petrodiesel, however the degree to which biodiesel is more microbial friendly than petrodiesel is inconclusive. Previous studies of soil microbial community on contaminated sites failed to reveal the dynamic changes of soil microbial communities. This laboratory study compared the effects of petrodiesel and three types of biodiesel on soil microbial communities in sandy loam soils. Contaminated soil samples were investigated at day 0, day 7 and day 180 to evaluate their effects on the composition and function of soil microbial communities. Biolog EcoPlates™ were used to test the microbial community functions based on carbon utilization while soil microbial composition were addressed by 16s rRNA gene sequencing of V3-V4 regions. Results suggested that biodiesels were not statistically different from petrodiesel in terms of their adverse impacts on soil microbial communities. In conclusion, our results suggested that biodiesels should not be automatically considered as harmless substitutes for petrodiesel and that metagenomic effects in soils persist beyond 6 months post contamination.

TU034

Evaluation of riparian groundwaters quality using microalgal response to pollutants

O. CHAMSI, ECOLAB UMR5245 CNRS UPS INPT; E. Navarro, CSIC - Spanish National Research Council / Dept. Recursos marins renovables; J. Sanchez-Perez, S. Sauvage, ECOLAB UMR CNRS UPS INPT; F. Comin, CSIC   Spanish National Research Council; I. Antiguedad, University of Pais Vasco; J. Bodoque, University of Castilla La Mancha; J. Charcosset, ECOLAB UMR CNRS UPS INPT; E. Pinelli, ECOLAB UMR 5245 CNRS UPS INPT

Contamination of ecosystems by pesticides, pharmaceuticals and trace metals becomes a major environmental problem. Freshwater algae are well known bio-indicators of river pollution but diatom indices do not allow to evaluate the specific effects of the contaminants. Their sensitivity to pesticides differ markedly among microalgae species and therefore the toxicity data for multiple species need to be efficiently obtained. In the present work, we measured the growth of the three lotic dominant species *Desmodesmus subspicatus*, *Nitzschia palea* and *Navicula pelliculosa* by an automated fluorometric microplate assay to evaluate the groundwater and river quality in four riparian wetlands in the south-west of Europe (Monbéqui (France), Saragossa (Spain), Bidasoa (Spain) and Toledo (Spain)). Four campaigns of water sampling were realized during contrasted hydrological conditions under different pedo-climatic conditions in agricultural area. Pesticides, pharmaceuticals and metals concentration were measured by HPLC-MS or ICP-MS. PCA, ANOVA and co-inertia analysis results showed that algal growth was different between freshwater and groundwater. As expected, the green alga was sensitive to alkalinity, SO_4, O_2 and pH whereas diatoms were positively sensitive to silica concentration and dissolved organic carbon (DOC). Besides, the green alga responded positively to the metals Co and Ni and negatively to *S*-triazines, terbutylazine and their metabolites. At last, the pharmaceuticals benzoylecgonine and carbamazepine/irbesartan/valsartan induced growth stimulation of *N. palea* and *N. pelliculosa*, respectively. Same records for pharmaceuticals were observed for the other three sites, excepted Bidasoa. Both extensive sampling and data analysis makes our approach a new useful bio-indicator for preliminary investigation of groundwater quality in order to predict the best location of quality water for human consumption (ATTENAGUA project).

Can trends in wildlife populations revolutionise our understanding of the impacts of chemicals on the environment? (P)

TU035

Can post mortem data be used to monitor population health in response in the barn owl?

L. Walker, Centre for Ecology & Hydrology; E.D. Potter, NERC Centre for Ecology & Hydrology / Lancaster; M.G. Pereira, Centre for Ecology & Hydrology / Lancaster; R. Shore, Centre for Ecology & Hydrology (NERC)

The Predatory Bird Monitoring Scheme (PBMS; http://pbms.ceh.ac.uk/) is national long-term project that monitors contaminant residues in a range of avian predator species. Each bird that is submitted to the scheme is given a post-mortem examination during which approximately 60 macroscopic observations and measurements are made. The information gathered during this examination could potentially be used to monitor health status of the birds at the time of their death or at a particular stage of their development. Previously we have focused on examining health indicators for the sparrowhawk, *Accipiter nisus*. We were able to establish baseline "norms" for indicators that could be broadly categorised as indicators of change in: (i) population demography because of altered recruitment, survival and mortality (measures were sex ratio, proportion of first-year birds, and proportion of deaths from starvation or disease); (ii) change in nutritional status (measures were body weight, fat score, condition index) that may be a pre-cursor for subsequent population impacts, (iii) physiological stress (as measured by fluctuating asymmetry) that may be an indicator of fitness. In the current study we investigated whether these population health indices could be applied to barn owls, *Tyto alba.* We were able to establish baseline "norms" in the form of Shewhart charts. For example the mean proportion of birds that were female was 48% with a prediction interval of 38-59%, and so years in which the prediction interval was exceeded would indicate unusual years. For the majority of indices studied adult and first-year birds could be considered collectively. For many indices females and males needed to be studied separately due to sexual dimorphism. Females had significantly heavier mean body weights than males (287g vs 258g) but there was extensive over lap in the prediction intervals for the two sexes. Prediction intervals for the percentage of birds with low fat deposits were 23-65% and 28-75% for females and males, respectively. The level of kurtosis within 10th primary feather weight precluded this metric from being used to investigate fluctuating asymmetry. This study shows that the proposed population health indices generally can be reported for barn owls. Establishing these population health indices can then be used to provide an early warning of whether chemical or other stressors are affecting the demography of barn owl populations.

TU036

Identifying suitable marine biomonitors in South Africa: Mussels vs Whelks

C. Sparks, Cape Peninsula University of Technology / Conservation and marine sciences; W. Samuels, Cape Peninsula University of Technology / Department of Conservation and Marine Sciences

Over the last three decades there has been a significant decline in marine pollution monitoring-related studies in South Africa. Very little research has been conducted to assess the prevalence of imposex in whelks and also very few studies have been conducted on comparisons between contaminants in different marine invertebrates at the same sites. The current study was conducted in July 2017 to compare metal bioconcentration between mussels (*Mytilus galloprovincialis*) and whelks *(Burnupena lagenaria*) as well as measure imposex prevalence in *B. lagenaria* at Bloubergstrand, Granger Bay and Green Point, Cape Town, South Africa. This was done in order to identify suitable bioindicators of ecotoxicity by determining whether the mussels and whelks bioaccumulate metals in the same way and to assess imposex prevalence in whelks (as an indicator of tributyltin contamination). The concentrations of metals (Al, Cu, Zn, Fe, Cr, Mn, CO, Ni, Mo, Cd and Pb) were measured in intertidal sediment, *M. galloprovincialis* and *B. lagenaria* and imposex prevalence recorded in *B. lagenaria*. Results showed that the highest prevalence of imposex in whelks and metal concentrations were recorded Granger Bay, an area of high boating activity. The most important result was that the whelks had higher bioconcentrations of metals than the mussels at all sites. Identifying biomonitors should be linked to purpose of investigation before selection of species, and mussels have been considered 'ideal' biomonitors of contamination in South Africa. Given the ubiquitous distribution of *B. lagenaria* along the South African coast, which is not the case for *M. galloprovincialis* that only occurs on the west and south cast of the country, the proposal is made that *B. lagenaria* could be considered as alternative bioindicators of ecotoxicity of contaminants in the region.

Recent developments in environmental risk assessment for pollinators (P)

TU038

Behavioural effects of imidacloprid, a neonicotinoid insecticide, on bumblebees (bombus terrestris)

J.S. Paus-Knudsen, University of Oslo / Department of Biosciences; H.A. Sveinsson, University of Oslo / Department of Physics; K. Borga, Department of Bioscences, University of Oslo / Department of Biosciences; M. Grung, A. Nielsen, University of Oslo / Department of Bioscience

Bees are increasingly facing multiple and interacting threats. One of the threats that have received increased attention lately is neonicotinoids: a group of systemic neuro-active pesticides that disturb the transmission of signals in the insect´s nervous system. In just a few years neonicotinoids have become the most widely used insecticide in the world, and protect a variety of crops against invertebrate pest. Despite being used in relatively small quantities, several studies have shown sub-lethal effects of neonicotinoids on honeybees(*Apis mellifera*) exposed to neonicotinoids in field-realistic doses. However, ecological and physiological traits vary among bee species and studies on honeybees may not provide satisfactory predictions for negative effects on other bee species. Using bumblebees, *Bombus terrestris,* the present study developed a new experimental method to quantify how chronic dietary exposure to the neonicotinoid imidacloprid affects learning, locomotor activity and consequently the ability to forage and thus pollinate in a non-*Apis* species. Bumblebees were exposed to three different dosages of imidacloprid through artificial nectar (sugar water), ranging from field realistic levels (1 mg/L and 10 mg/L) to distinctly higher levels (100 mg/L) in a chronic exposure regime, lasting for eight days. To assess whether imidacloprid influences learning, the bumblebees' ability to discriminate between blue nectar-filled (rewarding) and yellow water-filled (non-rewarding) artificial flowers were tested systematically in a flying arena. The bumblebees were tracked by cameras, allowing for analysis of the flowers choices, locomotor activity and all the flowers visited during numerous, simultaneous foraging bouts. This study shows the successful application of a new method to track bumblebee behaviour. Further, the study shows that learning and locomotor activity are negatively affected, in a dose-dependent manner, when bumblebees are exposed to imidacloprid. Moreover, we show that field-realistic doses of imidacloprid have negative effects on bumblebees.

TU039

Sensitivity of honeybee larvae to PPPs and impact analysis based on EFSA Bee GD *

R. Becker, BASF SE Agrarzentrum Limburgerhof; J. Lueckmann, Rifcon GmbH

* on behalf of the ECPA NTA & Bee Working Group Based on EU Regulation 1107/2009/EC the current regulatory risk assessment on bees has to address the risk on honeybee larvae or honeybee brood. In July 2013 the European Food Safety Authority (EFSA) published a guidance document on the risk assessment of plant protection products on bees (EFSA 2013). This document is intended to provide guidance for notifiers and authorities in the context of the review of plant protection products (PPPs) and their active substances under Regulation (EC) 1107/2009 (EC 2009). The first objective of this poster is to summarize all available industry data, for active substances and formulated products on honey bee larvae testing according to e.g. OECD 237 and OECD 239, in order to gain an overview of these results and the selectivity of different product groups. As a first step in the risk assessment, EFSA requires a screening step which consists of the calculation of risk quotients (ETRs) for honey bee larvae. This considers exposure routes for the in-field (PPPs applied as sprays) and off-field (PPPs used as seed treatments and granules) scenarios. Where a substance or use should not pass one of the screening level risk quotients, EFSA offers the possibility for refinement in a tier I risk assessment. This includes the refinement of exposure estimates from the screening step and also additional exposure routes, such as the exposure to flowering weeds in field margins and adjacent flowering crops. The second objective of this poster is to evaluate the impact of the proposed screening and tier I risk assessments on the pass/fail rate of currently available active substances and formulated products which is an ability of the scheme to correctly identify compounds of potential concerns and consequently screen out those of low concern. The aforementioned analysis follows the principles described in the ECPA impact analysis (Miles and Alix 2013) and compared the first approach with the outcome based on laboratory data. \n

TU040

Honeybee brood studies according to Oomen and OECD GD 75: Is there a difference of the brood termination rate under semi-field and field conditions*

J. Lueckmann, Rifcon GmbH; R. Becker, BASF SE Agrarzentrum Limburgerhof; S. Schmitzer, IBACON GmbH; B. Szczesniak, Eurofins Agroscience Services Ecotox GmbH

*on behalf of the ICP-PR Bee Brood Working Group and the Bee Brood Working Group of the German AG Bienenschutz Based on EU Regulation 1107/2009/EC the current regulatory risk assessment on bees has to address the risk on honeybee larvae or honeybee brood. According to the new "EFSA Guidance Document on the risk assessment of plant protection products on bees (*Apis mellifera*, *Bombus* spp. and solitary bees)" (EFSA 2014), both, the Oomen bee brood feeding test (Oomen *et al.*, 1992) as well as the OECD Guidance Document 75 (2007; OECD GD 75) are given as the two higher tier options to refine the risk on honeybee brood if concern is raised in tier 1. Both methods focus on the brood termination rate (hereafter BTR) as the key endpoint. While the Oomen brood test investigates an artificial and worst case acute or chronic oral exposure scenario with a test item spiked feeding solution administered inside the hive (Lückmann & Schmitzer 2015) brood studies according to OECD GD 75 under semi-field conditions rely on a realistic contact and oral exposure scenario to bees comprising contaminated nectar and pollen after overspray of a bee attractive crop. As the evaluation of historical data from semi-field studies according to OECD GD 75 showed a strong variability of the control BTRs (Becker *et. al* 2015), the performance of OECD GD 75 bee brood studies under field conditions was regarded as an option to get more reliable BTR data (Becker *et. al* 2015, Giffard & Huart 2015). The present poster compares control BTRs from Oomen feeding studies with BTRs obtained from OECD 75 semi-field and field trials and consider explanations for observed variances. Moreover, the possibilities and limitations of the three methods will be discussed.

TU041

Does assessing of all brood cells of a hive reduce uncertainty and increase reliability of Semi-field honeybee brood studies (OECD GD 75)?

H. Bargen, G. Gonsior, M. Kleinhenz, B. Szczesniak, Eurofins Agroscience Services Ecotox GmbH; S. Knaebe, EAS Ecotox GmbH / Ecotox Field

The OECD guidance document 75 (2007) introduced a semi-field test method to assess the effects of PPPs on honeybee brood. The assessment of bee brood development over one brood cycle is conducted by mapping cells. It starts from the egg stage and the fate of individual cells is followed until hatch. For this purpose, pictures are taken at defined stages of the development cycle and compared to the development in separate control hives. Three parameters in regards to brood development are assessed and evaluated: brood termination rate (BTR) (number of the marked cells where a termination of the bee brood development was recorded, expressed as percentage of the sum of all marked cells), brood compensation index (an indicator for the compensation of bee brood losses) and brood index (an indicator of the bee brood development, facilitates a comparison between different treatments). Due to the high variability of BTRs within treatments and high control mortality in several studies no definite conclusions regarding effects on brood were possible (Pistorius *et al*. 2012). To address this variance, effort was taken by the ICPPR bee brood working group and AG Bienenschutz to improve the method, compare historical data and give recommendations for future testing (Pistorius *et al*. 2012, Becker *et al*. 2014). Despite fulfilling all recommendations, high variability in brood termination and high control mortality is still evident in some of the conducted studies. For those studies data interpretation and conclusions are questionable. According to Wang and Görlich (2017) one reason for the variability of brood termination rates is that the evaluation of brood development is based on a limited, defined number of cells containing eggs. According to their opinion results might be different if all cells would have been chosen. They suggested that it would reduce the uncertainty to a minimum. In this poster the recommendation of Wang and Görlich (2017) is used to find out if uncertainty can be reduced by evaluation of all cells. The brood data used were collected according to OECD 75 during a semi-field trial in *Phacelia tanacetifolia*. The results of assessments of brood development of a whole colony (3 replicates, untreated) are compared to the results were 200 cells per hive were used. The parameters compared are: BTR, brood and compensation indices.

TU042

Ecotoxicological studies with bumble bees - latest developments and method improvement

L. Franke, Eurofins Agroscience Services Ecotox GmbH / Ecotoxicology Field; O. Klein, Eurofins Agroscience Services Ecotox GmbH / Ecotox Field; J. Fricke, Eurofins Agroscience Services Ecotox GmbH / Ecotoxicology Field; J. Sorli, TRIALCAMP SLU; T. Vollmer, Eurofins Agroscience Services EcoChem GmbH / Field Ecotoxicology; S. Knaebe, EAS Ecotox GmbH / Ecotox Field

The publication of the proposed EFSA risk assessment for pollinators resulted in an increasing demand for experiments with non-Apis pollinators. As bumble bees (*Bombus terrestris* L; Hymenoptera, Apidae) are commercially available and their biology is well-known, they can be used for ecotoxicological semi-field and field trials. Also, they provide important pollination services, e.g. in greenhouse cultures, and consequently impacts of pesticides on bumble bees have already been tested for years. Currently, an ICPPR Non-Apis working group is developing a standardized method for semi-field studies with bumble bees. Based on the protocols of the ICPPR working group, several semi-field studies have been conducted. The central endpoint in these higher tier studies is the colony reproduction success (production of young queens), as the production of sexuals is essential for the maintenance of a healthy bumble bee population. However, assessing the production of young queens in semi-field trials is challenging. Many variables influence the number of produced queens, such as the right timing for the termination of the study or the condition of the colonies at study start. Based on data collected in the past years, different strategies to reduce the variability in the production of young queens were evaluated. Also, several parameters influencing this most important endpoint were analyzed. We tried to answer some open questions concerning the colony reproduction success, such as how the experimental set-up can influence queen numbers and queen weights, how high the natural variation between colonies is and how the selection of bumble bee colonies for the studies can be improved.

TU043

Higher-tier risk refinement of solitary bees in the field - is the well-known 'focal species' concept a suitable approach?

J. Lueckmann, M. Faupel, J. Ludwigs, Rifcon GmbH

According to EFSA (2013) bumble bees and solitary bees have to be considered in addition to honey bees in the risk assessments. However, suitable testing methods in the lab are currently partly available only (e.g. for acute contact & oral bumble bee testing, acute contact solitary testing) or under development (e.g. chronic oral bumble bee testing, acute oral solitary testing). Regarding appropriate species for solitary bees EFSA (2013) proposes *Osmia cornuta* or *O. bicornis* as test organisms for the risk assessment, and higher-tier semi-field testing with *Osmia* as proposed by the ICPPR non-*Apis* working group has been proved to obtain sound results. However, experiences from current field studies on *Osmia* show that exposure of adults and larvae is not necessarily given as these solitary bee species have a pronounced polylectic feeding behaviour that can result into a low exposure to a test substance (*i.e.* not being a real worst-case). In order to address this problem, the refinement of worst-case solitary bee risk assessments under realistic field conditions may be achieved by using a 'focal species' concept, where most appropriate focal solitary bee species can be identified to represent a worst-case choice per crop, application time and country/zone. Whereas this approach is well-known for bird and mammal risk assessment it has not been yet applied for solitary bees. Here, we present the idea of a 'focal species' concept for solitary bees, its needs, refinement options, advantages and limitations.

TU044

Non-Apis (Bombus terrestris) versus honeybee (Apis mellifera) acute oral and contact sensitivity - Preliminary results of ECPA company data evaluation

A. Dinter, Cheminova Deutschland GmbH & Co. KG / Global Regulatory Sciences; A. Alix, Dow Agrosciences / Risk Management; J.C. Becker, New York State Dept. of Environmental Conservation / Biology; P. Campbell, Syngenta / Environmental Safety; M. Miles, Bayer CropScience UK / Environmental Safety; E. Pilling, Dow Agrosciences / REgulatory Sciences; N. Ruddle, Syngenta Ltd / Product Safety; A. Sharples, FMC Agricultural Solutions; G. Weyman, ADAMA; L. Oger, ECPA

A preliminary data evaluation was conducted by ECPA companies to compare the sensitivity of bumblebees (*Bombus terrestris*) with the sensitivity of honeybees (*Apis mellifera*). For the evaluation about 70 data sets were available for contact exposure and about 50 data sets for oral exposure. The data sets comprised insecticides, fungicides, herbicides in about equal numbers plus a few other substances. The preliminary ECPA company data evaluation of LD50 values indicates lower or similar contact sensitivity of bumblebees vs. honeybees. Similarly, lower or similar oral sensitivity of bumblebees vs. honeybees was determined with one exception for an insecticide that indicated higher acute oral bumblebee sensitivity compared to honeybees. For this insecticide, higher tier data indicates no negative impact on bumblebees at the maximum intended use rate. Overall, the ECPA company data evaluation indicates that bumblebees are not more sensitive than honeybees based on acute toxicity assessment.

TU045

Bumblebee (Bombus ssp.) 10 day feeding laboratory test design: First results from an ICP-PR ring test

N. Exeler, Bayer AG, Crop Science Division; N. Hanewald, BASF SE / Ecotoxicology; C. Jenkins, Envigo; H. Krueger, EAG Laboratories; A. Zicot, SynTech Research / Ecotoxicology; E. SOLER, TRIALCAMP SLU / Ecotoxicology; A. Molitor, Eurofins Agroscience Services GmbH; S. Vinall, Mambo-Tox Ltd; K. Amsel, BioChemagrar GmbH; S. Haupt, IBACON GmbH; S. Kimmel, Innovative Environmental Services (IES) Ltd. / Ecotoxicology

A decline in some pollinator species has led to an increasing pressure on political decision makers and regulatory bodies and resulted in a change of existing risk assessment paradigms and testing approaches. The published and already revised EFSA GD on the risk assessment of PPP on pollinators includes apart from the honeybee also bumblebees and solitary bees. In the need to address long term effects on bumblebees, the ICP-PR Non-Apis working group designed a ring test protocol to develop a first-tier chronic feeding test for bumblebees. Based on the recently published honeybee 10 day chronic feeding test guideline OECD 245 and the bumblebee acute oral toxicity test guideline OECD 247 a 10 day feeding test was set-up using dimethoate as reference substance. The response of adult *Bombus* ssp. workers to the test chemical Dimethoate EC400 (Perfekthion) was evaluated within a 10 day chronic exposure scenario. The test item was provided *ad libitum* for a period of 10 days. During the exposure phase bumblebees are kept individually in cages – "single housing". Bumblebees do not share food via trophallaxis and need to be fed individually. Furthermore, single housing prevents hierarchy fights (among the queen-less BB workers) potentially introducing mortality. Mortality and behavioral abnormalities in the test groups were observed and recorded daily and compared to the untreated control groups. The endpoints calculated were: LC_{50} (median lethal concentration) and LDD_{50} (median lethal dietary dose) values after 10 days and if possible the NOEC (no observed effect concentration) and NOEDD (no observed effect dietary dose). First results indicate that with this method reproducible results were obtained. The mortality in the control groups seem not to exceed 15 % (evaluation currently ongoing) and the overall food consumption allowed for a proper evaluation of the intended endpoints.

TU046

Standardization of method to test toxicity on stingless bees

T. Roat, UNESP Universidade Estadual Paulista Júlio de Mesquita Filho / Biology; R. Nocelli, UFSCar / Departamento de Ciências da Natureza Matemática e Educação; O. Malaspina, UNESP Universidade Estadual Paulista Júlio de Mesquita Filho / Departamento De Biologia, Centro de Estudos de Insetos Sociais

Brazil is the country with the greatest diversity of bees in the world. The Brazilian bee fauna consists of 5 families: Andrenidae, Apidae, Colletidae, Halictidae, and Megachilidae, with 1,576 species described. The family Apidae consists of 3 subfamilies: Xylocopinae, Nomadinae, and Apinae. The subfamily Apinae comprises 19 tribes, among them the Meliponini, commonly known as "stingless bees". However, several species have been included in the list of endangered animals and there are many reports indicating a decrease in the abundance of all native species. To ensure protection of this huge diversity and continued growth of agriculture, it is important for Brazil to have a system to evaluate which compounds are safe for bees and efficient in agriculture. Current toxicity studies in bees are carried out with the *Apis mellifera*, but stingless bees present a very different behaviour and biology. The oviposition process, the internal care of the hive, the feeding system, the nest building materials and the size of the hives are quite variable. However, there are no specific methods for toxicity tests to stingless bees. So, in our laboratory we are developing and standardizing methods to test the toxicity of pesticides to species of stingless bees. We test the Acute Contact Toxicity Test protocol of OECD guidelines (214) established to european honeybee for the stingless bees *Scaptotrigona postica* and *Melipona scutellaris*. For this, we used three active ingredients and the toxic standard dimethoate. The individuals were kept in 250 mL cages (ten bees were placed per cage, such that each treatment contained thirty bees from three colonies), fed in groups through microtubes (1.5 mL) punched in extremities, and kept in a chamber of biochemical oxygen demand (BOD) at 29 ± 2 °C, relative humidity of 70 ± 10% and in constant darkness. The diet used was composed of 50% (w/v) aqueous sugar solution. Our observations show that to perform the Acute Contact Toxicity Test for stingless bees some adaptations in OECD (214) are necessary, like to adjust the temperature of the incubator (29 ± 2 °C instead of 25 ± 2 °C), anesthesia should be done by cooling and the time should be adequate for each species. The development of these tests will allow the development of safer strategies for the protection of biodiversity and, at the same time, support the expansion of agriculture, which is an important socio-economic activity in the region.

TU047

A method for a solitary bee (Osmia spp.) first tier acute oral laboratory test: an update

I. Roessink, Alterra / Environmental Risk Assessment; N. Hanewald, BASF SE / Ecotoxicology; C.W. Schneider, BASF SE; N. Exeler, Bayer AG, Crop Science Division; E. Noël, SynTech Research; A. Schnurr, BioChemagrar GmbH; A. Molitor, Eurofins Agroscience Services GmbH; E. SOLER, TRIALCAMP SLU / Ecotoxicology; S. Kimmel, Innovative Environmental Services (IES) Ltd. / Ecotoxicology; J. Van der Steen, Alveus AB Consultancy

The recently updated EFSA draft honey bee Guidance document also specifies other hymenopteran pollinators, like solitary bees and bumble bees, as groups to take into consideration when assessing the risk of plant protection products to pollinators. However no validated test protocol and consequently no extensive data set is available to compare sensitivities of other relevant pollinators to those of honey bees. Within the current project of the ICP-PR Non-Apis working group a start was made to develop a first-tier acute oral test for *Osmia* spp. bees. Based on the honey- and bumble bee guidelines OECD 213 and OECD 247 an acute oral test was designed using dimethoate as reference substance. *Osmia bicornis* and *Osmia cornuta* were housed individually and fed a known amount of test volume per dosage. First results indicate that with this method reproducible results were obtained. In these tests, control mortality never exceeded 12 percent. Furthermore, sensitivities of *O. cornuta* and *O. bicornis* appeared to be rather similar, although *O. cornuta* showed a slightly less sensitive response, (which might be) due to its larger bodyweight. Hence, the LD_{50}values after 96 hours ranging from 2.6 – 7.1 ug a.i./ug bee indicate that a validated and workable methodology has been set up and a test guideline is within reach.

TU048

2 Years of Solitary Bee Semi-field Ring Testing and Final Conclusions (ICPPR Non-Apis Working Group)

S. Knaebe, EAS Ecotox GmbH / Ecotox Field; N. Exeler, Bayer AG, Crop Science Division; L. Franke, J. Fricke, Eurofins Agroscience Services Ecotox GmbH / Ecotoxicology Field; M. Frommberger, Julius Kuehn Institut; T. Jütte, Julius Kuehn Institute; S. Kimmel, Innovative Environmental Services (IES) Ltd. / Ecotoxicology; O. Klein, Eurofins Agroscience Services Ecotox GmbH / Ecotox Field; J. Lueckmann, Rifcon GmbH; H. Giffard, Testapi; A. Rossbach, Tier3 Solutions GmbH / Field team; C.W. Schneider, BASF SE; A. Schnurr, BioChemagrar GmbH

The publication of the proposed EFSA risk assessment guidance document of plant protection products for pollinators highlighted that there are no study designs for non-Apis pollinators available. Since no official guidelines exist for semi-field testing at present, a protocol was proposed and two years of ringtests were conducted in 2016 and 2017 to develop a general test set-up. The ringtest design was based on the EFSA guidance document, OEPP/EPPO Guideline No. 170 and results of discussions regarding testing solitary bees during the meetings of the ICPPR non-Apis workgroup in 2015, 2016 and 2017 followed by a workshop in 2017 to harmonise methodology. Ringtests were conducted with two representatives of a solitary bee species (*Osmia bicornis* L and *Osmia cornuta* Latr; Hymenoptera, Megachilidae). These species are polylectic and can forage on a diverse spectrum of flowering crops. They are nesting in cavities. Both are common species in Europe, commercially available and are widely used for pollination services. Several laboratories participated in the higher-tier ring tests. 8 semi-field tests were done in 2016and 9 in 2017. Two treatment groups were always included in the ringtest: an untreated control and dimethoate as a toxic reference item (optional other i.e. brood affecting substances (fenoxycarb)). In the study design adult bees were exposed in the tunnels during their reproductive period. Adult bees, as well as their offspring, were exposed to the treated pollen and nectar during development. Relevant endpoints for this study design are observations of the flight activity in front of the nesting units, nest occupation (i.e. number of nesting females), the production of complete cells and cocoons per female, the brood

termination rate during the larval development as well as the success of emergence of their progeny (F1-generartion) in the following year. Based on the results of the ringtests over 2 years a draft protocol is available together with recommendations for the methodology needed. This includes how the cocoon incubation and hatching of bees can be synchronised with the onset of flowering, how fit solitary bees are out of season and which substance at what rate can be used as reference item for brood studies.

TU049

Predicting wild bee sensitivity to Acetylcholine Esterase (AChE) inhibitors utilizing a trait based phylogenetically controlled approach

T. Pamminger, BASF SE, Agrarzentrum Limburgerhof / Ecotoxicology; N. Hanewald, BASF SE / Ecotoxicology; C.W. Schneider, BASF SE; J.C. Becker, New York State Dept. of Environmental Conservation / Biology; M. Bergtold, BASF SE

Plant protection products (PPP) play a vital role in modern agricultural practice. Nevertheless, their potential off-target effects on managed (e. g. Apis mellifera) as well as wild (most non-Apis species) bees have emerged as an intensively discussed topic. In current risk assessment *A. mellifera* is proposed as a surrogate species to cover potential adverse effects of PPPs on non-Apis bee species. However, as robust and scientifically sound information regarding the sensitivity of non-*Apis* bee species are scarce the validity of this approach has been challenged. As a first step to address this question we have compiled a comparative data set of the *Acetylcholine Esterase* (*AChE*) inhibitors sensitivities of 21 bee species, covering five of seven currently recognized bee families. This data set was complemented with information on bee bodyweight, a trait likely influencing bee sensitivity to PPP exposure. Our phylogenetic controlled analysis shows that bee bodyweight is a robust predictor of bee sensitivity to *AChE* inhibitors and confirms that *A. mellifera* is particular sensitive to this class of PPPs. In contrast, many stingless bee species, are comparatively resilient to *AChE* inhibitors, especially when controlling for body weight. We discuss the consequences of these findings in the context of the global non-Apis bee risk assessment debate in Europe and the Americas.

TU050

New approaches in testing of pollinator exposure under realistic conditions - Methods and recent experiences

M. Persigehl, Tier3 Solutions GmbH; U. Zumkier, Tier3 Solutions GmbH / Regulatory Science; A. Rossbach, Tier3 Solutions GmbH / Field team; C. Wolf, Tier3 Solutions GmbH

With the growing concern for insect populations and an increased awareness of the importance of pollinators in the public opinion as well as in the regulatory context reliable and robust methods are required to measure exposure to residues in pollen and nectar. The methods used need to adequatley reflect the properties of the tested substance and the circumstances of the application as well as potential influences of behavioural aspects such as foraging behaviour. Also, residue kinetics of a given substance have to be considered and must be reflected in the time points used for sampling. Here, we present recently employed approaches for studies which measure exposure to residues. Methods discussed include the determination of residues as part of (semi-) field studies with bees in pollen, nectar and honey, studies on foraging behaviour as well as methods to adequately determine residues for non-standard uses, such as home and garden uses, ornamentals and granules with a slow release formula. The different methods are compared and advantages and potential pitfalls are illustrated.

TU052

Normative Instruction 02/2017 - Brazilian risk assessment of pesticides to bees

F. Viana-Silva, C. Dias, L. Borges, K. Cham, C. Tonelli, R. Oliveira, A. Alves, IBAMA / DIQUA CGASQ; R. Rebelo, IBAMA / CCONP

Globally there are increasing concerns about possible decline in pollinators which requires that efforts be made in the direction of identifying its possible causes and in establishing policies for protecting pollinators. The Brazilian Institute for the Environment and Renewable Natural Resources (Ibama) is responsible for environmental assessments in the context of pesticide registration in Brazil. Since 2011 Ibama is implementing the risk assessment of pesticides in Brazil and one of the actions undertaken is the establishment of appropriate risk assessment procedures to protect pollinator insects against pesticides effects. In this context, it was published in February 2017 the Normative Instruction 02 (NI 02/2017) that establishes procedures to risk assessment of pesticides to pollinators. This is the first Brazilian specific regulation based on a risk approach, and in July 2017 Ibama published a Manual of Environmental Risk Assessment of Pesticides to Bees which explains in an accessible way how the normative should be applied. NI 02/2017 is widely based on US/Canada's approach, which means that it focuses on *Apis mellifera* data; the models used for screening are Bee-REX and AgDrift; tests required for tier 1 are the same and there is one scheme for foliar applications and other for soil/seed/trunk treatments. But there are few modifications: 4 tiers, the last one being post-registration monitoring; use of a safety factor of 10 for non-*Apis* bees; residue trials must be performed in Brazil and for tier 2 a crop grouping is considered. With this normative Ibama expects that pesticides be used efficiently without incurring unacceptable risks to bees. Although Ibama has a full framework for risk assessment established for honeybees there are still gaps in knowledge and research needs for ensuring that procedures to protect bees can be improved, especially regarding native bees. Hence, a matrix of selection for Brazilian bee species was proposed for selecting native species for use in pesticide risk assessment. This matrix provided the basis for electing meliponines (stingless bees) as a priority group. In the near future Ibama intends to assess the need of changes in the risk assessment procedure, eventually including a stingless bee as a representative species.

TU053

How the new Brazilian risk assessment framework for bees works

K.d. Coelho, ADAMA BRASIL / Regulatory Affairs; G. Weyman, ADAMA

The Environmental Assessment of pesticides in Brazil is performed by the Environmental Institute (IBAMA) and comprises two aspects: Environmental Hazard Potential Assessment and Environmental Risk Assessment. The Hazard assessment has been established since 1990 but the Risk Assessment, although required since 1996, only started to be implemented by IBAMA in 2012 and has been developing further since then. Due to numerous global discussions on the decline of pollinators, in February 2017 Ibama published the first ruling ("normative") to establish guidelines, requirements, and procedures for a systematic risk assessment scheme of pesticides for pollinators in Brazil. Further guidance for the scheme was issued later in 2017. The Brazilian overall approach is similar to EPA, but there are nuances in the Brazilian scheme regarding which active ingredients must be tested, and additional considerations for future which must be understood. Using hypothetical data from a mixture and a single formulation, we will show how the new Brazilian scheme (the "normative") works for two use patterns, foliar and soil application, considering the main aspects of Tiers 1 and 2 of the risk assessment. Furthermore, the main points of this Brazilian risk assessment framework for bees will be compared with those adopted by other countries, considering both similarities and differences. Keywords: Pollinators, bees, risk assessment, Brazil

TU054

An epidemiological study about an effect of neonicotinoids residues on honey bee colony survival in Japan

Y. Kameda, Chiba Institute of Technology / Creative Engineering; E. Fujita, K. Tanaka, Chiba Institute of Technology

Neonicotinoid insecticides are widely used in Japan. Seven neonicotinoid insecticides such as Imidacloprid, Acetamiprid, Thiocloprid, Clothianidin, Dinotefuran, Thiamethoxam and Nitenpyram are popular. Their usage began from the beginning of 1990 and is increasing till 2008 in Japan. Recent annual total usage of seven insecticides is not increasing, approximately 400 tons per year in Japan. However, very little is known about their occurrence, their behaviors and their ecological risk in Japanese environment. Especially, there is little known about the exposure and ecological risk of neonicotinoids to wild bees in Japan though these neonicotinoid pesticides are considered to be one of the reasons for losses of bees in EU, Canada and the US. It should be noticeable that the residual levels of neonicotinoid pesticides in foods are much higher than those in EU and the US and that some news reported that losses of bees and honeycombs occurred recently in Japan. The aim of this research is to reveal ecological risk assessment of honeybees including colony survival in Japan by ELISA analytical methods. The exposure assessment is conducted by neonicotinoids residue concentrations in adult honeybees, pupae, pollen and honey. These samples were collected from beekeepers around in Japan. Information about condition of colonies was also collected from beekeepers. Wild honeycombs were also collected. The six neonicotinoids were detected in all samples including honey, pupae and adults. Especially, more than ten times higher concentrations were detected in some of honey bee samples than those reported by previous reports in Europe, Canada and America. All colonies where adult honeybees were exposed by high concentrations were evaluated as abnormal condition such as CCDs, massive fatalities and sacbrood disease. Moreover, possibility of abnormality of colonies was strongly dependent on residue concentrations in adult honeybees. It was very interesting that EC50 values of colony abnormality, derived from this epidemiological research, were not much different from LC50 of adult bees. The values and the ELISA screening techniques could be one of easy warning values for beekeepers which indicate possibility of colony abnormality.

TU055

Thiamethoxam Honey Bee Large Scale Colony Feeding Study - Design and Interpretation

N. Ruddle, Syngenta Ltd / Product Safety; H. Thompson, Syngenta Ltd / Environmental Safety; J. Overmyer, Syngenta Crop Protection, LLC. / Environmental Safety; C. Elston, Syngenta Ltd; M.A. Feken, Syngenta / Ecological Risk Assessment; S. Bocksch, Eurofins Agroscience Services Ecotox GmbH / Ecotox Honeybees; P. Thorbek, Syngenta / Environmetal Safety; M. Hill, Eurofins AgroScience Services Inc

Colony feeding studies were originally developed to directly assess the insect growth regulating properties of insecticides and designed to determine mode of action rather than effect levels. More recently there has been regulatory interest in conducting colony feeding studies to determine the pesticide level in nectar substitute (sucrose solution) which leads to colony-level effects, thereby allowing for comparison with residue concentrations detected in pollen and nectar from treated and untreated crops. In 2016, a honey bee colony feeding study was conducted with thiamethoxam with the aim of providing a robust colony-level endpoint for comparison with residues in pollen and nectar. Analyses of the colony data indicate there were clear significant effects at the highest concentration of 100 µg/Kg for many colony parameters and overwintering survival. At 50 µg/Kg, despite a few transient differences for pollen stores, overall colony strength and overwintering survival were similar to the control, confirming the NOAEL as 50 µg/Kg. The NOEL was determined to be 37.5 µg/Kg. To assess the potential risk to honey bees from exposure to thiamethoxam and metabolite CGA322704 (clothianidin) residues in pollen and nectar, the NOEL and NOAEL can be compared to measured residues in treated or succeeding crops. In a treated oilseed rape multi-exposure study (Pilling et al., 2013) the maximum thiamethoxam residues found in pollen and nectar were 1.0 µg/Kg and 3.0 µg/Kg, respectively. The residues of CGA322704 were below the 1.0 µg/Kg LOQ. In an on-going study, residues in pollen and nectar in untreated succeeding crops of sugar beet were also found to be low. The maximum thiamethoxam residues in pollen and nectar were 2.6 and 0.55 µg/Kg, respectively. A maximum CGA322704 residue of 6.3 µg/Kg was detected in pollen, while residues in nectar were less than the 1.0 µg/Kg LOQ.The colony NOEL and NOAEL concentrations are an order of magnitude greater than the maximum residues in succeeding crops and a treated crop. The colony NOEL and NOAEL provide the basis by which to evaluate the potential risk of thiamethoxam residues detected in pollen and nectar. It also provides additional support for the lack of effects reported in field studies following exposure of colonies to levels of thiamethoxam in pollen and nectar of seed treated crops that are an order of magnitude lower than the no effect level observed in this study.

TU056

Alteration of the alternative splicing pattern in honeybees' nervous system genes as a tool to test pesticides toxicity

P. Decio, UNESP Universidade Estadual Paulista Júlio de Mesquita Filho; T. Roat, UNESP Universidade Estadual Paulista Júlio de Mesquita Filho / Biology; O. Malaspina, UNESP Universidade Estadual Paulista Júlio de Mesquita Filho / Departamento De Biologia, Centro de Estudos de Insetos Sociais; M. Soller, University of Birmingham

Evidence-based knowledge on pesticide-effects on pollinators, such as honeybees, has become mandatory in many countries. It is important to establish lines of action approved internationally to provide farmers and policy-makers more information about the applications of pest management programs. With this in mind, this work evaluated whether sublethal doses of the insecticide thiamethoxam, the fungicide carbendazim, and the herbicide glyphosate would be capable of altering the alternative splicing pattern of the Elav (embryonic lethal abnormal visual system) and Dscam (Down syndrome cell adhesion molecule) genes, which have an important role in the formation of nervous system. Elav encodes proteins commonly used as neuronal markers in metazoans, which has action on post-transcriptional regulation and is required for differentiation and maintenance of the nervous system. Whereas that, Dscam gene can suffer alternative splicing from a highly variable region and be able to generate more than 38,000 isoforms and it is important for growth and connection of mushroom bodies, a center of learning and memory, for the expansion of dendritic fields. Based on this, we injected 2 µL of each of the pesticides (0,01 mM Thiamethoxam, 2 mM Carbendazim, 47 mM Glyphosate) to the abdomen of forager bees. After 24 hours, the brains were dissected for RNA extraction. We analyzed alternative splicing of cDNA made from mRNA by reverse transcription. Then, we performed PCR with one P32 γ-ATP radioactively labeled primer for Elav and Dscam. Because the PCR products have very similar sizes but differ in sequence, we digested the PCR products with restriction enzymes and then separated these fragments on denaturing polyacrylamide gels. It was not possible to observe a differentiated pattern of splicing for Elav neither for Dscam, comparing the control groups with the bees exposed to pesticides. The doses used and the exposure time in our study was not sufficient to indicate these genes as biomarkers in *Apis mellifera*. However, further studies are needed, exploring different doses, contamination routes, and increasing the exposure time to verify if these pesticides are capable of altering the alternative splicing pattern of genes directly related to the nervous system. (Fapesp: 2015/22368-5).

TU057

Non-uniform distribution of treated sucrose solution via trophallaxis by honeybees affects variability of homing success rate, gene expression and mortality among replicates

L. Jeker, Swiss Bee Research Center / Agroscope; V. Christen, University of Applied Sciences and Arts Northwestern Switzerland

We compared the impact of the feeding regime group dosing with 10 bees versus group dosing with two bees per cage on the variability of the homing success rate, gene expression and mortality. Based on our own observations and the recently published publication (Brodschneider, R. et al.)^[1] it seems that food sharing via trophallaxis might lead to a non – uniform distribution of the tested sucrose solution between caged bees. This can cause high variability on measured parameters among group members, replicates and treatments. For homing success rate and gene expression endpoints, bees were orally exposed to different sub-lethal concentrations of thiamethoxam (TMX) at 0.1, 0.3 or 1 ng/bee, based on the homing flight ring-test protocol. For mortality, bees were exposed orally to dimethoate at 0.033, 0.07, 0.1, 0.13, and 0.35 µg/bee, based on the acute oral toxicity test guideline OECD 213. For both methods, the treatment-feeding regime, was conducted with ten bees/cage and two bees/cage. Homing flight success rate, at 1ng TMX/bee, was significantly lower with ten bees compared to the two bees approach. A large variability of success rate and gene expression among treatment replicates was found in the ten bees feeding group. Acute toxicity data with dimethoate showed that group feeding scheme with ten bees per cage resulted in higher mortality when compared to two bees (at same dosing levels). As consequence, the LD50 value is higher for the latter. High variability of homing success, gene expression or mortality rate in the ten bees feeding scheme is most likely caused by inhomogeneous dose distribution among bees, or either by over- or under dosing of single bees within replicates. A more accurate and uniform dosing distribution can be expected between 2 bees resulting in less variable data between runs, replicates and treatments. We highlight that feeding in smaller groups of honeybees should be discussed and considered to minimize the trophallaxis dependency regarding food distribution in group dosed honeybees. Moreover, to compare endpoints of toxicological studies with single dosed wild bees for regulatory purposes. <br clear="all" /> [1]Brodschneider, R., Libor, A., Kupelwieser, V., Crailsheim, K., 2017: Food consumption and food exchange of caged honey bees using a radioactive labelled sugar solution - PLOS ONE | https://doi.org/10.1371/journal.pone.0174684

TU058

Modelling and validation of honeybee foraging behaviour for the pesticide risk assessment

M. Wang, WSC Scientific GmbH / Dept Efate Modelling; C. Dietrich, WSC Scientific GmbH

In recent years a number of population models have been developed for honeybees and some have been used for pesticide risk assessment. While the in-hive development of honeybees is relatively well understood and can be validated relatively easily in models, the accurate estimation of exposure is more complex and more difficult to validate. In particular, foraging behaviour, which is included explicitly only in very few models, plays an integral role for exposure, since it determines to what extent foragers collect nectar or pollen from treated or untreated crops and other habitats, or if they find alternative food sources. Foraging behaviour is also tightly related to weather. We therefore evaluate how foraging behaviour can be implemented and validated in a honeybee model simulating natural conditions, with particular focus on the risk assessment of pesticides and on the protection goals formulated in the recently published honeybee guidance.

TU059

Automated waggle dance decoding

M. Wang, WSC Scientific GmbH / Dept Efate Modelling; J. Kleinmann, A. Görlich, WSC Scientific GmbH

In honeybee field studies EFSA recommended in its latest guidance that field studies should ensure that the 90^{th} exposure percentile is met. Practically, it is challenging to confirm where honeybees actually foraged. In recent years a variety of methods have been developed and tested to establish a kind of foraging maps, based on waggle dance observations analyses, harmonic radar or RFID chips. Most of these, however, can realistically be used only based on relatively few individual bees. We therefore explored options for an automated analysis of waggle dance in honeybees. The system should facilitate the use of standard hives and should be usable without a computer in the field. We evaluate the reliability of the method.

TU060

How to increase test power and understand risk in refined honeybee trials

A. Görlich, WSC Scientific GmbH; M. Wang, WSC Scientific GmbH / Dept Efate Modelling

For honeybee semi-field and field studies EFSA defined SPGs (specific protection goals) in its latest guidance document on the risk assessment of plant protection products (PPP) on bees. Detrimental effects on colony size as a result of PPPs should not exceed a 7% threshold to not endanger the fulfillment of the ecosystem services provided by honeybees. The measurement of effects on colony size as small as 7% is often difficult to achieve due to high uncertainty and variability both reducing the test power. By applying a modified field methodology and test design the test power can be increased substantially thus allowing to conduct field studies that are able to reach the SPGs. For the semi-field study colonies with sister queens of equal strength were used. From these a subset of colonies was selected based on hive assessments, which started approximately four weeks prior to exposure, by selecting those colonies that would be similar during the exposure phase. During the whole study the colony strength was assessed by photographing all bees in hives (all frames and walls). Additionally, to include also the number of foragers in the assessment, hives were weighted with and without bees. To avoid an influence of the time of the day on the number of foragers counted with photography all colonies were photographed in parallel at the same time of the day. All frames of all hives were also photographed to assess brood development and to obtain a full overview of the condition of each hive at each time point. It is shown that by applying a refined, new field methodology and test design for field studies on honeybees the test power refering to the number of adults can be increased. Assessments of complete hives, including adults and all cells, make it possible to gain a detailed insight into the development of colonies and hive parameters over the course of time. Environmental factors and their influence on different hive parameters can be assessed and used to explain how these parameters either alone or in conjunction with plant protection products have an impact on the strength and development of honeybee colonies.

TU061

The potential for immune activation and possible consequences for bees upon exposure to microbial pest control agents

B. Jones, M. Whittaker, Applied Insect Science Ltd

Microbial pesticides are unlikely to cause disease in non-target insects due to a lack of specific pathogenicity. However, simply exposing the insect to a microbe has the potential to activate subclinical responses that can lead to colony level effects. For example, injection with a non-pathogenic, microbial immune elicitor induces a massive antimicrobial peptide response in bumblebees and honey bees. This immune response lasts several days and is costly to maintain. These costs are demonstrable through trade-offs between immunity and other life-history traits such as learning and longevity. In addition, immune activation alters many aspects of normal colony functioning, such as changes in foraging activity, decreased queen attendance, modified feeding behaviour, increased production of sexuals and forced ejection. Crucially, many of these effects only become apparent in the colony, and show specificity between bumblebees and honey bees. The established paradigm uses laboratory trials as a 'worst-case' scenario before progression to higher tier field trials, which may mask the downstream immunological impacts on endpoints such as longevity. Evidence exists for immune activation in insects via oral exposure with non-pathogenic bacteria. We therefore propose the need to establish whether oral and cuticular contact with microbial pesticides can induce the immune system in bees. Should immune activation be confirmed in the laboratory in the absence of lethal effects, higher tier field trails may be required to reveal the consequences within the colony.

Environmental effects of metals: Improvements to risk assessment by considering speciation and bioavailability (P)

TU062

Assessment of Levels of Some Heavy Metals in the Organs of West African Dwarf Goat and Beef Cattle in Ogbomoso, Nigeria

A.A. Giwa, Cape Peninsula University of Technology / Department of Pure and Applied Chemistry; O.A. ADESOYE, Ladoke Akintola University of Technology Ogbomoso / Department of Pure and Applied Chemistry; F. Wewers, Cape Peninsula University of Technology / Chemistry

The dangers inherent in the exposure to heavy metals present in food products especially meat, have aroused widespread concern for food safety and human health. With increasing human activities and anthropogenic pollution sources, there has been deposition of large amounts of various toxic metals in the food material which ultimately make their passage into the tissue. This study aims at assessing the levels of five heavy metals (lead, cadmium, zinc, copper and iron) in organs of West African dwarf goat and beef cattle slaughtered in Ogbomoso metropolis, Nigeria. Chevon and beef samples of heart, intestine, liver, muscle and tripe were collected from both sexes of two breeds of cattle and West African dwarf goats. Raw and cooked samples were digested and analysed using the method described by the Association of Official Analytical to determine the levels of the metals by Atomic Absorption Spectrophotometric technique. Results show that there were significant differences ($p< 0.05$) in the concentrations of the metals in the different parts and sexes of the animals studied. The estimation of the non-essential metals in the investigated samples indicated the following range; lead: 1.11- 6.00 mg/kg and Cadmium: 1.25- 6.52 mg/kg while that of the essential metals are Zinc: 1.27- 7.65 mg/kg, copper: 17.00- 72.30 mg/kg and iron: 98.93- 352.00 mg/kg. The results also revealed that the concentrations of lead, cadmium and Iron exceeded the stipulated permissible limits. Higher-than-limit concentrations are observed more in the various parts of cows than in bulls of the two cattle species. There was, however, no significant difference ($p=0.05$) in the amount of these metals accumulated by both the Buck and Doe. There was a major reduction in the results obtained for cooked samples when compared with raw samples for all the metals analysed. From the various data obtained, it can be concluded that all the five metals are present in all the samples analyzed and their average concentrations are significantly high in most of the samples. Cooking lowered the amount of the potentially toxic metals in the meat samples.

TU063

Assessment of metal bioaccessibility, bioavailability and toxicity in soil using the earthworm

P.Y. Robidoux, AGAT Laboratories, Ltd / Specialty services Division; Z. Omouri, INRS-Institut Armand-Frappier

Simultaneous contamination of ecosystems by various substances sets a challenge as regards to environmental assessment. One of the aspects is appraisal of pollutant bioavailability. Such an analysis was performed on a mixed contaminated site. The earthworm *Eisenia andrei* was exposed to different soils according to a contamination gradient. An integrated approach including a suite of biomarkers and chemical analyses was adopted to determine site toxicity. Parameters of the antioxidant system (catalase [CAT] and superoxide dismutase [SOD] activity), an enzyme of detoxifi cation metabolism (glutathione S-transferase activity [GST]) as well as acid phosphatase (AP) activity and lysosomal membrane fragility of coelomocytes (neutral red retention time, NRRT) were used as tools. Overall toxicity endpoints (lethality, body weight change, reproduction) were assessed. Lethal effects were detected in some soils whereas chronic endpoints significantly decreased. A significant response of time-growing extent and consistency was recorded for SOD from to 2-28 days, whereas effects on other enzymatic markers were low and temporally inconsistent. NRRT also was significantly decreased after 28 days concurrently to a body weight loss of the worms (30-42 %) as well as a complete impairment in reproduction at 56 days. These results are revealing of early sub-lethal biological alterations in connection to contaminant toxicity and bioavailability. Bioaccessible bismuth (Bi) concentrations (using KNO_3 soil extraction) were correlated to SOD activity and suggest an important contribution to the overall toxicity. Bi is used increasingly to replace lead in several industrial applications including the production of alloys and munitions formulations. However, little information is available on the environmental fate and ecological effects of Bi. This paper also summarizes the acute toxicity (LC50=416 mg Bi/kg) and biaccessibility of Bi, and describes bioavailability and chronic effects of bismuth on the earthworm *Eisenia andrei*. In reproduction tests, adult earthworms were exposed to natural sandy soil spiked with Bi citrate. Results indicate that Bi significantly decreased reproduction parameters at concentrations ≥ 75 mg Bi/kg and 0,005 mg bioaccessible Bi/kg. Bismuth had little effect on phagocytic efficiency of adult earthworm coelomocytes. After 28 days,Bi concentrations in earthworm tissue increased up to 21.2 mg Bi/kg and reaching a stationary state at 212 mg Bi/kg of soil.

TU064

Assessment of subcellular metal-binding ligands in white suckers (Catostomus commersonii): are all the metals accumulated in the heat-stable fraction (HSP) detoxified by binding to metallothioneins?

N. Urien, INRS-ETE / Centre Eau Terre Environnement; S. Jacob, P.G. Campbell, P. Couture, Université du Québec, INRS / Centre Eau Terre Environnement

Metal exposure may lead to accumulation and toxicity in aquatic species. Once metals enter living organisms, they can penetrate into their cells and cause deleterious effects. Alternately, metals can be detoxified by binding to molecules designed to sequester them and prevent them from exerting their toxic effects, such as metallothioneins (MT) and metallothionein-like peptides (MTLP). MT and MTLP are mainly found in the subcellular cytosolic HSP (heat-stable proteins) fraction, generally obtained after homogenization, differential centrifugation and heat-denaturation steps. It is normally hypothesized that metals present in the HSP fraction are detoxified. To confirm this hypothesis, the nature of the metal-binding ligands found in the HSP fraction needs to be determined. Thus, the aim of this work was to investigate the ligands binding metals (As, Cd, Cu and Se) in the HSP fraction from hepatic cells of white suckers collected in a reference lake and in a lake subject to multi-metallic contamination. After isolation of the HSP fraction, we used size exclusion chromatography coupled to an inductively coupled plasma mass spectrometer (SEC-ICP-MS) to separate biomolecules present in the HSP fraction and to quantify the associated metals. For each metal, higher concentrations were measured in the HSP fraction of the exposed fish than in the reference fish, but overall, metal-handling strategies did not vary between the reference and exposure fish, with the exception of As. For Cd and Cu, a major peak was observed after a retention time of 16 minutes, corresponding to the retention time of MT, suggesting that these two metals were reasonably well detoxified and regulated in these fish by binding to MT. In contrast, for Se, a major peak was observed at 27 min indicating that Se was not bound to MT but rather to a biomolecule with lower molecular weight. Finally, regarding As, two major peaks were observed in the reference fish (25 and 27.5 min), whereas in exposed fish a major peak was identified at 29.5 min, suggesting the potential induction of a specific ligand to bind As in exposed white suckers. For future work, the identification of the Se and As binding biomolecules would be of great interest to determine if these metals are detoxified or if, conversely, the biomolecules are metal-sensitive and their binding to Se or As represents a threat for the health of fish.

TU065

Assessment of Toxicological Impact of Anthropogenic activities on Onitsha Stretch of River Niger in Southeastern Nigeria.

A.C. Udebuani, Federal University of Technology / Department of Biotechnology; J.I. Nwajuba, Federal University of Technology Owerri / Department of Biotechnology; p. Abara, federal univeristy of Technology Owerri / Biology

The impact of anthropogenic activities on an urban stretch of a major river in Nigeria with respect to endocrine disrupting compounds and heavy metal concentrations was investigated. Three sampling points were selected along the Onitsha stretch of River Niger, based on the inlets of different tributaries into the river. Heavy metal contents of the water samples were analyzed after acid digestion, while the endocrine disrupting compounds were analyzed using gas liquid chromatography. The result obtained showed that six heavy metals with varying concentrations were obtained in the order of Zn>Pb>Hg>Cd>Cr>Ni. The HPI and MI values were far above the critical values. Results also showed EDCs obtained to include PAH, phthalates, PCDDs, PCDFs, PBDEs, bisphenol A and PCBs. This study established that Onitsha stretch of River Niger contains varying concentrations of heavy metals and EDCs. The stretch of that river is highly polluted, and anthropogenic activities are highly impacting negatively on the river. There is therefore need to regulate the activities of people, especially the influx and disposal of pollutants into this surface water.

TU066

Bioaccumulation, DNA damage and metallothionein expression in plants grown on heavy metal contaminated soil supplemented with sewage sludge

M. Jaskulak, Czestochowa University of Technology / Institute of Environmental Engineering; A.O. Murtaś, Czestochowa University of Technology / Department of Infrastructure and Environment; A. Grobelak, M. Kacprzak, Czestochowa University of Technology / Institute of Environmental Engineering

Many anthropogenic activities have contributed to a release of contaminants, including heavy metals, into the environment. Since plants cannot leave polluted areas, it is, therefore, essential to possess a vast range of defence mechanisms that can reduce the toxic effects of heavy metals (HM). Contamination of soil and water with HM not only decreases the growth of plants but since metals can be accumulated in plant tissues they cause a severe threat to animals and humans the food chain. Identification of plants response mechanisms to contamination is becoming a prime objective in research since this knowledge can provide a solution for soil contamination and metal accumulation in plants. Studies on plants have demonstrated the ability of specific proteins - metallothioneins (MTs) to hyperaccumulate heavy metals, and play a significant role in their detoxification and overall oxidative stress. The physiological roles of MTs are not completely understood and much is still unknown concerning their characterization in many higher plant species. The aim of the study was to evaluate the effects of fertilization of contaminated with HM soil by sewage sludge on the genotoxicity levels and the expression of metallothioneins in plants shoots and roots. The toxicity assessment was conducted using selected measurement endpoints: germination index, roots length, the severity of DNA damage, chromosome aberrations and the expression level of metallothioneins. *Sinapis alba* L. was chosen as a model plant for this experiment. Plants were grown for 28 days in a growth chamber where they were exposed to soil contaminated by HM from metallurgical activities and to contaminated soil amended with different concentrations of sewage sludge. The study showed the effects of sewage sludge on the level of genotoxic effects caused by heavy metals as well as on MT expression. As such, a significant increase in the expression levels of MT was observed in plants grown under metal stress. The differences showed statistically significant changes between related conditions which means that presented assay can be used as a sensitive stress marker for phytoremediation process.

TU067

Chronic toxicity assessment of Ni contaminated rivers in Japan using Ceriodaphnia dubia for development of biotic ligand model for Japanese surface waters

H. Watanabe, M. Noguchi, T. Misaki, National Institute for Environmental Studies / Center for Health and Environmental Risk Research; N. Nakayama, National Institute for Enviornmental Studies / Center for Health and Environmental Risk Research; M. Ishikawa, National Institute for Environmental Studies / Center for Health and Environmental Risk Research; S. Oda, National Institute for Environmental Studies / Center for Environmental Risk Research; K. Matsuzaki, H. Yamamoto, National Institute for Environmental Studies / Center for Health and Environmental Risk Research

Ni is one of industrial essential chemicals and have been widely detected in Japanese river. US and EU have already established the water quality standard/criteria for aquatic life protection; however, it is still under development in Japan. In metal toxicity assessment, bioavailability of metals is an important factor and Ni bioavailability models (i.e. biotic ligand model (BLM)) for both acute and chronic toxicity have already been available for plant, invertebrates, and fish. They were generally established based on the data of European or US surface waters (hard water in general), which have different water chemistry from Japan (soft water, in general). Since water chemistry parameter (e.g. Ca, Mg, Na, K, pH, natural organic carbon) highly influence on metal toxicity, we should check applicability of the existing BLMs on Japanese surface waters or develop our original BLM based on the data of Japanese surface waters. To collect Ni toxicity data in surface waters, we collected 45 river water samples from Ni contaminated rivers all over Japan and conducted the daphnid reproduction test using *Ceriodaphnia dubia*, which is one of the most sensitive species to Ni and recently came into use as test species to evaluate surface waters and industrial effluent in Japan. We used The Windermere Humic Aqueous Model (WHAM7) for speciation calculation. Ni toxicity were predicted using the existing chronic Ni bioavailability model for *C. dubia* established by De Schamphelaere et al. (2006). Except for uncontaminated upstream samples, the daphnids demonstrated typical toxic symptom of Ni (delayed lethal toxicity) and reproduction inhibition levels were correlated with Ni concentration suggesting that Ni is the representative toxicants in the collected samples. However, in several stations, other metals (such as Zn) may also contribute the toxicity thus we should carefully interpret the mixture toxicity.

TU068

Comparing metallic elements in corals from South Africa and the Mascarene Basin

V. van der Schyff, North-West University / Unit for Environmental Sciences and Management; R. Choong Kwet Yive, University of Mauritius / Chemistry; H. Bouwman, North-West University / Unit for Environmental Science and Management

Coral reefs are one of the most bio-diverse biomes on earth. One of the many dangers that coral reefs face is the accumulation of metals and metalloids in skeleton and tissues of the colonies. No knowledge exists on the state of metal and metalloid contamination in corals from the Western Indian Ocean (WIO). Fragments of four soft- and five hard coral genera were collected from five sites in the WIO. Sodwana and Aliwal Shoal constituted the coastal sampling localities from South Africa. Three Mauritian outer-islands in the Mascarene basin (Agalega, Rodrigues, and St Brandon's Atoll) were the selected oceanic sampling sites. Eighty-one coral fragments were collected and analysed for 31 metallic elements using ICP-MS. The corals collected from South Africa contained a higher concentration of most of the metals that were analysed compared with the Mascarene Island samples. Corals without symbiotic algae could only be collected from the South African reefs, and contained the highest concentration of metalloids. Soft corals exhibited a different relative composition pattern of metals than hard corals. Alkaline earth metals, as well as Fe, and U predominated in the hard corals. Soft corals contained relatively higher concentrations of most of the post-transitional metals that were analysed. *Sinularia* is the coral genus with the most elements at the highest concentrations. *Pocillopora* from SBR had very high concentrations of Fe and Cr, possibly due to several shallow shipwrecks in the atoll. Most of the elements tested had lower concentrations in the WIO than in certain regions of the Great Barrier Reef and the Red Sea. Iron was consistently higher in all corals collected during this study than in corals from other studies. Some metals, such as Cu, Ni, and Cd, affect fertilization success of corals. Very high concentration of Ni was reported in *Sinularia* (1300 mg/kg dm)from Sodwana. As ocean temperature rises and ocean acidification increases, metals can become more bioavailable to corals, requiring further study.

TU069

Cytochrome P450, fat and ageing: new insights into metal toxicology

N. Rai, Orebro University / The Life Science Centre,s School of Science and Technology; P. Olsson, Orebro University / The Life Science Center-Biology; J. Jass, Orebro University / SCHOOL OF SCIENCE AND TECHNOLOGY

Toxic metals are among the most persistent environmental pollutants worldwide and have been implicated in metabolic disorders, including diabetes, obesity and neurological diseases. Long-term exposure to metals increase the susceptibility of healthy individual to hematological and developmental disorders. Several genetic markers including metallothioneins, heat shock proteins and oxidative stress related genes have been used to analyze metal stress in different organisms. Other biomarkers for metals include cytochrome P450 (CYPs) a class of xenobiotic metabolizing enzymes that can transform compounds to either non-toxic or carcinogenic metabolites. Studies have shown that CYPs can metabolize important fatty acids and regulate lipid metabolism. Both CYPs and fatty acid metabolism have been implicated the ageing process and lifespan regulation, however the mechanism has not been explored in details. *Caenorhabditis elegans* is an excellent model to study stress response mechanisms induced by metals due to some functional similarities with humans. Our aim was to study the mechanism behind the metal induced CYPs and fatty acid metabolism alterations leading to regulation of lifespan in *C. elegans*. Transcriptomics, viability, lifespan, gene expression analysis and RNA interference were used to explore the interconnection between the CYPs, fatty acid metabolism and lifespan of *C. elegans* following metal exposure. *C. elegans* were exposed to metal contaminated environmental sample and lab reconstituted 12 metal mixture during post hatching larval stages (L1 to young adult). Transcriptomic analyses showed upregulation of *cyp-33A1*, *cyp-35B1* and *cyp-35B2* genes on exposure to both the metal mixture and environmental samples, but the upregulation was above 15 fold in the metal mixture exposed nematodes. Fat staining with Nile red also showed significant increase in the level of stored fats on metal mixture and environmental sample exposure. Further, fatty acid metabolism related genes such as *fasn-1, pod-2, acs-2* and *fat-5* were also altered on exposure to both the metal mixture and environmental sample. Our results show that metals alter the CYPs and fatty acid metabolism and can have further implications on the lifespan of *C. elegans*. Understanding the interplay of CYPs and fatty acid metabolism can unravel possible mechanisms of metal induced onset of several diseases and their detrimental effect on the longevity of exposed individuals.

TU070

Determination of the effects of platinum in the oyster (Crassostrea gigas) using cell and tissue level biomarkers

R. MEDRANO, University of the Baqque Country; M. Abdou, UMR5805 EPOC / Geochemistry; M. Soto, University of the Basque Country / Zoology and Animal

Cell Biology, Research Centre for Experimental Marine Biology and Biotechnology PIEUPVEHU; U. Izagirre, University of the Basque Country UPV/EHU / CBET Research Group Dept Zoology and Animal Cell Biology Faculty of Science and Technology and Research Centre for Experimental Marine Biology and Biotechnology PIE; J. Schäfer, Université de Bordeaux; B. Zaldibar, University of the Basque Country UPV/EHU / Department of Zoology and Animal Cell Biology. Centre of Experimental Marine Biology & Biotechnology

Platinum (Pt) is a trace metal present in very low concentrations under natural conditions, but since the 1970ies the strongly increased industrial use of Pt, especially for car catalytic converters, has totally modified its global biogeochemical cycle increasing its presence in many natural compartments. Oysters have been widely used as sentinel organisms in environmental biomonitoring programs for decades because of their sedentary way of life and ability to accumulate pollutants with little metabolic transformation. The present work addresses the effects of Pt on the Japanese oyster (*Crassostrea gigas*) at low levels of organization, such as cellular and tissue. For this, oysters were exposed to different Pt concentrations (Control = 0 ng.L^{-1}; Low = 50 ng.L^{-1}; Medium = 100 ng.L^{-1} and High = 10 µg.L^{-1}) for 3 (T3), 7 (T7) and 28 (T28) days. The condition index of each individual was calculated as well as the gametogenic development stage. In addition, the histopathology of the oysters' digestive gland was studied, including atrophy levels, tissue structure and parasite prevalence among other anomalies. Different histochemical parameters such as lipofuscins and neutral lipids were also measured and combined with autometallography to detect the location and quantity of Pt. An increase of autometallographical black silver deposits in T7 and T28 was detected for the highest exposure concentrations. Moreover, significant increase in lipofuscin content occurred at all exposure times. On the other hand, neutral lipid levels showed a significant decrease at T28 for the exposure conditions Medium and High. Only minor and non-significant alterations occurred at histological level. This experiment has shown that short-term (28 days) exposure to relatively high Pt concentrations in seawater do not induce alterations at histological levels but lower levels of biological organization such as cellular (lipofuscin accumulation, neutral lipids deployment) are impaired in oysters.
Acknowledgements: Work funded by, Basque Government (IT810-13), UPV/EHU (UFI 11/37). EU FP7 Ocean 2013.2 Project SCHeMA (Project-Grant Agreement 614002), IdEx University of Bordeaux.

TU071

Ecological Risk Assessment of Trace Metal Contaminated Tropical Estuarine Sediment, Southwest Nigeria

A. Usese, University of Lagos, Nigeria / Department of Marine Sciences; O.L. Chukwu, University of Lagos Nigeria / Marine Sciences; R. Naidu, The University of Newcastle / Global Center for Environmental Remediation (GCER), Faculty Science and information Technology; M.M. Rahman, The University of Newcastle / Global Centre for Environmental Remediation GCER, Faculty of Science; S. Islam, The University of Newcastle / Global Centre for Environmental Remediation Faculty of Science and Information Technology,; E.O. Oyewo, Nigerian Institute of Oceanography and Marine Research / Victoria Island, Lagos

Lagos lagoon, the largest of the eight lagoons that make up the lagoon systems of Nigeria has been under intense pressure from several anthropogenic influences over the years. This study evaluates the level of contamination and potential ecological risk of trace metal (Cd, Co, Cr, Cu, Fe, Mn, Ni, Pb, and Zn) concentrations in surface sediment from 15 sites in Lagos lagoon during the wet and dry season by an Agilent 7500c (Agilent Technologies, Tokyo, Japan) Inductively Coupled Plasma Mass Spectrometry (ICP-MS). With only few exceptions, the concentrations of trace metals in the order Fe ? Mn ? Zn ? Cu ? Cr ? Pb ? Co ? Ni ? Cd rarely exceeded threshold element levels for the protection of aquatic life. Risk analysis using contamination factors (CF) and Enrichment factor (EF) suggests very significant enrichment from Zn and Cd as well as a high degree of contamination (C_d) from Cd (16.88-21.56) at locations closest to urban runoffs, industrial activity, domestic and solid waste dumps. Estimated pollution load index (PLI), geochemical accumulation (Igeo ? 0) index as well as the applied sediment quality guidelines (SQG) values by the World Health Organization (WHO) and United State Environmental Protection Agency (USEPA) indicates low to moderate degree of contamination from sediment metals concentrations and the unlikely risks to ecological receptors during the study period.

TU072

Effects of culture medium on metal toxicity and new approach for ecotoxicology assessment

G. Pascual, Tohoku University / Civil and Environmental Engineering; I. Garcia, N. Tanaka, O. Nishimura, Tohoku University / Architecture Civil and Environmental Engineering

Pollution of aquatic ecosystems is a global problem affecting the biological continuity of living organisms. In this context, metals is a group of pollutants occuring naturally in the environment, however human activity increase its concentration in the environment. Therefore, for environment surveillance purposes, metals entering aquatic ecosystems are regulated by water quality guidelines. The last one is based on the results obtained in toxicology tests using aquatic organisms, nonetheless the reported medium Effective Concentration (EC50) of a tested metal in a species widely varies. Here, our first purpose was to study the effects of culture medium on metal toxicity. Based on these results, our second purpose was to propose a new approach for the evaluation of metal toxicity on microalgae avoiding the interference of culture medium. In this study, we evaluated the toxicity of copper (Cu), lead (Pb) and zinc (Zn) on the microalgae *Pesudokirchneriella subcapitata*, since they are considered to be more sensitve to chemicals compared with other aquatic organisms such as fish. Cu and Zn were chosen as study metal species, since they play an important role on biological activity. However, for the other study metal, Pb, any positive, biological funtion has not been reported. All tests were run in transparent microplate (96 wells), and pH of test solutions was adjusted at 6.5. The algae growth was determined measuring the fluorescence (435/685 nm). In the first experiment, the microalgae was exposed for 72 hours to each metal using three different types of culture medium, OECD medium, modified OECD medium (mOECD) and Bold Basal Medium (BBM). In the second experiment, the microalga was exposed in a simplified test medium (destilled water buffered with MOPS and NaOH) only for 6 hours, as nutrients available for algae were limited. In the first experiment condition, the EC50 after 72hours were 140, >1200 and 293 µg/L for Cu, Pb and Zn in OECD medium, respectively, in mOECD, they were 34, 219 and 134 µg/L, respectively and in BBM, they were >300 µg/L in all the cases. In the second experiment, the obtained EC50 after 6h were 150, 189 and 88 µg/L for Cu, Pb and Zn, respectively. The obtained EC50 of the metals differed between culture mediums. So the composition of culture medium affect the metal toxicity. Hence, we suggest that the simplified test medium may be an appropiate alternative to evaluate metal toxicity preventing interference of culture medium.

TU073

Environmental diagnosis of water and tilapia Oreochromis niloticus of the Tenango dam, Puebla, Mexico.

M. Munoz-Najera, G. Barrera Escorcia, Universidad Autonoma Metropolitana Iztapalapa / Hidrobiologia; P. Ramirez Romero, U.A.M. Iztapalapa / Hidrobiologia

Human population has seen the deterioration of resources derived from\n overexploitation and contamination by anthropogenic activities, an example of\n this is water. Due to the growing demand for this resource, associated to\n population growth, industry, livestock, and agriculture, dams have been built to satisfy these needs. In Mexico, some of them were constructed in sites\n that over time were decleared protected natural areas. Populations\n settled on its banks to make use of the water, as well as of the\n organisms linked to these aquatic bodies, which represents economic sustenance\n for the inhabitants. On the other hand, it is common to use the water for\n various purposes, many of which contradict each other. Such is the case of\n the Tenango Dam, in Puebla, México, which is used for fishing, irrigation,\n recreation and electric power generation, among other purposes. A study was\n carried out to evaluate the Tenango Dam water and tilapia quality. Five field\n trips were made in 2015. Physicochemical parameters were recorded: pH,\n dissolved oxygen and temperature; as well as nutrients: nitrites, nitrates\n and phosphorus; also metals: cadmium, chromium, copper and lead were\n determined in both water and tilapia. Results indicated that the\n physicochemical parameters are within Mexican admissible ranges. Nitrite and\n phosphorus exceeded the acceptable limit for urban use and protection of\n aquatic life. Lead and chromium in water exceeded the limits in four\n collections, and tilapia, only in two of them. Cadmium and copper registered\n in water behaved similarly exceeding in two seasons the levels allowed by\n Mexican law, while in tilapia, cadmium only exceeded the acceptable limits\n for consumption in two seasons. Based on the concentrations of nutrients and\n metals, it is concluded that water of the Tenango dam is not suitable for\n urban use, nor for the protection of aquatic life and tilapia should not be\n consumed. These levels of contaminants could represent a risk to the life\n associated with this artificial water body. The diverse uses and the absence\n of a management strategy have deteriorated the dam's water quality and also the tilapia as a resource associated with it; finally, this situation compromises\n the integrity of an aquatic body included in a site declared as a protected\n natural area.

TU074

Estimation of Target Hazard Quotients and Potential Health Risks of Some Heavy Metals from Lipsticks in Nigeria.

O. Otitoju, federal University Wukari / Department of Biochemistry; A.C. Udebuani, Federal University of Technology / Department of Biotechnology; T. Otitoju, University of Nigeria Nsukka / human nutrition and dietetics

Heavy metals have been implicated as a causal factor in literally any health problem including infertility and cancer. Their presence in most cosmetic products may pose more harm than envisaged. Lipsticks are common beautifying cosmetics used by young and matured ladies for the purpose of appearing attractive. However, the presence of heavy metals in most lipsticks may help to predict the possible risk associated with the use of these products. The main objective of this paper is to evaluate the hazard quotients of heavy metals due to daily ingestion or use of lipsticks among users and also to evaluate target cancer risks due to its use. This study was carried out in Wukari, Nigeria, samples of different lipsticks and lip glosses of many colours and texture were collected and analyzed for heavy metals contents (lead, Arsenic, Chromium, Cadmium and Mercury) using Atomic Absorption Spectrophotometer (AAS). The result of the mean concentration of the heavy metals are as follows; Lead, ranges between(2.65-7.40 ± 0.17) mg/kg;

Arsenic concentration range between (0.55-1.53 ± 0.26) mg/kg and chromium was 0.04-0.16 ±0.02) mg/kg. Cadmium and arsenic concentrations were below detectable limit of 0.001mg/kg) while mercury concentration ranges between 0.04-0.61 ± 0.01) mg/kg. Calculated target hazard quotient (THQ) was highest in mercury with the value of 560.59 and the lowest value was obtained in Arsenic with 1.43×10^{-3}. However, target cancer risk (TR) was highest for Lead with the value of 2.05×10^{-2} and the lowest value for cadmium was 6.32×10^{-6}. This study has shown that some lipsticks products popularly used in Nigeria contain high concentration of heavy metals such as Lead, Arsenic, Chromium, Cadmium and Mercury and they have high cancer risk. Therefore, public health awareness on the risk associated with the use of these cosmetic samples should be carried out.

TU075

Fatty acid profile of Cerastoderma edule and Scrobicularia plana affected by copper sulphate exposure

A.d. Mesquita, Department of Biology & CESAM - University of Aveiro / Department of Biology and CESAM,; F. Gonçalves, University of Aveiro / Department of Biology and CESAM; J.C. Marques, University of Coimbra / MARE, Dep. of Life Sciences, Coimbra University; A.M. Gonçalves, MARE, Dep. of Life Sciences, Coimbra University/Biologia Department & CESAM, Aveiro University

At the past 30 years were recorded an intensive practice in the use of fertilizers and pesticides, mainly in the European Mediterranean region, that, in particular cases, exceeded the limits of regular legislations established by the European Union. The widespread use of these chemicals compounds and the pressure over agricultural fields near valuable ecologically coastal areas conducted to the implementation of monitoring plans to the recovering of aquatic ecosystems. Copper sulphate is used in industrial activities, but also it is much used in pesticides formulations, with application in agricultural activities, namely in rice farms to control pests. Studies reported that copper may affect biochemical processes, such lipid metabolism of some organisms, although specific changes in fatty acid (FA) profiles are still unknown. Nowadays, bivalve species are used in ecotoxicological bioassays due some particular characteristics, such as the wide distribution, ecological relevance, the capacity to filter and ingest large volumes of sediment particles and water and ease handling in the field and in the laboratory. Therefore, this work aims to determine toxic effects and changes in fatty acids profile composition of the two marine bivalve species *Cerastoderma edule* and *Scrobicularia plana* when exposed to copper sulphate, considering small (medium body size = 1.97 cm and 3.47 cm, respectively) and big (medium body size = 2.45 cm and 4.20 cm, respectively) size classes. In a first phase organisms were exposed under laboratorial conditions to copper sulphate to determine lethal concentration; at a second phase, it was compared the FA profile and the nutritive quality of both species and size classes at the field and in the lab. Our results state *C. edule* is more sensitive to copper sulphate (LC50 = 0.818 (0.595–0.987) mg/L; 1.129 (0.968–1.289) mg/L, to big and small organisms, respectively) than *S. plana* (LC50 = 2.563 (2.229–2.903) mg/L; 4.705 (3.540–12.292) mg/L, to big and small organisms, respectively). Furthermore the last one presents greater abundance and variety of FA and essential fatty acids (EFA), namely DHA and EPA, rates than *C. edule*. Still, big size class of both bivalve species is the most affected by the contaminant.

TU076

Heavy metals in soil and vegetables of allotment gardens in the Cape Town, South Africa

M. Kweza, F. Wewers, T. Oosthuysen, T. Farrar, A. Giwa, Cape Peninsula University of Technology / Chemistry

Increased industrialization has resulted in an unprecedented dissemination of toxic substances, among which are heavy metals, in the environment. Heavy metals are persistent environmental contaminants which ultimately accumulates in soil with possible translocation into the tissue of vegetables, thereby posing a potential risk to human health. While most research focus on major agricultural areas, less attention has been paid to the accumulation of heavy metals in home gardens, schools and rural areas where subsistence farming is increasingly used in South Africa as a means of poverty alleviation and increasing food security. This study was conducted to investigate the concentration of selected heavy metals in soil, water and vegetables from allotment gardens in informal settlements around Cape Town, South Africa. Thereby assessing the health risk associated with the consumption of vegetables grown in the informal agricultural sector. Soil, water and vegetables were sampled during winter and summer seasons from the study areas and were analyzed for heavy metals (Pb, Cd, Mn, Zn, Cr, Cu, Ni, Fe and Co) using Inductively Coupled Plasma (ICP). Results showed that there are is no significant seasonal variation ($p < 0.05$) in the physico-chemical parameters of soil and water samples. The soil and water pH are slightly acidic, ranging from 6.30 to 6.90, and 5.60 to 7.00, respectively. Soil organic matter ranges from 1.7 to 13.5%. Results for water indicated that there was concentration fluctuation during winter and summer, with summer concentrations ranging from 0.062 to 0.947 mg/L, while in winter the range was 0.002 to 2.347 mg/L. Soil heavy metal concentrations ranged from (0.59 -1209.95 mg/kg) in winter and (0.52 -1127.41 mg/kg) in summer. For both seasons the metal concentration in soil increases in the order; Cd < Co < Ni < Cr < Pb < Mn < Zn < Fe. The concentrations of all the elements in soil and water samples were within the permissible limits set by WHO and FAO. The concentration of heavy metals in vegetables were generally higher in summer (ranging from (nd – 116.26 mk/kg) than in winter (ranging from nd – 144.28 mg/kg), with the general trend being in the order; Cd < Ni < Pb < Co < Cu < Cr < Zn < Mn < Fe. In general, the below-ground vegetables such as brinjals and green peppers exhibited lower accumulation tendencies than above-ground and leafy vegetables such as cabbage and spinach.

TU077

High-selenium lentils offer a nutritional solution to combat arsenic poisoning in Bangladesh

J. Smits, University of Calgary / Ecosystem & Public Health Faculty of Veterinary Medicine; R. Krohn, University of Calgary / Dept. of Ecosystem & Public Health, Faculty of Veterinary Medicine; E. Akhtar, International Centre for Diarrhoeal Diseases, Bangladesh / Nutritional Biochemistry; B. Vandenberg, University of Saskatchewan / Plant Sciences; R. Raqib, International Centre for Diarrhoeal Diseases, Bangladesh / Nutritional Biochemistry

Background: Worldwide, the major chronic environmental threat to human health affecting over 100 million people, is daily exposure to naturally high levels of arsenic through drinking water and food, notably rice. Malnutrition increases the toxicity of arsenic. Low blood selenium specifically, increases the risk of arsenic-induced skin lesions and other manifestations of arsenic poisoning. Selenium, an essential element that interacts antagonistically with arsenic in the body, has been shown to decrease body burdens of arsenic and reduce arsenic-induced atherosclerosis in animals fed high selenium diets. *Objectives:* To reduce arsenic absorption, and therefore arsenic-associated toxicity in highly exposed people, through a dietary intervention with naturally high selenium lentils. This treatment is especially practical for populations already consuming lentils on a daily basis, as in the region notorious for chronic arsenic poisoning, the Indogangetic plains of northeast India and Bangladesh. *Methods*: For six months in a double-blind study, 400 participants with tube well As levels from 100 to 1200 ppb based on atomic absorption spectroscopy (AAS) analysis (WHO limits; 10ppb for the west and 50 ppb in other regions) ate the same variety of lentils with high (0.854ppm) or low (0.029ppm) selenium because of the soil where they were grown. Urine, stool and hair samples were collected before, during, and at the end of the study, to determine arsenic levels and other physiological responses. *Major outcomes:* Mixed model statistical analyses determined that people consuming the high selenium lentils excreted significantly more arsenic though their urine ($p< 0.05$) than those on the low selenium lentils, but there were no differences in stool As concentrations. Considering females only, there was a trend towards a difference in hair As on the 2 diets, Hair As decreased by 0.20 ppm in the high selenium lentil group, whereas it increased by 0.49 ppm in the low selenium group ($p=0.07$). *Summary*: This study provides evidence of the potential effectiveness of a simple, whole food solution of consuming lentils naturally high in selenium to reduce absorption of arsenic from water and food.

TU078

Metals removal from water for hazard classification

G. Burton, University of Michigan / School for Environment and Sustainability; M. Hudson, University of Michigan / School of Natural Resources and Environment; R.F. Carbonaro, Mutch Associates, LLC / Civil and Environmental Engr; K.J. Rader, Mutch Associates, LLC; S. Baken, European Copper Institute; E.R. Garman, NiPERA / Ecotoxicologist

Metals usually enter aquatic ecosystems in an oxic environment and associated with particles. It is important to understand this initial fate process in regards to partitioning, speciation and resulting biological effects. In addition, current European Union regulations and the global GHS system mandate a hazard evaluation, which includes the assessment of Rapid Degradation (greater than 70% within 28 days), which for metals equates to metal removal from the water column. The Transformation/Dissolution Protocol (OECD 29) is an established method that was modified to examine metal removal from the water column under oxic conditions. The modifications include the addition of a small amount of sediment, and the inclusion of a resuspension event. We conducted a series of laboratory evaluations to address the following questions: Are copper (Cu) and nickel (Ni) removed from the water column of freshwater systems and if so, what is the rate of removal? How do various test method conditions affect metal removal, using OECD method 29? What sediment characteristics affect metal removal and which show a reasonable worst case (RWC) condition? What is the mechanism for metal removal, and are metals released into overlying waters upon subsequent resuspension? Method parameters evaluated included: sediment type and loading rate, pH control, metal loading rate, pre-incubation of sediment, and resuspension. Chemical analyses included dissolved Cu, Ni, and Fe, dissolved oxygen (DO), pH and AVS-SEM of sediments. Multiple dried vs. non-dried sediments were tested in batch reactors for both 96 h and 28 d tests. Dry Buffalo River sediment, a sediment with reasonable worst-case properties for metal binding, typically removed 70% Cu and Ni from the water column at 1 mg/L loading. Incubated sediments removed metals significantly faster than non-incubated sediments ($p < 0.03$). Higher sediment loading rates removed metals faster as expected. Sediment type and loading rates affected pH, which started at 6.0. Cu removal (96 h) and resuspension (1 h post 96 h) resulted in no significant increase in Cu, but did elevate Fe concentrations. The results show that 70% of Ni and Cu is removed from the water column using this test modified OECD 29 test method, using a variety of sediments and conditions.

TU079

Modelling the chronic toxicity of copper to fish at low pH

S. Baken, European Copper Institute; K. De Schamphelaere, Ghent University (UGent) / Applied Ecology and Environmental Biology

Bioavailability models account for the effects of water chemistry on metal toxicity to biota. They are a cornerstone of the environmental risk assessment of many trace metals, including copper. In this context, it has often been assumed that toxicity of dissolved copper to fish increases with decreasing pH. However, some studies show that this relationship may only be valid above pH 7. Below pH 7, the chronic toxicity of dissolved copper to fish seems to be independent of pH. Existing bioavailability models use the well-known mechanistic concept of the Biotic Ligand Model (BLM), but this modelling framework seems to have difficulties to reproduce the observed relationship of copper toxicity versus pH. This study was set up to refine the bioavailability models for chronic copper toxicity to fish, in order to better reflect the observed relationship between chronic copper toxicity and pH. The available chronic copper toxicity data to fish were reviewed. A new bioavailability model was developed using the concept of a generalized bioavailability model (gBAM). This semi-empirical model assumes a log-linear relationship between pH and effect concentrations (ECx) expressed as free cupric ion activity ($Cu2+$) and links it to the geochemical speciation model WHAM7 to predict toxicity on a dissolved copper basis. The existing bioavailability models were evaluated and compared to the newly developed gBAM. The underlying assumptions, advantages and limitations of each model are identified. Conclusions and implications for modelling copper bioavailability to fish are discussed.

TU080

Novel In-situ Toxicity Assessment of Sediment Capping Effectiveness in Deep Water

G. Burton, University of Michigan / School for Environment and Sustainability; M. Hudson, University of Michigan / School of Natural Resources and Environment; S. Nedrich, A. Rentschler, University of Michigan; K. Thiamkeelakul, University of Michigan School for Environment and Sustainability; S.S. Brown, The Dow Chemical Company / Environmental Remediation and Restoration

A former mining site has been the subject of intensive restoration for the past few years, with significant focus on disconnecting mine spoils from groundwater and managing the quantity and quality of runoff. A remaining task is to ensure that concentrations of zinc in surface water of a large pit lake are reduced below water quality standards. An investigation was conducted to compare the efficacy of selected capping materials for decreasing Zn dissolution during periods when the hypolimnion is anoxic and acidic (pH=5.5). Capping materials were selected based on results from laboratory batch testing and included AquaBlok, limestone, and limestone + bonechar. Experimental field tests implemented novel methodologies, using Limnocorrals (LC) to isolate water columns above various capping treatments, simulating lake-mesocosms. Simultaneous in-situ and ex-situ toxicity tests were conducted using *Daphnia magna*, *Hyalella azteca*, and *Chironomus dilutus*. Test organisms were protected from temperature shock by pre-acclimating over 24 hrs and then deploying the test chambers in a Toxicity Assessment Container System (TACS), which protected the organisms from warm surface waters until reaching the bottom sediments and colder water. Test organisms were exposed to surficial sediments in reference LC or capping materials and overlying water. Ex-situ testing was conducted in waters and/or sediment cores collected from the bottom of each LC, and these tests were done at the same temperature as the in-situ TACS exposures (15 to 19 C, depending on deployment period). Results from in-situ testing demonstrated the usefulness of the TACS and provided similar results to the ex-situ testing. Preliminary results suggest organism survival is similar between in-situ capped and reference sediments; however, supplemental ex-situ analyses will help determine whether capping performance and toxicological response is indicative of site specific characteristics (sedimentation, sediment type, cap layer attenuation) and/or cap specific indices (permeability, adsorption, ion exchange capacity). Results provided for more effective decision-making, with reduced uncertainty, than standard laboratory and chemistry only approaches.

TU081

REEchangE - Rare Earth Elements Ecotoxicology in a Changing Environment

H. Tien, Hamburg University of Applied Sciences/University of the West of Scotland; A. Hursthouse, University of the West of Scotland / School of Science; S. Heise, Hamburg University of Applied Sciences / Life Sciences

REEchangE focuses on the anthropogenic release of the rare earth elements (REE) lanthanum and gadolinium to the environment and their potential risk within ecosystems. REE are increasingly applied e.g. in green technology, and consequently also emitted to the environment. But there is a diversity of potential polluting sources of which little is known, and no regulatory environmental framework for immissions exists so far. In addition, a review by Herrmann et al. (2016) demonstrated the considerable lack of reliable data for La toxicity in the aquatic environment. Considering their future use, release, and environmental fate, an evaluation of environmental risk from lanthanum and gadolinium will have to be based on information on exposure pathways, exposure and effect concentrations. The project REEchangE addresses these topics in the following ways: (1) by studying the toxicity to aquatic organisms. Results on ecotoxic responses obtained for *Aliivibrio fisheri* and *Raphidocelis subcapitata* so far are in the same range as literature data, and show a higher toxicity of Gd compared to La. Effect concentrations are of the same magnitude as for cadmium. (2) by substance flow analysis (SFA) for La and Gd, exemplarily performed for Germany. Information has been collected from published work for a variety of potential sources for La and Gd in rivers and lakes. Additionally, water and sediment samples have been analysed at specific locations. Current data point to wastewater and specialised industries as prominent sources of emission. (3) by investigating the impact of changing environmental parameters (pH, redox, salinity) on the bioavailability of particle bound La and Gd. In a microcosm, a battery of miniaturized biotests will be applied to monitor the toxicity responses in overlying water and sediment. This includes tests with *Aliivibrio fisheri*, *Vibrio proteolyticus Arthrobacter globiformis* and especially *Daphnia magna*. Additionally, speciation and bioavailability of the La and Gd are examined following a procedure by Simpson et al. (2014), applying a cascade of different filters and a chelating resin. The presented poster will depict the current results of the microcosm experiments along with the information on bioavailability based on biotests and speciation data.

TU082

Sediment characteristics of natural and anthropogenic origin and their possible association with benthic macroinvertebrates in a minimally affected river in South Africa.

C. Wolmarans, H. Pienaar, G. Van Niekerk, NorthWest University School of Biological Sciences / Zoology

Sediment characteristics generally entail metals, minerals, organic content, elements, particle size conductivity and pH. The origin of metals in sediment may originate from anthropogenic activities including mining, industries, agriculture as well as aerial deposition. Limited attention is however given to the contribution that natural occurring phenomena play in the concentration of metals in the sediment. Weathering of minerals originating from the primary lithology can on the one hand add to the metals in sediments and on the other hand to the particle size composition. Although it is well known that chemical pollutants, due to anthropogenic impacts, act as a major determinant for the macroinvertebrates composition in surface waters the influence of the above mentioned components in a pristine river is less known. The aim of this investigation was firstly to determine the sediment characteristics and secondly to establish which of these characteristics have a significant impact on the macroinvertebrate community structures in the Marico River, South Africa. Sediment was collected from the upper 7cm of the substratum at various sites, dried and sieved using an Endocott dry-sieving system to collect fractions < 2000µm and < 50µm. The total sediment samples >2000µm and clay fraction samples, 50µm were subjected to metal, scanning electron microscopy and minerals by X-ray diffraction analyses. Element anlyses were done by means of an FEI Quanta 250 FEG ESEM microscpe equipped with an integrated Oxford Inca X-Max 20 EDS. Macroinvertebrates present in the benthos were collected for 15 minutes using a standard sweep net, preserved in 90% ethanol and identified up to family level. RDA redundancy analysis was constructed to investigate the distribution of macroinvertebrates Forty two families of which the vast majority associated with particles >2000µm, were found. Sediment particle sizes < 2000µm had a detrimental effect on the biodiversity. No significant correlation was demonstrated between variation in temperature, pH, and electrical conductivity and both diversity and abundance of macroinvertebrates. Although relatively high concentrations of selected metals were present in the sediment, it was largely from geological origin and most probably not bioavailable.Therefore, it can be concluded that, under these conditions, sediment particle size, played the decisive role on the distribution and abundance of macroinvertebrate taxa.

TU083

The effect of copper sulphate on the antioxidants enzymes activity of two size classes of Cerastoderma edule

A.d. Mesquita, Department of Biology & CESAM - University of Aveiro / Department of Biology and CESAM,; S.M. Marques, University of Aveiro; J.C. Marques, University of Coimbra / MARE, Dep. of Life Sciences, Coimbra University; F. Gonçalves, University of Aveiro / Department of Biology and CESAM; A.M. Gonçalves, MARE, Dep. of Life Sciences, Coimbra University/Biologia Department & CESAM, Aveiro University

Anthropogenic activities, such as agriculture or industrial activities are the main source of pollution contributing for the degradation of water quality and thus affecting the living organisms of the aquatic systems. Copper is often released into the aquatic systems, and may affect these ecosystems and its communities. Copper sulphate is a copper-based formulation, used in the agriculture practices to control pests. The main aim of this study is to determine the effects of copper in the antioxidant defence system of an important commercial bivalve species, *Cerastoderma edule* in two size classes. In this work was observed the behaviour activity of the organisms during the exposure time to copper sulphate and subsequently it was determined the antioxidant enzymatic activities of GST, GRed and GPx in the muscle tissue (foot). Moreover, lipid peroxidation was evaluated

through thiobarbituric acid reactive substances (TBARS) measurement in the muscle tissue. The results showed changes in the behaviour and enzymatic activity at the different copper sulphate concentrations to both size classes. Moreover, according to TBARS levels, lipid peroxidation possibly occurred on the big size class of *C. edule*. The muscle tissue (foot) showed to be a good tissue to use in biochemical analysis to detect response to the exposure to toxicants.

TU084

The impact of single metals and mixtures in nature: a microcosm experiment

M. Van Ginneken, University of Antwerp, Dept. Biology / Biology (SPHERE); R. Blust, L. Bervoets, University of Antwerp / Department of Biology (SPHERE Research Group)

Evaluating ecological risks of metal-contaminated systems remains an important challenge. While laboratory experiments with metal mixture exposure are receiving more attention in the literature, little research has examined the interaction of natural stressors with metal mixtures. In the lab, we already performed experiments on *Asellus aquaticus*, exposing this freshwater isopod to a combination of metal mixtures and temperature stress. This way we could study effects on the individual level and relate metal accumulation to relevant sublethal endpoints (e.g., growth rate, feeding rate). The present study, a microcosm experiment in a greenhouse, was designed to gain more insight into the effects of these metals on populations and communities. Small ecosystems with several species of macroinvertebrates were exposed to Cd, Cu, Pb and a mixture of these three metals under semi-natural conditions. In each bucket, we placed *Asellus aquaticus*, *Daphnia magna*, *Chironomus riparius* (midge larvae), *Physa* sp. (Mollusca), *Elodea nuttallii* (macrophytes) and *Raphidocelis subcapitata* (algae). The theoretical metal concentrations were 1.5 µg/L Cd, 70 µg/L Cu, and 72 µg/L Pb. Half of the medium was renewed weekly. The effects of the metal mixtures and natural stressors were examined after 4 and 8 weeks, on the individual level (total metal accumulation, survival, shoot and root length), the population level (species densities, biomass) and the community structure (diversity, evenness). Preliminary results show a high variability between replicates. We observed no significant differences in species densities between the metal treatments after 4 or 8 weeks. After 4 weeks, we found that Cu and the tertiary mixture negatively affected shoot and root length of *E. nuttallii* compared to the control treatment. However, after 8 weeks, we did not find these significant differences. As we could not find any significant effects of the metals at the end of the experiment, further research focused on sublethal factors or with a longer exposure duration is needed.

TU085

The influence of soil properties on lead bioavailability and toxicity to Enchytraeus crypticus

L. Zhang, VU University Amsterdam / Animal Ecology; C. van Gestel, Vrije Universiteit Amsterdam / Ecological Science

Soil properties are important factors modifying metal bioavailability to soil organims and subsequently affecting the metal toxicity. The present study aimed at investigating the bioavailability and toxicity of lead to the potworm *Enchytraeus crypticus* in the soils with different properties. Six soils with a wide range of properties were spiked with $Pb(NO_3)_2$ at 9 concentrations of Pb to determine the effects of soil properties on Pb bioavailability and toxicity to *E. crypticus*. Survival and reproduction after 21 d exposure were related to total, 0.01 M $CaCl_2$ extractable and porewater Pb concentrations in the soil and internal Pb concentrations in the surviving animals. pH_{CaCl2} and $pH_{porewater}$ decreased with increasing total Pb concentartion for 6 soils, but pH decrease was much stronger for the soils with lower CEC and OC contents. Sorption of Pb from the $CaCl_2$ extracts could be well described by a Freundlich isotherm (R^2 = 0.96-0.99) and Freundlich sorption constant K_F increased linearly with increasing cation exchange capacity (CEC) (R^2 = 0.86) or organic carbon content (OC) (R^2 = 0.76). Pb bioaccumulation in the enchytraeids was soil-dependent, but differences between soils almost disappeared when relating Pb bioaccumulation to available Pb concentration in soils. Toxicity values varied greatly among soils, with median lethal concentrations (LC50) based on total Pb concentrations ranging from 246 to >3092 mg Pb/kg dry soil. LC_X on the basis of total Pb concentration increased linearly with increasing CEC (R^2 = 0.70-0.90) or pH_{CaCl2} (R^2 = 0.87-0.94). The differences in Pb toxicity among soils could be explained from $CaCl_2$ extractable Pb concentrations in soil (R^2 = 0.97) and internal Pb concentrations (R^2 = 0.97). Median effective concentrations (EC50) based on total Pb concentrations varied 12-fold among soils from 81 to 1008 mg Pb/kg dry soil. EC_X on the basis of total Pb concentrations increased linearly with increasing pH_{CaCl2} (R^2 = 0.70-0.94). The variation in EC50 was best explained by differences in the $CaCl_2$ extractable Pb concentrations in the soils (R^2 = 0.94). In general, pH was an important soil property affecting LC50, EC50 and internal Pb concentrations in enchytraeids, as Pb availability, internal Pb, mortality and reproduction were inversely related to soil pH. Soil properties should be taken into account during the ecological risk assessment of metals in contaminated soils.

TU086

Toxicity evaluation of soils sampled in the vicinity of an Aluminum smelter in Montenegro using the Ames, Bioluminescence and DR-LUC bioassays

A. Perovic, University of Montenegro, Faculty of Natural-sciences and Mathematics / Biology; S. Perovic, J. Vukic, University of Montenegro Faculty of Naturalsciences and Mathematics; D. Sukovic, Centar for Ecotoxicological Investigations; H.A. Leslie, Institute for Environmental Studies VU Amsterdam

This study conducted as a part of the national project ECOTOXI that main topic was testing applicability of several bioassays in assessment of cause-effect relation between levels of organic environmental pollutants in soils and its toxic and mutagenic response on samples organic extracts. Samples was collected in vicinity of Aluminum Plant Podgorica and pools of red sludge in Zeta plain. This area with intensive industrial activity is also reach with agriculture and is just in 5km distance from Podgorica (Capital of Motenegro). Waters of several rivers in this area, which are significantly influenced by municipal wastewaters of tree cities, are abundantly used for watering crops in area. We tested mutagenic potential of samples organic extracts in the Ames test, on bacterial strain *Salmonella typhimurium* TA98, acute toxicity on bioluminescent bacteria *Vibrio fisheri* and concentrations of possible dioxins present in the samples by DR-Luc test on rat H4IIE hepatoma cell line. The obtained results indicate a strong mutagenic effect of organic pollutants mixture in tree samples collected near the Aluminum Plant and pools of red sludge, what was significantly in correlation with the recorded concentrations of dioxins in the DR-Luc test and with measured concentrations of polycyclic aromatic hydrocarbons, which exceeded the maximum allowable prescribed concentrations. Two of tree samples, with high response, were in agricultural area. Iven if it is a clear trend of decrease of mutagenic effects as well as reduction of the concentration of dioxins and PAHs with increasing distance of sampling sites from the Aluminum Plant and pools of red sludge, almost all samples shoved a certain elevated level of mutagenic activity, which may be a consequence of the impact of multiple sources.

Safe by Design: responsible and innovative research for safe and sustainable chemistry (P)

TU087

In silico approaches to screen and design safer chemicals

E. Papa, A. Sangion, P. Gramatica, University of Insubria / Department of Theoretical and Applied Sciences (DiSTA)

The prohibitive economic and social cost of testing, necessary to provide extensive information on fate and effects of existing chemicals to humans and the environment, highlights the need to focus on rational and safe design of chemicals before synthesis (i.e. Safe by Design – SbD approach). This approach applies the principle of green chemistry “Design safer chemicals and products,” and is useful to prevent hazardous substances from being developed and entering the environment, as well as to build safer alternatives to existing hazardous chemicals. While in the last decades computational chemistry and *in silico* models have been widely and successfully applied in the design of drugs with desirable pharmacological activity, these strategies have not yet been applied extensively in the design of sustainable, “safe by design” industrial chemicals as well as no real guidelines exist at the regulatory level. Modelling approaches based on Quantitative Structure-Activity Relationships (QSARs) rely on the assumption that biological activities/properties of chemicals are intrinsically dependent on the molecular structure. Endpoints like for instance toxicities, physico-chemical properties as well as biotic and abiotic degradations can be predicted starting from models based on molecular descriptors of the chemical structure, which serve as basis to develop the SbD approach. Therefore, *in silico* strategies such as the aforementioned QSAR (and QSAR-like) models and multivariate analysis (MVA) can be successfully applied to screen undesired properties of large sets of chemicals in order to identify potentially hazardous compounds or safer alternatives. In this poster we show different examples of QSAR models mainly implemented in the software QSARINS and available in the freely distributed QSARINS-Chem module to screen “safe” from “unsafe” compounds on the basis of different endpoints of scientific and regulatory interest. Different classes of emerging pollutants were investigated using *in silico* models, such as Flame Retardants (FR), Personal Care Products and Pharmaceuticals (PPCPs) and nanoparticles. All the presented strategies support the identification of safer alternatives to chemicals that are screened by QSAR as undesirable from their molecular structure. The QSAR approach, applied for SbD before the chemical synthesis, provides concrete opportunities to increase the sustainable use of chemicals and to reduce the need for *a posteriori* remedial actions.

TU088

Application of chemometric methods and QSAR models to support pesticide risk assessment starting form ecotoxicological datasets

F. Galimberti, ICPS   International Centre for Pesticides and Health Risk Prevention / Public Health; M. Luini, ICPS / Public Health; A. Moretto, Università degli Studi di Milano; E. Papa, University of Insubria / Department of Theoretical and Applied Sciences (DiSTA)

Recently the International Center for Pesticides and Health Risk Prevention (ICPS) of Milan-IT, together with the Wageningen University and Research Centre of Wageningen-NL, worked on a data collection project commissioned by the European Food Safety Authority (EFSA). The aim of the project was to investigate the comparability of the EC_x approach (Effect Concentration affecting a percentage x of test organisms) to the current NOEC approach (No Observed Effect

Concentration), both derived from chronic and long term studies of a data sets of 70 active substances of plant protection products (PPP). The new Regulation for the authorization of PPPs requires that ecotoxicological endpoint values, derived from chronic or long-term studies submitted by the Applicant, are reported as EC_{10} or EC_{20} as well as NOEC. NOEC endpoints have been recently criticized since their values strongly depends on the experimental study design, whereas EC_x values are considered more appropriate since they take into account the whole concentration-response curve. Ecotoxicological data gathered from 70 active substances' approval dossiers were collected and stored into a database, and then analyzed to derive NOEC. Adequate statistical models were selected and used to calculate EC_{10}, EC_{20}, and EC_{50} with confidence intervals. In the present work, quantitative methods and models based on Structure-Activity Relationships (i.e. QSARs) were used to validate the Effect Concentrations of the active substances of the selected pesticides, and to predict missing data. The whole approach is mainly oriented to the aquatic environment, and can provide useful information to screen the potential undesired toxic effects of new pesticides, and of alternatives to existing active substances, starting just from the chemical structure.

TU089

Influence of coatings in the bioaccumulation of TiO2 and CeO2 nanoparticles in rainbow trout

M. Fernandez-Cruz, INIA - National Institute for Agricultural and Food Research and Technology / Environment; I. Rucandio, CIEMAT; A. Garnica-Soto, INIA National Institute for Agricultural and Food Research and Technology; A. Valdehita, Instituto Nacional de Investigación y Tecnología Agraria y Alimentaria / Environment; E. Conde, CIEMAT; J. Navas, INIA - National Institute for Agricultural and Food Research and Technology / Environment; F. Torrent, Universidad Politecnica de Madrid / Escuela Superior de Ingenieros de Montes; D. Hernandez-Moreno, INIA / Environment

In the framework of FP7 Project GUIDEnano we investigated the effect of different metal nanoparticle (NP) coatings (synthetized from PlasmaChem GmbH, Germany) on the the toxicity and bioaccumulation of these NPs trying to establish some relationships between coatings and the related effects. Bioaccumulation studies with rainbow trout have been performed for CeO_2 NPs and TiO_2 NPs of 4-8 nm uncoated and coated with citrate or polyethylene glycol phosphoric acid ester (PEG). OECD Test Guideline (TG) 305 (diet administration) has been followed. Fish (5±1 g weight) were fed for 10 days with a diet spiked with 100 mg/kg of the NPs dispersed in water. A control group fed with pellets containing the vehicle (water) was tested in parallel. This uptake phase was followed by a depuration phase of 42 days. Whole fish, stomach and intestine were collected at different time points (0, 10, 11, 17, 24, 38 and 52 days). In addition at the end of the uptake and depuration phase liver and gills were also collected. The levels of the metals in these tissues were measured by inductively coupled plasma mass spectrometry after an acid digestion. During the treatment and depuration phase, no signs of toxicity and no differences in fish growth or in the hepatosomatic index among groups were recorded. At the end of the uptake phase levels of Ti could be measured in stomach, gills and liver without differences among TiO_2 NPs. A difference was observed for the uncoated NP for which Ti levels in the fish were higher than for the other coated NPs. Ti levels reached basal values already in the first day of depuration indicating a very fast elimination of these NPs from the organism. Higher levels of Ce with respect to the control group could be measured at the end of the uptake phase in stomach, intestine and gills but not in liver. Ce levels were found in fish treated with the coated NPs but not in the group treated with the uncoated NPs. Levels of Ce could be measured the first day of depuration in stomach and intestine of fish treated with CeO_2 NPs coated with citrate whereas Ce was only detected in the stomach of fish treated with CeO_2 NPs uncoated or coated with PEG. After 7 days of depuration, Ce residues reached basal levels indicating a lack of accumulation of these CeO_2 NPs. These results indicate a different behavior for the CeO_2 NPs and TiO_2 NPs. No relationship could be observed between the coating and the observed effects. **Acknowledgements:** EU FP7 project 604387 GUIDEnano.

TU090

Colloidal characterization of nano-enabled products for the restoration of works of art: environmental fate of nano-ingredients

E. Badetti, University Ca Foscari of Venice / Environmental Sciences, Informatics and Statistics; V. Cazzagon, University Ca Foscari of Venice / Department of Environmental Sciences Informatics and Statistics; E. Semenzin, Ca Foscari University of Venice / Department of Environmental Sciences Informatics and Statistics; A. Bonetto, University CaFoscari Venice / Environmental Science, Informatics and Statistics; A. Brunelli, University Ca Foscari of Venice; E. Giubilato, M. Picone, University Ca Foscari of Venice / Department of Environmental Sciences Informatics and Statistics; A. Volpi Ghirardini, University Cà Foscari Venice / Department of Environmental Sciences Informatics and Statistics; C. Giuliani, G. Di Carlo, CNR ISMN; M. Salzano De Luna, M. Lavorgna, CNR IPCB; A. Marcomini, University of Venice / Department of Environmental Sciences Informatics and Statistics

The development of highly innovative techniques and technologies for artworks preservation is providing conservators with new engineered nanomaterials (ENM) and ENM-based formulations that can enhance performance and technical sustainability of art materials [1].However, the human health and environmental impacts that may potentially emerge from these new materials and/or techniques are still little known and requires an adequate assessment and management of potential risks [2,3]. ENM formulations are increasingly preferred for conservation interventions compared to the corresponding bulk materials formulations because of their small size and enormous specific surface area that favour their interaction with the material to be conserved/restored. But the small size, coupled with their capacity to adsorb biomolecules and interact with biological receptors, can increase the transport to reach sub-cellular locations leading to potentially higher localized concentrations and toxicity. A lot of factors such as size, shape, surface coating and the fact that these particles are subject to fast alteration, make complicate the elucidation of the interaction mechanisms of these nanomaterials with the artefact material and the surrounding environment including the nano-bio interaction. In this context, in the frame of the EU H2020 NANORESTART project, innovative nano-enable formulations for the conservation and restoration of modern and contemporary artworks have been provided, following a Safe-by-Design (SbD) approach. The safety of the new formulations was investigated by applying both EU CLP self-classification approach for mixtures (ECHA, 2017) and experimental in vivo and in vitro ecotoxicological tests. In order to better understand the key interactions occurring between ENMs and the biological medium used for the tests, the colloidal characterization of the new formulations was performed by means of Dynamic Light Scattering (DLS) and Centrifugal Separation Analysis (CSA) techniques. Moreover, possible releases from outdoor conserved works of art were also investigated by immersion tests, simulating the exposure of the treated materials to the worst applicable environmental conditions (e.g. rain, humidity, temperature).

TU091

Considerations for Safe Innovation: The Case of Graphene

M.V. Park, RIVM / GZB; J.T. Quik, RIVM / DMG; E.A. Bleeker, W. Brand, RIVM / VSP; F.R. Cassee, RIVM / DMG; M. van Elk, RIVM / GZB; I. Gosens, RIVM / DMG; W.H. de Jong, RIVM / GZB; J.A. Meesters, RIVM / VSP; W. Peijnenburg, RIVM / Center for Safety of Substances and Products; R.J. van de Briel, RIVM / GZB; A.J. Sips, RIVM / VSP

Safe-by-design in chemistry may positively contribute to a circular economy by avoiding the introduction of hazardous substances. Specifically in nanotechnology the terms "Safe innovation" and "Safe(r)-by-design" are currently popular, referring to the goal of considering safety aspects already at an early stage in the innovation process of (nano)materials and nanoenabled products. We specifically look at the case of Graphene and investigate the possibilities of considering safety aspects during various stages of the innovation process. Based on this we suggest that in the first stages a clear description of the production processes and substances involved is needed in order to identify potential for exposure. After this the standardization of the production process becomes important in order to reach a more reliable exposure assessment and enable use of exposure reduction measures where needed. Furthermore we outline what information on graphene is already available for assessing potential human and environmental hazard, exposure, and risks. For example a first indication of the hazard of an (intended) product can be obtained by collecting information on a limited number of physicochemical properties of the intended graphene product: dimensions, shape and surface properties. In addition, we recommend further steps to be taken by various stakeholders to promote the safe production and safe use of graphene. We emphasize that a safe and time-efficient innovation process is only possible under the conditions of clear and timely communication between innovators, scientists, risk assessors and regulators.

TU092

Safer-by-Design framework for supporting Small and Medium Enterprises early in sustainable innovation for nanomedicine

M. Schmutz, C. Som, EMPA Technology & Society Lab

One of the hot topics in nanomedicine is the use of nanobiomaterials for drug delivery. On the one hand, nanobiomaterials have various expected advantages compared to its bulk material: 1) decreased doses, 2) possibility to cross biological barriers, 3) increased drug efficacy, 4) reduction of side effects, and 5) targeted drug delivery. On the other hand, the nanosize brings new challenges for risk assessment.nanomedicine is complex, and combines knowledge from different fields. It is at the junction among pharma, medtech, biotech, nanotech and chemical companies which are important economic and social player in Europe. In this context, the GoNanoBioMat project aims to facilitate SMEs in Europe in the decision making for developing and producing safer and sustainable polymeric nanobiomaterials for drug delivery. To do so, the consortium designed a Safer-by-Design framework for supporting the needs of SMEs in an early stage of innovation. The framework comprises sustainable material design considering the whole life cycle of polymeric nanobiomaterials, environmental and human health risk assessment. Difficulties in the nanomedicine field arise at different levels which are at the research, regulatory and manufacturing levels. Nanomedicine is still considered as a young field and needs further research to better understand the interactions of nanomaterials at the bio-interface and to find out which are the critical quality attributes (link between physico-chemical properties and toxicity, product safety, quality and purity). Furthermore, there are difficulties in reproducing environmental and human health experiments for assessing the related risks and having batch-to-batch uniformity. Finally, notifying bodies are behind development because of the uncertainties arising from this field. Therefore, it seemed important to include in the framework the following aspects: safe material's design, human health and environmental risks, manufacturing, storage and transport and the regulations related to the topic at hand. At the end of the project, the Safer-by-Design framework will be used as a structural backbone for creating nano-specific guidelines in nanomedicine. These guidelines aim to facilitate the communication among the different stakeholders in the value chain and with regulators for safe and sustainable innovation.

TU093

Review of the applicability of early-stage sustainability methods integrating toxicity and environmental assessments

C. Frenandez Dacosta, University of Utrecht / Copernicus Institute; P. Wassenaar, National Institute for Public Health and the Environment (RIVM); I. Dencic, Corbion; M.C. Zijp, RIVM / Centre for Sustainability Environment and Health; A. Morao, Corbion; L. Shen, University of Utrecht / Copernicus Institute; E. Heugens, RIVM / Centre for Safety of Substances and Products; L. Posthuma, RIVM / Centre for Sustainability, Environment and Health

The chemical industry strives for the development of bio-based alternatives for present fossil-based chemicals driven by the transition to a bio-based economy. Key in this transition is "safe and sustainable by design", which means safety and sustainability are taken into account at the earliest possible development stages. Many sustainability assessment methods are developed for this purpose. The aim of this study is to evaluate a selection of 12 early-stage methods (ESMs), their applicability, the relevance for bio-based chemicals and the coherence of their outcomes, using bio-based lactic acid as a retrospective case study. The selected methods contain at least one of the following themes: energy, climate change, eutrophication, land use, human toxicity and ecotoxicity. These six themes were considered as most relevant for the case of bio-based lactic acid. The selected early-stage methods point to the right hotspots concerning energy and climate change, which is promising for application during process design. In general, the selected ESMs define simple environmental and toxicity indicators that have lower data requirements and are faster to implement than full assessment methods. However, the results they provide have intrinsically a higher level of uncertainty. Besides, the ESMs existing in the literature do not meet important criteria for utility. They are often not clear in the definitions of the environmental and toxicity indicators neither transparent in background data sources and not up-to-date. Important limitations of the selected ESMs are 1) narrow life cycle scopes (excluding the environmental impacts of material and biomass feedstock production) and 2) omission of some environmental aspects relevant to bio-based materials and toxicity aspects in general. Within this study, we pinpoint limitations and positive aspects of several early-stage sustainability methods. Based on this exercise we identify and propose successful elements of existing methods to be included in a framework that supports the assessment of safety and sustainability in early development phase.

TU094

Liquid organic hydrogen carriers (LOHC) - comparative hazard assessment

M. Markiewicz, Technical University of Dresden / Sustainable Chemistry Group; Y. Zhang, Dresden University of Technology / UFT Centre for Environmental Research and Sustainable Technology Department Sustainable Chemistry; S. Stolte, University of Bremen / UFT Centre for Environmental Research and Sustainable Technology Department Sustainable Chemistry

Renewable energy stored in LOHC systems could replace fossil fuels yet their environmental impacts are largely unknown. This technology is still relatively new, and requires R&D efforts to optimise its performance to commercially attractive levels. This opens the possibility to proactively design the carriers for increased operational and environmental safety. A preliminary, comparative hazard assessment was performed using automotive diesel oil as a reference. The biodegradability and acute/subchronic (eco)toxicity using: enzymes (acetylcholine esterase), cell lines (IPC-81), bacteria (*Vibrio fischeri*), algae (*Raphidocelis subcapitata*), freshwater plants (*Lemna minor*) and invertebrates (*Daphnia magna)* were investigated. Test set included LOHC systems based on quinaldine, ethyl-, propyl- and butylcarbazole. For each LOHC system three forms of the carrier were assessed: H_2-lean, H_2-rich and partially hydrogenated. Low to moderate (eco)toxicity, comparable to automotive diesel oil, was observed for the quinaldine LOHC system. No effect occurred in aquatic tests for H_2-lean alkylcarbazoles due to unstable exposure. The H_2-rich forms were moderately cyto-/ecotoxic. High cytotoxicity was observed for partially hydrogenated alkylcarbazoles, with the effect increasing with the chain length. Alkylcarbazole LOHC systems were generally more toxic than diesel oil. None of the LOHC chemicals show appreciable biodegradation except quinaldine. Further biodegradability test under less stringent conditions are needed to investigate potential persistence. Additionally, hydrophobicity of H_2-lean and intermediate forms of alkylcarbazoles (log D 3.6-4.8) indicates that they might be bioaccumulative. Nonetheless, undeniable socioeconomic benefits come from the fact that LOHC energy systems can operate on renewable energies. Moreover, this LOHCs are more favourable in the terms of handling and transportation safety. The composition of LOHC chemicals is much better defined than it is in case of fossil fuels, which facilitates standardisation or quality control. This study also showed that many of the standard (eco)toxicity testing approaches are not well suited for LOHC systems showing moderate to high hydrophobicity as it is the case for diesel oil.

TU095

1-Octanol and 2-Butanone as biofuel candidates - Using "Green Toxicolgoy" for abiofuel development

H. Hollert, RWTH Aachen University / Institute for Environmental Research; S. Heger, Institute for Environmental Research RWTH Aachen University; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics; M. Du, Institute for Environmental Research, RWTH Aachen / Department of Ecosystem Analysis

The development and production of biofuels, and thus also the risk of a release in the environment, is rising. This implies an increased risk for a release into the environment. Aquatic systems are particularly considered to be vulnerable to fuel contaminations. (Eco)toxicological bioassays can be applied as screening tools during the early developmental phase of biofuels for obtaining information on potential hazardous properties. These bioanalytical tools can assess adverse effects of many substances on various organisms and endpoints and thus provide a rapid and reliable screening of potential biofuels for identification of potentially harmful biofuel candidates at a very early stage of product development. This testing strategy is part of a framework proposed by the new discipline of "Green Toxicology" which strives to move safety considerations of newly developed chemicals to the earliest possible moment of its lifecycle Aquatic toxicity is considered as one important ecological endpoint relevant for biofuels. Therefore, the investigation of aquatic toxicity of promising biofuel candidates focused on acute immobilisation of *Daphnia magna* and acute embryotoxicity and teratogenicity of *Danio rerio*. Moreover, genotoxicty of the biofuel candidates was also investigated in the Micronucleus assay with V-79 cells to assess the potential effects on human health. This study focuses on the investigation of two biomass-derived fuel candidates: 2-Butanone and 1-Octanol. Both substances are considered very promising alternative fuels. The toxicity testing revealed a very low acute and developmental toxicity for 2-Butanone compared to 1-Octanol. 2-Butanone induced acute toxicity and genotytixcity in concentrations >2 g/L and even teratogenic effects were found at 822 mg/L. 1-Octanol did induce effects in concentrations between 7-15 mg/L. The overall results indicate that 2-Butanone is not harmful for aquatic organisms and should be focused in the further biofuel development. For a further integration of this screening approach in the biofuel development, more biofuel candidates can be investigated and, thus, more detailed information on their potential toxicity can support the development and production of green biofuels. This work was performed as part of the Research Cluster "Tailor-made fuels from biomass" funded by the Excellence Initiative by the German federal and state governments to promote science and research at German universities.

TU096

Investigation of the toxic effects of new mixtures of deep eutectic solvents (DES) on the environment and human health

G. Mengotti, Heriot Watt University; A. Tagliati, Heriot Watt University / Institute of Life and Earth Sciences; E. Tagliavini, Heriot-Watt University; C. Samorì, University of Bologna; H. Johnston, D. Brown, Heriot-Watt University; T.F. Fernandes, Heriot-Watt University / Institute of Life and Earth Sciences

The development of environmentally benign and green synthetic protocols, due to the growing concern over the environment, has brought to the necessity to find greener, readily biodegradable and low cost solvents. This new concept of green chemistry has recently led to the synthesis of *Ionic Liquids* (ILs), from which have evolved in few years the deep eutectic solvents (DESs).[1] These compounds are obtained mixing two components: a quaternary ammonium salt (e.g. ChCl) with different hydrogen bond donors, in such a ratio that the resulting substance has a significantly lower melting point than that of each individual component. DESs have proved to be environmentally sustainable and alternative to the conventional organic solvents in synthetic chemistry, able to increase efficiency of organic transformations. Those solvents have attracted widespread academic and industrial interests, and have found almost unanimous worldwide approval. Cosmetics has become in the last years one of the most profitable industries. The majority of cosmetics are composed of chemicals, generally as emulsions. Given the ease of synthetize DESs, along with their low cost, it is thought to a possible use of them in the formulation of cosmetic and beauty products. Some of these DESs contain nitrogen (N), which can be used as fertilizer in the growth of the crops. Another field of interest could be the agriculture as well: some of these solvents can be prepared as *gels*, meaning a gradual release of the substance in the ground. Toxicological studies on ChCl+Glycerol and ChCl+Levulinic Acid (never studied before) on algal species of the genus *Symbiodinium* and on skin *in vitro* cells have been carried out in order to extend the limited knowledge about toxicity at environmental and human level, as well as the biodegradation pathway of this family of solvents. Preliminary results show extremely low toxicity on *Symbiodinium clade B*, known to be highly sensitive to environmental stress, for all the tested mixtures. Algae growth and Reactive Oxygen Species (ROS) production, a general indicator of stress, it is indeed not affected by all the tested compounds in the order of gL^{-1}. Results from the present study indicate an expected safer

behaviour of those solvents with respect to others marketed known. [1]Khandelwal, Tailor, Kumar, 2016. Deep eutectic solvents (DESs) as eco-friendly and sustainable solvent/catalyst systems in organic transformations. Journal of Molecular Liquids 215 345–386

New frontiers in Life Cycle Inventory data collection and modelling (P)

TU097

Predicting environmentally beneficial production pathways for chemicals with neural networks

J. Kleinekorte, RWTH Aachen University, Institute of Technical Thermodynamics / Institute of Technical Thermodynamics; M.R. Tillmanns, A. Sternberg, RWTH Aachen University / Institute of Technical Thermodynamics; A. Bardow, RWTH Aachen University

Life Cycle Assessment (LCA) has gained wide acceptance as methodology to evaluate the environmental impact of chemical processes. However, LCA requires detailed data on mass and energy balances. Data is usually limited for processes in stages of early development. For these cases, predictive LCA approaches are required. Current predictive LCA approaches employ solely molecular descriptors to estimate the environmental impacts of products. Thus, the choice between different production pathways towards the same molecule cannot be resolved. Therefore, we propose a neural network-based approach that uses both molecular and process descriptors. The resulting neural network is able to distinguish between various production pathways for the same product while still employing only data available at early stages of development such as stoichiometry. We estimate 5 impact categories including, e.g., cumulative energy demand (CED) or climate change (CC). The novel approach is compared to a neural network trained with molecular descriptors only. The results show that integrating process descriptors increases the coefficient of determination from 0.37 to 0.65 and from 0.39 to 0.65 for CED and CC, respectively. The route-specific prediction is illustrated for methanol production from CO_2 versus natural gas. It is shown that neural network models can serve as an initial screening tool for identifying environmentally beneficial new production pathways.

TU098

A Study on the development of Food LCI DB and PCR for estimating environmental footprint in South Korea

S. Cho, SMaRT-ECO / Sustainable Business Div.; S. An, S. Seol, I. Kim, SMaRT Eco Corporation

As a result of the Paris Climate Convention adopted in December 2015, 195 countries in the world were required to implement greenhouse gas reduction, and Korea also proposed a 37% reduction target compared to BAU. In accordance with the environmental regulation trends of developed countries, Korea is also not free from environmental regulations at the time of preparation for active response. Under the Single Market of the EU, product environmental regulations starting from automobiles in 2000 expanded to include food in 2020, requiring the disclosure of high-quality environmental information on foods. As a result, the development of a high-quality environmental information database is accelerated, and the EU has created an ILCD Data Network to induce DB registrations in each country. However, Korea's agricultural and livestock LCA DB does not meet the ISO requirements, it is time to revise. The purpose of this study is to develop the LCA database for the estimation of the environmental footprint (PEF) of major domestic food exports to Europe and to use the common protocol and food - specific guidelines (PCR) to estimate environmental footprint , And aims to obtain EPD certification of food. To do this, we benchmarked the protocol and PCR for the Korean conditions by examining the cases of the calculation guidelines of the developed countries. In the future, it is meaningful to construct a database that can be used as basic data for obtaining PEF certification for foods exported to Europe and overseas.

TU099

Transition from ILCD To Environmental Footprint: changes in the database structure, format, nomenclature, methods and other adaptations.

S. Fazio, EC-JRC; O. Kusche, Karlsruhe Institute of Technology; E. Diaconu, JRC European Commission; R. Pant, European Commission / Institute for Environment and Sustainability; S. Sala, European Commission - Joint Research Centre / Bioeconomy unit

In 2013 a Communication from the Commission to the European Parliament (COM/2013/0196) established the Environmental Footprint (EF) scheme. The common methods to measure and communicate the life cycle environmental performances for EF have been defined in a specific EU recommendation (2013/179/EU). Within this framework, the International reference Life Cycle Data system (ILCD) format, developed since 2007, along with a simplified set of compliance rules called “ILCD Entry Level Requirements” has been recommended as a baseline for data development in the EF scheme. However, in the development of the EF methodology, nomenclature and recommended Life Cycle Impact Assessment (LCIA) Methods have been partly changed and adapted to fulfill the scope. Beyond that, the reference data contained in the ILCD package was found to contain some format, syntax and conceptual errors which were inherited from several data providers over time. Therefore a new database has been developed. Errors have been fixed, new files have been developed, and redundant or obsolete files have been deleted. The content of this presentation represents a synthesis, recalling general considerations or decisions, that have been applied for specific impact categories, and technical details with respect to each impact category, documenting specific choices made when implementing the characterization factors as well as problems/solutions encountered in the course of this implementation. Furthermore, a list of changes made from the ILCD to the EF package, beyond the LCIA methods recommended (i.e. Elementary Flows, Flow Properties, Unit Groups) and complementary objects defining the compliance (i.e. stylesheets, schemas) are described, in a change log file available through the EPLCA website. Among the above mentioned items the overall changes occurred in the ILCD-EF transition phase can be resumed as following: - 1242 obsolete or wrong elementary flows have been deleted /mapped - 560 new elementary flows have been created - Around 55.000 characterisation factors are different (this is mainly due to the introduction of new methods, and regionalization of some of them) - 37 duplicated flows have been eliminated - 275 wrongly categorized flows have been assigned to the proper category - 218 wrong, duplicated or useless flow properties have been deleted and mapped to the remaining ones - 35 unit groups have been deleted, one new has been created.

TU100

New tools for Environmental Footprint data checking and sharing: Soda4LCA, ILCD validator and Registry for the node management

S. Fazio, EC-JRC; E. Diaconu, JRC European Commission; O. Kusche, OkworX; R. Pant, European Commission / Institute for Environment and Sustainability; S. Sala, European Commission - Joint Research Centre / Bioeconomy unit

Several tools for Life Cycle Inventory data development, validation, sharing and registration to the Life Cycle Data Network (LCDN) have been released by the EC since 2007, and improved after the official launch of the LCDN (2014). All those tools were originally meant for the International Life Cycle Data (ILCD) scheme, Since 2013 after a specific EC Communication (COM/2013/0196) the Environmental Footprint (EF) scheme has been developed and led to significant changes in the structure of the ILCD/LCDN. During the development of EF-compliant data, the tools had to be adapted and improved to fulfill the new requirements. Particularly the following tools and software have been changed: - ILCD validation tool: software for the compliance assessment of datasets (format syntax, archive structure, nomenclature, links and orphaned items, categorization, etc.) - soda4LCA: software for distributing data based on the ILCD data format, with search and management functions, including the data registration in the LCDN - LCDN Registry: online registration facility that can deal with data from different nodes running on soda4LCA, and meant to make available only fully compliant data (while the nodes can host also intermediate data) The changes that have been applied can be summarized as follows: - ILCD validation tool: additional validation profiles added for EF scheme. Checks against different parameters for Elementary Flows, location IDs, new LCIA methods, Flow Properties, Unit Groups and schemas, according to the changes made in the DB structure - soda4LCA: new access profiles are available for data stocks. The developer can now select entire data stocks and restrict the access only to authorized users. The entire data stock can be now downloaded directly, while before it was possible only at the single dataset level. The registration form includes a statement for the use of data within the EF framework. Declaration of compliance in the registration phase and possibility of multiple registration in more than one registry at once (a dataset can be both ILCD and EF compliant and therefore registered in two registries with different compliances declared). - LCDN registry: a dedicated registry has been developed for EF, with new functions. The compliance scheme is now visible (before was implicit since only ILCD was possible), possibility to register entire batches of data at once (before each single dataset had to be registered manually). Search interface improved.

TU101

Improving the consistency and the accuracy of water inventories of chemical sites in PlasticsEurope LCIs in the perspective of the applicability of the impact assessment method AWARE

M. Baitz, thinkstep / LCA; G. Castelan, PlasticsEurope / LCA

The ISO 14046 standard has clarified the definitions of water use and of water consumption which is the part of water withdrawn from a drainage basin not returned back to the same drainage basin. These only definitions have enabled PlasticsEurope to fix a number of important inconstancies in its eco-profile program, where use and consumption were sometimes confused. Further to this short term action, in perspective of enabling the application of the latest consensual water assessment method AWARE to the water consumption, PlasticsEurope and thinkstep have collaborated to improve the consistency of the data collection phase of water flows for the various projects of the program. For the purpose of good water management in a chemical plant, plant managers need to have a good knowledge of all the water flow inputs, their origin (lake, river, public supply, underground…), their treatment, in what equipment or process they are employed like for example in a cooling towers or being injected in the chemical process like in steam cracking. They must know the post use-treatment and where all the outputs end (back to the river, evaporated, in the public sewage network, in the product…). For the purpose of consistency of the Life Cycle inventory phase, it is then very important to report these collected operational flows in the ILCD input and output flows the right and same way whoever the LCA practitioner is. This will be the basis for the calculation of the consumptive water (output minus input within the same drainage basin) and the application of AWARE. The presentation elaborates on the various operational use of water in a chemical plant and the link to the life cycle inventory phase and ILCD flow names. This has been added to the PlasticsEurope methodology for calculating eco-profiles. It is expandable or adaptable to all kind of industrial sites. A similar work would need to be conducted on all datasets so that the consistency of water inventory gets improved in both foreground and background data, enabling a better comparability of water footprint in cradle to gate LCA. The presentation aims to attack LC(I)A water experts and scientists as well as people applying the water methods in practice to exchange on challenges, relevancy of aspects and to align on a continuous improvement of water data, regionalization efforts and method improvement in the future, to inspire broad application.

TU102

Methodological improvements by dynamic approaches for the life cycle assessments of buildings

K. NEGISHI, CSTB; L. Barna, INSA Toulouse / LISBP; Y. Pigné, Universite du Havre; T. Navarrete-Gutierrez, LIST; N. SCHIOPU, A. Lebert, CSTB; T. Gibon, Luxembourg Institute of Science and Technology (LIST) / Environmental Research and Innovation; E. Popovici, LIST; E. Benetto, Luxembourg Institute of Science and Technology (LIST) / Environmental Research and Innovation

Nowadays in France, environmental and energy rules for the construction sector are based on environmental performances of products assessed through LCA methodology. However, the actual practice of LCA is lacking of temporal dimension whereas the temporal evolution during the long lifetime span of buildings has non-negligible impact on overall LCA results. A new framework of LCA method was recently proposed by L.Barna et al (2016) and A.Shimako et al (2017) with a particular attention to the process and supply chains dynamics (web tool DyPLCA, http://dyplca.pigne.org/), aiming at calculating time dependent environmental interventions and the related impacts of toxicity and climate change. The aim of this study is to investigate the environmental performances over a large time span of two low-energy single houses, one on concrete and one on timber. The time dimension was integrated on both LCA steps (LCI and LCIA) using the framework cited above. The implementation of dynamic LCA took several steps. Buildings life cycles were first modeled in SimaPro 8.02 with ecoinvent 2.2 to calculate the conventional LCI. The calculated technological and environmental interventions matrix was then used with DyPLCA web tool for temporal LCI calculation. The temporal characterization of the product system considered two parts. Temporal characteristics of the foreground system were related to the building construction, materials replacement and renovation activities during the building's life time of 100 years. The temporal characteristics of background processes were previously integrated in a dedicated database and used with DyPLCA tool. The temporal LCI, i.e. environmental interventions distributed in time, was then used for climate change impact calculation in function of time. Two indicators were calculated in function of time: mean temperature change and radiative forcing. The new method allows considering fossil and biogenic carbon for climate change proposes without clinging on to fixed characterization factors and time horizon and without compensation as done in conventional method. The use of dynamic LCA framework in our case study is justified by a deeper understanding and a more consistent analysis of environmental impacts of buildings. However, simulation time and memory usage for dynamic LCI calculation can be a principal limitation for the practice of dynamic LCA.

TU103

Carbon footprint from Brazilian soybeans based on spatially-explicit life cycle inventories, including land use change

N. Escobar, University of Bonn / Institute for Food and Resource Economics ILR; J. Godar, Stockholm Environmental Institute

That location matters when it comes to quantifying environmental impacts of agricultural products is proven by the increasing number of case studies within the LCA literature. Authors tackle the influence of spatial variability by capturing differences in agricultural practices, transport options and industrial processing sites in the life cycle inventory (LCI). This information is, however, incomplete when quantifying impacts of agricultural commodities that are produced in large amounts and traded worldwide, e.g. soybean. Despite the efforts from the Input Output (IO) community to trace these impacts along global supply chains, this usually requires the use of aggregated resource consumption and emission data for environmental extensions, which provides little detail on the technological and logistic factors contributing to overall ecological footprints. From the LCA perspective, total nation's supply is made of thousands of individual life cycles from farm to gate, for which LCI data is not frequently available. The Trase platform allows for real-world pathways of several internationally traded commodities to be annually mapped, from producing regions to destination countries. In view of its great potential for LCA purposes, our goal is to implement a carbon footprint module able to deliver results on CO_2-eq. emissions associated, on the one hand, to annual production of soybean supplied from Brazil for the period 2010-2015; on the other hand, to every individual supply chain embodied in the whole supply of seed, oil, and cake to the international market. These include the following life cycle stages: land use change (LUC), soybean farming, domestic transport, export, and crushing, dealing with allocation challenges. In this way, our approach represents the convergence between top-down Multi-Regional IO analysis and bottom-up Attributional LCA. Preliminary results highlight the relative contribution of the sub-stages that occur further down the supply chain, mainly LUC, for which considering sub-national scales is crucial in the quantification of climate change impacts. Outcomes support the argument that importing countries of soybean-based commodities should take responsibility on deforestation and associated carbon emissions, provided that spatial explicit data is available. This transparency tool is meant to provide science-based evidence to the ongoing debate on global responsibility, while assisting supply chain management and governance decisions.

TU104

Carbon Footprint Projections for Japan Using Computable General Equilibrium

Y. Ichisugi, Tokyo City University; T. Masui, National Institute for Environmental Studies; N. Itsubo, Tokyo City University

In 2015, Science Based Targets (SBT) has been paid attention to the world. The targets adopted by companies reducing greenhouse gas (GHG) emissions to keep global temperature below 2 degrees increase from that of preindustrial revolution. Approximately 300 companies in the world declared to follow their targets. The target to the mitigation would be based on the calculated results of Integrated Assessment Models (IAM) such as Asia-Pacific Integrated Model (AIM), Integrated Model to Assess the Global Environmental (IMAGE). However, these results usually don't consider the entire supply chain, because of the differences of the aims of application. In contrast, Life Cycle Assessment (LCA) considers the entire supply chain. On the other hand, most LCA studies use current environmental data and normally don't include the dynamism of system. Therefore, this study aimed at the development of a dynamic evaluation of environmental impact method based on life cycle thinking to use both advantages of IAM and LCA. Currently, we try to develop the database using AIM developed by the National Institute for Environmental Studies (NIES). That model uses Computable General Equilibrium (CGE) which can estimate economic efficiency in the future, based on price mechanism in the market. In this study, we collect fundamental data using LCA database and estimate GHG emissions in the future considering the supply chain among industrial sectors. We estimated GHG emissions in 2005 as a tentative result in Japan. The total emission is approximately 1.13E+08t-CO_2eq. We confirmed the validity compared with the existing report published by the ministry of the environmentin Japan. In the future, we will estimate environmental impact projection considering the scenario like Shared Socioeconomic Pathways (SSP).

TU105

Network LCA as a tool to enhance data collection and usage in a value chain

C.L. Hohenthal, VTT Technical Research Centre of Finland; M. Myllysilta, S. Majaniemi, VTT Technical Research Centre of Finland Ltd

Keywords: LCA, data collection, value chain Life cycle assessment as defined by the ISO (14040) consists of four phases. First, the goal and scope are defined, after that the inventory analysis is performed followed by the life cycle impact assessment. At the end, the results are interpreted. The inventory analysis includes the data collection. There are many challenges related to inventory analysis. Firstly, it is seen as the most time consuming phase of every Life cycle assessment study. This is because the data are collected from various sources and the sources might be from different organization than the commissioner of the study. Secondly, the data provider might hold their data confidential. This is because the recipes might be secret, i.e. the raw materials and the amounts of raw materials and/or the amount of energy consumed in the production process reveals the cost structure of the product. Thirdly, the data provider typically gets no benefit from delivering data and putting a lot of effort to collect and get together the data. This may decrease the motivation to deliver data. The fourth aspect is the unwillingness of data providers to reveal their performance indicators to competitors, if considered that their environmental impacts are bigger than those of the competitors. The idea of network LCA is to tackle all the above mentioned four challenges. The main impact of network-LCA is to produce from the confidential source data of a company network level results, e.g. carbon footprint, which may be delivered to all network members openly. At the same time, all the network members can perform a local LCA computation to study their own local footprints. In other words, network members can independently run test and investigate the impacts of the changes e.g. on material choices or manufacturing methods both locally and at the network level. This feature is also beneficial for policy planners who want to see the big systemic picture and formulate their action plan based on the observed data. The data needed for the life cycle assessment can be provided via a web form, which has certain built-in features to speed up the data gathering process. The web form supports predefined parameter lists and it is also possible to add new parameters to the existing lists. Also the compatibility with impact categories such as impact on greenhouse gas emissions or resource depletion is hereby ensured.

TU106

Developing guidelines for elementary flow nomenclature

A. Edelen, ORISE; W. Ingwersen, US EPA

In general, a *flow* in life cycle inventory data refers to an input or output to a process. Flows may be of two broad types: elementary flows or intermediate (known as "technosphere") flows according to ISO 14044 (ISO 14044 2006). *Elementary flows* may be defined as materials, energy or space that are used directly from the environment or released directly back into the environment. Life cycle assesment (LCA) data providers are currently not using a common list or system of elementary flows. An early activity within the UNEP-SETAC Life Cycle Initiative was the creation of a recommended list of flow exchanges by the Data Availability and Data Quality Workgroup (de Beaufort-Langeveld et al. 2003). Elementary flows in all life cycle inventory and life cycle impact assessment sources used in a model must correspond, or match, in order to build a functional LCA model. Edelen et. al. 2017 formulated recommendations on formating and management based on a critical review of elementary flows from eleven LCA sources. These recommendations have been used to categorize flow information into three components and flow metadata into six components. These structured components of flows allows for systematic analysis and structuring of flow components through a knowledge organizational structure (KOS). The ISO 14048 standard was used to structure the different flow and metadata components as exculsive, inclusive or user-defined nomenclatures. The KOS is maintained in a user friendly, publically accessible interface through the US EPA terminology services. This research presentation will focus on describing the benefits of the KOS approach and the tools used to develop the nomenclature system and provide an example application of the KOS to current elementary flow nomenclature. References [1] de Beaufort-Langeveld A, Bretz R, Hischier R, Huijbregts M, Jean P, Tanner T, van Hoof G (2003) Code of life-cycle inventory practice. SETAC Press, Pensacola, FL [2] Edelen A, Ingwersen W, Rodriguez C, Alvarenga R, de Almeida AR, Wernet G (2017) Critical review of elementary flows in LCA data. INT J LIFE CYCLE ASS. http://dx.doi.org/10.1007/s11367-017-1354-3 [3] ISO 14044 (2006) ISO 14044: Environmental management--Life cycle assessment--Requirements and guidelines. International Organization for Standardization, Switzerland

TU107

Building a Life Cycle Inventory of stormwater pollutant fluxes: model evaluation for a separate residential urban catchment

E. Risch, IRSTEA Montpellier / UMR ITAP; P. Roux, Irstea / ITAP ELSA-PACT; R.K. Rosenbaum, National Research Institute of Science and Technology for Environment and Agriculture - Irstea / UMR ITAP; C. Sinfort, ITAP, Irstea, Montpellier SupAgro, Univ Montpellier / ELSA Research group and ELSA-PACT Industrial Chair

Current life cycle assessment studies of urban wastewater systems (UWS) substantially underestimate impacts of these systems to receiving waters by not including stormwater pollution generated from the impervious surfaces of urbanized catchments. To this date, UWS are typically modelled with average discharges of treated effluents in dry conditions. In recent work, untreated stormwater discharges were shown to be significant on the freshwater ecotoxicity impact at year and event scales. Stormwater pollution typically shows a high spatio-temporal variability owing to (i) a variety of anthropogenic activities/sources within the urban catchment and (ii) rainfall specificities of local climates. The links between urban land uses, associated activities and stormwater pollution are missing in existing LCA methodology and warrant further developments. In order to address this issue we propose to implement a fate model for pollutant emissions from relevant urban sources within the life cycle inventory (LCI) of an urban catchment. The main objective of the proposed framework is to provide site-dependent LCI of stormwater pollutant fluxes for residential urban catchments with separate sewer networks. Major urban sources contributing significantly to stormwater pollution are defined and linked to the urban structure. The model hierarchy is built on four levels from micro-scale (elementary urban surfaces) to meso-scale (city). Urban sources within the catchment contribute to stormwater pollution by emitting pollutants following either (i) a direct deposition route to urban surfaces (e.g. brake wear, metal roof corrosion) or (ii) an atmospheric emission followed by a partial deposition (e.g. diesel exhaust gases from vehicles). The resulting build-up of pollutants on elementary urban surfaces was modelled for each primary source. During storm events the wash-off and transport of available pollutants via runoff were calculated for different urban surfaces. Stormwater fluxes were aggregated at wider scales (block, neighborhood and city) using a semi-distributed dynamic rainfall-runoff model SWMM. The proposed framework was evaluated on a virtual urban catchment under two contrasted climates with different rainfall distribution. Pollutant fluxes from urban surfaces were analysed and compared for each climate over a one year period. Stormwater LCI results showed a site-dependency under a given climate, and a minor sensitivity to rainfall distribution.

The added value of using invertebrate species in ecotoxicology: new insights for environmental risk assessment (P)

TU108

Tissue specific 32P accumulation and consequent biological effects in bivalve molluscs

E. Vernon, The University of Plymouth / School of Biological & Marine Sciences; J.T. Smith, University of Portsmouth / School of Earth and Environmental Sciences; A.N. Jha, Plymouth University / Biological Sciences

1. Introduction The aquatic environment is the final recipient of anthropogenic contaminants, including radionuclides. Despite the growing concern over presence of radionuclides in the aquatic environment, there has been only limited studies to determine potential biological responses in aquatic invertebrates following exposure to environmentally realistic levels of radiation doses. This integrated study determines and compares tissue specific bioaccumulation, doses delivered and induced biological damage in two species of mussels, the freshwater species *Dreissena polymorpha* (DP) and marine *Mytilus galloprovincialis* (MG), following exposures to an important radionuclide, phosphorus-32 (^{32}P). **2. Materials and methods** The study involved 10 day exposures of mussels to ^{32}P of varying dose rates (i.e. 0.10, 1.0 and 10.0 mGy/d) taking into account a current no-effect screening value of 0.24 mGy/d (European Commission). The first set of studies determined ^{32}P accumulation in specific mussel tissues (i.e. adductor muscle, digestive gland, mantle, gills and 'other'), internal mussel water (water inside the mantle cavity), shell and faecal matter using scintillation techniques. From this bioaccumulation study, we were able to highlight key tissues of interest; the digestive gland for example, received the greatest proportion of ^{32}P independent of mussel species. In the next set of studies, a suite of biological responses or biomarkers were investigated in digestive gland and gill cells. This included the induction of DNA damage (Comet assay) and repair response (Gamma-H2AX), the induction of micronuclei (MN) and the expression of key stress related genes (i.e. SOD, CAT, GST, HSP70/90). **3. Results and discussion** Our findings highlighted DNA damage and MN induction at radiation doses as low as in 0.1 mGy/d in digestive gland (MN = also in gill) in both species, below the screening benchmark. Furthermore, compared to fresh water (DP), marine bivalve (MG*)* displayed greater DNA damage (both tissues) across all ^{32}P treatments. This study highlights that (a) radionuclide activity concentrations in a biological system cannot be predicted by surrounding environmental media (b) with regards to bioaccumulation of waterborne contaminants, whole body measurements may mask the tissue specific nature of radionuclide uptake, and (c) the importance of adopting a multi species, multi biomarker approach when assessing the possible effect of contaminants in the aquatic environment.

TU109

Endocrine disruption in Mytilus galloprovincialis: Is ethinylestradiol a vitellogenin inducer?

L. Fernández González, P. Sanchez Marin, University of Vigo / Ecology and Animal Biology; G. Grueiro Noche, S. Muniategui Lorenzo, University of A Coruña / Analytical Chemistry Department; A. P Diz, University of Vigo / Biochemistry Genetics and Immunology; r. beiras, University of Vigo / Toralla marine sciences station (ecimat)

Vitellogenin (Vtg), the egg-yolk precrusor in female oviparous animals, is a common biomarker of estrogenicity widely used as an indicator of endocrine disruption in aquatic environments. Nevertheless, in the case of mollusks, it is still unclear if the synthesis of Vtg is regulated by steroid hormones as in the case of vertebrates. In the case of the synthetic hormone ethinylestradiol (EE2) the results of the studies are inconclusive. The aim of this work is to verify whether the synthetic estrogen 17α-ethinylestradiol (EE2) induces the production of Vtg in *Mytilus galloprovincialis* at two exposure times and different nutritional regimes. For that, we used a shotgun label-free proteomics approach by high resolution LC-MS/MS to identify and quantify Vtg in mussels gonads. In this way, we can verify if the energetic balance is a key confusing factor in Vtg production. The relationship between the maturity state of mussels and their Vtg levels was studied as another possible confusing factor. Mussels from uncontaminated area in Galicia (Spain) were collected in autumm/winter, corresponding with early gametogenesis stage . Mussels were exposed during 4 and 24 days to 100 ng L^{-1} of EE2 to assess whether Vtg synthesis was induced by EE2. During exposure, mussels were fed three times per week with two different regimes: a low regime (equivalent to 0.29 % of mussel dry weigth per day), or with a high regime (equivalent to 5.55 % of mussel dry weigth per day), representing negative and positive energy balance respectively. For the low feeding regime, shotgun proteomics identified an detected Vtg only in female gonads. The results showed an increase in Vtg levels in mussels exposed for 4 days to 100 ng/L EE2 compared to the solvent control, althought this increase was not statistically significant. In mussels exposed for 24 days to 100 ng/L EE2, Vtg levels were not higher in the exposed organisms than organisms exposed to a solvent control. These results suggest that EE2 does not induce Vtg in *M. galloprovincialis*. However, it is possible that Vtg synthesis was impaired by the fact that organisms were in negative energy balance. The results of the experiments done at a higher feeding regime (currently being analysed) will confirm this result. A significant correlation was found between Vtg levels and the maturation state of female mussels, indicating that maturation state is a confounding factor for the application of Vtg levels in endocrine disruption studies.

TU110

Integrating natural processes in environmental hazard assessments of the oil sands

D. Nunes Cardoso, CESAM, University of Aveiro / department of Biology & CESAM; J. Pestana, CESAM & University of Aveiro / Biology; S. Goncalves, Department of Biology & CESAM - University of Aveiro / Department of Biology and CESAM; A.M. Soares, University of Aveiro / department of Biology & CESAM; F.J. Wrona, University of Calgary / Department of Biological Sciences; S. Loureiro, Universidade de Aveiro / Biology

The Athabasca oil sands deposits in northern Alberta, Canada are a naturally occurring mixture of bitumen, sand, clay and other minerals. Bitumen, which is a heavy and extremely viscous oil, is mined and then subsequently refined to produce gasoline, diesel and other hydrocarbon-based products. Moreover, the naturally occurring Athabasca Oil sands deposits are a source of both physical and chemical stressors to regional rivers that flow through the deposit. Physical stress on aquatic biota from natural bitumen results from hillslope erosion processes and slumping of material into the rivers, while chemical stress arises from bitumen-derived contaminants entering the waters. To fully understand the ecological and cumulative effects of oil sands mining activities on aquatic ecosystem water quality and associated biological structure and function, there is a need to evaluate the effects of naturally occurring bitumen in the aquatic environment. The main objective of this study was to evaluate the possible ecotoxicological effects associated with the slumping of river bank material (i.e. oil sands deposit that naturally enters the river systems through fluvial geomorphological processes). A series of inter-related laboratory ecotoxicological assays were conducted using benthic and pelagic aquatic invertebrates exposed to oil sands material collected from four different sources in regional rivers (SP, ATB, STB and ELLs). All ecotoxicological results were complemented with the chemical analysis of metals, naphthenic acids (NAs) and polycyclic aromatic hydrocarbons (PAHs) to understand the possible effects that this material will induce when in contact with aquatic systems. All tested organisms responded negatively to the presence of oil sands material through either exposure to contaminated liquid media or through sediment contamination with solid oil sands material. A pattern of toxicity was also observed, where the SP source material was the less toxic and ELLs material being more toxic. These results corresponded with the chemical analysis which showed the ELLs sample having high levels of PAHs and NAs. In summary, tests revealed that oil sands material affected model organisms under laboratory exposures, especially in samples with more NAs and PAHs content.

TU111

Genomic DNA methylation level : a stress molecular marker in the species Gammarus fossarum ?

P. Cribiu, ENTPE, IRSTEA LYON; a. chaumot, O. Geffard, Irstea / UR MALY Laboratoire Ecotoxicologie; J. Ravanat, CEA Grenoble; T. Bastide, ENTPE / UMR 5023 LEHNA; A. Devaux, S. BONY, INRA - CNRS / UMR LEHNA USC INRA IGH ENTPE

Genotoxic evaluation has been developing for a couple decade among ecotoxicological assessment approaches in the aquatic field. It offers some prospect for understanding delayed effects on the offspring and the population dynamics (provided genetic mutations affect gametic genome). However, the modification of the DNA sequence by genetic mutation (as a result of primary DNA damage) is not the only impact of toxic substances on the genome. For example, epigenetic effects, defined as hereditary effects on the DNA function, may add up to mediated effects by genetics way. Among these marks, DNA methylation is extremely studied by scientists. As such, it is important to examine epigenetic changes in the context of population level perspective. This provides a complementary approach to effects on the primary structure of the genome for taking into account the time interval between the exposure to environmental contaminants and their effects. This issue is a real challenge in ecotoxicology. Moreover, epigenetic allows improving the understanding of changes of life history traits (reproduction, growth and development). These parameters are suitable for assessing the toxicity and are known to be regulated by epigenetic. Finally, epigenetic could also help better understand the variability of effects related to experimental conditions between the studies. Therefore, epigenetic marks have an innovative nature for the evaluation of environmental risks. In this regard, we have investigated the measurement of genomic DNA methylation level as a possible stress biomarker in the ecologically relevant species *Gammarus fossarum*. First, the basal level was explored by studying the difference of overall DNA methylation between male, female and juvenile. Then, we evaluated the effects of natural factors on genomic DNA methylation level as starvation and exposed to different temperatures (8, 12 and 18°C) for 7 days and 1 month. To know the variability of DNA methylation in populations of *Gammarus* fossarum, three French populations from the same genetic strain, were studied. Lastly, the epigenetic biomarker response to a chemical stress was evaluated in a field experience. We encaged gammarids stemming from a reference unpolluted station in sited impacted by various human activities.

TU112

iNVERTOX: Characterising individual metabolomic variability of the freshwater invertebrate, Gammarus pulex

T.H. Miller, Kings College London / Analytical and Environmental Sciences; J. MacRae, The Francis Crick Institute / Metabolomics; N. Bury, University of Suffolk / Division of Diabetes and Nutritional Sciences; S. Owen, AstraZeneca / Safety Health Environment; L. Barron, Kings College London / Analytical and Environmental Science

The (pseudo)persistence of emerging contaminants and other organic micropollutants in the environment represents a risk for the organisms that are exposed to them. 'Omics technologies are providing a powerful tool within environmental toxicology to better understand the effects of these exposure scenarios. However, metabolomics is an emerging field and to interpret the use of metabolite data for the understanding of toxicological responses is challenging. We need to know what is 'normal'. The variability in individual metabolomes for a species, or a "background metabolome" should be established to determine possible confounding factors such as age, sex and moulting (among others) that may influence data interpretation. Thus, we have characterised the effect of these factors on the metabolic variability in the freshwater invertebrate, *G. pulex*. Herein, an analytical method is presented for the extraction and non-target analysis[1] of the metabolome in *G. pulex*. Briefly, a dual phase liquid extraction was used followed by HILIC-HRMS to enable detection and annotation of metabolic features extracted from individual animals. Animals collected from the field were analysed immediately and compared to animals that were extracted after a fixed period of acclimatisation to laboratory conditions. The results indicated that sex, moulting stage and acclimatisation period affected the metabolic variability and factors that are likely to influence metabolomic analyses should be investigated to aid understanding of pathways involved in effect-based studies. Furthermore, it may be prudent to pre-select animals based on these factors to reduce inherent variability in the data. Overall, the characterisation of metabolic variance for invertebrates along with the use of metabolomics shows a very powerful approach for understanding adverse effects that may be associated with environmental contaminants.

References. 1) Zhang, T., et al. (2012). *Analytical chemistry*, 84(4), 1994-2001.

Keywords: metabolomics; invertebrates; pharmaceuticals; modelling

TU113

Ecotoxicological effects of the insecticide Imidacloprid on amphipods along pollution gradient in a river

V. Svara, Helmholtz Centre for Environmental Research GmbH / Effect-Directed Analysis; M. Krauss, Helmholtz centre for environmental research - UFZ / Effect-Directed Analysis; T. Luckenbach, Helmholtz Centre for Environmental Research UFZ / Department Bioanalytical Ecotoxicology

Many aquatic organisms are sensitive to man-made chemicals in the water. However, some species tolerate the occurrence of toxic chemicals and at the same time benefit from the nutrients that are often abundant in polluted waters. Such is also an amphipod species *Gammarus pulex* (Crustacea, amphipoda), which can be found throughout a pollution gradient of a stream. In our research, we investigated whether *G. pulex* individuals have an ability to adapt to the pollution gradient and to survive likewise in pristine and in polluted sites or whether different conditions select for different phenotypes within the population. *G. pulex* individuals were sampled at different sites along a pollution gradient in the river Holtemme (Saxony-Anhalt, Germany). Sites were characterized with respect to pollution burdens of water and amphipod tissues. Amphipods from three sampling points were brought alive to the lab and exposed to the insecticide Imidacloprid, which was found in water and animal samples beforehand. Lethal time for 50 % (LT50) values were determined and movement activities and moulting rates were recorded. Mortalities in the Imidacloprid treatments clearly differed for amphipods sampled at the different sites; animals from more polluted sites were more sensitive to the toxicant and also exhibited reduced movement and moulting activities. We examined whether these differences in toxic sensitivities were related to differences in toxicokinetics of Imdacloprid by quantifying imidacloprid tissue levels after different times of exposure to the compound but toxicokinetics of imdacloprid proceeded similarly in animals from different sites. Population genetics approaches (sequence comparisons of a DNA stretch of the cytochrome oxydase I (COI) gene and comparisons of 9 microsatellite loci) revealed that differences in conditions between sites did not lead to the separation of distinct subpopulations suggesting that survival of individuals at each site is based on individual acclimation and not on adaptation to specific conditions by a distinct subpopulation.

TU114

Antennae Regeneration of the Marine Amphipod Parhyale Hawaiensis as a Possible Endpoint in Ecotoxicology - Preliminary Data

O. Diehl, P. Assano, G. Umbuzeiro, School of Technology, UNICAMP / LAEG

Parhyale hawaiensis is a marine amphipod of worldwide circumtropical distribution and has been used in acute ecotoxicological tests. *P. hawaiensis* is able to regenerate its appendages, limbs and tissues after an injury or lost during the entire course of their life. Regeneration can be used as an ecotoxicological endpoint to assess potential teratogenic compounds and their impact on stem cells. Studies suggest that *P. hawaiensis* has local progenitor cell in each part of body It was already been demonstrated that *P. hawaiensis* has a fast regeneration of thoracic limbs, whitin a week, but no information on antannae's regeneration was found. Thus, the aim of this study was to obtain data on regeneration of antennae of *P. hawaiensis* to determine the viability this endpoint on toxicity tests. On day one left antennae of six months old organisms were amputed with sterilized tweezers, each organism transferred to recipients containing 100 mL salt water and a picture of each organism was taken under an stereomicroscope. Each test consisted of 20 organisms, 10 males and 10 females. During this period, organisms were feed three times a week, the necessary conditions of salinity, temperature, aeration, substrate and luminosity were provided. Four independent experiments were performed. The organisms were monitored daily until all of them undertook full regeneration. At that time, another picture was taken to determine the difference between antennae lenght (mm) before and after full regeneration. Antennae regeneration occured from 7 to 20 days (n=80) after amputation and males and females behaved differently, Males took more time than females to complete regeneration. Lenght of the regenerated antennae varied from 50 to 80% of the original appendages to both male and female. Next steps will be the exposure of organisms to selected toxicants to verify their ability of affecting the regeneration process in the developed experimental conditions. *Acknowledgement:* Conselho Nacional de Desenvolvimento Científico e Tecnológico" (CNPq-PVE Process: 400362/2014-7) for funding and PIBIC for undergrad fellowship. Amanda dos Santos e Gabriel Rampazzo Magalhães for technical contribution.

TU115

Added value of community approaches in environmental risk assessment

M. Hammers-Wirtz, T. Strauss, Research Institute gaiac / gaiac - Research Institute for Ecosystem Analysis and Assessment; A. Toschki, Research Institute gaiac

Community studies are an ecologically relevant tool to assess effects of stressors on population and community level. With these kinds of studies direct as well as indirect effects on populations can be addressed under environmentally relevant conditions. Furthermore in community studies like aquatic mesocosms, terrestrial model ecosystems (TME) or field studies, a variety of non-standard species interacting with each other and their abiotic environment are included and can be evaluated. Aquatic mesocosm studies have been used as higher tier tool in risk assessment of plant protection products in the EU since the 1990ies. In the last decades, they have been proven to be able to indicate potential risks for non-standard species that cannot be covered by the current lower tier studies. However, community tests were often criticised for their high variability and low statistical power. In the last decade, sampling methods have been optimized and a pragmatic approach for MDD categorization has been developed to evaluate effects with regard to their statistical power. Furthermore, in the last years there is an increasing concern that current risk assessment is related to a single product while in the environment the populations are exposed to a multitude of different plant protection products. In principle, in those community test systems also multiple mixtures or typical sequences of products can be tested. Due to the characteristic of the cur-rent risk assessment procedure sequences of different products are not yet considered. In conclusion, community studies are often noticed only as tools to defend single plant protection products without recognizing their outstanding ecological value. These studies are still the most realistic approach to assess effects on population and community level under realistic environmental conditions. The position and the order of these highly informative studies in risk assessment should be rethought. We suggest further options to integrate community approaches in risk assessment: 1) as a screening test system even in lower tier testing to get a broader idea about the relevant effects on ecosystem structure and function; 2) as monitoring tool for products which passed risk assessment to check up on community level effects; 3) as monitoring tool for typical sequence scenarios of different products which will be used together in one crop. Here exemplary results of community studies and a screening study will be presented.

TU116

Metal pollution and macro-invertebrate communities in the Olifants River, Western Cape, South Africa

J. Lucas, Cape Peninsula University of Technology / Department of Conservation and Marine Sciences; R.G. Snyman, Cape Peninsula University of Technology / Biodiversity and Conservation; J. Odendaal, Cape Peninsula University of Technology / Department of Environmental and Occupational Studies

Freshwater ecosystems are considered among the most threatened, as a result of current trends in water utilization globally. Within the Western Cape, 76% of rivers are polluted and run the risk of irreversibly losing their ability to support ecosystems and biodiversity. Anthropogenic activities all have an effect on water quality and quantity. Numerous pollutants result from these activities, with metals being particularly prevalent in most urban rivers. These pollutants are known to affect freshwater macro-invertebrate communities. In South Africa, SASS5 (South African Scoring System for Invertebrates) is a river health index that studies the invertebrate assemblages within specific riverine microhabitats, and can assist in showing the relationship between water quality and macro-invertebrate communities. The Olifants River in the Western Cape, is not only recognized as a hotspot for freshwater biodiversity, but seen by many, until the previous decade, to be the last pristine river along the South African coastline. It is also one of the three main feeding rivers to the City of Cape Town for fresh water. Despite increasing urbanisation, the last State-of-Rivers Report for this river was published in 2006. It has therefore become crucial to investigate the current degree of pollution within this river, as well as the general integrity of the system. This study aimed to determine the degree of metal pollution along the length of the Olifants River, as well as to investigate the effects of pollution and land use on the invertebrate communities, using SASS5. Water and sediment samples were collected seasonally at 5 sites from upper to lower reaches, acid digested and analysed with an ICP-AES for metal concentrations. Invertebrates were also sampled seasonally, identified and scored according to SASS5 sensitivity scores. An Average Score Per Taxon (ASPT) was calculated for each site. The results showed a general trend of increasing sediment metal concentrations, land use practices and habitat alterations, with concomitant decreasing ASPT's, from site 1 to site 5, indicating a loss of certain sensitive species at the most impacted downstream sites. Although metal pollution was found to be relatively low, a cocktail of pollutants, coupled with structural alterations, are clearly impacting the health and integrity of this river system. A future study should focus on organic pollutants, as agriculture is one of the main land use practices in the area.

TU117

QWATER - Bioassay integration under the European Water Framework Directive?: A step towards an ecological approach

M. Martinez-Haro, IREC-Instituto de Investigación en Recursos Cinegéticos / Department of Life Sciences; P. Acevedo, IREC-Instituto de Investigación en Recursos Cinegéticos; A.J. Pais-Costa, MARE-EBD; L.R. Vieira, ICBAS & CIIMAR, University of Porto / Department of Populations Study Lab of Ecotoxicology; J.M. Neto, MARE; M. Taggart, University of the Highlands and Islands / Environmental Research Institute; N. Álvarez-Ospina, Universität Potsdam; L. Guilhermino, ICBAS & CIIMAR University of Porto / Department of Biology; R. Ribeiro, Universidade de Coimbra / Life Sciences; J.C. Marques, MARE Marine and Environmental Sciences Centre

The Water Framework Directive (WFD) is the most important piece of water legislation in Europe. It aims at ensuring the 'good water status' of EU water bodies and includes both chemical and ecological status. To achieve and assess a 'good ecological status', the WFD advocates the integration of various lines of evidence, and demands a set of low-cost tools and techniques to deliver appropriate data. The WFD accounts for chemical and ecological evaluation, but there are no requirements regarding the establishment of cause-effect relationships in the assessment of environmental quality. Under this approach, the European Marie Curie QWATER project was aimed to gauge the ecological relevance of integrating short-term toxicity bioassays and biomarkers into quality elements in the WFD, as these may contribute to our ability to assess and manage EU water bodies. Ecologically relevant in situ cost-effective toxicity bioassays were use in a battery of in situ bioassays using representative species for several key functions in the ecosystem. Biomarkers, determined on the individuals used for the bioassays, were also integrated. Principal component analyses (PCA) were performed independently for each source of information, in order to improve the interpretation of the resultant PCA-factors in biological terms, and to verify whether the integration of 'quality elements' (bioassays and biomarkers) did (or did not) strengthen the robustness of the standard Ecological Quality Status approach used to assess water quality. Results shown some discrepancies in the water quality determined from each independent factor, i.e., the sampling sites were not equally ordered by all factors. Therefore, only by interpreting the values of all PCA-factors together is possible to perform a complete assessment of water quality status. The European regulatory authorities are presently in the phase of implementing the WFD based on community level approaches all over Europe. Interestingly, bioassays developed here and biomarkers, are available tools to be introduced as new feasible, cost-effective and sensitive protocols in the WFD. It might then become pertinent to stand for the combined/complementary use of ecological indices, biomarkers, and bioassays in the assessing the Ecological Quality Status of water bodies. Overall, this project allowed us to efficiently contribute to national and international efforts focused in evaluating the water quality in European water bodies.

TU118

Chronic testing of mayfly and stonefly species - Development of a new approach

M. Brüggemann, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology; K. Hund-Rinke, Fraunhofer IME / Department of Ecotoxicology; K. Schlich, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology; C. Schaefers, Fraunhofer-Institut / Ecotoxicology

Aquatic organisms, especially lotic invertebrate species originating from running waters, are exposed to releases of plant protection products which are mainly used in agriculture. Since lotic invertebrate species are regarded to be very sensitive but are hardly considered in chronic ecotoxicity testing, we developed a test system in order to investigate chronic effects on mayfly and stonefly species. After successfully development of a test method for stonefly larvae *Protonemura sp.*,the next step was the establishment of a method for testing of mayfly species *Epeorus sp.* In the developed test system, contrary to usual indoor stream systems, not the water body itself, but test vessels inside test containers are circulated, thereby creating a target flow. The test containers are filled with medium and contains ten replicates each. Each replicate is a small cage, which serves as individual compartment for individual testing of one test organism. The surroundings are adapted to the natural habitat of the test organisms. As endpoints growth, emergence and mortality are observed during a 21 day exposure period. For the studies we use field collected larvae which are adapted to laboratory conditions before test start. In a first step the test conditions were adapted to the requirements of mayfly larvae. Therefore testing of different media was performed. It turned out that, contrary to testing of stonefly larvae, which is performed in Cu-reduced dilution water, moderately hard reconstituted water (according to EPA) works best for testing of mayfly larvae. Instead of Tetramin®, which was used in stonefly testing, the green algae *Desmodesmus subspicatus* was used for feeding of mayfly larvae. In addition, while stonefly larvae were acclimated for 48 hours, the acclimation period was extended to seven days before test start. Under these conditions mayfly larvae showed an acceptable mortality of test organisms. The next step is to perform a test with the test substance Imidacloprid which will be exposed to mayfly larvae for 21 days. Afterwards sensitivity of mayfly and stonefly larvae to Imidacloprid will be compared. The new testing method can provide toxicity data of chronic testing with different aquatic insect larvae, which can be used for a SSD (Species Sensitivity Distribution) approach. The developed test system, the results of the performed tests as well as a comparison between testing of mayfly and stonefly species will be presented.

TU119

Toxic effects of a carbamate insecticide on a non-target freshwater gastropod: active ingredient versus commercial formulation

P. Cossi, CONICET - UBA; L.T. Herbert, CONICET - UBA / Departamento de Química Biológica Laboratorio de Ecotoxicología Acuática Invertebrados Nativos.; M. Yusseppone, CONICET - UBA / Departamento de Química Biológica Laboratorio de Enzimología, Estrés Oxidativo, y Metabolismo.; M.d. Ríos de Molina, IQUIBICEN CONICET Universidad de Buenos Aires / Departamento de Química Biológica; A.F. Pérez, CONICET - Universidad Maimónides / CEBBAD Laboratorio de Invertebrados Marinos.; G. Kristoff, UBA-CONICET / Departamento de Química Biológica Laboratorio de Ecotoxicología Acuática Invertebrados Nativos.

Carbamate insecticides are commonly used in agriculture for crop protection exerting their toxicity through the inhibition of the enzyme acetylcholinesterase. In Argentina, the maximum concentration of carbaryl (CAR) detected in surface and subsurface drainages was 45.7 µg L^{-1}. In this study, we evaluated the subchronic toxicity of environmental concentrations of the active compound and a commercial formulation of CAR on biochemical and reproductive parameters in *Biomphalaria straminea*, a freshwater gastropod native to Argentina. Five treatments were included in this study: dechlorinated tap water, acetone in dechlorinated tap water (solvent control), CAR active compound (dissolved in acetone) in dechlorinated tap water at 12.68 and 126.8 µg L^{-1}, and the equivalent to 126.8 µg L^{-1} CAR of a formulation (dissolved in dechlorinated tap water) with 85% of the active compound. The concentrations used were chosen so as to have the same molarity as azinphos-methyl, an insecticide previously used in our laboratory. In bioassay 1, eight glass vessels per treatment were used with six snails each. After 14 days of exposure, homogenates were made with the organisms' soft tissues (pool of five snails per vessel). In the supernatant fraction, the following parameters were measured: cholinesterases (ChEs), carboxylesterases (CEs) with two substrates, glutathione S-transferase (GST), glutathione (GSH), superoxide dismutase (SOD) and catalase (CAT). In bioassay 2, ten containers per treatment were used with a single recently-laid egg mass each. The time and success of hatching were registered and, after one month, the survival of the offspring was evaluated. In both bioassays, CAR solutions were renewed every 48 h based on previous stability studies. The active compound caused an increase in the activity of SOD with both CAR concentrations (28 and 83%, respectively, compared to the solvent control). The formulation, besides increasing SOD activity (72%), augmented GSH levels by 23% and inhibited CAT activity by 47% (compared to the water control). Regarding the reproductive endpoints analyzed, no toxic effects were found neither with the active compound nor the formulation. Our findings show that a subchronic exposure of *B. straminea* to CAR, active compound or formulation, does not affect the primary target, ChEs. However, other toxicity pathways, in which antioxidant enzymes are involved, seem to be affected by this insecticide, mainly by the commercial formulation.

TU120

Toxicity of lanthanides to freshwater microcrustaceans

M. Muna, National Institute of Chemical Physics and Biophysics / Department of Materials and Environmental Technology; A. Lukjanova, H. Vija, A. Kahru, I. Blinova, National Institute of Chemical Physics and Biophysics / Laboratory of Environmental Toxicology

The application of lanthanides (Ln) in different sectors of the world economy has significantly increased during the last two decades. This process has been accompanied by emissions into the environment via different pathways. The anthropogenic anomalies of Ln in soil, surface water, groundwater and even in tap water have already been registered. The disruption of the natural biogeochemical cycle of Ln increases the risk of biota being exposed to elevated concentrations of Ln. However, the ecotoxicological effects of these elements and their fate in the environment are still insufficiently understood. The toxic concentrations reported in the literature, e.g., for *Daphnia magna*, noticeably vary presumably due to different test conditions. For this study, acute ecotoxicity testing of La, Ce, Pr, Nd and Gd nitrates to freshwater crustaceans *Daphnia magna* (48 h) and *Thamnocephalus platyurus* (24 h) were performed in synthetic freshwater and natural lake water. Also, long-term (21 days) exposure of *D. magna* (OECD 211) in lake water was included. It was shown that the Ln fractionation between two main phases (precipitated and settled or remained in the water column) changed during the tests depending on (i) water composition, (ii) nominal concentration, (iii) exposure time, and (iiii) tested chemical element. Therefore, nominal concentrations were used for toxicity calculations. Acute toxicity of investigated Ln to both crustaceans was similarly low. $E(L)C_{50}$ values obtained in synthetic freshwater varied from 18.2 to 34.6 mg Ln/L for *T. platyrus* and 18.5–31.1 mg Ln/L for *D. magna*. Gd was the most toxic to both species, however, difference between $E(L)C_{50}$ values for Gd and other Ln was statistically significant ($p< 0.05$) only in *T. platyrus*. In the lake water, bioavailability of Ln was much lower: mortality of exposed organisms did not exceed 25% at the largest tested concentration (50 mg Ln/L). In contrast to acute assay, the 21 day chronic test performed in the lake water showed high Ln toxicity to *D. magna* (0.2 to 0.5 mg Ln/L). It was revealed that mortality was a more sensitive endpoint than reproduction. Differences between LC_{50} of individual Ln were not statistically significant. Thus, our results support the hypothesis that different lanthanides have a similar mechanism of toxicity in crustaceans. This work was supported by Estonian Research Council grant IUT23-5.

TU121

Relevance and suitability of invertebrates swimming behavior as sub-lethal endpoint to be considered for ecotoxicological investigation

s. morgana, V. Piazza, C. Gambardella, E. Costa, F. Garaventa, M. Faimali, CNR ISMAR

Ecotoxicology is aimed to assess, monitor and predict the effect of contaminants in the environment. Looking for new and alternative approaches in this discipline has become of increasing importance. Furthermore, within the 3Rs approach (reduction, refinement and replacement) the basic idea is to reduce the use of vertebrate organisms and to refine the procedures to minimize pain, suffering, and distress. To achieve this goal, ecotoxicology needs analytical tools, able to detect organisms ' responses even at very low toxicant levels. At the CNR-ISMAR laboratory, it has been developed an innovative automatic recording system, namely Swimming Behavioral Recorder system (SBR system), coupled with an advanced image processing software. Nowadays, the SBR system has been used to record and track the swimming speed of different marine invertebrates, including cnidarians, crustaceans, rotifers and echinoderms. In 10 years of research, SBR system has proved to be sensitive to a wide range of contaminants, such as metals, organic compounds, micro and nanomaterials, both polymeric and not, and even environmental matrices such as sediment elutriates. The amount of robust and significant data produced supports the suitability of this methodology to be applied to aquatic invertebrates. Here we reported a brief summary of SBR's applications, which show the relevance, sensitivity and versatility of the swimming speed alteration of marine invertebrates as ecotoxicological endpoint. These data also contribute to support the hypothesis by which behavioral endpoints, such as swimming alteration, represents attractive approach that should be taken into account in ecotoxicological risk assessment.

TU122

Benefits of Using Ecologically and Economically Valued Invertebrate Species for Ecotoxicological Analyses: Potential Phototoxic Effects Comparing a Freshwater Vertebrate and Invertebrate

E.N. Vebrosky, Louisiana State University / Department of Environmental Sciences; W. Xu, Louisiana State University AgCenter / Renewable Natural Resources; L.M. Basirico, Louisiana State Univeristy; C.G. Lutz, Louisiana State University AgCenter / Renewable Natural Resources; K. Armbrust, Louisiana State University / Environmental Sciences School of the Coast and Environment

In Louisiana, crayfish are not just a standard invertebrate species found in bayous and rice fields but also a staple in the cuisine and culture. Over 82 million pounds of crayfish are harvested annually, resulting in a $45 million industry; therefore, they are both ecologically and economically valued in the region. In areas, such as Louisiana, where an invertebrate species is of such importance, incorporating that species into ecotoxicology testing may benefit the overall risk assessment for the chemical in question and any potential effects to the organism itself. For example, dicloran is the active ingredient in the fungicide Botran™, which is used throughout Louisiana on sweet potatoes; the toxic and phototoxic impacts of dicloran were analyzed using a vertebrate and invertebrate species (fathead minnows, *Pimephales promelas*,and red swamp crayfish, *Procambarus clarkii*). Fathead minnows showed negative impacts at concentrations as low as 0.1 mg/L and >90% mortality at 0.75 mg/L and red swamp crayfish showed negative impacts at concentrations ranging from 0.50-1.0 mg/L; the effects at similar concentrations show that *P. clarkii* is a useful, nontraditional organism to be used for ecotoxicological analyses in areas such as Louisiana where they are of such high importance. The use of crayfish, or other valued invertebrates, in ecotoxicology testing are additionally beneficial as they do not require IACUC approval and can likely be spawned in labs.

TU123

Impacts of anti-cancer drugs on freshwater rotifers at environmentally realistic concentrations

N.B. Martins, University of Minho, Department of Biology & CBMA / Department of Biology; A. Pradhan, University of Minho / Department of Biology; F. Cassio,

C. Pascoal, University of Minho / Centre of Molecular and Environmental Biology CBMA Department of Biology

As human population increases, the presence of emergent chemical contaminants (ECCs) in freshwaters increases. ECCs have shown to be persistent and bio active, reaching the freshwater aquatic systems mostly untreated, where their fate and behavior is little understood. Anti-cancer drugs are among of the ECCs of concern due to their high cytotoxicity and increasing usage. The administration of drugs in cocktails, instead of single drug treatment, make the assessment of the environmental risk of these compounds a difficult task with much information lacking on sub-lethal effects on aquatic species. We used two cytotoxic drugs aiming at linking their effects on the reproduction inhibition of the rotifer *Brachionus calyciflorus* with processes of oxidative stress. The rotifer was exposed to a range of concentrations of a antimetabolite (5-Fluorouracil; 5FU) and a cytotoxic antibiotic (Doxorubicin; DOX) alone and in mixtures. The results showed that 5-Fluorouracil had a stronger effect (EC_{50}=0.074 mg L^{-1}) on the population growth rate then Doxorubicin (EC_{50}=13 mg L^{-1}) and toxicity effects were detected at environmentally relevant concentrations. Two concentrations of each drug were chosen for binary mixtures and two concentrations per drug were used to assess reactive oxygen species (ROS) accumulation and plasma membrane damage with epifluorescence microscopy. In the presence of low concentrations of 5FU, there was a reduction of the toxicity induced by DOX indicating possible antagonistic effects between both drugs. At concentrations, as low as EC_{05} we found accumulation of ROS in a dose dependent manner showing a clear connection between ROS accumulation and the toxicity of these compounds. Furthermore, this indicates that even in concentrations with no apparent effect on the rotifer reproduction, cellular effects were found with possible consequences for the community at the long term.

TU124

Development in vitro and in vivo methods of measuring acetylcholinesterase and general esterases in aquatic invertebrates

Y. Cao, University of Copenhagen / Department of Plant and Environmental Sciences; M. Gottardi, University of Copenhagen / Plant and Environmental Sciences; N. Cedergreen, University of Copenhagen / Department of Plant and Environmental Sciences

Quantification of acetylcholinesterase (AChE) and other esterase activities are important in terms of assessing the toxic mechanism of organophosphate and carbamate insecticides. There are several techniques for measuring AchE and general esterases (GE) activity involving spectrophotometric or fluorescence detection of transformation products. In this study, we tested four methods to detect AchE and GE activity *in vitro* and *in vivo* in the two aquatic invertebrate species: *Daphnia magna* and *Chironomus riparius*. The aim of the study was: 1) to compare the efficiency and selectivity of the four methods, 2) to compare *in vitro* with *in vivo* measurements and 3) to compare the inherent esterase activities of *D. magna* and *C. riparius*. The four assays were: 1) AChE-assay using acetylthiocholine iodide (ATCI) as substrate, 5,5'-dithio-bis(-2-nitrobenzoic acid) (DTNB) as chromogen, measuring the production of 5-thio2-nitrobenzoic acid; 2) AChE-assay using acetylcholine bromide (ACh) as substrate, measuring resorufin production; 3) GE-assay using 1-naphthyl acetate (1-NA) as the substrate, measuring 1-naphthol production and 4) GE-assay using 4-methylumbelliferyl butyrate (4-MUB) as the substrate, measuring 4-methylumbelliferone production. Michaelis-Menten curves were created for all substrates, where it was possible. The results showed that the GE-assay using 4-MUB measured general esterase activities well both *in vitro* and *in vivo*. The GE-assay using 1-NA and AChE-assay using ATCI-assays could only be used *in vitro*, while the AChE-assay using resorufin formation could not be used either *in vitro* or *in vivo*. The maximal GE-activities *in vitro* in *D.magna* and *C.riparius* were 345±44 and 151±51 nmol min^{-1} mg^{-1} protein, respectively, when using 1-NA and 295±8 and 60±13 when using 4-MUB, hence, showing comparable activities across substrates. Focusing only on AChE-activity *in vitro* the maximal activities were 13.2±0.3 and 52.3±1.1 nmol min^{-1} mg^{-1} protein in *D. magna* and *C. riparius*, respectively, making *C. riparious* the species with the highest activity. Turning to the *in vivo* measurements, the GE-activities were 49.1and 17.4 nmol min^{-1} mg^{-1} protein for *D.magna* and *C. riparius*. The results of GE-assays using 1-NA and 4-MUB are similar. The AChE-assay could not be conducted *in vivo*. The GE-assay using 4-MUB, however, could be conducted *in vitro* as well as *in vivo*. The GE-activity in *D.magna* was higher while the AChE-activity in *D.magna* was lower compared to *C.riparius.*

TU125

Factors influencing bioaccumulation of metals and pollutants in corals

V. van der Schyff, North-West University / Unit for Environmental Sciences and Management; H. Kylin, Linköping University / Department of Thematic Studies Environmental Change; H. Bouwman, North-West University / Unit for Environmental Science and Management

Bioaccumulation is the total accumulation of contaminants in and by an organism from all sources and routes of uptake. Bioaccumulation is normally defined as the sum of the mechanisms of bioconcentration (contaminants obtained from water only), and biomagnification (contaminants obtained from food). Corals pose a conundrum to classifying uptake mechanisms, due to their particular growth forms. Biomagnification in corals can occur through both filter and suspension feeding. It is also known that metals in corals will consistently reflect metallic element composition of the water of a particular area – hence, bioconcentration. Other methods of metal uptake are difficult to assign to one of these categories. Metal particles in suspension in the water are trapped by the defensive mucus layer and ingested by the coral colony as 'food'. This might be seen as biomagnification. However, biomagnification is traditionally associated with trophic transfer through prey items, and thus the pathway does not fit the normal description. In this case, we propose that this route of uptake be called "particulate vectored accumulation". Corals can also include other elements into their skeleton lattice by substitution of Ca^{2+} with other divalent metallic elements. "Latticine inclusion" might be an apt novel term for this occurrence. The crystalline structure of the $CaCO_3$ coral skeleton differs between hard and soft corals, being either aragonite or calcite. Different metals may bind differentially with the different crystalline structures. Small metal particles in suspension can also simply become lodged in the pores and cavities of the skeleton, particularly hard corals, where it may become part of the eventual skeleton by overgrowth. This pathway might conceivably be considered as 'particulate bioconcentration'. Zooxanthellate algae are known to accumulate metals differently from their hosts. We surmise that the intricacy of the symbiosis between algal cells and coral tissue will make it difficult to apportion relative metal contributions (and therefore toxicity) to each of the two symbionts. These potentially different routes of uptake of elements and pollutants may complicate ecotoxicological studies of corals, but may also indicate new avenues of investigation and explanation.

TU126

Survival, metabolic rates and locomotory activities of a groundwater-obligate copepod species under long-term exposures to tetrachloroethylene

T. Di Lorenzo, Institute of Ecosystem Study of the CNR Firenze; L. Piccini, University of Florence, Department of Earth Sciences; D. Galassi, University of LAquila; G. Messana, Institute of Ecosystem Study of the CNR; M. Saenz, PRIET CONICET, National University of Luján; W.D. Di Marzio, CONICET-PRIET / PRIET

Tetrachloroethylene (TCE) is a contaminant frequently found in groundwater of industrialized areas worldwide. The degradation of this chlorinated aliphatic hydrocarbon (CAH) is often incomplete in groundwater and takes several decades. Contamination from TCE is considered persistent and difficult to remediate, due to its high density that favors a gravity-driven vertical infiltration into groundwater bodies. Through means of the Water Framework Directive the European Union has demanded Member States to provide TCE threshold values (TV) for assessing groundwater body quality. In Italy, TCE TV is 1.1 μg/L in groundwater bodies. Studies on surface water species have shown that TCE causes oxidative stress in different organisms, however the effect of this contaminant on groundwater-obligate species has not been investigated to date. More importantly, the effect that 1.1 μg/L TCE may have on groundwater species under chronic exposures is unknown. In this study, we investigated the effect of 1.1 μg/L TCE on survival, oxygen consumption, and locomotory activities of a groundwater-obligate copepod species (*Moraria* sp.) under different time exposures. The specimens required for the trials were collected in the Antro del Corchia Cave (Tuscany). We measured the individual-based oxygen consumption of this species as a proxy of possible metabolic reactions to long-term (> 4 days) exposures to TCE at 8.0° C, i.e. about the mean annual temperature of groundwater in the cave. To this end, we used a sealed glass microplate equipped with planar oxygen sensor spots with optical isolation glued onto the bottom of 80-μL wells (Loligo Systems, Denmark) integrated with a 24-channel fluorescence-based respirometry system (SDR Sensor Dish Reader, PreSens, Germany). The system allows simultaneous measurement of 20 replicates and 4 controls. Survival and locomotory activity assessments were performed by counting the number of alive individuals and measuring the number of moving animals in 5 mL glass vials each containing 20 individuals.

TU127

MOLECULAR AND BIOCHEMICAL BIOTRANSFORMATION RESPONSES IN OYSTERS Crassostrea brasiliana (Lamarck, 1819) EXPOSED TO PYRENE AND FLUORENE

F.L. Zacchi, Universidade Federal de Santa Catarina / Departamento de Bioquímica; M.N. Siebert, Universidade Federal de Santa Catarina / LABCAI Bioquimica; J.J. Mattos, Universidade Federal de Santa Catarina / NEPAQ, Departamento de Aquicultura; F. Nunes, UFSC / Departamento de Aquicultura; I.M. Martins dos Reis, Labcai UFSC; G. Toledo e Silva, Universidade Federal de Santa Catarina / LABCAI Bioquimica; C.E. Piazza, Universidade Federal de Santa Catarina / LABCAI, Departamento de Bioquímica; T.B. Pessatti, J.F. Tisca, C.C. Parenti, Universidade Federal de Santa Catarina / LABCAI Bioquimica; B.N. Othero, Universidade Federal de Santa Catarina / Departamento de Aquicultura; B. Righetti, Universidade Federal de Santa Catarina / NEPAQ-CCA; M.C. Bicego, University of Sao Paulo - USP / Department of Physical Oceanography, Oceanographic Institute; S. Taniguchi, Universidade de Sao Paulo / Oceanographic Institute; S.T. Sasaki, University of Sao Paulo - USP / Oceanographic Insitute; A. Bainy, Universidade Federal de Santa Catarina / Bioquimica

Polycyclic aromatic hydrocarbons (PAHs) constitute a class of widely distributed organic pollutants in aquatic environments. PAHs affect organisms due to its carcinogenic, mutagenic and/or teratogenic characteristics. Once the PAHs enter the cell, they require a multistep metabolic activation by specific enzymes that participate in biotransformation reactions. The aim of this study was to evaluate biochemical and molecular biotransformation responses of the oyster *Crassostrea brasiliana* exposed to pyrene (50 mg.L^{-1} and 100 mg.L^{-1}) and fluorene (100 mg.L^{-1} and 200 mg.L^{-1}), after two time periods of exposure (24 h e 96 h). The half-life times of both PAHs were quantified by fluorescence in the aquaria exposure water and the transcription levels of phase I (*CYP1-like*, *CYP2-like*, *CYP2AU1* and *CYP356A1-like*) and phase II (*GST-like, GSTm-like* and *SULT-like*) biotransformation genes, EROD, GST and GSTm activity, were evaluated in gills. The half-life time of pyrene (100 mg.L^{-1} = 2 h and 12 min) in water was lower than fluorene (100 mg.L^{-1} = 5 h and 54 min). These results might be related to the higher lipophilicity of pyrene, facilitating its influx through the plasma membrane into the intracellular medium. After fluorene exposure, transcript levels of *CYP2AU1* gene were higher in 200 g.L^{-1} (96 h). Transcript levels of all genes were higher in oysters exposed to 100 mg.L^{-1} of pyrene (24 h). Besides, *CYP2AU1* (24 and 96 h); *GSTO-like* (24 and 96 h) and *SULT-like* (24h)were higher in oysters exposed to pyrene 50 mg.L^{-1}. EROD and GSTm activities were higher in oysters exposed to 100 mg.L^{-1} of pyrene (96 h). These results suggest an important role of phase I and II biotransformation genes and enzymes in pyrene metabolism. This study contributes to the identification of new biomarkers of PAHs contamination in *C. brasiliana*. Also evidences a possible participation of these genes and enzymes in pyrene biotransformation metabolism. In addition, it suggests the participation of *CYP2AU1* gene in the biotransformation process of PAHs in gills of *C. brasiliana*.

TU128

BIOCHEMICAL AND CELLULAR RESPONSES OF THE CRAB PACHYGRAPSUS MARMORATUS TO EVALUATE THE ENVIRONMENTAL CONTAMINATION OF THE LIVORNO HARBOUR (ITALY) AND OF AN ADJACENT MPA

I. Caliani, F. Bellucci, M. Vitale, University of Siena / Department of Physical, Earth and Environmental Sciences; M. Fossi, University of Siena / Department of Physical Sciences, Earth and Environment; S. Fratini, University of Florence / Department of Biology; C. Pretti, CIBM Centro Interuniversitario di Biologia Marina; S. Casini, University of Siena / Scienze Fisiche della Terra e dellAmbiente

The main purpose of the present investigation was to assess the toxicological status of Livorno harbour in Italy and of an adjacent MPA, through the responses of a set of biomarkers in the crab *Pachygrapsus marmoratus*. This investigation is part of the IMPACT project (Port Impact on Marine Protected Area: cross-border co-operative actions), which has the purpose to debug cross-borders management plans to actually protect the Marine Protected Areas. Male and female crabs were collected from the different areas: Livorno harbour, considered a polluted area, and the marine protected area “Secche della Meloria”, located just a few miles from the Livorno harbour, where we intended to explore the eventual adverse effects of port contamination. A battery of biomarkers was employed to assess neurotoxic effects (acetylcholinesterase, AChE activity), energy metabolism (isocitrate dehydrogenease, IDH; lactate dehydrogenease, LDH), oxidative stress (lipid peroxidation, LPO; glutathione S-transferase, GST; glutathione peroxidase, GPX; glutathione reductase, GR; catalase, CAT; glutathione, GSH) and DNA damage (erythrocytic nuclear abnormalities,ENA assay) in the crabs. The levels of trace elements and PAHs were also evaluated in the sampled specimens. Results showed that the crabs sampled at Livorno harbour are exposed to contaminants able to cause oxidative stress and genotoxic effects. LPO and ENA assay showed a statistically significant difference between specimens collected at Livorno harbour and the samples coming from the MPA. The average values of LPO were about three times higher in crabs sampled in Livorno harbour in comparison with that sampled in the MPA. The results trends are not influenced by the sex and the female showed higher values of biomarkers in comparison with the males. The crab *P. marmoratus*, used for the first time as a bioindicator to investigate the toxicological status of a port and an MPA area by the use of a multi biomarker approach, was found to be a good sentinel species to monitoring coastal marine environment.

TU129

Toxicity of titanium on the mussel Mytillus galloprovincialis

S.T. Costa, Aveiro University & CESAM; R. Monteiro, University of Aveiro CESAM; F. Coppola, Department of Biology & CESAM - University of Aveiro / Biology; R. Freitas, University of Aveiro / Departamento de Biologia CESAM; C. Vale, CIIMAR University of Porto; M.E. Pereira, CESAM University of Aveiro / Department of Chemistry

Titanium (Ti) is at forefront of research related to nanomaterials. Due to their physical and chemical properties, Tinanoparticles ($nTiO_2$) are widely used in various industries and materials, such as additives in pharmaceuticals and food colorants, toothpastes, solar cells, sunscreens, cosmetics and boat paints. With the increasing production and use of $nTiO_2$, Ti has been inevitably released into aquatic systems through wastewater treatment plants, surface run-off, direct inputs and atmospheric deposition. The increasing input of $nTiO_2$ in the aquatic environment has raised concerns about the toxicity of Ti to inhabiting organisms. Once in the aquatic environment $nTiO_2$ interact with the surrounding water components, including other contaminants, which may change the availability of Ti to organisms, namely the ability to penetrate into cells which may result in toxicity. In the present study the mussel species *Mytillus galloprovincialis* was used to evaluate the impacts caused by the exposure of Ti (II) solutions with the initial concentrations of 5µg/L, 50µg/L, 100 µg/L of Ti (II) (). Biochemical (oxidative stress related biomarkers, metabolic capacity and energy reserves) markers, after 96 hours and 14 days exposure periods, were evaluated. The obtained results revealed significant alterations in contaminated mussels, varying with the concentration and time of exposure. Mussels exposed to Ti presented lower metabolism, represented by lower electron transport system (ETS) activity, which decreased along exposure time (decreased their metabolic capacity), leading to the maintenance of their glycogen (GLY) and protein (PROT) contents. Moreover, contaminated individualsactivated their antioxidant defences increasing the activity of superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx) and glutathione S-transferases (GSTs), which still were not enough to prevent cellular damages (revealed by the increased of lipid peroxidation in mussels exposed to Ti).

TU130

Comparing interspecific Artemia responses to chronic zinc exposure

A.P. Costa, Marine and Environmental Sciences Centre / Faculty of Sciences and Technology, University of Coimbra; I. Varo, CSIC Spanish National Research Council / Biology, culture and pathology of marine species; M. Martinez-Haro, IREC-Instituto de Investigación en Recursos Cinegéticos / Department of Life Sciences; P. Almeida Vinagre, WavEC - Offshore Renewables / Marine Environment and Public Policies; m. sanchez, CSIC / Wetland Ecology

The invasive species *Artemia franciscana* is displacing native *Artemia* (*A. salina* and *A. parthenogenetica*) from eastern Atlantic coasts and across the Mediterranean region. The Aveiro and Odiel saltpans (highly contaminated) are the few remaining sites with native Artemia. Most studies indicate that pollution generally favours invasion of exotic species. However, recent studies suggest that local adaptation to the contaminated conditions by the native population may prevent colonization by *A. franciscana.* Under this context, the sublethal toxicity of zinc was assessed in natural populations of *A. parthenogenetica* from the highly contaminated Odiel estuary (southern Spain), and in *A. franciscana* from a less contaminated area (Cadiz bay, Spain, SW Spain). The Zn concentration used in our experiments (0.2 mg/L) was the double of that recorded in water from the Odiel saltpans to make our results as relevant as possible to real field conditions. Cysts were hatched in seawater and nauplii (A. parthenogenetica) or separated in couples (*A. franciscana*) according to their groups (control and treatment) and a set of reproductive parameters were examined. Results showed that *A. franciscana* performs better (higher survival and growth) than *A. parthenogenetica*. Both species experienced significant slower growth and higher mortality when exposed to Zn, but not significant effects were found in final size. Regarding reproductive parameters, Zn exposure increased offspring production of both *Artemia* species when compared to control. However, native *Artemia* showed, generally,a better reproductive performance (higher number of broods and offspring production; lower % non-viable nauplii) than *A. franciscana*. The results of this work highlight competitive advantages of native species (*A. parthenogenetica*) from contaminated areas to prevail under the selective pressure of abiotic factors as environmental pollution. Based on these results the highly polluted Odiel estuary would not be a refuge for native Artemia populations as suggested by the theory of local adaptation. **Keywords:** Artemia species; Local adaptation; Sublethal exposure; Zinc contamination.

TU131

Promising invertebrate species as model organism in ecotoxicology: ephyrae stage of the jellyfish Aurelia sp. and Sanderia malayensis

E. Costa, C. Gambardella, V. Piazza, CNR ISMAR; S. Lavorano, Costa Edutainment spa Aquario di Genova; M. Faimali, F. Garaventa, CNR ISMAR

In aquatic toxicology, one of the most important steps is the selection of suitable model organisms, able to provide information on the acute and chronic toxicity of marine pollutants. In this contest, invertebrates species are being used extensively in laboratory tests for their usefulness for seeking mechanistic links between effects occurring at the individual level and consequences for higher levels of biologic organization. In addition, compared to vertebrates they are also easy to maintain under laboratory conditions, widely distributed and ecologically relevant. Although Cnidarian jellyfish (Scyphozoans) are known to play an important role in marine food webs and are often conspicuous components in marine ecosystems, they are not yet employed in routine ecotoxicology. The aim of this current investigation is to suggest the use of two new invertebrate species of the jellyfish *Aurelia* sp. and *Sanderia malayensis* as model organisms in ecotoxicological bioassays. A series of experiments were carried out in laboratory controlled conditions, in order to characterize some experimental parameters that can influence the Frequency of pulsation (Fp) of ephyrae a new behavioural end-point identified as potentially sub-lethal response for this innovative invertebrate model for ecotoxicological testing. After these preliminary tests, ephyrae were exposed to a wide range of potentially toxic compounds (metals, surfactants, pesticides, nano-materials and harmful algae, emerging compounds), in order to evaluate the potential of ephyrae jellyfish in ecotoxicology. The experiments allowed to identify two end-points (sub-lethal, frequency of pulsation and acute, immobilization) with different levels of sensitivity and to optimize the use of an automatic recording system of swimming marine invertebrates (Swimming Behavioural Recorded e SBR), already employed with other biological models. In addition, the comparison of the EC_{50}

values obtained exposing ephyrae jellyfish to different toxic compounds and materials such as nanoparticles and microplastics with those obtained with other marine invertebrates, highlights that ephyrae are an interesting and promising invertebrate model with a very high ecological relevance to be used in ecotoxicological investigations.

TU132

Paracentrotus lividus and Artemia sp.: never too old model organisms to give new end-points

s. morgana, C. Gambardella, M. Faimali, F. Garaventa, CNR ISMAR

In the last few years it has become increasingly important the contribution of ecotoxicological assays to the environmental monitoring, as a fundamental integration of chemical analyses. In environmental risk assessment, in order to fulfill several regulatory requirements, such as the 3R principles (reduction, refinement and replacement), the development of novel approaches to reduce and eventually substitute the use of vertebrate species results to be paramount. Swimming alteration is one of the most frequently used behavioral responses in aquatic ecotoxicology and its evaluation has proved to be a valuable endpoint in ecotoxicological studies with aquatic organisms. Behavioral responses have proven their usefulness in evidencing impacts of chemicals at environmental concentration that do not necessarily cause mortality; therefore, behavioral endpoints are less invasive than traditional acute tests, but still sensitive and more ecological relevant. In this work, we reported a novel research on the use of swimming behavior of two "old" marine model invertebrates in ecotoxicology, the crustacean *Artemia* sp. and the echinoderm, *Paracentrotus lividus*, as sub-lethal endpoint. In detail, we optimized and improved an automatic recording system, namely Swimming Behavioral Recorder system (SBR), by developing i) a new swimming speed alteration test using for the first time sea urchin early stages; ii) a new short-term test based on the evaluation of the swimming speed alteration of *Artemia* nauplii incubated at 39 °C (± 1) for only 6 hours. Thanks to a modern video-based technology, this study provides novel perspectives and future applications applied to two well-known marine model invertebrates, meeting regulatory and market demands, including the reduction in using vertebrate species and the need for early warning technologies.

TU133

Application of sea-urchin embryo test in the effect directed analysis approach for the evaluation of WWTP effluent in an estuarine media

L. Mijangos, University of the Basque Country UPV/EHU / Department of Analytical Chemistry; M. Krauss, T. Schulze, W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; H. Ziarrusta, University of the Basque Country UPV/EHU / Department of Analytical Chemistry; M. Olivares, O. Zuloaga, University of the Basque Country UPV/EHU / Plentzia Marine Station (PiE-UPV/EHU) & Dep Analytical Chemistry; r. beiras, University of Vigo / Toralla marine sciences station (ecimat); A. Prieto, N. Etxebarria, University of the Basque Country UPV/EHU / Plentzia Marine Station (PiE-UPV/EHU) & Dep Analytical Chemistry

Since target chemical analysis often cannot explain the cause-effect relationship between certain contaminants and the observed effects in organisms, effect directed analysis (EDA) can be applied to unravel the drivers of toxicity in complex mixtures. In this study, the sea-urchin embryo test (SET) was implemented for the first time in a EDA approach in order to evaluate an estuarine environment influenced by the effluent of the main waste-water treatment plant (WWTP) of Bilbao. WWTP sample (225 L) was extracted with SPE and submitted to a sequential LC-UV fractionation methodology based on two different columns: a Nucleodur C_{18} column (21 fractions were collected) and an aminopropyl column (15 fractions). Two endpoints were used to determine the toxic effects after 48 h: the growth rate of the larvae and the rate of skeletal malformation. 6 levels (n=3) of dose-curve were prepared in units of relative enrichment factor (REF, final volume of 3 mL of filtered seawater with 0.1 % of DMSO). Non-target analysis was performed by means of UHPLC-Qexactive Plus MS in positive and negative modes with a C18 column. Toxic compounds were identified using MS2 spectrums, Metfrag and Compound Discoverer (Thermo) interfaced to MZmine. Among the collected C_{18}-fractions, only fraction 13^{th} (F13) showed a clear toxicity and, therefore, it was tested separately to establish the concentration-response model. The curve-dose response of the raw sample (EC_{10}= 10 REF and EC_{50}=19 REF) could be explained by the contribution of active F13 (EC_{10}=14 REF and EC_{50}=39 REF). Regarding the chemical analysis, among the final candidate list (206 compounds), mebendazole (an antihelminthic agent) was confirmed chromatographically with standards. Nervertheless, a sequential fractionation of F13 was also carried out with an aminopropyl column, which showed a different orthogonality compared to C_{18} column, and the resulting 15 fractions were also submitted for further bioassays and data-dependent analysis. Overall, the results of this work suggest the possibility of addressing a kind of specific toxicity in sea-urchin embryos owing to the determination of only one toxic fraction and the contaminants identified in that fraction. *Acknowledgement.* This work was financially supported by the Ministry of Economy and Competitiveness through the project CTM2014-56628-C3-1-R. L. Mijangos is grateful to the Basque Government and H. Ziarrusta to the Spanish Ministry for their predoctoral fellowships.

TU134

Plausibility of Daphnia magna model to evaluate eicosanoid pathway related toxicity

S. Lee, Seoul National University / System Toxicology Research Center; M. Cho, Korea Institute of Toxicology; S. Yoon, W. Kim, Korea Institute of Toxicology / System Toxicology Center

Eicosanoids are biologically active, oxygenated metabolites of C20 polyunsaturated fatty acids and are synthesized through cyclooxygenase, lipoxygenase or cytochrome P450 epoxygenase pathway. As signaling molecules, they are important for diverse physiological systems such as inflammation, allergy, pregnancy, pain perception and blood pressure control. Therefore, they could be the important target for toxicant or drugs such as ibuprofen and aspirin. However, invertebrate model to evaluate eicosanoid pathway related toxicity has not yet been developed well though similar eicosanoid pathway was expected in invertebrate species. Therefore, the purpose of this study was to identify genetic information of key genes of eicosanoid pathway in *Daphnia magna*, a widely used invertebrate model, and to evaluate the transcription levels of the genes after the exposure to eicosanoid targeted drugs. To this end, we collected amino acid sequences of eicosanoids from other species and then compared the sequences in water flea genome database. After the BLAST and alignment, the genetic information of 10 key eicosanoid genes, such as *pla2*, *cox*, *pgd2a* and *pge2*, was identified. After that, *Daphnia magna* was exposed to the eicosanoid pathway targeted drugs, i.e., ibuprofen, indomethacin, celecoxib and acetaminophen at 0.25, 2.5 and 25 μM. Then, we analyzed transcription of the genes by qPCR system. In *D. magna*, transcription of *pla2* gene was commonly down-regulated by all the eicosanoid targeted drugs. Interestingly, some genes, such as *pgd2* and *gpx1*, were responded to certain specific drugs, celecoxib and ibuprofen, respectively. Through this study, we found that the nine of ten genes were expressed by exposure to the eicosanoid targeted drugs in *D. magna*. We believe that these results partially indicate the plausibility of *D. magna* model to evaluate the effects of chemicals on eicosanoid synthesis pathway. Also, the genes expressed in this study could be used as suitable biomarkers for the eicosanoid related toxicity assessment.

TU135

Responses to single chemical and pulse exposures of two monophyletic Daphnia species under a multi-generation approach

G. Araujo, Universidade de Aveiro / Biologia; A.M. Soares, University of Aveiro / department of Biology & CESAM; D.M. Abessa, Universidade Estadual Paulista - UNESP/CLP / Marine Biology and Coastal Management; S. Loureiro, Universidade de Aveiro / Biology

Plenty human activities can be related to environmental contamination (e.g. industries, agriculture). Those contaminants may have continuous or pulse sources and can affect organisms from natural habitats. In different latitudes even phylogenetically close related species may present divergent chemical tolerance. Therefore, it was used in this study the model species from temperate areas *Daphnia magna* and the tropical species *Daphnia similis*. Most studies rely on short standard acute and chronic tests. To enhance veracity, we performed a chronic multi-generation exposure (nine generations) to lead (0.05 mg/L Pb) under two different dietary exposures (regular and restricted) and, regarding chemical mixtures of natural environments, pulse exposures (on Pb acclimated *daphnids*) to the fungicide mancozeb were also accomplished. Organisms from F6 were changed to a clean media for recovery for three generations. To monitor acclimation, standard acute immobilization tests to $K_2Cr_2O_7$ (to check for sensitivity), to the metal Pb and to mancozeb were made. Since size is crucial on organisms' tolerance, neonates' body length was also measured. No difference on *daphnids* sensitivity was spotted among generations, excepting *D. similis* from recovery period under food restriction. However, Pb tolerance increase is seen on both species. Regarding mancozeb exposure (Pb acclimated), no difference between treatments is seen under regular food. However, opposite outcomes are shown under food restriction, such as *D. magna* increasing and *D. similis* decreasing tolerance to mancozeb. Adverse outcomes regarding recovery was shown, *D. magna* relied on genetic adaptation, since it kept a higher tolerance to Pb, even after three generations under recovery, and *D. similis* relied on physiological acclimation, being similar to organisms from control treatment. Food restriction also decreased reproduction and increased neonates' body length (both species). The opposite outcomes under food restriction is crucial regarding natural environments and the natural fluctuating amount of nutrients. Therefore, results indicate that *daphnids* are capable of acquiring resistance to Pb under a long-term exposure, being an essential data on chemical mixtures contaminated habitats, since their tolerance to other chemicals can vary (as shown for mancozeb). And also, opposite outcomes regarding monophyletic species indicates that it is not accurate to use species from different climates to estimate toxicity.

TU136

Chronic effects of BPA, BPS, and BPSIP in Daphnia magna

Y. Hong, B. Jeon, I. Ryoo, j. Lee, K. Ji, Yongin University

Since bisphenol A (BPA) can act as a weak estrogen agonist and disrupt steroidogenesis, concerns on adverse health effects are increasing. In response to the regulatory pressures to eliminate BPA in plastics, bisphenol S (BPS) and

4-hydroxyphenyl 4-isoprooxyphenylsulfone (BPSIP) have been frequently used in manufacture of thermal paper and plastic containers. However, there is paucity of information on their chronic toxicity using aquatic invertebrates. In the present study, chronic toxicity of BPA, BPS, and BPSIP were evaluated using *Daphnia magna* in accordance with OECD Test Guideline 211. The endpoints for the long-term exposure were survival, reproduction, and growth. Compared to the control group, the body length was significantly decreased in *D. magna* exposed to 1 mg/L of BPA, 5 and 10 mg/L of BPS, and 1 mg/L of BPSIP ($p< 0.05$). The results showed that BPA, BPS, and BPSIP could induce endocrine disruption related to the growth in aquatic invertebrates, and the effective concentration of BPSIP was similar to that of BPA. With increasing use of these alternative compounds, more monitoring program in aquatic environment and study of toxicity mechanism appeared to be necessary. Acknowledgement: This study was supported by the National Research Foundation of Korea (Project NRF-2015R1D1A1A01056628).

TU137

Oxidative effects of mono-(2-ethylhexyl)-phthalate on Daphnia magna in both molecular and population level

Y. Kim, KIST Europe / Environmental Safety Group; Y. Seol, KIST-Europe / Environment Safety group

Mono-(2-ethylhexyl)-phthalate (MEHP) is the metabolite of di-(2-ethylhexyl)-phthalate (DEHP), which is widely used in the industry as plasticizers. According to previous studies, DEHP inhibits molting hormone. In addition, MEHP is highly persistent and bioaccumulative in environment and living organisms. However, little is known about mechanism of MEHP as an endocrine disrupting chemical in aquatic organism such as *Daphnia Magna*. The aim of this study was to elucidate the linkages between toxicity test result and oxidative stress of MEHP. We studied the effects of oxidative stress as molecular initiating events on *Daphnia magna*. We observed the changes in different levels of the lipid peroxidation, glutathione S-transferases (GSTs), catalase (CAT) and superoxide dismutase (SOD) in the treated daphnids. This study showed the molting rate, reproduction rate and growth of daphnids during chronic (21 d) test in order to link the activities of reproduction system to antioxidant responses. Further study is needed to clarify how MEHP leads to dysfunction of endocrine system of *Daphnia magna*.

TU138

Are Daphnia magna and Chironomus riparius acute responses comparable?

B. Ponti, ChemService Srl; R. Bettinetti, University of Insubria / DiSTA; D. Garagna, ChemService s.r.l.; F. Casarotto, University of Insubria; M. Neri, ChemService srl - Controlli e Ricerche

Laboratory ecotoxicity test results predict the responses of organisms with varying degrees of accuracy. Traditionally, the acute toxicity on aquatic invertebrates is estimated by exposing for 48 hours young cladocerans of *Daphnia magna* (OECD test guideline n. 202, 2004), taking advantage of its well-experienced sensitivity and reliability for a huge number of known and unknown toxicants. The 48 hours test on *Daphnia magna* conducted according to OECD 202 is listed as a data requirement in EU Regulation 284/2013 on plant protection products as well as in Reg EC 1907/2006 on chemicals (REACH). In recent years even the use of first instar larvae of *Chironomus riparius* (Insecta, Diptera) has been proposed (OECD guideline n. 235, 2011) to be used to complement existing Test Guidelines for chironomid chronic toxicity assays (OECD test guidelines 218, 219, 2004) (OECD Test Guideline n. 233, 2010). *Chironomus riparius* is generally used to test the quality of sediments but during its first days of life it freely swims in the water column before becoming benthic for the other three larval stages. As a matter of fact, the Chironomus acute test can be a useful tool to control the conditions/sensitivity of the breeding organisms in the lab Therefore it can be of interest to understand its relevance when compared with the answers of other organisms, belonging to other taxa and with different life cycles. In order to compare the responses of *Daphnia magna* and *Chironomus riparius* when exposed to the same contaminant, we carried out different exposures using three different substances: two reference items (potassium chloride, and potassium dichromate, commonly used to test sensitivity of *C. riparius* and *D. magna* respectively) and an unknown toxicant (a fatty acid C14-C20). Preliminary results indicate possible differences in responses of both test organisms; if confirmed by definitive tests these observations may represent a warning when carrying out acute toxicity tests on water medium, confirming the importance to test different trophic levels and showing the need to further investigate the use of the acute test on *Chironomus riparius* according to OECD 235 to assess the acute toxicity on aquatic organisms.

TU139

Analysis of mixtures of bisphenol A and UV filters Octocrylene and OD-PABA on Chironomus riparius using a specific RT-PCR array

A.M. González, UNED / Mathematical Physics and Fluids; J. Martinez-Guitarte, UNED / Fisica Matematica y de Fluidos

Ana-Belén Muñiz-González, José-Luis Martínez-Guitarte.\nGrupo de Biología y Toxicología Ambiental. Facultad de Ciencias, UNED. Madrid (Spain)\nKeywords: UV filters, BPA, RT-PCR array \nThe ultraviolet (UV) filters are main components of many personal care products (PCPs) that are extensively used. They are organic compounds defined as emergent contaminants, which are increasing their presence in the environment because their use in recreational and industrial activities. Described as endocrine disruptors in vertebrates, their effects on invertebrates have been poorly studied, especially in mixtures. On the other hand, Bisphenol A (BPA) is as plasticizer used in packaging and other industrial products with confirmed endocrine disruption activity. In this study we have used two common UV filters, octocrylene (OC) and 2-ethylhexyl 4-dimethylaminobenzoate (OD-PABA), and BPA to mimic the putative mixtures resulting from PCP and interaction with plastic of PCP containers. These mixtures can reach the biota of freshwater ecosystem so the main objective was studying of the effects that the mixtures can have on an invertebrate with a relevant role in the food chain of these ecosystems, Chironomus riparius. C. riparius is a dipteran with aquatic larvae frequently used in toxicity tests. Fourth instar larvae were exposed for 24 hours to single compounds and to binary and ternary mixtures. Expression profile of 42 genes was analyzed by retrotranscription and Real-Time PCR using a specific array covering a number of relevant metabolic pathways like endocrine system, immune response, stress response, detoxification mechanisms and apoptosis among others. Using an array could improve the toxicological evaluation of the cellular effects of the compounds favoring the identification of new molecular biomarkers useful for ecological risk assessment and toxicity tests. The methodology used to design the array can be used with other species improving also our knowledge about the mode of action of these compounds. \nThis work has been funded by the Ministerio de Economía y Competitividad, CICYT (SPAIN), CTM2015- 64913-R.\nA.B.M.G is the receiver of a pre-doctoral contract from the National University of Distance Education (UNED).

TU140

Genetic variability in tolerance to microbial insecticides in Chironomus riparius

M.D. Bordalo, University of Aveiro; A. Rodrigues, University of Aveiro / Biology Department and CESAM; I. Lopes, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro; J. Pestana, CESAM & University of Aveiro / Biology

Natural populations are constantly facing a large array of environmental stressors, from both natural and anthropogenic origin, which represent a strong selective force shaping the behaviour, physiology and morphology of organisms. Genetic diversity acts as a key component of population response to perturbation and on their potential to adapt to changing environmental conditions. Ecological risk assessment needs to couple quantitative genetic analysis with ecotoxicological studies in order to understand the mechanisms underlying evolution of tolerance. Research on genetic variation regarding tolerance to contaminants has been mostly performed with clonal lineages. An alternative methodology has been applied for sexually reproducing organisms, by comparing sensitivities of siblings (family genotypes) and estimating genetic variation in fitness traits. However, additional environmental stressors are not usually considered, limiting the predictive capabilities and determination of tolerance costs across different scenarios. The aim of this work was to assess genetic variation in tolerance in the aquatic insect *Chironomus riparius* exposed to microbial insecticides. A *C. riparius* population was established in the laboratory by crossbreeding five populations, in order to ensure sufficient levels of genetic diversity. By employing a full-sib family split design, this study presents a quantitative genetic analysis among families of *C. riparius* across different environments (microbial insecticide exposure under two levels of salinity). Each egg mass was considered a single family (full-sib genotype). Within each family, larvae were randomly allocated to all treatments. Emergence rate, time to emergence and imagoes weight were used as endpoints. Relationships between genetic variation, life-history traits and fitness costs in response to microbial insecticides, and the potential of a key aquatic insect species to evolve tolerance to these compounds will be discussed as well as an evolution of disease resistance on aquatic species. The study also highlights the suitability of *C. riparius*, a model organism in aquatic toxicology, for quantitative genetic analyses.

TU141

Effects of Amitraz on Chironomus riparius: life history and biochemical responses

H.R. Monteiro, University of Aveiro / Department of Biology and CESAM; J. Pestana, CESAM & University of Aveiro / Biology; S.C. Novais, Polytechnic Institute of Leiria / MARE IPLeiria; A.M. Soares, University of Aveiro / department of Biology & CESAM; B. Devreese, Ghent University / Laboratory for Protein Biochemistry and Biomolecular Engineering; M.F. Lemos, Instituto Politécnico de Leiria / MARE IPLeiria

Amitraz is a very effective formamidine insecticide used in agriculture to control fruit trees and cotton pests. Due to its widespread use and high direct application rates, there is an elevated risk of contamination of adjacent aquatic ecosystems. However, the information of amitraz toxicity to aquatic non-target invertebrates is scarce. In this study, the toxicity of amitraz to the midge *Chironomus riparius* (Meigen) was assessed in terms of life history responses using standard ecotoxiciological tests and biochemical endpoints using oxidative damage, antioxidant defenses, energy metabolism, and neuromuscular toxicity biomarkers. Chronic exposure to amitraz contaminated waters (28 days; 10, 20, 40, 80, and 160 $\mu g\ L^{-1}$) resulted in impairment of *C. riparius* emergence and developmental rates (reduced larval growth and delayed emergence), with male development time being the most sensible endpoint (LOEC of 40 µg L^{-1}). Short-term exposures (48 h; 0, 10, 40, and 160 µg L^{-1}) to amitraz induced glutathione peroxidase activity and a decrease in catalase activity. Additionally, amitraz exposure caused a decrease in lactate dehydrogenase activity and a significant increase in electron transport system activity, both energy metabolism associated biomarkers. Regarding oxidative damage biomarkers, lipid peroxidation increased in *C. riparius* larvae exposed to 160 µg L^{-1} of amitraz while there was a significant decrease in DNA damage levels at 10 and 40 µg L^{-1} treatments. The present results reveal possible biochemical targets of amitraz toxicity and suborganismal responses associated with amitraz exposure. The assessment of biochemical biomarkers may support the interpretation of toxic responses observed at organism level and therefore in the assessment of the ecological effects of environmental contamination. Moreover, it increases the information available on the outcomes of amitraz exposure in freshwater invertebrates, and underlines the importance of risk assessment studies of formamidine pesticides. Aknowledgements: This study had the support of the Fundação para a Ciência e a Tecnologia through project PROTEOME (PTDC/AAG-MAA/1302/2014), co-financed by COMPETE (POCI-01-0145-FEDER-016773).

TU142

Multigenerational exposure of Folsomia candida to copper agrochemicals: conventional and nano-pesticides

C. Malheiro, Department of Biology, University of Aveiro / Biology; A.R. Silva, University of Aveiro / Dept.of Biology & CESAM; D. Nunes Cardoso, CESAM, University of Aveiro / department of Biology & CESAM; J.T. Neves, University of Aveiro / department of Biology & CESAM; P. Silva, Universidade de Aveiro; J. Ulčar, University of Ljubljana / Department of Biology; F.J. Wrona, University of Calgary / Department of Biological Sciences; A.M. Soares, University of Aveiro / department of Biology & CESAM; S. Loureiro, Universidade de Aveiro / Biology

Agricultural practices include the use of agrochemicals for crop maintenance and enhanced production, although soil contamination may result. Inorganic agrochemicals, like copper pesticides, have been used in a range of agricultural applications, which may result in environmental problems. More recently, nanopesticides were introduced in the market with the intent to improve efficacy and decrease environmental negative effects. However, the chronic ecotoxicological effects of nanopesticide exposure on soil biota are not well known since related environmental hazards are most often assessed using only the active ingredients. Moreover, the multigenerational effects of long-term, chronic exposure of soil organisms to agrochemical applications are unknown. The objective of this study was to evaluate the impact of long-term, multigenerational exposure of the soil collembola *Folsomia candida* to conventional and nanoparticle formulations of copper pesticides. Two formulations were assessed: Kocide® 3000 (nano form) and Champion® WP (conventional), as well as the pure active ingredient $Cu(OH)_2$ in spiked LUFA 2.2 soil. The effects of multigeneration exposure to the Cu pesticides were assessed using two soil treatments: 1) Cu spiking performed only at the beginning of the experiment and collembolan responses (survivorship, reproduction) measured for three generations (i.e., aging soil exposure); and, 2) Cu spiking performed at the start of each new cohort for three generations (i.e., renewal soil exposure). After three generations in both soil treatments, the surviving collembola were moved to uncontaminated soil for three generations to assess their recovery potential. Similar response patterns were observed in the two soil treatments for all three Cu formulations. Exposure to aging soils revealed an increasing tolerance across generations of *F. candida*. In contrast, in treatments with renewed Cu spiking, the collembolan populations showed ongoing sensitivity to Cu exposure. In both treatments, after being moved to clean soil, all treatment populations showed some recovery by displaying increased reproductive output. Copper forms presented difference between them in the long term exposure. This study further emphasises the importance of using multigenerational approaches to obtain more ecological relevant evaluations of environmental risk associated with chronic exposure to soil agrochemicals.

TU143

Effects of multiple environmental stressors on Eisenia fetida coelomocytes: cell viability and different behaviour of amoebocytes and eleocytes

N. García-Velasco, University of the Basque Country / Zoology and Animal Cell Biology, Research Centre for Experimental Marine Biology and Biotechnology PIEUPVEHU; E. Urionabarrenetxea, University of the Basque Country / Zoology and Animal Cell Biology, Research Centre for Experimental Marine Biology and Biotechnology; M. Soto, University of the Basque Country / Zoology and Animal Cell Biology, Research Centre for Experimental Marine Biology and Biotechnology PIEUPVEHU

Earthworm immune cells (coelomocytes) have become a target system in ecotoxicology due to their sensitivity against a wide range of pollutants. Moreover, endpoints measured in coelomocytes retrieved from exposed *Eisenia fetida* offer rapid and accurate information to predict impairments caused by pollutants at longer exposure times and higher complexity levels (organism, population). Since soils are subjected to multiple environmental stressors (i.e., temperature raises, acidification, organic matter depletion, new pollutants) it is of great interest to assess how those stress scenarios pose changes in earthworms at cellular level. Coelomocytes compose a heterogeneous cellular group where two major cell subpopulations are distinguished, amoebocytes and eleocytes. However, the behaviour of those subpopulations against different stressors is still unclear. Hence, the aim of the present work was to address the effects of different stressors (increase in temperature, low OM, model and emerging –nanoparticles- contaminants) on *E. fetida* coelomocytes by assessing mortality and changes in the relative proportion of subpopulations (amoebocytes, eleocytes). For that, earthworms were maintained under low OM content (6% vs. 10%), thermal stress (19ºC vs. 26ºC) and pollution conditions (Cd: 5-25 mg/kg dw-, Ag NPs: 0-100 mg/kg dw) in OECD soil during 3 days. After exposure of earthworms, coelomocytes were retrieved and viability was assessed in microplate through Calcein AM assay. In addition, flow cytometric analysis was used to determine mortality of coelomocytes and changes in the relative proportion amoebocytes/eleocytes. Coelomocytes extruded from earthworms maintained at low OM and higher temperature showed lower cell viability, but no changes were recorded in the relative proportion of amoebocytes and eleocytes. Exposure to Cd provoked higher mortality in eleocytes while Ag NPs caused more mortality in amoebocytes. Thus, we can conclude that the response of the different subpopulations was dependant on metal form and was enhanced by environmental factors (increased temperature and low OM). These results reinforce the potential of endpoints measured at cellular level for an accurate soil health assessment in a global warning scenario. Acknowledgements: Basque Gov (IT810-13), Univ. Basque Country (UFI 11/37) and MINECO (Nanosilveromics Proj).

TU144

Toxicity of abamectin and difenoconazole, pure and formulated, to Folsomia candida

L.P. Figueiredo, University of São Paulo USP; G. Mainardi, Vrije Universiteit / Department of Ecological Science; C. Lima, Vrije Universiteit Amsterdam / Animal Ecology; E. Espindola, University of São Paulo USP / Hydraulics and Sanitation; C. van Gestel, Vrije Universiteit Amsterdam / Ecological Science; D. Roelofs, Vrije Universiteit / Department of Ecological Science

The progressive increase in the use of pesticides has been accompanied by effects at different levels of biological organization, implying losses of species and consequently of ecosystem services. Among the species utilized in terrestrial ecotoxicological tests, the springtail *Folsomia candida* (Collembola, Isotomidae) is one of the species suitable for assessing side effects on detritivorous soil arthropods. In Brazil, the acaricide abamectine and the insecticide difenoconazole are widely used in agriculture, but little data is available about their possible side effects on the soil community. The objective of this work therefore was to evaluate the effect of abamectine, pure and in the formulation Kraft®, and of difenoconazole, pure and in the formulation Score®, on the reproduction of *F. candida* using a standard Lufa 2.2 soil. Juvenile *F. candida*, with age 10-12 d, were exposed following the standardized ISO and OECD test guidelines. The results were analyzed by analysis of variance (ANOVA) followed by Dunnett's test at 5% significance level. Median lethal concentration (LC_{50}) was calculated using Trimmed Spearman Karber (TSK) and EC_{50} and EC_{10} values for effects on reproduction were estimated using a logistic model. The results showed that both pesticides were more toxic in the formulation than when applied as pure active ingredient. For abamectin dosed as the formulation Kraft® EC_{50} was 1.0 (0.17-1.8) mg/kg dry soil, while it was 6.3 (1.8–11) mg/kg dry soil for the pure active ingredient. For difenoconazole applied as the formulation Score®, EC_{50} was 53.5 (40.0-67.0) mg/kg dry soil while no effects on springtail reproduction were seen at concentrations of the pure active ingredient up to 333 mg/kg dry soil. The data indicate that it is essential to test commercial formulations along with tests on pure active ingredients. It is currently unknown which component of the formulation causes the increased toxicity. Therefore, we are applying gene expression analyses to mechanistically underpin increased toxicity levels caused by the tested formulations.

TU145

Terrestrial arthropods as indicators of environmental pollution

V. Lesch, North-West University; H. Bouwman, North-West University / Unit for Environmental Science and Management

In recent years, the use of and interest in terrestrial arthropods as indicators of environmental pollution has increased. Arthropods are diverse, with over 31 000 species described. Terrestrial arthropods are relatively easy to sample, and collection normally has less ethical restrictions then for higher animals. We reviewed the literature. We found relatively few general studies on arthropods as indicators of environmental pollution. However, those that did worked on spiders, bees, earthworms and ants. In addition, most studies favoured predatory species, since bio-magnification occurs and can give insight into the extent of pollution in the area. In most studies, the sampling sites were close to old mines, or the studies were comparisons of arthropods from different locations. Published literature on terrestrial arthropods not only focuses on whole body utilization but also organ specific studies, as well as research on the use of arthropod products (such as honey and spider webs) as matrixes for analyses. Most arthropods have a close association with soil, foliage, and air, representing pollution concentrations in the immediate environment. Each arthropod species occupy a unique niche and in doing so represent the concentration or degree of pollution in different ways. Each species, therefore, indicates pollution from different perspectives. However, there is still a lack of sufficient data on most terrestrial arthropods as indicators. We discuss a number of possible predatory taxa, such as dragonflies, spiders, wasps, and beetles.

TU146

The impact of chlorpyrifos and its formulations on the acetylcholinesterase activity in non-target soil organisms

D. Fojtová, Masaryk University, Faculty of Science, RECETOX / Research centre for toxic compounds in the environment RECETOX; J. Vašíčková, Masaryk University / Faculty of Science RECETOX; J. Hofman, Masaryk University, RECETOX / Faculty of Science, RECETOX

The Ecological Risk Assessment of pesticides requires data regarding their effects to terrestrial non-target species. Commercial pesticides formulations, however, contain a significant proportion (> 90%) of so-called inert ingredients, which may greatly enhance or lessen the toxicity of a formulation. Chlorpyrifos is a broad-spectrum organophosphate insecticide that is used globally for crop protection and pest control and as many other active ingredients of pesticides is applied formulated into a suitable product. The objective of this study was to investigate the impact of the technical active ingredient (a.i.) chlorpyrifos and its four commercial formulations (Dursban® 480 EC, Pyrisimex ® 480 EC, Pyrifos ® 480 EC, Nurelle® D) on the acetylcholinesterase activity in snail *Helix aspersa* and earthworm *Eisenia andrei*. The difference in sensitivity of tested organisms towards above mentioned pesticides was assessed by the *in vitro* exposures at range of concentrations 5-300 ug/L tissue in the different fractions of organisms homogenates (head and haemolymph in snails; head and whole body in earthworms). The data from the *in vitro* study with the technical active ingredient and formulations showed AChE inhibition in a concentration dependent manner. The most sensitive responses to pesticides formulations exposures were found in *H. aspersa* haemolymph and *E. andrei* whole body homogenate. Among the tested pesticides, the inhibitory effect (based on the IC50s comparison) increased in the following order a.i. < Nurelle < Dursban < Pyrisimex < Pyrifos (earthworm head tissue) and a.i.< Dursban < Nurelle < Pyrifos < Pyrisimex (snail haemolymph). This study showed that the formulated pesticides caused significantly higher AChE inhibition compare to the technical a.i. in both model organisms. The data suggested that the *in vitro* exposure studies have predictive value for sensitivity to insecticides. Risk assessment based on the on active ingredient toxicity might not be sufficient and toxicity testing of both a.i. and commercial formulation provide more realistic reports on the overall ecotoxicological impact of pesticides on sensitive non-target organisms. Keywords: chlorpyrifos, acetylcholinesterase, non-target organism, pesticide

TU147

Ariadna spider as a good candidate bioindicator of heavy metal contamination in the Namib Desert

E. Conti, University of Catania / Department of Biological, Geological and Environmental Sciences; G. Liberatori, G. Liberatori, G. Protano, F. Nannoni, University of Siena / Department of Physical, Earth and Environmental Sciences; G. Costa, University of Catania / Department of Biological, Geological and Environmental Sciences; I. Corsi, University of Siena / Physical, Earth and Environmental Sciences

Human activities are strongly affecting natural ecosystems and native species have been proposed as bioindicators for pollution monitoring and assessment. The present study is the first attempt to use *Ariadna* spiders as indicators of trace metals contamination in the Central region of the Namib Desert. Mining activity is the biggest contributor to Namibia's economy in terms of revenue and several trace elements as well as semi-precious gemstones and minerals are main products. Therefore, their released and potential contamination of specific Namib mining areas cannot be excluded. Various spider populations belonging to undescribed *Ariadna* species are widespread in gravel plains within the Central Namib Desert. Being sit-and-wait predators, *Ariadna* spiders spend their life in individual tunnels dug in the soil, so resembling the behaviour of ground-dwelling spiders known to be strong metal accumulators in terrestrial ecosystems. In the present study, we collected 60 specimens of three *Ariadna* populations (20 spiders from each site) in austral summer 2016, along a N/S and W/E transect at various distances from main mining areas of the Namib Desert. Depth and diameter of entrance burrow and body weight of each spider were recorded. Trace metals analysis were conducted in spider's whole body as well as in soils samples collected around spider's burrow. Oxidative stress parameters, CAT, GST and MDA were analysed in soft tissue of spiders and neurotoxicity assessed by measuring cholinesterases activity (ChE). Entrance diameter and depth of burrow seems to be affected by the distance from mining areas. Levels of Zn, Cd, As and Cu resulted higher in *Ariadna* body compared to levels found in soils around their burrows. On the opposite levels of Pb, V, Cr, Co and Ni were 1 or 2 order of magnitude lower in spiders than in soils. Similar trends in such levels and biological responses as CAT, GST and ChE were observed among sites and based on various distance from the mining area. Such preliminary results support the recognition of *Ariadna* spider as a good candidate as bioindicator of trace metals contamination in Namib Desert.

TU148

Effect of spray drift reduction techniques on pests and predatory mites in orchards and vineyards

D. Fornasiero, N. Mori, P. Tirello, A. Pozzebon, C. Duso, University of Padova / DAFNAE; E. Tescari, Dow AgroSciences Italia srl; R. Bradascio, Dow AgroSciences Italia srl / RD; S. Otto, Italian National Research Council

Spray drift of pesticides has a negative impact on aquatic ecosystems and the environment, including damage to non-target organisms. Particularly, the drift of some insecticides can have detrimental effects on beneficial arthropods such as predatory mites. According to a recent EU Directive, the reduction of spray drift is required for a sustainable use of pesticides, yet without reduction of efficacy against pests. In this framework, eight field trials were conducted from 2012 to 2014 in two typical growing areas of Verona district (Northern Italy), four on apple orchards and four on vineyards. The aim of these trials was to evaluate, for two spray drift reduction techniques: 1) the spatial patterns of in-field droplets, 2) the efficacy against key pests on apple and grape (Cydia pomonella and Lobesia botrana respectively), 3) the side effects on predatory mite populations. Four insecticides: chlorpyrifos, chlorpyrifos-methyl, methoxyfenozide and spinetoram, were applied with three different spraying techniques: high-drift nozzles (Albuz, ATR 80 yellow), low-drift nozzles (Albuz, TVI 80015 green), and high-drift nozzles with an anti-drift adjuvant (rapeseed oil). Results showed that the two spray drift reduction techniques effectively increased droplets amounts next to sprayer, reducing potential drift on both apple orchards and vineyards and were generally as effective as standard nozzles without additional side effects on beneficial arthropods. Results suggest that the use of spray drift reduction techniques such as low-drift nozzles and anti-drift adjuvants can be effective in managing key pests and also in decreasing the environmental impact of using insecticides. Full article in: Crop Protection 98 (2017) 283-292, DOI:10.1016/j.cropro.2017.04.010.

Microplastics in freshwater and terrestrial systems - fate, monitoring and biological interactions (P)

TU149

Freshwater organism can recognize microplastics as microplastics

S. Kim, Y. Chae, D. Kim, Konkuk University; Y. An, Konkuk University / Department of Environmental Heath Science

The plastics are slowly weathered into nano- (< 100 nm) and micro- (< 5 mm) sized particles owing to physical, chemical, and biological processes in the environment. In the present study, we observed the behaviour of freshwater organisms and evaluated whether they recognize and response to microplastics. Adult zebrafish was exposed to the different concentration of microplastic (MP, 250-300 μm) and food materials (F) (20 mg MP, MP20; 20 mg F, F20; 10 mg MP + 10 mg F, MP10F10). The behaviour patterns were recorded and quantified. Diving beetle fed the adult zebrafish, which exposed under MP10F10 and F20 conditions, and the ingestion rate was quantified. The number of capturing patterns were counted as 21±4, 8±5, and 14±3 under F20, MP20, and MP10F10 conditions, meanwhile the spitting patterns were determined as 0.0±0.0, 2.8±1.3, and 0.2±0.4, respectively. Ingestion rate of diving beetle on control group was calculated as 0.63±0.10 zebrafish wet mg/sec. The exposure group, which fed the MP10F10 exposed zebrafish, showed the significant decreasing ($p < 0.05$) of ingestion rate (0.55±0.08 zebrafish wet mg/sec) during 591±85 seconds. On diving beetle, the MP were only found at crop organ until 720 min after ingestion, and did not transfer to another organ. The digestive organs, especially crop, seemed to separate the microplastic as indigestible food. We concluded that the freshwater organisms recognize the microplastic, and exhibit the defence behaviour *This research was supported by Basic Science Research Program through the National Research Foundation of Korea (NRF) funded by the Ministry of Science, ICT, and future planning (2016R1A2B3010445).*

TU150

Microplastic shedding from functional textiles

C. Jönsson, Swerea IVF AB / Energy and Environment; S. Schellenberger, Stockholm University / Department of Applied Environmental Science (ITM); P. Mellin, Swerea KIMAB AB; O. Levenstam, University of Borås; A. Hanning, Swerea IVF AB; S. Roos, Swerea IVF AB / Energy and Environment

Microplastic pollution of marine environment is an environmental issue which is intensely discussed on a global level. Synthetic based textiles contribute to microplastic pollution of the marine environment. Besides littering and the size-shape effects that microplastics have when being exposed to humans and animals they provide an additional vector for chemical pollutants, i.e. possibly providing a new entering mode into organisms of pollutants already existing in the sea. But fibers generated from consumer articles such as textile garments might carry chemical pollutants due to different chemical treatments. This study investigates the fiber loss of polyamide (PA) and polyester/cotton blend (PES/CO) textiles that were functionalised with durable water repellent (DWR) treatment. The chemical treatment consists of polymers that are based on per- and polyfluoroalkyl substances (PFAS). *Question 1*: Do we have release of fluorinated fibers from functional textiles? *Question 2*: What is the amount of fluorinated fibers lost during the washing which can have an impact on the environment? The microscopic investigation identified particles as well as fiber fragments generated during the simulation of industrial washing (Gyro wash). Fiber fragments were identified with REM and the EDX analysis of showed fluorine (F) as part of the fiber surface composition. The results will be further verified using combustion ion chromatography (CIC) of shedded fibers with and without DWR treatment. These results will then be used to model a scenario simulating for instance Swedish consumption and use of DWR treated outdoor garments and the total exposure of fluorine contaminated fibers to the environment, from washing via waste water treatment to the recipient. This study proved that functional textiles can contribute to the release of microplastic pollution due to the formation of synthetic fiber fragments during washing. The presence of fluorine in the fiber surface composition confirmed that these fibers still contained traces of the functional DWR treatment. Released into the environment these fibers might be exposed to long term degradation processes which would finally cause the formation of persistent environmental pollutants. In addition the results of this study suggest that similar mechanism might be relevant for textile containing other functional coatings such as flame retardants, softeners or dyes as well.

TU151

Fate of 14C-labelled Calcium Poly(styrene sulphonate) (CaPSS) Microplastic in waste water treatment at environmentally relevant concentrations

M. Hüben, Fraunhofer IME / Ecological chemistry; D. Hennecke, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Ecological chemistry; M. Simon, Fraunhofer IME / Applied Ecology; M. Kruse, Fraunhofer IME, Institute for Molecular Biology and Applied Ecology; A. Kroll, GAB Consulting GmbH; A. Häusler, GAB Consulting GmbH / Environmental Fate and Modelling; E. Wischerhoff, Fraunhofer IAP Institute for Applied Polymer Research

Wastewater is one of the exposure pathways of microplastic into the environment. Microplastic enters the wastewater e.g. as an ingredient of cosmetics or from specific pharmaceutical applications. Wastewater treatment plants (WWTPs) are generally considered to remove microplastic from the wastewater stream and to protect the receiving river. However, there is not much information to prove this assumption experimentally at environmental relevant concentrations. This is due to the fact that so far no sensitive detection systems are available to analyse organic polymers in a complex sludge matrix at such low concentrations. In view of these limitations, the aim of this work was to determine the fate of a model polymer, crosslinked polystyrene sulfonate (PSS), in a simulated WWTP using radiolabeled material. PSS is a polymer which is widely used as an ion exchange resin in various applications. The polymer is insoluble in water and is not degraded in the human body. Calcium loaded PSS (CaPSS) was synthesized in a procedure downscaled from an industrial method with ^{14}C-radiolabelled styrene monomer. This is a key step in the entire project as the radioactivity of the monomer interferes with the polymerization reaction. The resulting ^{14}C-polymer was characterized by comparison with commercial non-labelled CaPSS to prove success. The ^{14}C-radiolabelling enables detection in sludge matrix as well as the determination of potential water soluble degradation products and $^{14}CO_2$ from mineralization. A mass balance was established to identify the most relevant processes for the fate of CaPSS in WWTPs. Due to the high sensitivity of ^{14}C-detection, the test can be performed at realistic/environmentally relevant concentrations. As the detection limit of ^{14}C-microplastic in environmental matrices is currently orders of magnitudes below that of non-radiolabelled polymer particles, this study can serve as an example how future studies on the general topic “microplastics in the environment” can be supported.

TU152

Microplastics in the environment: Evaluating the risks and identifying knowledge gaps

E.E. Burns, University of York / Chemistry; A. Boxall, University of York / Environment Department

The past ten years has seen increasing scientific and public concern over the harmful effects of microplastics (MPs) in the natural environment. In 2010, < 10 scientific publications contained the word ‘microplastic’ while this number had risen to around 170 in 2016. Alongside this, there have been significant policy and regulatory developments around the use and emissions of MPs. We present the results from a systematic review of the published literature to attempt to answer the question ‘what is the evidence that microplastics adversely impact freshwater and marine systems?’ In answering this question, we explore the evidence-base for a number of assertions made by the broader community around MPs in the environment. We have summarized the global coverage of microplastic occurrence studies in both aquatic and sediment compartments. We found that many of the occurrence studies employ unsuitable analytical confirmation methods which may lead to high error rates and limit data interpretation. In many ecotoxicology studies, effects were not seen at the highest concentrations investigated while others reported impacts on molecular level endpoints, feeding, fish eggs, reproduction, growth, tissue inflammation and mortality. Studies have also assessed the potential for MPs to act as a vector for hydrophobic organic compounds to accumulate in organisms. No conclusive evidence was found in the literature to support this theory, instead most studies exploring this effect disprove the hypothesis while a few are inconclusive due to flaws in the experimental design and interpretation. Comparison of monitoring and effects data indicates that concentrations of MPs currently detected in the environment are orders of magnitude lower than those where effects/no-effects are observed in the laboratory. Moreover, enough ecotoxicity data is now available to begin building species sensitivity distributions. We demonstrate that based on current data concentrations of MPs measured in the environment are not high enough to elicit the effects reported from laboratory studies. There is however a mismatch between the size ranges and types of MPs used in laboratory ecotoxicity tests and those detected in the environment. There is an urgent need to address this mismatch by performing better quality and more holistic monitoring studies alongside environmentally relevant effects studies. Only then will we be able to determine whether these materials are having real impacts or not.

TU153

A cost-effective methodology for separation of microplastics from freshwater systems

M.O. Rodrigues, Department of Biology & CESAM - University of Aveiro / Department of Biology; F. Gonçalves, University of Aveiro / Department of Biology and CESAM; H. Nogueira, Universidade de Aveiro / Department of Chemistry; J.C. Marques, University of Coimbra / MARE, Dep. of Life Sciences, Coimbra University; A.M. Gonçalves, MARE, Dep. of Life Sciences, Coimbra University/Biologia Department & CESAM, Aveiro University; N. Abrantes, University of Aveiro / CESAM/DAO

Plastics, one of the most demand material worldwide, are considered one of the most emerging aquatic pollutantsdue to their ubiquity, high persistence and insufficient management. Especially, microplastics (< 5 mm) are of scientific and social apprehension as they can reach high densities, derive from a variety of sources and can interact with biotic and abiotic environment. Currently, the concerning about the occurrence of microplastics (MPs) in freshwater systems has been increasing, notwithstanding there is no unified method for MPs separation in these systems. This result in inaccuracy data that differs in quality and resolution, not allowing data comparison between different studies (large-scale spatial and temporal comparisons). Hence, this work aims to assess the effectiveness of distinct separation methods as an attempt to identify and establish the most cost-effective method. For that, artificial samples containing eleven plastics belonging to the most common types of polymers (e.g. low/high-density polyethylene, polypropylene, polystyrene, polyvinyl chloride, polyethylene terephthalate) were prepared (secondary MPs) and subjected to distinct methods. These methods included density separation methods using sugar, olive oil and zinc chloride, as well as organic matter degradation methods usinghydrogen peroxide (wet peroxide oxidation) and multienzymatic detergent (enzymatic digestion). The samples were then undergoing the detection, quantification and identification of polymers using a dissection microscope and Fourier transform infrared spectroscopy (FTIR). Several criteria were considered in order to achieve the aims of this work: efficiency of density separation and organic matter degradation, the total mass of recovered polymers, cost of each procedure, the time spent with each method, the simplicity and the quality of recovered polymers. Based on this multi-criteria approach, this study concludes that the wet peroxide oxidation with addition of zinc chloride was the most cost-effective method. This method should be used in future studies of monitorization of MPs in aquatic systems, notwithstanding the use of hydrogen peroxide must be cautious and only applied when necessary.

TU154

Applicability of remote sensing methods for indirect mapping of microplastic distribution within aquatic ecosystems

S. Piehl, University of Bayreuth / Department of Animal Ecology I; E.C. Atwood, RSS Remote Sensing Solutions GmbH; M. Bochow, Helmholtz Centre Potsdam GFZ German Research Centre for Geosciences; J. Franke, RSS Remote Sensing Solutions GmbH; F. Siegert, Ludwig Maximilians University of Munich / Department of Biology; C. Laforsch, University of Bayreuth

Recently, there have been intensified research efforts to get reliable information about sources, sinks, and transportation pathways of microplastic in aquatic environments. Due to the high spatiotemporal variability of these systems, our knowledge of those aspects is still limited. Earth remote sensing is a key technology within the field of environmental monitoring, providing a unique tool for large area observationsof water parameters such as suspended particulate matter, chlorophyll-a or colored dissolved organic matter. Since floating microplastic is probably influenced by the same transport mechanisms as non-motile plankton or particulate matter, we tested the hypothesis of a spatial correlation between microplastic and specific water parameters. In situ water parameters, microplastic (5mm – 250 µm), and in situ derived spectral reflectance measurements (ASD FieldSpec) were taken during field campaigns at three different river mouths (Trave and Elbe in Germany, Po in Italy). Microplastics from surface waters were sampled with a manta trawl. Organic material was removed by enzymatic digestion and wet peroxide oxidation, and the remaining microplastics were analyzed down to plastic type with FT-IR as well as SWIR spectroscopy. Initial regression analysis results comparing microplastic with water parameters will be presented, and their suitability as indicators for microplastic abundance is discussed. Furthermore, simultaneously obtained remote sensing data for the river Elbe will be linked with in situ derived microplastic data and water parameters. If significant relationships between microplastic and water parameters exist, remote sensing of water parameters as an indicator for microplastic abundance would provide a cost-effective monitoring tool, able to cover much larger areas than feasible with ship-based point measurement.

TU155

Coastal accumulation mapping of microplastic particles emitted from the Po River, Italy: Integrating remote sensing, in situ sample collections and ocean current modelling

E.C. Atwood, RSS Remote Sensing Solutions GmbH; F.M. Falcieri, CNR - ISMAR; S. Piehl, University of Bayreuth / Animal Ecology I; M. Bochow, Helmholtz Centre Potsdam GFZ German Research Centre for Geosciences; M. Matthies, University of Osnabrueck / Institute of Environmental Research; J. Franke, RSS Remote Sensing Solutions GmbH; S. Carniel, M. Sclavo, CNR - ISMAR; C. Laforsch, University of Bayreuth; F. Siegert, RSS Remote Sensing Solutions GmbH

Plastic pollution in inland waters and the open ocean is a long recognized problem for marine wildlife, coral reefs, the fishing industry and shipping transport safety. Microplastics, defined as particles < 5 mm, form a considerable portion of this pollution and have recently received increasing public attention following recent discoveries that not only can these particles be ingested by planktonic animals, but also outnumber natural food items in some ocean areas. Ingested particles can induce negative survival effects as well as serve as introduction vectors for accumulated persistent organic pollutants (POPs) or carcinogenic plastic additives into the base of the food chain, potentially leading to many seafood products consumed by humans. Research has mainly concentrated on marine systems, and while a growing number of studies focus on freshwater lakes, river systems have to date received little attention. In particular, riverine plumes as an important influencing factor for the input and distribution of microplastics into coastal ocean areas remain largely unexplored. Here we present a study of the accumulation of microplastic particles emitted by the Po River along the Adriatic coastline in northern Italy. We posit that river-induced coastal microplastic accumulation can be predicted using a hydrodynamic model, supported by remote sensing data from Landsat and Sentinel-2A. Model accumulation maps were validated against *in situ* sampling at 9 beaches (analyzed particle size range: 1-5 mm). Hydrodynamic modelling suggests that the amount of discharged particles is only semi-coupled to beaching rates. Object tracking revealed that beaching of emitted particles was strongly mouth dependent and relatively low (less than 25% of all released particles from a given river mouth), primarily occurring within the first five days. The southernmost Po River mouth posed an exception, where more released particles (94%) were found to beach over an extended period of time and along a longer stretch of coastline. Comparison with remote sensing based accumulation maps and validation against *in situ* beach sampling are discussed. The presented methodology lays the groundwork for developing an operational monitoring system to assess microplastic pollution being emitted by a major river and its distribution along adjacent coastlines as well as into the open ocean.

TU156

Cause and effect of the plastic industry in South Africa as a developing country

C. Verster, North-West University - School of Biological Sciences / Environmental Sciences and Development

In 2017, the South African plastic industry has grown with 1.9%, compared with 2016 (Plastics SA). Although legislation is in place to promote recycling and sustainable use of natural resources, the recycling of plastic based materials is done predominantly by corporate initiative. Many South Africans believe that the country is lagging in terms of recycling. South Africa has however achieved a recycling rate of 41.8% in 2016, of which most is done post-consumer. Plastics SA has set an ideal of no plastics to landfills by 2030, and plans are being set in place to achieve this goal. Inadequate waste disposal infrastructure and protocols, especially in informal settlements causes large quantities of unrecycled plastic to end up in aquatic systems and subsequently in the marine environment. Beaches surrounding estuaries are heavily polluted with macroplastics which calls for greater prevention and clean-up efforts. Much effort is spent corporately to reduce South Africa's 'plastic footprint', but efforts in terms of microplastics are trailing. We collected and filtered 46 fresh water samples from various localities in and around Gauteng, the most densely populated province in South Africa. High levels of plastic pollution were found in almost all samples. Up to 40 plastic particles (> 20 µm) per litre was found in surface water of the Vaal River, a major river in the country's largest drainage basin flowing through industrialised areas. These levels are comparable to high levels of microplastic pollution found in European rivers. The growing plastics industry in South Africa requires excellence in clean-up and recycling to reduce the negative impacts on the environment and create a viable plastic sector.

TU157

Understanding the distribution and fate of microplastics in a tertiary sewage treatment plant in the UK

R.M. Blair, S. Waldron, University of Glasgow; C. Gauchotte-Lindsay, University of Glasgow / Infrastructure and Environment; V. Phoenix, University of Strathclyde / Civil and Environmental Engineering

Microplastics (MPs; < 0.5 mm) are classified as contaminants of emerging concern but currently are not regulated by water quality standards. Microplastics are highly diverse and their distribution in the environment is highly variable in space and time, making their quantification and risk assessment difficult. Further, their monitoring and regulation are hindered by limited empirical data, particularly of fresh- and wastewater systems as important pathways of land-based contaminants to oceans. Here, a study was conducted in a tertiary sewage treatment plant in the UK (Glasgow, Scotland) to assess the presence of MPs in the system and the effect of treatment stage in removing these contaminants before discharge into recipient freshwaters. The MPs were extracted from 5-L samples from each treatment stage, using H_2O_2 digestion and vacuum filtration through 1.2 µm GF filter. Characterisation and quantification of MPs was carried out by light microscopy followed by detailed chemical analysis of representative subsamples via SEM-EDS and FTIR-ATR. Microplastics were present in wastewater samples collected at all treatment stages, and abundances generally decreased from inflow to outflow. Further, high variability in MP abundances was evident across sampling dates. Chemical characterisation by SEM-EDS revealed that 94% of analysed pieces were C-based materials, but only 25% were confirmed plastics based on FTIR-ATR results. In conclusion, the tertiary treatment process evaluated here efficiently removes MPs entering the system but small quantities may still be discharged into the environment. Further, visual characterisation with light microscopy may result in overestimation of MPs due to misidentification of cellulose and other microdebris as plastics. Therefore, analysis of chemical composition through a combination of detailed analytical techniques is crucial for improved accuracy of results. This study contributes to understanding what methods are needed to extract and identify MPs from environmental samples, and the spatio-temporal data generated provide understanding of what needs to be monitored and where controls should be implemented.

TU158

Weathering-induced changes in the effects of microplastic particles and their leachates

A. Jahnke, Helmholtz Centre for Environmental Research - UFZ GmbH / Cell Toxicology; C.D. Rummel, Helmholtz Centre for Environmental Research GmbH - UFZ / Department of Bioanalytical Ecotoxicology; D. Kühnel, Helmholtz-Centre for Environmental Research / Bioanalytical Ecotoxicology; M. Schmitt-Jansen, UFZ - Helmholz Ctre Environm. Research / Department of Bioanalytical Ecotoxicology; B. Escher, Helmholtz Centre for Environmental Research GmbH - UFZ / Cell Toxicology; M. MacLeod, ITM - Stockholm University / Department of Environmental Science and Analytical Chemistry

Studies on the potential effects of microplastic (MP) particles in the aquatic environment are numerous. However, many laboratory studies apply spherical, pristine particles, which may be of limited relevance given UV light irradiation, mechanical stress, salinity, biofilm growth and other factors that can influence the weathering of the particles in the environment. The overall aim of the JPI Oceans-funded project WEATHER-MIC is to assess the impacts that weathering has on the transport, fate and effects of MP particles and their leachates. We summarize recent results on potential effects. **(1.) Impact of MP particles on organisms**: We have exposed copepods, daphnia and algae to different fractions of virgin and weathered MP as well as particle-free leachates under controlled conditions. From the observation of apical endpoints in the acute toxicity assays, concentration-response relationships for the different fractions can be deduced. A critical evaluation of the suitability of the applied test protocols for the assessment of adverse effects of MP will be presented. **(2.) Influence of ageing plastic and leachates on biofilm structure and function:** Natural biofilms (containing bacteria, algae and fungi, embedded in extracellular polymeric substances) grown in microcosms on different types of aged and pristine polymeric substrates have been studied to observe the influence of weathering on the attachment and succession of biofilms. Sum parameters (biomass, pigment profiles, photosynthesis) and sequencing data were studied. **(3.) Mixture effects of leachates from the most common polymers:** Cell-based bioassays have been applied to study mixture effects of additives and degradation products of the polymers liberated during weathering of plastic material in artificial seawater in agitated UV chambers. The chemicals in the seawater leachates were enriched by solid-phase extraction or chemicals were directly extracted from pristine particles by ultrasonic-assisted solvent extraction. The concentrated leachates and solvent extracts were then dosed into cell-based bioassays, covering i) cytotoxicity; ii) activation of metabolic enzymes, e.g. via binding to the arylhydrocarbon receptor; iii) specific, receptor-mediated effects such as estrogenicity; and iv) adaptive stress responses such as oxidative stress. The results may help to understand effects caused by additives and parent compounds opposed to the degradation products liberated from the UV-weathered plastic.

TU159

Occurrence and characteristics of fine microplastics in sewage water, domestic water, sewage treatment water and river water by coagulation and FT-IR microscopy method

Y. Kameda, Chiba Institute of Technology / Creative Engineering; N. Yamada, T. Yasuda, Chiba Institute of Technology

The occurrence of Microplastics (MPs) is of great concern in aquatic environment, especially ocean. Many current studies evaluate MPs ranged from 100 µm to 5 mm. However, MPs used in personal care products and other industrial processes are reported to be smaller according to previous reports. Because MPs are very important substances to current economic activities, new materials for MPs will be needed such as cellulose. Though alternative of the materials is mainly conducted in personal care products, effect of the volunteer actions and various regulations on decrease of MPs in aquatic environments has not been evaluated. It is necessary to reveal their sources such as sewage water, sewage treatment water, wastes in aquatic environments and so on. This research shows occurrence and characteristics of MPs of more than 10 µm size in sewage water, sewage treatment water and river water by coagulation and FT-IR microscopy method developed in our laboratory. MPs in the various contaminated water were collected by a plankton net whose mesh size is 10 µm. The collected particles were separated by a density separation method. After that, MPs in the collected particles were separated by coagulation process. Finally, the MPs were passed through a membrane and were identified by stereoscopic microscope for larger MPs as well as finer MPs by FT-IR microscopy. MPs are characterized by materials, size, color and multi-regression analysis by FT-IR spectrum data. Based on these data, contribution of MPs from personal care products to total MPs concentration will be discussed as well as estimation of sources of MPs in various water samples.

TU160

Detection of micro-paint particles and microplastic in harbour soil samples using FPA-µFTIR-Imaging-FTIR

A. Vianello, Aalborg University / Civil Engineering Department - Section of Water and Environment; M. Simon, N. van Alst, F. Liu, Aalborg University / Civil Engineering Department; K.B. Olesen, Aalborg University / Department of Civil Engineering; J. Vollertsen, Aalborg University / Civil Engineering Department

Microplastic (MP) pollution is nowadays recognized as a global concern affecting both marine, freshwater and also the terrestrial environment. Beside microplastic pollution, also micro-paint particles (MPP), originated by the degradation and abrasion of painted surfaces (buildings, ship-paint materials) have been reported in some recent studies. As many paints also contain organic compounds and heavy metals used as biocides aimed to inhibit the growing of biota on painted surfaces, these particles could be even more harmful to the environment and wildlife than microplastics. Here we present some results obtained from the analysis of soil samples collected in a recreational boat facility in the North of Denmark. A “Microplastic-based” approach has been used, extracting the particles from the soil matrix using air-assisted density separation followed by FPA-µFTIR-Imaging analysis. This approach allows to identify and quantify microplastics and micro-paint particles down to 10-20 µm in size. Surficial soil samples were collected along three transects located in different areas of the shipyard. The samples, previously sieved (5000–500 µm and 500-10 µm) were submitted to flotation using $ZnCl_2$ followed by sample cleanup using enzymes and H_2O_2 oxidation to remove organic matter. The analysis was carried out using FPA-µFTIR-Imaging spectroscopy and the data were processed with a dedicated software (MPHunter) developed at Aalborg University. The first results highlighted a high micro-paint and microplastic particles contamination. The total MP and MPP concentration were 222,500 particles Kg^{-1}, while the estimated mass was 17.1 mg Kg^{-1}. The most abundant polymers/paints detected were polyester (30%), acrylic coating (32%), alkyd coating (10%) and polyethylene (7%). The particle size distribution showed the most abundant size ranges were between 20 - 40 µm and 40 - 80 µm. The high MP and MPP concentration measured in the sample highlights the harbor areas are potential hotspots for the accumulation and further spreading of MP and MPP in the terrestrial and aquatic environment. Microplastic and micro-paint particles were successfully extracted and detected in a recreational harbor area using a state of the art analytical approach, including multi-step sample preparation and FPA-µFTIR-Imaging analysis, the most suitable spectroscopic technique for an unbiased MP detection and quantification.

TU161

Runoff of microplastics from agricultural soil: a study in a semi-arid area

R. Hurley, NIVA - Norwegian Institute for Water Research; T.C. Schell, IMDEA Water Institute / Ecotoxicology; A. Rico, IMDEA Water Institute / Aquatic Ecotoxicology; M. Vighi, IMDEA Water Institute / Earth and Environmental Sciences; L. Nizzetto, NIVA

More than 90% of microplastics (MPs) present in raw wastewater are captured by wastewater treatment plants and much of this is incorporated into the sludge phase. Therefore, the use of sludge as a fertiliser for agricultural soils may be a relevant source of MPs in the terrestrial ecosystem and, through runoff, in surface waters. The fate of MP applied with sludge is strongly dependent on weather conditions (rain, temperature, etc.). The objective of this study was to determine the fate and distribution of MPs in the agricultural soil-water system. The study was performed in an experimental farm of IMIDRA (Instituto Madrileño de Investigacion y Desarrollo Rural, Agrario y Alimentario) located in central Spain, in an area characterised by semi-arid climate: hot and dry summer, low rainfall (about 450 mm per year) which is concentrated in spring and autumn. Suitable devices for runoff collection (modified Pinson collectors) were placed on three different plots with different MPs treatments: (i) soil never treated with sludge (control), (ii) soil treated with sludge in the past (in 2013), and (iii) soil treated with sludge at the start of the experiment (November 2017), according to usual agricultural practices. Besides sludge application, soil characteristics (composition, texture, etc.) were comparable in the three plots. Sludge was applied early November and the plots were sown with barley. After each relevant rainfall event, runoff water was collected and filtered in-situ and to isolate the MP fraction. Soil samples were taken in all plots at the start of the experiment, as well as 3, 6 and 12 months after the start of the experiment. To determine the vertical MP transfer within the soil, soil cores were divided into three fractions (0-5, 5-10, 10-15 cm). Separated plots receiving the same sludge treatments and soil conditions were used to sample earthworms and to study potential accumulation and MP impacts over the soil fauna. The content of MPs in runoff water, soil and biological samples were extracted using organic matter digestion (soil and organism samples), density separation (soil samples), and filtering (all samples). MPs were identified visually and characterised chemically using FTIR. Preliminary results of this experiment, which can be used to quantify fluxes and emissions of MPs in agro-ecosystems under semi-arid conditions, are presented.

TU162

Microplastics in wastewater and freshwaters: a case-study in the Henares river watershed (Central Spain)

T.C. Schell, IMDEA Water Institute / Ecotoxicology; R. Hurley, Nowegian Institute for water research; A. Rico, IMDEA Water Institute / Aquatic Ecotoxicology; M. Vighi, IMDEA Water Institute / Earth and Environmental Sciences

Wastewater is considered to be one of the major sources of microplastics (MPs) entering surface waters. Although a high retention potential of wastewater treatment plants (WWTPs) for MPs in raw wastewater has been reported, the removal efficiency seems strongly dependent on particle characteristics and treatment processes. MPs not retained by WWTPs are directly discharged into the aquatic environment through WWTP effluents, whereas the majority of MPs are assumed to be retained and accumulated in the sewage sludge. Runoff, after application of sewage sludge to agricultural fields, may consequently serve as an additional source of surface water contamination by MPs. Therefore, the aims of this study were (i) to evaluate the occurrence of MPs in surface waters, and (ii) to ascertain the MP contribution of WWTP effluents. The study was carried out in the Henares River watershed (Central Spain). Five WWTPs with differing dimensions (population equivalents between 10,000 and approx. 400,000), differing influent types (domestic, industrial, or both mixed), and differing treatment processes were selected. Wastewater inflow, outflow, and sludge (humid and dried) were sampled during two different seasons (summer and autumn). In addition, river water and sediment samples were taken in three different seasons (spring, summer, and autumn) at three differently impacted sites: i) low human impact; ii) high agricultural impact; and iii) high mixed impact (urban, agricultural, industrial). MPs in river water and wastewater were divided into four fractions by filtering a suitable amount of water through plankton nets of different mesh sizes (from 300 to 20 mm). In order to assess the MP concentration and composition in the samples, solid substrates (sludge and river sediment) were subjected to an organic matter removal treatment, followed by density extraction. Subsequently, those samples, as well as the liquid samples (river and wastewater), were filtered onto filter papers to visually identify the MP content and then chemically characterize their polymer composition using FTIR spectroscopy. Finally, the most relevant characteristics of the watershed (total population, characteristics and location of WWTPs, agricultural land use patterns including sludge application, etc.) will be integrated into the study’s findings to deduce their importance for MP contamination. The preliminary results of the distribution and characterization of MP in different matrices are presented.

TU163

Microplastics occurrence and composition in drinking water from a Norwegian urban area

a. gomiero, International Research Institute of Stavanger / Enviromnent; G. Skogerbø, IVAR; K. Øysæd, A. Vatland Krøvel, International Research Institute of Stavanger

Microplastics as a potential health and environmental problem has gained increasing attention recently. Microplastic is defined as plastic pieces smaller than 5 mm in diameter, and the sources of microplastic are many. State of the art literature reports that microplastics are ubiquitous worldwide. While several authors report fragments of different polymers being observed practically in all environmental compartments of marine, freshwater and terrestrial ecosystems; others point out the accumulation of micro- and nanometric sized plastic particles thorough the marine and terrestrial food webs posing the risk of marine and terrestrial life and ultimately the human health. Despite of these studies point out the occurrence of micro plastics in freshwater systems including surface and groundwater basins, very little in know about the occurrence of microplastics in the drinking water and their implications on human health. According to the WHO men should consume 3 L and women should consume 2.2 L of beverage per day. Most of these beverages consist of tap water, or drinks derived from tap water (such as coffee, tea, or reconstituted juice). The risk of plastic uptake from drinking water is currently unpredictable and furthermore, these plastic particles are in addition to plastics potentially consumed in other sources, such as sea salt, beer, and food and seafood. The research tastks of the present work were: Map published and available literature, develop and optimize a standardized fast, sensitive protocol for sampling and quantification of nano/microplastics particles in drinking water and finally. Analyze and possibly

detect microplastic particles in drinking water supply systems with special focus on different polymeric composition and size fractions. Study area was the Rogaland area (Norway) populated by approx 110.000 inhabitants. Samples of drinking water were collected every two week for ten months contemporary from the supply water system collection point as well as in different sites of the urban area. A fast and sensitive method based on a GCMS-pyrolysis was developed. Polyethylene, Polypropylene and Polyvinyl chloride were the most recurrent polymers. Levels ranged from 0,02 to 16 ng/L. Time and space related trend are presented.

TU164

Macro and Micro(plastics) in the Environment of Some French rivers

V. Verney, CNRS -ICCF / Photochimie-CVP; G. BISSAGOU KOUMBA, UCA-ICCF; F. Delor Jestin, Sigma-ICCF; Z. Dominink, H. Askanian, CNRS-ICCF; J. Peiry, E. Roussel, O. Voldoire, CNRS-Geolab; A. Schaal, L. Durantou, Observatoire du Microplastique; M. Liboiron, Memorial University of Newfoundland

It is now known that the vast majority of microplastics found in the seas and oceans originate from lands. In such a process freshwater environment (rivers and riverbanks) play a major role. It is therefore necessary to imagine the scenario that a used plastic, becoming a waste after use, will be found in the environment if it has escaped to a waste treatment stream. Its stay in the environment can persist for a very long time and this waste will then be exposed to a set of environmental constraints (UV, rain, wind, mechanical erosion, ..) which will continue and amplify its degradation, leading to its fragmentation The work we have undertaken consists of: 1. Mapping the density, the mobility and the chemical composition over time of macroplastics present on the banks of an experimental site of the Allier River, and linking it to the density of the vegetal areas. 2. Set up on site a controlled pollution to follow its fate along the time 3. Analyze the composition of microplastics extracted from the sediments, especially at the entrance of the abandoned channel, where it may exist some vortices of flow. These first three points are the topic of the Plasticcages project supported by the CNRS[1, 2]. 4. Collect and analyze the composition of microplastics in the surface waters of different french rivers (Allier, Charente, Loire, Touvre, etc.). To do this, we rely on citizen science operations, in particular thanks to the contribution of the babyleg sampling net [3,4], which makes it possible to multiply samples and analyzes. *1 Occurence of plastic litters in the Allier river in France. Vincent Verney,Gaëlle Bissagou Koumba, Alexandre Garreau, Florence Delor-Jestin, Erwan Roussel, Olivier Voldoire, Jean-Luc Peiry; To be published 2- https://www.researchgate.net/project/PLASTICCAGES 3- Compromise agency, the case of babylegs, Max Liboiron, Engaging Science, Technology and Society 3(2017), 499-527 4- http://lapagaiesauvage.org/laboratoirecitoyen/*

TU165

Spatial and temporal trends of microplastics in an urbanized Canadian river

M.S. Ross, T. Bujaczek, S. Kolter, MacEwan University / Department of Physical Sciences; D. Locky, MacEwan University / Department of Biological Sciences

Microplastics are ubiquitous contaminants in the marine environment, but quantitative data on their presence in the freshwater environment is sparse. This study investigates the occurrence, composition, and potential sources of microplastic contamination in the North Saskatchewan River, an urbanized river flowing through the city centre of Edmonton, Alberta, the fifth largest city in Canada. Surface water samples were collected monthly during the summer of 2017 using plankton nets with 53µm mesh. Samples were collected from seven sites throughout the city, including sites both upstream and downstream of the city and potential point sources (i.e., a wastewater treatment plant, WWTP). Microplastics were found in all samples, and at some sites concentrations exceeded those reported in other urbanized rivers. Various coloured fragments, films, beads, and fibers were identified, with the majority of microplastic contamination being in the form of fragments. Both the total concentrations and the proportion of each type of plastic varied with distance downstream and size class, suggesting changing inputs as the river flows through the city. No differences in either concentration or composition were found between sites upstream and downstream of the WWTP, suggesting a lack of significant input to the river. Moving forward, the chemical composition of suspected microplastics will be investigated using Raman microspectrophotometry. This work represents one of the first studies on the occurrence of microplastics in the freshwater environment in Western Canada and will provide a baseline for future monitoring studies.

TU166

A Historical Sediment Record of Microplastics in an Urban Lake, London, UK

S. Turner, University College London / Geography; A.A. Horton, Centre for Ecology and Hydrology; N. Rose, University College London / Department of Geography

A historical record of microplastics extracted from a radionuclide (^{210}Pb and ^{137}Cs) dated sediment core from a London lake provides novel data on the long-term deposition of microplastic waste in freshwater systems. Microplastic particle abundance and calculated accumulation rates are indicative of plastic usage in the 20th century. Concentrations of microplastics extracted from the sediment, by sieving and floatation using dense liquid separation range from 30 to 880 particles per kg of dried sediment. Fibres dominate the assemblage of microplastic particles identified in the time-resolved sediment samples. Polystyrene microplastic particles were identified and are found in post-1950s sediment and up to the present day. An increase in microplastic concentration is evident in recent sediments (post 2000) but a peak in concentration is also observed in late 1960s-1970s age sediment. Raman spectroscopy of selected particles and fibres provides compositional data on the fibres and particles found in the sediment. The size and nature of microplastic particles found in the sediments, as well as the historical geography of the catchment, suggests that atmospheric deposition has been an important vector of plastic transport to the lake. Microplastic analysis of temporally well-resolved lake sediment sequences will greatly assist in quantifying the historical flux of microplastic waste into the environment and should be included in future analyses to enable calculation of catchment-based budgets of microplastic contamination.

TU167

Microplastics from sewage treatment works and storm water outfalls discharging into the Victoria Harbour, Hong Kong SAR

C. Mak, Y. Tsang, The Chinese University of Hong Kong / School of Life Science Environmental Science; K. Chan, The Chinese University of Hong Kong / Life Sciences

We conducted surveys of microplastic pollution in the surface waters and sediments from Deep Bay, Tolo Harbour, Tsing Yi, and Victoria Harbour in HKSAR. In the microplastics survey (June 2015 to March 2017), the averaged concentrations of microplastics in local coastal waters and sediments respectively ranged from 51 to 27,909 particles per 100m^3 and 49 to 279 particles per kilogram. The highest concentration of micro-plastics (coastal water) was recorded as 35,642 particles per 100m^3 in March 2016 in Victoria Harbour (West Kowloon). Therefore, we also study the effluents directly discharged from chemical enhanced primary sewage treatment works (Stonecutters Island STW) and secondary sewage treatment works (Sha Tin STW), and two stormwater outfalls (SWOs) (Kwun Tong Ferry Pier,New Yau Ma Tei Typhoon Shelter),which are potential microplastic pollution sources entering into the Victoria Harbour. Effluent samples from each of these sewer systems were collected in three weekdays per month and different seasons (December, March, June and September) to determine spatial, temporal (seasonal) variations of microbeads in treated sewage and stormwater discharges.The average concentrations of microbeads present in effluents from STWs and SWOs respectively ranged from 137,239 to 1,081,597 particles per 100m^3 (December 2016 to March 2017) that consider as moderate emission level. Biological samples (fishes and mussels) are also collected in two SWO for the assessment of microbeads abundance and composition in its digestive system. Microplastics of different shapes from sewage and biota (mainly fragments, lines, fibres, and pellets) were identified by means of Attenuated Total Reflectance-Fourier Transform Infrared Spectroscopy. Zebrafish exposed to microbeads individually would ingest different sizes of polyethylene microbeads (10-22µm,45 to 54µm,90-106µm, 212-250µm & 500-600µm) and their digestive tracts and gill filaments were fully occupied by microbeads.Mixtures of microbeads in environmental related concentrations are used for expression profile of cytochrome P450 1A1(CYP1A1) and vitellogenin 1(VTG1) studies. Our objectives in microbeads exposure experiments in zebrafish adult (*Danio rerio*) are 1) the upper and lower size boundaries for microbeads ingestion (ingestion range:10 to 600µm), 2) amount of microbeads accumulated inside the digestive tracts, and 3) expression profile of oxidative stress-related gene (CYP1A1) and endocrine-related gene (VTG1).

TU168

Models for Data Synthesis, Sampling Design and Scenario Analysis: Some examples using the INCA-MP model of microplastic fate and transport in soils and surface waters

M. Futter, Swedish University of Agricultural Science / Aquatic Sciences and Assessment; J. Crossman, University of Windsor; J. Ledesma, V. Russo, E. Lannergård, SLU Swedish University of Agricultural Sciences / Aquatic Sciences and Assessment; L. Nizzetto, NIVA

Quantification and classification of microplastics in soils, sludge and surface waters is both time consuming and expensive. Ideally, measurement campaigns can be focussed on areas that are likely to provide the greatest returns on effort yet this is often difficult to accomplish in practice. Here, we show how INCA-MP, the Integrated Catchments model for Micro Plastics, the first published model of microplastic terrestrial fate, riverine transport and contaminant co-transport can be used to synthesize available data, identify knowledge gaps, plan monitoring, and perform risk assessments. Synthesizing available data involves collation of microplastic and proxy data. We show how proxy information, including timing and rate of biosolid application, event-based measurements of field erosion and high frequency water quality monitoring can constrain estimates of microplastic mobility in terrestrial and freshwater environments. Through the application of uncertainty analysis in INCA-MP, it is possible to identify the most sensitive pools and processes when making predictions of microplastic fate and transport. Furthermore, knowledge gaps related to these pools and processes can then be targeted for more intensive field sampling campaigns. As INCA-MP includes sophisticated routines for tracking the environmental fate of current and emerging micropollutants, the model can be used for risk assessment of co-transported contaminants. Such information is needed both for more targeted sampling campaigns and for communicating risks associated with microplastics in terrestrial and freshwater environments. We illustrate these concepts using data from Swedish and Canadian catchments. Our results show the importance of autumn storms and spring snowmelt for microplastic mobilisation to surface waters and highlight the potential knowledge gains associated with targeted sampling of riverine sediments, constructed wetlands and waste treatment facilities.

TU169

Occurrence and concentration of microplastics in an urban river

C. Campanale, C. Massarelli, G. Bagnuolo, Italian National Research Council; V. Uricchio, Italian National Research Council / Water Research Institute

The term 'microplastics' was first used in 2004 to describe very small fragments of plastic (~ 50 μm) in the water column and in sediments. In 2009, Arthur et al. proposed that microplastics should include all fragments < 5 mm. Over the past decade, microplastic debris in both marine and freshwater systems has become an emerging environmental issue. Although 70 – 95 % of the marine litter, including microplastics, come from land environment, studies of microplastics in freshwater systems are limited respect to those focused on marine habitats. Rivers and inland waters may transport microplastics to marine habitats and may be a novel vector for the downstream transport of organic persistent pollutants suggesting an overlooked and potentially significant component of the global microplastic life cycle. Herein we report results from a monitoring study with the main objective of evaluate the occurrence and concentration of microplastics in an italian urban river and assess the hypotheses that microplastics amount could vary in response to temporal and seasonal trends. In order to monitor the trend of microplastic concentrations, two seasonal sampling campaigns have been planned (February and April 2017). Superficial waters samples were collected with three surface plankton nets fixed in the middle of the river simultaneously for two different time slot (11:00-13:00 and 13:00-15:00) for a total of six replicates for each campaign. After sample extraction and purification, validation of visually based microplastics identification was achieved using pyrolysis-gas chromatography-mass spectrometry (Pyrolysis GC-MS). The composition of microplastic was studied in term of size, shape, color and polymer type. Results from the six replicates are expressed as mean values (± DEV. ST.) of number of particles per cubic meter (p/m^3). Microplastics were found in each net sample for a total amount of 22152 items collected, photographed, enumerated and categorized. Sample concentrations ranged from 3.52 to 13.43 p/m^3 showing significantly higher abundances during February than April campaign (Mann−Whitney U Test = 18.00; p-value = 0.028). A total of five polymer have been characterized: PE, PP, PS, PVC and TDI-PUR. All samples contained at least three polymer types: PE, PP and PS. PE accounted for 77% of the total particles identified, followed by PS (12%), PP(10%), PVC (0.9%) and PU (0.4%).

TU170

Removal of 10-500 μm microplastics from wastewater effluent by disc filter

M. Simon, Aalborg University; N. van Alst, Aalborg University / Civil Engineering Department; K.B. Olesen, Aalborg University / Department of Civil Engineering; F. Liu, J. Vollertsen, Aalborg University / Civil Engineering Department

In this study the efficiency of a disc filter to remove microplastic particles from wastewater effluent was evaluated. The size range of particles addressed was 10-500 µm and the identification technology was micro-FTIR imaging spectroscopy applying a focal plane array (FPA). Effluent wastewater was collected at the wastewater treatment facility at Grindsted, Denmark operated by Billund Spildevand A/S. The treated wastewater was sampled before and after the disc filter by filtering on site onto 10 µm stainless steel meshes using a custom made large-scale water sampling device. The filtered volume of effluent wastewater before the filter was 200 L and 1.6 m^3 after the filter. The residue collected on the filters containing a mixture of organic matter, inorganic particles and microplastics was subjected to a purification procedure including enzymatic digestion, chemical oxidation and flotation in order to eliminate the sample matrix and extract the microplastics. Non-degradable particles were stored in ethanol, and a fraction of the ethanol particle suspension transferred to a transmission window to quantify particles by infrared imaging technique. The entire window was scanned to create a mosaic with 3.3 µm pixel resolution on the FPA. The spectra of all particles in a scan were analyzed to quantify their chemical composition and to determine whether they were of plastic, and if so, of which plastic material. The spectral analysis was carried out with a semi-automated IR spectra analyzer software developed at Aalborg University, Denmark. The size and shape of plastic particles were recorded and their mass was estimated. Preliminary data shows that the removal efficiency of the disc filter was 96 % in terms of both mass and particle number. The material composition of plastic in the sample before the filter (polyesters, polyethylene and acrylates) was somewhat dissimilar to the composition in the effluent sampled after the filter (polyester and polystyrene).

TU171

PlasticBudget - Project on the environmental assessment of microplastic emissions

N. Thonemann, Fraunhofer Institute for Environmental, Safety, and Energy Technology UMSICHT

Shortly after the introduction of many types of plastics (e.g. polystyrene, polyethylene terephthalate, polypropylene) in the 30s, 40s and 50s, first traces of plastics in the environment have been detected. Packaging, pellets and parts of a kitchen sponge were found in the stomachs of seabirds; Whales and seals were caught in polypropylene cords. By weathering and fragmenting larger plastic objects (macroplastic) into smaller pieces (microplastics), plastic waste in the environment seems to be gradually disappearing. However, as recent research shows, microplastic is found in freshwater, on beaches and in open water, in the deep sea and in the Antarctic ice. It is taken up by organisms and passes on in the food chain. The long dismantling periods suggest that microplastic and plastic waste will accompany, if not outlast, mankind for a long time to come. Although the number of publications on microplastics has risen in the last two decades and the topic has entered the social discourse, there are still many research gaps on sources, pathways, amounts, sinks, accumulation spaces, adsorption and absorption of pollutants as well as damaging effects on organisms and humans. The project PlasticBudget is aiming to close some of the above-mentioned research gaps. Taking into account the relevance that plastic litter has gained in recent years in the environmental discussion, the assessment of the environmental impact of those emissions is needed. Macro- or microplastics' emissions have an impact on ecotoxicity (for example, when birds or fish confuse plastic with food) and human toxicity (for example by eating food, which contains microplastics). The resulting environmental impacts could be, for example, the extensively documented dying of marine organisms by microplastics, the danger of massive aggregation of tiny plastic particles in the food chain, or even the negative aesthetic impact associated with plastic in the environment. Corresponding midpoint and endpoint indicators and associated characterization methods, as well as standardization to a reference value (e.g. by the production volume of the specific plastic), are therefore developed in the PlastikBudget-project and discussed in expert dialogues as well as workshops.

TU172

How do we know that microplastics are different from natural particles in their effects on biota?

Z. Gerdes, M. Ogonowski, E. Gorokhova, Stockholm University / Department of Environmental Science and Analytical Chemistry (ACES)

Microplastics (MP) have been identified as a potential environmental hazard, which has motivated a wide range of effect-studies, testing different combinations of polymers, sizes and shapes. However, risk assessment of MP exposure, in the lower size range < 100um, is today hampered by both the lack of data regarding their presence in the environment and the inadequate experimental design of many effect-studies. A crucial issue in designing such studies is to include control particles that are ubiquitously present in the environment and represent a background variability with regard to suspended solids. Such controls, allow effects caused by plastic particles to be separated from those induced by particles, *per se*. This separation is crucial for testing MP-specific effects, as many test organism are suspension-feeders that have evolved in turbid aquatic environments. To highlight and provide examples of the importance of particle controls in MP-effect studies we will present: I) results of a review on effects of particle suspensions (MP, or natural nutritionally inert particles), and II) case studies employing control particles. The data synthesis strongly suggest that particle controls are essential for identification of MP-specific effects, so that MP impacts can be assessed based on ecological soundness.

TU173

Influence of environmental conditions on the sorption of organic pollutants to microplastic particles

S. Seidensticker, J. Lamprecht, P. Grathwohl, Eberhard Karls Universität Tübingen / Center for Applied Geoscience; C. Zarfl, University of Tuebingen / Center for Applied Geoscience

The ubiquitous contamination of all environmental compartments with microplastic particles is extensively discussed in both science and public. Large numbers of microplastics have been measured in effluents of wastewater treatment plants. These particles might act as sorbent and transporter for frequently occurring wastewater contaminants and are hence a factor that needs to be considered if the environmental fate of pollutants is examined. Some contaminants, among those also micropollutants like pharmaceuticals, can be charged under certain pH conditions. While it is known for a wide variety of natural particles that charged compounds sorb only little or not at all, sorption interactions between microplastics and charged compounds have not been analysed so far. Thus, the aim of this study was to clarify the sorption behaviour of dissociating compounds to microplastic particles. We measured the equilibrium partitioning between 19 typical wastewater contaminants (pharmaceuticals, personal care products, pesticides) and microplastics at three different pH values (4, 7, and 10). The investigated compounds showed a wide variety in their physico-chemical properties, e.g. a log K_{OW} range between 0.1 and 5.8 and pK_a-values from 1.6 to 13.9. We performed batch experiments with fourteen ionizable and five non-ionizable substances. In all experiments equilibrium was reached after two days. Measured log K_{PE} for the neutral species ranged from 0.75 to 4.00. The uptake of contaminants varied according to their hydrophobicity. Sorption of ionizable substances is strongly influenced by the pH while non-ionizable substances showed a partitioning independent of pH. For sorption into polyethylene, the amount of accumulated pollutants is principally dominated by the neutral fraction, while the charged species did not contribute. Thus, with increasing pH sorption of acids decreased while the sorption of bases increased. Whereas electrostatic interactions between charged species and polyethylene could not be detected, this might be different for other polymers, such as polystyrene and polyamide.

TU174
Influence of microplastics on transport of organic contaminants in soil
T. Hüffer, S. Slawek, T. Hofmann, University of Vienna / Department of Environmental Geosciences
The worldwide production and usage of mainly disposable plastic has increased from 1.7 million tons in 1950 to 299 million tons in 2013 [1]. Consequently, plastic wastes are deposited in the environment and persist due to long durability and limited recovery [2]. Polyethylene is one of the mass-manufactured polymers that is found in the terrestrial environment, used in many different sectors, including agricultural mulches, composites and package material [3]. To date, microplastics have been mainly studied in marine and freshwater systems, while there is hardly any data on microplastic occurrence, fate, and effect in terrestrial environments [2]. Low-density polyethylene (LDPE) foils, that may become brittle due to insolation, are used in large amounts on agricultural areas to protect crops, suppress weeds, regulate the temperature and retain irrigation water in the soils [1]. In soil microplastics may affect the transport of hydrophobic organic pollutants and pesticides, as they can be preferentially sorbed by polymer particles with large surface to volume ratio compared to sorption by natural sorbents [1, 4]. The strength of sorption as well as the relevant molecular interactions depend on the properties of the sorbent and the sorbate [5]. The aim of this study was to investigate the influence of microplastics on the transport of organic contaminants in a soil under varying aqueous conditions. [1] K. Duis, A. Coors, Environ. Sci. Eur. 2016, 28, 2. [2] M. Rillig, Environ. Sci. Technol. 2012, 46, 6453. [3] M. Beg, S. Kormin, M. Bijarimi, H. Zaman, Adv. Polymer Technol. 2015, 35, 21521. [4] A. Bakir, S. Rowland, R. Thompson. Mar. Pollut. Bullet. 2012), 64, 2782. [5] T. Hüffer, T. Hofmann, Environ. Pollut. 2016, 214, 194.

TU175
Influence of polystyrene microplastics in combination with organic pesticides on the giant rams-horn snail Marisa cornuarietis: behavioral and biochemical responses
S. Krais, University of Tubingen / Animal Physiological Ecology; H. Schmieg, Tübingen University / Animal Physiological Ecology; E.E. May, University of Tuebingen / Animal Physiological Ecology; A.S. Ruhl, TU Berlin / Department of Water Quality Control; H. Köhler, University of Tubingen / Animal Physiological Ecology; R. Triebskorn, University of Tuebingen / Animal Physiological Ecology
Due to the increasing demand for and usage of plastic products during the last decades, the quantity of globally produced synthetic polymers rises continuously resulting in high amounts of plastic debris of all sizes in the environment. Very small-sized particles and fibers (< 5 mm) which are defined as microplastics result either from degradation of macroplastics or are produced as primary microplastics which are contained e.g. in cosmetics. Microplastics are of particular interest in ecotoxicology, because they can interfere with organic substances like pesticides or pharmaceuticals, transport them into food chains and modulate their toxicities. In addition, they can mechanically affect exposed organisms. Whereas in the past, most of the studies on microplastics have focused on the marine environment, there is still little knowledge about the occurrence and impacts of microplastics in freshwater ecosystems. The aim of this study is to examine possible influences of polystyrene particles in combination with different organic pesticides on the behavior and biochemical responses of the giant rams-horn snail (*Marisa cornuarietis)*. Snails were exposed to 10.000 polystyrene particles per liter (cryogenically milled, < 100 µm) in combination with different concentrations of the pesticides cypermethrin, methiocarb and thiacloprid. In order to quantify the observed behavioral responses, five categories of behavior were defined, which are "crawling", "attached to the wall", "attached above the water surface", "inactive on the ground" and "retracted with closed operculum". All snails were individually categorized twice a day for nine days. The results make evident that snails exposed to cypermethrin significantly changed their behavior between the first (day 1-4) and second (day 5-9) observation period, independent of the test concentration. In the first period, they were often categorized as "attached to the wall" or "attached above the water surface", whereas in the second observation period, these snails were mainly classified as "inactive" or "retracted". As biochemical endpoints we study oxidative stress (lipid hydroperoxides, superoxide dismutase), proteotoxicity (Hsp70 level) and neurotoxicity (inhibition of acetylcholinesterase). The analyses, however, are still in progress. The study is part of the joint research project MiWa ('microplastics in the water cycle') funded by the German Federal Ministry of Education and Research (support code: 02WRS1378).

TU176
Effects of artificial weathering on polypropylene microplastics
V. Fernández-González, Universidade da Coruña / Grupo Química Analítica Aplicada (QANAP),; G. Grueiro-Noche, Universidade da Coru / Grupo Química Analítica Aplicada (QANAP); J.M. Andrade-Garda, Universidade da Coruña / Grupo Química Analítica Aplicada (QANAP); P. López-Mahía, Universidade da Coruña / Analytical Chemistry Department; D. Prada-Rodríguez, Universidade da Coruña / Grupo Química Analítica Aplicada (QANAP); S. Muniategui, Universidade da Coruña / Analytical Chemistry
Microplastics are defined as plastic materials or fragments with diameter below 5 mm. These microplastics occur due to the release of manufactured (primary) microplastics in various products and the breakdown of larger plastic litter (secondary microplastics). The time required for plastic to degrade under natural conditions is estimated to be on the order of hundreds to thousands of years, being photo-oxidation by UV light its primary degradation pathway. The small fragments of microplastic created by weathering are detrimental to ocean ecosystems for multiple reasons. In the frame of the BASEMAN project (JPI Oceans) 9 natural microplastics of different polymers materials, in two size presentations (100-500 µm and pellets ≤1 mm) were artificially weathered. A pilot-scale simulated weathering system (dry conditions and simulated marine conditions), using UV/Vis metal halide lamps, was deployed. This study focuses on the characterization of the changes that an accelerated artificial weathering process produces in polypropylene microplastics using ATR-FTIR. This could be useful to identify real plastics and microplastics found in marine environment, and understand how aging affects the surface and chemical structure of this material. New absorption peaks can be seen, that reveal changes in the main structure of the microplastics. Some indexes were calculated as the ratio of these IR bands to a reference peak that indicate oxidized carbon in the plastic hydrocarbon chain. It is worth to note that the direct identification of the type of polymer is usually not possible. Weathering-related changes in the IR spectra difficult the correct identification of the polymer when are compared with the IR polymer library. Moreover SEM microscopy was also done in order to characterize further the weathering process. SEM images showed that the microplastics experienced mechanical erosion and weathering. These results reveal that an adequate pretreatment of the sample along with an adequate customized polymer library that contains real/weathered polymers spectra are highly recommended for the adequate monitoring of microplastics in the environment. **Aknowledgements**: Program of Consolidation and Structuring of Units of Competitive Investigation of the University System of Galicia (Xunta de Galicia) partially financed by ERDF (ED431C 2017/28). Ministry of Economy and Competitiveness: PCIN-2015-170-C02-01 BASEMAN-JPI Oceans and, project CTM2016-77945-C3-3-R (ARPA-ACUA).

TU177
Freshwater microplastics and effect of conditioning on pollutant and chemical transfer potential
K. Reilly, The University of Birmingham; J. Sadler, The University of Birmingham / Department of Geography and Environmental Science; I. Lynch, University of Birmingham / Geography Earth Environmental Science
The presence of microplastics in the environment has received increasing scientific and societal interest over recent years. Following this, there has been a range of scientific studies and discussions on impacts that microplastics are having in the environment and how we can mitigate this, leading to changes in legislation, although more is needed. Micro, and recently nano, plastics have been shown to have a range of detrimental effects on various organisms in both field and laboratory studies. Effects are typically dose-dependent and include reduced feeding and successful reproduction, change in organism's behaviour and decreased survival. This study aims to build on current research on the effects of freshwater microplastics, focussing on the micro and nano plastics potential to transfer chemicals in the environment. The formation of a corona on plastic particles changes their surface characteristics which could lead to a change in how biota and chemicals interact with the plastics. In this study we compared the effect that plastic conditioned under different scenarios can have on the interaction with *Daphnia magna* (a keystone species). We assessed this using several parameters including: (1) quantifying the proteins and polysaccharides secreted by the organism's before and after exposure, (2) quantifying uptake of plastic in the individual organisms, and (3) observing changes in behaviour (reproduction and movement). We explored these effects for pristine plastics and plastics combined with chemicals commonly found in the environment, including 17α ethynylestradiol and detergents. The effect that competitive binding of biomolecules naturally secreted by Daphnia have on both adsorption and desorption of the target chemicals on the plastic's surface was a key element of this study, to ascertain how chemically contaminated microplastics may be part of a more complex pollution issue in the environment. This study could help to explore the issue of combined stressors using parameters that can be controlled in the laboratory. Through this, we can assess the impact of assumptions about test conditions, and the impact of over-simplification of standardised test media, on the resulting data regarding the Trojan-horse potential of micro and nano plastics. Building on this data we include recommendations to improve the environmental realism of the laboratory conditions to make more accurate exposure assessments for environmental modelling in the future.

TU178
Exposure to conventional but not biodegradable microplastics impacts fitness in Daphnia magna
Z. Gerdes, Stockholm University / Department of Environmental Science and Analytical Chemistry (ACES); M. Puranen, Stockholm University; M. Ogonowski, E. Gorokhova, Stockholm University / Department of Environmental Science and

Analytical Chemistry (ACES)
Conventional, oil-based polymers are considered as the major source of microplastic pollution, whereas biodegradable polymers (bioplastics) have not attracted much attention as sources of microplastics. However, given that production of bioplastics is increasing and that biodegradation is slow under ambient conditions, it is likely that they enter the aquatic environment in the same way as the oil-based plastics. Therefore, it is important to understand potential environmental impacts of both polymer types. We compared effects of exposure to polylactic acid (PLA; bioplastic) and polystyrene (PS; oil-based polymer) on primary life history traits in the crustacean *Daphnia magna*, a standard model species in ecotoxicology. To exclude particle effects caused by food dilution and thus identify microplastic-specific effects, kaolin clay was used as a reference treatment. In total, four treatments were included: PLA, PS, clay (reference), and control (food only). The exposure was conducted over 21 d using a plankton wheel to keep test particles and algae in suspension for comparable exposure concentrations. In the PS treatment, we observed high mortality, decreased feeding rate and reproductive output compared to all other treatments. These effects were not caused by toxic monomers of styrene or additives leaching out of the polymer, which was demonstrated in a follow-up test with the PS leachates. By contrast, no significant effects were found in the daphnids exposed to PLA compared to the reference treatment. Thus, a significantly higher toxicity of the conventional polymer was observed, whereas effects of the biodegradable microplastics were similar to those caused by the ubiquitously occurring clay particles. More studies are needed to identify the mechanisms of PS toxicity and to confirm the observed ecotoxicological differences between the polymer types using different test materials. To evaluate toxicity of these materials, it is essential to include a reference treatment as a benchmark.

TU179

Effects of polystyrene microplastics in different life stages of brown trout (Salmo trutta f. fario)

H. Schmieg, Tübingen University / Animal Physiological Ecology; S. Krais, University of Tubingen / Animal Physiological Ecology; F. Rezbach, University of Tuebingen / Animal Physiological Ecology; A.S. Ruhl, TU Berlin / Department of Water Quality Control; H. Köhler, University of Tubingen / Animal Physiological Ecology; R. Triebskorn, University of Tuebingen / Animal Physiological Ecology
The widespread use of plastic products in our daily life has led to a constant increase in the production of synthetic polymers. In consequence and also resulting from the longevity of plastics, high amounts of plastic debris can be found worldwide in aquatic and terrestrial environments. In general, plastic items smaller than 5 millimeters are defined as microplastics. Primary microplastics are produced for different purposes and are, for example, contained in many cosmetic products. Abrasion and fragmentation of larger plastic items lead to the formation of secondary microplastics. Up to now, most studies investigating effects of microplastics in organisms concentrate on marine ecosystems, whereas knowledge on effects of microplastics in freshwater organisms is still scarce. The aim of our study is to investigate effects of polystyrene microplastics (cryogenically milled granules, fractionated to < 50 µm, up to 100.000 particles/L), also in combination with organic pollutants (pharmaceutical, pesticide), in different life stages of brown trout (*Salmo trutta* f. *fario*). For that purpose, we conducted a fish early life stage test (FELST) according to OECD 212. The endpoints of interest were heart rate, hatching success and mortality. After consumption of the yolk sac by the fish larvae, we additionally investigated the level of oxidative stress by means of the ferrous oxidative xylenol orange assay (FOX-assay). In a second experiment, we examined effects of polystyrene particles (< 50 µm, 10.000 particles/L) alone and in combination with the pesticide methiocarb in juvenile (11 months old) brown trout. In this experiment, the mortality rate, biometric parameters, the level of oxidative stress, the induction of the 70 kD stress protein (Hsp70) and the inhibition of acetylcholinesterase were under investigation. Furthermore, we examined histopathological effects in gills and in guts of the trout. First results showed no effect of microplastics on mortality and biometric values of either larvae or juvenile brown trout. Further analyses are still in progress. The present study is part of the joint research project "MiWa" (microplastics in freshwater systems) funded by the German Federal Ministry of Education and Research (support code: 02WRS1378).

TU180

Daphnids in distress? Acute and chronic effects of primary and secondary microplastics on three species of Cladocerans

G. Jaikumar, CML Leiden University / CML; N. Brun, CML Leiden University / Conservation Biology; J. Baas, Centre for Ecology & Hydrology / Centre for Ecology and Hydrology; M.G. Vijver, CML Leiden University / Conservation Biology; T. Bosker, CML Leiden University / Ceter for Environmental Sciences
Microplastics (< 5mm) are ubiquitously distributed in the environment, causing increasing concern in recent years. The two predominant types of microplastic differ in shape and origin: primary microplastics (PMP) are intentionally produced as micro-particles for commercial applications, whereas secondary microplastics (SMP) are formed by the environmental breakdown of large plastics. Information regarding effects of microplastics on freshwater ecosystems is limited.In the present study, the acute and chronic effects of microplastics on three Cladoceran species, *Daphnia magna*, *Daphnia pulex,* and *Ceriodaphnia dubia*, to both PMP and SMP was assessed. The acute toxicity was assessed at 18o, 22o, and 26o C, to determine the influence of temperature as an additional stressor on toxicity.\nAcute sensitivity of *D. magna* and *D. pulex* to both PMP and SMP, increased sharply with temperature, whereas that of *C. dubia* was stable across temperatures. *C. dubia* was the most sensitive species at 18o, followed by *D. pulex* and *D. magna,* which were of comparable sensitivity, however, the trend was reversed at 26o C. In addition, SMP and PMP had a similar effect on *D. magna* and *D. pulex* but PMP was more toxic to *C. dubia*. \nBoth PMP and SMP showed adverse effects on all three species during chronic exposure. Further, *C. dubia* was the most sensitive species followed by *D. pulex* and *D. magna*. All species were more affected by PMP than SMP during chronic exposure.The results of the current study indicate that exposure to microplastics has adverse effects on health and reproductive output of the species studied, although at relatively high levels of exposure, and that temperature as an environmentally relevant additional stressor has a major influence on species sensitivity to microplastics.

TU181

Evaluation of chronic toxicity of polystyrene microplastics on freshwater mussels

S. Magni, University of Milan / Department of Biosciences; F. Gagne, Environment and Climate Change Canada; C. Della Torre, State University of Milano / Biosciences; C. André, J. Auclair, H. Hanana, Environment and Climate Change Canada / Aquatic Contaminants Research Division; F. Bonasoro, University of Milan / Department of Environmental Science and Policy; A. Binelli, University of Milan / Department of Biosciences
The annual global plastic production follows a positive trend and plastic pollution represents an emerging worldwide issue. In particular, microplastics (MPs), plastic fragment smaller than 5 mm, are potentially dangerous for aquatic community because their ability to be accumulating in the tissues of biota. MPs can reach the aquatic environment through the Wastewater Treatment Plants (WWTPs) or afterwards the degradation of macroplastics. Considering that few studies, especially in freshwater environment, have been conducted about the adverse effects of MPs, the aim of our study is the evaluation of chronic toxicity of these contaminants on the freshwater mussel *Dreissena polymorpha* using a multi-biomarker approach. As MP standards we choose two different beads of polystyrene, one of the most common MP classes detected in the environment, with a size of 1 and 10 µm. On the basis of the daily great release of MPs from WWTPs, we tested the following mixtures (MIXs) of polystyrene MPs: MIX1, which contained 2 millions/L of 10 µm MPs and 2 millions/L of 1 µm MPs, and MIX2, which contained 500,000/L of 10 µm MPs and 500,000/L of 1 µm MPs. Therefore, mussels were exposed for 7 days in static conditions to the MIXs and to related control; every 3 days we collected from each tank the mussels to assess both chronic toxicity and uptake of polystyrene MPs. We evaluated the adverse effects by monitoring end-points of cellular stress, as the activity of antioxidant and detoxifying enzymes, oxidative damage, cyto-genotoxicity and neurotoxicity (analyses in progress). To evaluate the uptake of polystyrene MPs in the exposed mussels, exploiting the reflection of MPs, we collected hemolymph and then fixed the whole soft tissue for cryostat sectioning. We then observed the samples using the confocal microscopy. Despite we found both sizes of polystyrene MPs in the hemolymph and soft tissues of mussels, we did not obtain significant increase of tested biomarkers compared to control, excepted for a significant increase of CAT activity at the end of exposure for mussels exposed to MIX 2. The lack of adverse effects induced by tested MPs could suggests that the toxicity of MPs could be mainly associated to the carrier role of MPs toward chemicals, or involved in metabolic pathways not detectable by our biomarkers. In addition, prolonging the exposure time the MP toxicity could be increased.

TU182

Polystyrene microplastic effects on the lipid peroxidation and antioxidant capacity in non- and temperature-stressed individuals of Dreissena polymorpha

A. Weber, N. Jeckel, C. Weil, S. Umbach, Goethe University Frankfurt am Main / Aquatic Ecotoxicology; N. Brennholt, German Federal Institute of Hydrology / Biochemistry and Ecotoxicology; G. Reifferscheid, German Federal Institute of Hydrology; M. Wagner, Norwegian University of Science and Technology / Department of Biology
Microplastic (MP) toxicity has been considered in numerous taxa including bivalves, which are of special interest due to their high filtration activity and therefore MP particle uptake. Previous studies in marine bivalves reported stress and inflammation processes in response to high levels of MP exposure, while data on freshwater species is missing. Therefore, we analyzed the effects of irregular polystyrene MP (< 63 µm) on the lipid peroxidation and antioxidant capacity in the freshwater bivalve *Dreissena polymorpha* both in a single and multiple stressor exposure regime. We exposed *D. polymorpha* to polystyrene MP at concentrations between 6.4 and 100,000 p mL^{-1} over 6 weeks at 16 °C. After the exposure, the midgut gland tissues were analyzed for malondialdehyde concentrations as indicator for lipid peroxidation (TBARS assay, thiobarbituric acid reactive substances) as well as for the remaining abundance of hydrophilic, non-enzymatic antioxidant substances (ORAC assay, oxygen radical absorbance capacity) – an estimate of the remaining antioxidant capacity. The analysis of lipid peroxidation

and antioxidant capacity did not indicate any increased stress levels in response to chronic MP exposure in *D. polymorpha*. In addition, the same experiment performed in a sub-chronic exposure (1, 3 and 7 d) did not reveal stress-induced effects either. Therefore, this study indicates that polystyrene MP does not induce a stress response in *D. polymorpha* in the current exposure scenario. In a more environmentally realistic scenario, bivalves will experience other stressors (e.g. increased water temperature) besides particulate matter. Thus, we hypothesize that a temperature-induced stress response can be modulated by MP exposure. To explore such a scenario further, we will present results from ongoing multiple-stressor experiments in which we expose *D. polymorpha* to MP at 16, 24 and 28 °C.

TU183

Tissue Translocation of Polystyrene Micro- and Nanoparticles in Daphnia magna?

C. Schuer, Goethe University Frankfurt / Dpt. Aquatic Ecotoxicology; S. Rist, DTU (Technical University of Denmark) / Department of Environmental Engineering; N.B. Hartmann, Technical University of Denmark (DTU) / DTU Environment; M. Wagner, Norwegian University of Science and Technology / Department of Biology

The last decade has seen a surge in research investigating various aspects of micro- and nanoplastics originating from plastic pollution in aquatic ecosystems. Aspects include occurrence, uptake, and potential effects in biota. Working with particles in a laboratory setting bears its own kinds of challenges, some of which had already been faced by researchers in the realm of nanotoxicology. Our knowledge about biota-particle-interactions is still limited and often based on early studies that – due to the infancy of the field – may have deficiencies in the experimental design and quality controls. One such example relates to the potential of plastic particles to cross the gut epithelium and translocate to other tissues. This phenomenon has been reported in the literature for the freshwater cladoceran *Daphnia magna* and – if true – is of toxicological relevance. To substantiate the limited available data, the aim of our study was to replicate these findings. We exposed neonate daphnids in a number of scenarios regarding particle concentration and exposure duration at two independent geographical locations using animals from two separate cultures. We expanded on the previous experiment by improving imaging through the addition of a fructose-based-clearing followed by investigation through confocal laserscan microscopy. We additionally applied the lipophilic dye nile red to localize lipid droplets. This step facilitated the identification of lipid droplets inside the tissue and could therefore associate fluorescence detected before staining to a respective tissue. Our findings potentially challenge previous publications that reported the translocation of both micro- and nanoparticles. This discrepancy may be based on false-negative results on our side or false-positive results in the earlier reports, both potentially caused by inadequate exposure settings during the investigative parts of the studies. We were unable to replicate these findings implying a tissue translocation of nano- and microplastics under conditions closely resembling those reported in the literature. This highlights that the replication of nano- and microplastics studies is important, especially if these have a high impact on the body of knowledge. Our study also demonstrates that attempts of replication are inhibited by a lack of transparency in reporting methodology and results. We were able to adapt a fructose-based clearing protocol to the use with high amounts of *Daphnia* samples.

TU184

Do terrestrial organisms, isopods Porcellio scaber and earthworms Eisenia Andrei, avoid microplastic contaminated soil?

A. Jemec Kokalj, University of Ljubljana, Biotechnical Fac. / Department of Biology; P. Zidar, University of Ljubljana / Department of Biology Biotechnical Faculty; G. Kalcikova, University of Ljubljana / Faculty of Chemistry and Chemical Technology

Microplastics (MP) can potentially enter the terrestrial environment *via* sewage sludge deposition on agricultural land. In some countries plastic bags are used as soil cover in home gardens and agricultural land to act as mulch film. Due to fragmentation of these plastic bags, secondary MP may enter the soil and can be further transported along the soil column by bioturbation. Despite the potential presence of MP in terrestrial environments, data regarding the effects of MP on terrestrial organisms are very scarce. In this study, we investigated if terrestrial isopods *Porcellio scaber* and earthworms *Eisenia andrei* avoid soil contaminated with microplastic. We tested microplastic extracted from facial scrub and fragmented from plastic bag. The mean size of MP from facial cleanser was 0.137 ± 0.051 mm, while the plastic bag MP was larger 8.80 ± 5.05 mm (with 62% of particles smaller than 5 mm). Microplastic was mixed into the soil at environmentally relevant concetration 4 mg/g dry weight (0.4 % w/w). The isopods were exposed individually and in groups of 10 animals. The test container comprised two identical polypropylene vessels connected with a fixed polypropylene tunnel to enable animal migration between the two vessels and covered with a lid. In individual exposure the position of each animal was recorded 10 times within the 48 h exposure period and the number of positions on each side was calculated. In group exposure, the isopods were inspected only after 48 h and the number of animals at each side was recorded. Earthworms were exposed in one test container that was divided when applying the control soil and MP contaminated soil. Before the animals (10) were placed into the test container the divider was removed. The number of animals on each side of the soil was counted after 48 h of exposure. Our results indicate that isopods show no preference or avoidance behaviour towards facial scrub microbeads or plastic bag microplastic contaminated soil. This was shown for both types of exposures, individual and group. On the other hand, earthworms clearly avoided the facial scrub contaminated soil but were not affected by plastic bag MP. It remains to be investigated how longer exposures to MP would affect the behaviour of terrestrial organisms. Also it is of interest how environmentally aged MP (e.g. coated with biofilm) would affect the organisms. Knowledge in this field is important to assess the potential hazard of microplastic deposited on soil.

TU185

Analysis of the Trojan horse effect of a mixture of microplastics and chlorpyrifos in an aquatic microcosm study

L. Deermann, Research Institute gaiac / gaiac - Research Institute for Ecosystem Analysis and Assessment; X. Chen, University College London; T. Strauss, Research Institute gaiac / gaiac - Research Institute for Ecosystem Analysis and Assessment; H. Hollert, RWTH Aachen University / Institute for Environmental Research

Microplastic particles (MP) are of concern in the aquatic environment because of their increasing amounts in production and release into the environment. Beside their physical adverse effects, MP can sorb hydrophobic chemicals, which can then be transported together into biota by the so called ‘Trojan horse effect’. In this study, a higher-tier study with a complex ecosystem community was performed with the aim to discover the Trojan horse effect by means of a laboratory aquatic microcosm study. The insecticide chlorpyrifos (CPF) was used sorbed to 5 µm polystyrene microbeads. Beside the control microcosms (C), an MP-control (MPC) group was treated with 4 mg MP/L. For two other treatment groups, the same concentration of MP was coated with nominal CPF concentrations of 0.5 µg/L (L) and 5 µg/L (H) in the water phase before application. With six replicates per treatment, all 24 aquaria constained 16 L water and a 3 cm sediment layer, both taken from outdoor ponds. The natural plankton community got enriched by the amphipod *Crangonyx pseudogracilis*. After a pre-treatment period of five weeks, the experiment run for eight weeks. A chemical analysis of CPF in the water phase of the stock solutions and the treatment groups L and H (day 14) was performed. Since CPF could not be detected in neither of them, a strong sorption of CPF to MP is indicated. Abundances of *Daphnia pulex* revealed higher population increments in MPC than in C, L and H, indicating higher reproduction rates in the first two weeks after application. Furthermore, body lengths of juvenile D. pulex remained nearly constant during the test period in all MP treatments (MPC, L, H) while they increased in the controls (C). Interpreting these results, MP might have led to higher reproduction rates as a stress response which were lowered when CPF was present. In this case, CPF must have become bioavailable to *D. pulex* after ingestion of MP. For *C. pseudogracilis*, total abundances increased the most in MPC, whereas L and H developed similar as C. As for the cladoceran, MP might have led to higher reproduction rates that were lowered by CPF. The Trojan horse effect has probably occurred since CPF could not be detected in the water phase but probably became bioavailable to at least two different species, according to the lowered abundance levels in L and H compared to MPC. Though, the reason why the presence of MP (without CPF) might have led to enhanced abundance levels still needs to be clarified.

TU186

Microplastics exposures of fish: internalization and effects on behavior and growth

c. vignet, Eawag / UTOX; X. Cousin, IFREMER / Laboratoire de Ressources Halieutiques de La Rochelle; R. Behra, Eawag / Department of Environmental Toxicology; L. Joassard, IFREMER; L. Sgier, Eawag; M. Bégout, IFREMER / Laboratoire de Ressources Halieutiques de La Rochelle; J. Cachot, University of Bordeaux / EPOC; K. Schirmer, Eawag / Environmental Toxicology

Awareness of the presence of microplastics, i.e. plastic particles ranging in size from 1 µm to 5 mm, in marine and freshwaters has recently risen but detection and quantification is challenging. Furthermore, whether they pose a risk to aquatic organisms is not yet clear. Fish, for example, have been demonstrated to ingest microplastics particles but the link between quantification of uptake and impact assessment has not yet been made. In this context, we are exploring methods for particles quantification upon feeding juvenile fish with regular food and microplastics and assess whether exposure impacts behavior and growth. For quantification of uptake, we hypothesized that it is possible to analyze the fish tissue by flow cytometry in combination with viSNE, which allows the 2D clustering of particles with different features according to the fluorescence measured. Exposure experiments were carried out for up to three weeks, using different types of microplastic particles and a wide concentration range. In the flow, when particles were mixed with fish tissue, flow cytometry/viSNE was able to differentiate particles natures, numbers and sizes. About 10% of added particles were internalized by the fish from all particles that floated or settled on the bottom. Particles ingestion resulted in a slight impact on behavior. Yet, floating particles were massively incorporated by the fish and significant numbers remained even after 24h of depuration. Based on this, we are currently exploring if continuous

feeding with microparticles contaminated food has consequences on juvenile fish growth. Taken together, our study demonstrates the power and limits of flow cytometry/viSNE for microplastics quantification in a complex biological matrix like fish. The setup could be extended to other types and forms of microplastics in different environmental matrices. Moreover, our study sheds light on ecological consequences that microplastics exposure might have on fish.

TU187

Microplastic ingestion by fish: a comparison of Thames Estuary and Firth of Clyde populations

A.R. McGoran, Royal Holloway; P.R. Cowie, Field Studies Council Scotland; P.F. Clark, The Natural History Museum; J.P. McEvoy, D. Morritt, Royal Holloway

This study compares the ingestion of microplastic by pelagic and benthic fish populations from two major UK watersheds: the Thames Estuary and the Firth of Clyde. A total of 760 fish from 20 species and 116 brown shrimp, *Crangon crangon*, were sampled. Individuals were examined under a dissection microscope and potential plastics were removed to be later identified by FTIR analysis. Out of 21 species, including both fish and shrimp, sixteen species from different trophic levels ingested plastics. Overall, between 33–47% of fish ingested plastic, mostly fibres (83% of potential plastics; before FTIR analysis). In addition, microplastics were also found in the stomach of a common prey species, *C. crangon*, but had ingested far less plastic than predatory fish species, such as the European flounder, *Platichthys flesus*. In the Firth of Clyde, benthic, flatfish ingested significantly more plastic than pelagic fish and other benthic fish. This may indicate that, in estuarine systems, plastics accumulate in the sediment.

TU188

Polystyrene microplastic uptake and effects on feeding behaviour and reproduction in the cladoceran Daphnia magna

B. De Felice, Università degli Studi di Milano; R. Bacchetta, University of Milan; P. Tremolada, University of Milano / Deprtment of Biomolecular Sciences and Biotecnology; M. Parolini, University of Milan / Department of Environmental Science and Policy

Plastic contamination is a well-known environmental problem as demonstrated by the huge presence of plastic debris ranging different sizes in diverse aquatic ecosystems worldwide. In recent years, the attention has been attracted to microplastics (MPs), small plastic particles (dimensional rangeDaphnia magnaaffecting food intake, growth and reproduction. First, we performed a 24 hours uptake and 48 hours release test to assess the ingestion and elimination rate of MPs in daphnids. Already after 1 hour of exposure we found that MPs fill up the digestive tract of daphnids at all the tested concentrations. On the other hand, release test demonstrated that even after 48 hours in a clear medium MPs were still found massively in the digestive tract of treated individuals. The lack of a complete release of MPs can cause the blockage of the digestive tract and starvation, leading the crustacean to the death. Moreover, these effects can negatively affect body growth, swimming activity and, consequently, have strong consequences on reproduction, as suggest by a standard 21-day reproduction test.

TU189

Uptake of differently sized microplastics in gut passage by different species of Daphnia

S. SUPIAN, University of Birmingham; I. Lynch, University of Birmingham / Geography Earth Environmental Science; J. Sadler, The University of Birmingham / Geography Earth Environmental Science

Plastic-debris, for example from textile weathering and washing, are increasingly accumulating in aquatic environments, and while they are now recognized as environmental pollutants, their impact on aquatic ecosystems is not yet fully understood (Jemec, 2016). Microplastics, which are synthetic polymers with a diameter smaller than 5 mm and extending down to the nanoscale, have a widespread occurrence and negative effects on different trophic levels have been described (Hollman, 2013). The freshwater crustaceans Daphnia inhabit all kinds of aquatic systems and forms part of the plankton community acting as algae grazers (filtrators) and predators (raptors) making them an important indicator species in the foodchain. The Daphnia family includes species ranging in size from *D. magna* (2.3-5.0 mm) to *D. galeata* (1.3-2.0 mm) which span a similar range of sizes as mciro and nanoplstics, thus suggesting that different members of this family may be differentially sensitive to or affected by different sizes of micro or nano plastics. This work presents a first analysis of the effect of Daphnia body and gut size on uptake of microplastics of different sizes. We investigated the ingestion and effects of polybead carboxylate microspheres (0.1, 1.0 and 10 µm) on freshwater cladocera of different body sizes (*D. magna*, *D. pulex* and *D. galeata*) after 24, 48, and 72 hours exposure to a range of mass concentrations (also compared on the basis of particle number) (Nasser, 2016). The hypothesis tested was that the size of microplastic particles preferentially taken up by the organisms will scale with organism size, due to differences in their gut sizes and filter feeding capabilities. We assessed the uptake, accumulation, and depuration of the microplastics in Daphnia species using stereomicroscope (Nikon SMZ800) measurements. Image analysis approaches (ImageJ and Matlab) were utilised to calculate gut area, and pixel density within the gut, in an effort to quantify particle uptake. This was correlated with fluorescence measurements using fluorescently-labelled microplasticss, corrected for the average fluorescence per particle. Rapid accumulation in the gastrointestinal tract was observed after exposure to all particle sizes in *D. magna*, with the smaller particle sizes being detectable in the guts of neonates of all three species. The comparative uptake and effects of polybead microplastics on *D. magna*, *D. pulex*, and *D. galeata* are presented here for the first time.

TU190

Determination of microplastics in mackerel stomachs by enzymatic digestion and µFTIR

G. Grueiro-Noche, Universidade da Coru / Grupo Química Analítica Aplicada (QANAP); V. Fernández-González, J.M. Andrade-Garda, Universidade da Coruña / Grupo Química Analítica Aplicada (QANAP); P. López-Mahía, Universidade da Coruña / Analytical Chemistry Department; D. Prada-Rodríguez, Universidade da Coruña / Grupo Química Analítica Aplicada (QANAP); S. Muniategui, Universidade da Coruña / Analytical Chemistry

Plastic is one of the most used materials in the world and is one of the most common and persistent pollutants in the oceans. In fact, plastics constituent 60-80% of marine litter. A particular fraction of plastic debris are the microplastics (particles ?5 mm). The presence and accumulation in the ocean is cause for concern for several reasons, one of the most important is that they can be ingested by marine biota [1]. Different studies have shown the effects on the biota, such as intestinal blockage, decreased mobility or death [2]. Microplastics can absorb persistent bioaccumulative and toxic compounds from seawater. Once ingested, the absorbed pollutants may be transferred to the respective organisms. A variety of methods has been developed to measure microplastics in biota. One important aspect of these analytical methods is the extraction of microplastics from interfering biomass. Many studies have employed one or more chemicals (KOH, H_2O_2) to dissolve the biomass, which can be destructive to the plastic particles and their surfaces and create interferences that were problematic for µ-spectroscopy-based analyses. Enzymatic digestion methods have been used to minimize damage to plastics [3]. An enzymatic digestion has been developed and optimized for digesting biological material without destroying microplastics. Different times and enzymes were tested to optimize the enzymatic protocol. In addition, the enzymatic protocol was compared with chemical digestion (KOH) for the treatment of mackerel stomachs. The optimized enzymatic protocol has been applied to isolate and quantify the microplastics debris present in the mackerel stomach. Identification and characterization of microplastics was done by µFTIR. **Acknowledgement:** Financial support is acknowledged to the Program of Consolidation and Structuring of Units of Competitive Investigation of the University System of Galicia (Xunta de Galicia) potentially co-financed by ERDF (ED431C-2017/28) and by the Ministry of Economy and Competitiveness (subproject PCIN-2015-170-C02-01 EU-Funded BASEMAN (JPI Oceans) and, project CTM2016-77945-C3-3-R (ARPA-ACUA). **References**: [1] V. Hidalgo-Ruz, L. Gutow, R.C. Thompson and M. Thiel, Environmental Science & Technology 46, 3060 (2012). [2] M. Cole, H. Webb, P. K. Lindeque, E.S. Fileman, C. Halsband and T. S. Galloway, Scientific Reports 4, 4528 (2014) [3] J. Wagner, Z-M. Wang, S. Ghosal, C. Rochman, M. Gassel and S. Wall, Anal. Methods, 9, 1479 (2017)

TU191

Microplastic contamination of the model system Weser-National Park Wadden Sea: an across-ecosystem approach

S. Moses, University of Bayreuth / Animal Ecology I; M. Loeder, I. Schrank, C. Laforsch, University of Bayreuth

For the first time worldwide, in the joint project PLAWES the pollution with microplastics of a large European river basin will be investigated on the example of the model system Weser-National Park Wadden Sea. PLAWES, as a pioneer study, is going to conduct an interdisciplinary and ecosystem overarching analysis concerning the contamination with microplastics from the headstreams to the North Sea, thereby considering exemplarily major point (e.g. wastewater treatment plants, combined sewer systems) and diffuse (drainage, atmosphere) sources and entry routes. The new insights are going to be included in a new modeling concept for the identification of primary transport mechanisms and accumulation zones of microplastics. Effects of microplastics on ecosystems of the Weser-Wadden Sea system will be investigated on both, aquatic invertebrates and the interaction of pathogens with microplastics in biofilms. The insights on ecologically relevant aspects are going to be used to assess the environmental effects of microplastics on the model system Weser-National Park Wadden Sea and to transfer these to other systems. Furthermore, the results will be used to develop novel teaching materials to provide an education platform for teachers, pupils and parents across Europe. Hence, PLAWES will generate unique data on the impacts of microplastics on a large European river basin and on environmental health. This will not only be instrumental for decision makers and stakeholders but also serve as focal point to develop science-based solutions.

TU192

Phothochemical fragmentation of freshwater (micro)plastics under UV irradiations

V. Verney, CNRS -ICCF / Photochimie-CVP; G. BISSAGOU KOUMBA, UCA-ICCF; F. Delor Jestin, Sigma-ICCF

We begin to understand and describe more and more the fate of a plastic waste arriving (and remaining) in the aquatic environment. Nevertheless, we still do not know many things, for example, the time scaling of the process from the abandonment of a waste, its arrival, and its persistence in the aquatic environment. During this period, the material will be exposed to various environmental aggressions that will initiate and spread the photoaging of the material. This scenario is accompanied by a physical fragmentation into particles of increasingly smaller sizes, and by a chemical functionalization due to the photo-oxidation of the macromolecular chains. Finally, the increase in both the specific surface area and the chemical functionality may influence strongly the interaction parameters with persistent organic pollutants. We have studied, in simulated laboratory conditions, the fate of various plastics fragments (Polystyrene, Polypropylene and Polylactic Acid) immersed in fresh water and UV irradiated. We worked either with real wastes (from post-consumer sector) or with model polymers totally free of additives. The polymers were chosen for their different physical properties. Polystyrene behaves like a glass (Tg = 104 ° C) at the temperatures of use, which is not the case of the PP (Tg = 0 ° C). Finally, PLA can start hydrolysis reactions. During the exposure time, solids and liquids (a small volume of water) are taken for analysis (melt rheology, ion and liquid chromatography). The main result is that in all cases there is a formation and leaching of short chain (1C, 2C, 3C) carboxylic acids (acetic and formic acids, lactic and glycolic in the case of PLA) all known to be markers of polymer degradation. In the absence of light no transfer of any acid in the liquid is recorded.

TU193

Characterization and Environmental Risk Assessment of Polymeric Cosmetic and Personal Care Ingredients

I. Davies, Personal Care Products Council / Science

Polymers have been developed to perform a variety of functions that are central to modern living. Cosmetic and personal care products (CPCPs) contain a wide array of polymeric ingredients which are identified by the International Nomenclature of Cosmetic Ingredients (INCI). An INCI name often represents several polymers with different physical and chemical properties. This often leads to one INCI named polymer existing as several physical forms. For example, polyethylene can exist as a solid plastic microbead or a non-plastic wax thickening agent. The presence of polymers in the environment, particularly plastics, is of growing concern, yet relatively little is known about the environmental risk these materials may pose or how this can be assessed. The CPCP industry therefore developed a risk-based prioritization framework for polymeric ingredients. Polymers are characterized by their physchem properties. Solid polymers, such as plastics, are prioritized for assessment since they are routinely detected in the aquatic and marine environment and have the highest potential to contribute to environmental litter. Environmental exposure and hazard of priority polymers is then assessed. When a polymer poses an unacceptable environmental risk, risk mitigation options are considered. A polymer's physchem properties provide insights into how environmental exposure and hazard can be assessed. A decision tree was developed linking physchem properties to methods for assessing polymers. Depending on a polymer's properties and how these may change in the environment, polymers are either assessed following existing polymer assessment guidelines (such as those prescribed by USEPA) or use of novel methods. The work presented provides a scientifically robust approach for accurately assessing the impact of polymers in the environment.

TU194

Toxicological effects of irregularly-shaped and spherical microplastics in a marine teleost, the sheepshead minnow (Cyprinodon variegatus)

J. Park, Korea Institute of Toxicology; J. Choi, Korea Institute of Korea KIT; Y. Jung, Korea Institute of Toxicology; S. Hong, Korea Institute of Ocean Science and Technology / Oil and POPs research group

Increasing worldwide contamination of the marine environment with plastics is raising public concern of potential hazards of microplastics to environmental and human health. Microplastics formed by the breakdown of larger plastics and thusare typically irregular in shape. The objective of this study was to compare the effects of spherical or irregular shapes of microplastics on the changes in organ distribution, swimming behaviors, gene expression, and enzyme activities in sheepshead minnow (*Cyprinodon variegatus*). Both types of microplastics were accumulated in the digestive system, causing intestinal distention. However, irregular microplastics decreased swimming behaviors (total distance travelled and maximum velocity) of sheepshead minnow, when compared to spherical microplastics. Both microplastics generated cellular reactive oxygen species, while molecular changes (transcriptional and enzymatic characteristics) of key genes and enzymes, respectively were differed. This study provides insights into environmentally relevant (fragmented) microplastics will help to improve understanding of their environmental impacts. \n Keywords: Microplastics, Sheepshead minnow, Behaviors, Gene expression\n \n

TU195

Assessment of the microplastic contamination in sediments from the French Atlantic coast

N. Phuong, Universite de Nantes; L. Poirier, Univeristé de Nantes / MMS; f. lagarde, Institute of molecules and materials of Le Mans / Institute of Materials and Molecules of Le Mans IMMM UMR CNRS; M. Déniel, Institut des molécules et matériaux du Mans; A. Kamari, A. Zalouk-Vergnoux, University of Nantes / MMS

The ubiquitous presence of MPs has been demonstrated by scientists for recent years. They are detected in all environmental compartments: air, freshwater, aquatic organisms and particularly in marine ecosystems. Sediments are known as the most contaminated environmental compartment. Thus, the aim of this study was to assess the MP contamination in sediment from the French Atlantic coast. Sediments were sampled at three locations (Pays de la Loire region, France) and in two seasons: October 2015 (beginning of autumn) and March 2016 (beginning of spring). Firstly, different protocols (with or without digestion step) were tested according to the literature and the most appropriate was validated spiking 25 g of sediment samples in triplicates with 4 different MP types: polyethylene (PE), polypropylene (PP), polyvinyl chloride (PVC) and polyethylene terephthalate (PET). MP extraction from sediments was performed using milliQ-water combined with centrifugation technic. After a filtration step, MPs were detected and identified directly on the membrane filters using µFTIR spectroscopy in reflection mode.Then, a test was performed in order to determine the replicate number required to obtain a satisfactory representativeness of the whole sampled sediment. For the sediment collected in the field, MPs were found in each location and for each season. Average levels ranged from 38 (± 46.72) to 102 (± 105.37) MP per kg of dry sediment (N = 10; 250 g). Ten different compositions of MPs were defined by µFT-IR with a high proportion of PE and PP, 38 and 23% respectively. Five MP types (PE, PP, PVC, polystyrene and polyester) represented more than 90% of MPs. Interesting information of MP characteristics supported the explanation of the source and also the long-time passed in marine environment. None of significant differences were found among six sample groups. This work provides the first dataset on the level of contamination in sediments from the French Atlantic Coast.

Derivation, Validation and Implementation of Environmental Quality Benchmarks (P)

TU196

Challenges in implementing legal frameworks for assessing water quality : the cases of the EU and Swiss approaches

N. Chèvre, M. Milano, E. Reynard, University of Lausanne / Faculty of Geosciences and Environment

Human activities have a great impact on river quality. Monitoring programs show that multiple chemicals are present in water and that physico-chemical properties and runoff/dissolution capacity of rivers evolve due to climatic changes. These changes can affect the aquatic ecosystems as well as the amount of useable water. It is therefore crucial to evaluate the state of river systems using a holistic approach. The European Commission established a framework to highlight rivers' ecological deficits and to enhance regional or local water management plans. In Switzerland, such a framework is currently under development. In this study, we compared both procedures and implemented them in a Swiss catchment dominated by agricultural activities. The goal was to identify challenges linked to the application of these approaches. Both frameworks highlighted that no section of the river currently meets a good environmental state and that the latter deteriorates as tributaries and wastewater discharge flow into the main riverbed. Chemical issues and water quality changes due to hydro-climatic variations and management strategies were also pinpointed. Both frameworks are thus able to highlight the main problems of the river and are consistent with each other. They are thus useful tools to survey the spatial and temporal evolution of rivers quality. However, several challenges remain, especially regarding the strategies to monitor and analyze chemicals, the definition of target values and conditions, the evaluation and integration of human-induced-pressures, and the overall evaluation of the rivers state. Development of integrated indicators is seen as a potential solution to explore river health and to provide efficient restoration measures by water managers.

TU197

Updating the Environmental Quality Standards for the EU priority substance chlorpyrifos

M. Wildi, Ecotox Centre CH / Ecotoxicologie; M. Casado-Martinez, Centre Ecotox; M. Junghans, B.J. Ferrari, Centre Ecotox EAWAGEPFL; I. Werner, Ecotox Centre Eawag-EPFL / Department of Anatomy Physiology and Cell Biology

Chlorpyrifos (CPY) is widely used as an active ingredient in insecticides. Since 2005 CPY is a priority substance under the EU Water Framework Directive (WFD) with an AA-EQS of 0.033 mg/L and a MAC-EQS of 0.1 mg/L. The aim of this study was to update the environmental quality standards (EQS) based on current data and the WFD method for EQS derivation published in 2011. Both AA-EQS and MAC-EQS decreased by more than one order of magnitude. The original AA-EQS was not derived based on available chronic ecotoxicity data but was set as MAC-EQS divided by a factor of 3, while the revised value of 0.00046 mg/L is based on a NOEC for *A. bahia* taken from the EFSA authorisation dossier and an assessment factor (AF) of 10. The original MAC-EQS was derived from mesocosm NOECs using an AF of 1. The revised MAC-EQS of 0.0044 mg/L is based on an HC_5 from a species sensitivity distribution (SSD) for crustaceans and insects using the lowest eligible AF of 5. The SSD reveals branchiopoda and amphipoda being the most sensitive taxonomic groups for CPY. A re-evaluation of old and new mesocosm data showed that using the available mesocosm data for EQS derivation is likely to be underprotective for amphipoda. The original EQS dossier from 2005 contains no specific EQS derivation for sediment. It was concluded that "Protection of sediment [is] covered by the QS referring to the pelagic community". The data search resulted in 17 acute and 4 chronic toxicity data for sediment organisms with effect data ranging from 0.324 mg/kg dw (acute) to 0.032 mg/kg dw (chronic). Acute data show that the amphipod *H. azteca* might be as sensitive to CPY as the insects *C. riparus and C. tentans* but chronic data are available only for insects. The resulting sediment $EQS_{sed,\ AF}$ of 0.32 µg/kg dw was derived by applying an AF of 100 on the chronic NOEC for *C. riparius*. For comparison, also the equilibrium partitioning method was used to derive an $EQS_{sed,EqP}$ from the revised AA-EQS. The application of this model including an AF of 10 that covers uptake by ingestion resulted in a $EQS_{sed,EqP}$ of 0.016 µg/kg dw. Wihtout this AF, the $EQS_{sed,AF}$ would be in the same order of magnitude as the calculated $EQS_{sed,\ AF}$. Based on our EQS update we strongly recommend to revise the current EQS values for CPY.

TU198

Lead exposures in European Freshwaters; are they a risk? A regulatory assessment accounting for bioavailability

I. Wilson, A. Peters, G. Merrington, wca; J. Chowdhury, International Lead Association / Senior Scientist -Environment

Lead (Pb) is a chemical for which one EQS has been set and is applied across all countries in Europe. A way in which the EQS (Environmental Quality Standard) is used in Europe is through a compliance assessment, effectively a comparison of the measured concentration of the chemical of interest in a water sample and the EQS. An indicative tiered compliance assessment of the Europe-wide bioavailable lead EQS of 1.2 µg L^{-1} (EQSbioavailable) was undertaken against regulatory freshwater monitoring data from six European member states and FOREGS database. A tiered approach to compliance assessment enables site-specific water chemistry to be accounted for, by correcting the measured dissolved metal concentrations in the water sample to a bioavailability-based concentration to be compared to an EQSbioavailable. In Tier 1 measured concentrations were compared against the EQSbioavailable. In Tier 2, Bio-met, a user-friendly tool based upon Biotic Ligand Models (BLMs) was used to account for bioavailability, along with the current European Water Framework Directive lead dissolved organic carbon correction (DOC-WFD) approach. The outputs from both approaches were compared to the Biotic Ligand Model (BLM), the final tier in the tiered approach. Lead exposures are relatively low across all regulatory datasets and FOREGS. At Tier 1, only 3.9 % of sites and samples assessed have a dissolved Pb concentration of greater than or equal to EQSbioavailable. Thus, the levels of compliance failure against the current Pb EQS in European freshwaters, as tested with regulatory monitoring datasets is relatively low. Of 8257 samples from 443 sites taken from regulatory monitoring records from six member states 61 samples showed an exceedance at Tier 3 (0.7%) with a maximum RCR of 5.32. For the FOREGS dataset, 2 % of the sites assessed assessed had a concentration greater than or equal to the EQS, and at tier 3, 1 site had a concetration failing to meet the EQSbioavailable. The waters showing the greatest sensitivity to potential lead exposures are characterized by relatively low DOC (< 0.5 mg L^{-1}), regardless of the pH and calcium concentrations. Whilst some risks due to lead are possible this is due to the combination of sensitive waters with elevated lead concentrations, as there is very little overlap of the distributions of site specific PNEC values and dissolved exposure concentrations.

TU199

Assessing compliance of European freshwaters for copper: accounting for bioavailability

A. Peters, I. Wilson, G. Merrington, wca; D. Heijerick, ARCHE; S. Baken, European Copper Institute

The importance of accounting for bioavailability in understanding the effects of metals has long been recognised in terms of setting environmentally relevant regulatory limit values. Attempts to deliver practical, routine methods to do so have been limited. For regulatory purposes in Europe an agreed Environmental Quality Standard (EQS) for a substance must be a fixed value: the same EQS is applied for all waters within the geographic area that is regulated. For metals, the EQS must be set for the bioavailable forms, and it is termed $EQS_{bioavailable}$. This study determines the levels of compliance of European freshwaters with a copper EQS, and evaluates the usefulness of a tiered approach to compliance assessment for copper. The first tier compares the dissolved metal concentration to a threshold, estimated using either regional or continental water chemistry data. At Tier 2, the bioavailable metal concentration is calculated using the physico-chemistry of the water body, and compared to the $EQS_{bioavailable}$. It follows that the thresholds at Tier 1 must be set at a level which will ensure protection of sensitive environments. The value of the threshold has important implications in terms of the financial costs of the compliance assessment. For copper, setting the thresholds at the same level for the whole of Europe (i.e. continental) would leave some countries with costly and unwarranted monitoring requirements. Deriving the threshold on a region or country specific basis enables effective use of resources without compromising on the level of protection. A very high level of compliance for copper is observed where bioavailability based thresholds are used for the implementation derived from regionally relevant water chemistry data (99.3%). Sites where elevated ambient background levels of copper are combined with very high bioavailability, principally when the waters have low DOC concentrations, are those most likely to be at risk due to copper exposures.

TU200

Are lead exposures a risk in European freshwaters? A map of EQS compliance assessment accounting for bioavailability

J. Chowdhury, International Lead Association / Senior Scientist -Environment; A. Peters, I. Wilson, G. Merrington, wca

Lead (Pb) is a priority substance for which the bioavailable Environmental Quality Standard ($EQS_{bioavailable}$) of 1.2 µg L^{-1} has been set under the European Commission Directive 2013/39/EU for application across all countries in Europe. In the present study, a tiered approach was applied to undertake a compliance assessment of the $EQS_{bioavailable}$ using the FOREGS database that includes paired data for water quality parameters and measured Pb concentrations from freshwater streams and rivers across Europe. In Tier 1 measured dissolved Pb concentrations were directly compared against the $EQS_{bioavailable}$. In Tier 2, Bio-met, a user-friendly tool based upon Biotic Ligand Models (BLMs) was used to account for bioavailability, along with the current European Water Framework Directive lead dissolved organic carbon correction (DOC-WFD) approach. The outputs from both approaches were compared to the chronic Pb Biotic Ligand Model (Pb BLM), the final tier in the tiered approach. The maximum Pb concentration in FOREGS is approximately 11 µg L^{-1}. At Tier 1 screening, only 16 (2.0%) water samples of the whole dataset (n=797) have Pb concentrations that are greater than the EQS of 1.2 µg L^{-1}. The exceedances further decreased to 3 (0.4%) and 1 (0.1%) upon accounting for bioavailability at Tier 2 and 3 respectively. The map of site-specific PNECs (predicted no effect concentrations) as calculated by the Pb BLM identifies that the most sensitive waters in the database are all those with extremely low concentrations of DOC (< 0.5 mg L^{-1}). The greatest frequencies of such sites are found in the alps and Norway. The lowest PNEC values for Pb are around 0.5 µg L^{-1}, and the WFD EQS value of 1.2 µg L^{-1} is equivalent to approximately the 4th percentile of the dataset. The results indicate that the European freshwater bodies with low anthropogenic pressure are unlikely to fail the compliance with the EQS, with the exception of very local situations such as historic mining sites.

Integrated approaches in ecotoxicology: bridging the gap between experimental toxicology and mechanistic modelling (P)

TU201

Modelling survival under chemical stress. A comprehensive guide to the GUTS framework

T. Jager, DEBtox Research / Dept of Theoretical Biology; R. Ashauer, University of York / Environment

Testing, analysing and predicting the lethal action of chemicals on organisms plays a central role in the fields of ecotoxicology and toxicology, both for scientific and regulatory purposes. The dominant approaches to deal with survival data are descriptive, focussing on standardised tests and simple summary statistics (such as the LC50). Such descriptive methods ignore the fact that lethal effects develop over time, thereby leading to biased assessments and precluding useful predictions to untested exposure scenarios. Making sense of toxic effects over time requires mechanistic models, and, more specifically, the explicit consideration of toxicokinetics and toxicodynamics (TKTD). For the endpoint survival, almost all existing TKTD models can now be viewed as special cases of an overarching framework: GUTS, the General Unified Threshold model for Survival. GUTS was conceived in 2010, and has subsequently gained a large user community. Furthermore, the model is receiving increasing interest from the regulatory field as it is expertly suited for the analysis of survival data, and for extrapolation across different exposure scenarios. With the increasing interest in GUTS, and the increasing interest in good-modelling practice, it is time for a more detailed treatise on this model framework. In a CEFIC-LRI funded project, we have prepared an extensive e-book on GUTS (which will be available for download, free of charge, January 2018). The book contains a detailed description of the model framework (concepts, underlying assumptions and mathematics) and its historical roots, as well as worked-out case studies, guidance for users of the model (or its results), and the results of a ring test for a range of software implementations. This book is the standard work on GUTS modelling, and the e-book format allows the contents to be kept up to date with the major novel developments in this area.

TU202

Dose response modelling in aquatic and terrestrial effect models

M. Wang, WSC Scientific GmbH / Dept Efate Modelling; K. Billau, WSC Scientific GmbH

In recent years mechanistic effect models including GUTS and DEBTox have been successfully used in the aquatic and terrestrial risk assessment of pesticides. These models offer the advantage that results from laboratory studies, usually conducted with constant exposure, can be translated to time variable exposure, which is more typical under field conditions. At present these models consider a threshold beyond which effects start to appear. Once this threshold is surpassed the amount of effect is calculated using a linear regression, i.e. effects increase linearly with increasing concentration. In other areas of the risk assessment dose-response modelling and the derivation of reliable dose-response curves has received much focus (see e.g. the new EFSA guidance on benchmark dose modelling). Therefore, it is investigated in the present study if these models can be improved by considering more typical dose-response curves, which often are sigmoid shaped. It is investigated when the specific shape of a dose-response curve affects the outcome of an assessment and how the magnitude of predicted effects is affected.

TU203

Investigating toxico-kinetics of emergent pollutants (PFASs) in the common sole (Solea solea) from in situ measurements and experimental data on PCBs within a DEB-based modelling approach.

F. Mounier, National Research Institute of Science and Technology for Environment and Agriculture - Irstea / UR EABX; V. Loizeau, IFREMER / UR Biogeochemistry and Ecotoxicology; L. Pecquerie, IRD / UMR LEMAR; P. Labadie, UMR CNRS EPOC Universite Bordeaux / UMR 5805 EPOC; G. Munoz, Universite de Montreal / Chemistry; H. Budzinski, CNRS / UMR 5805 EPOC; J. Lobry, Irstea / UR EABX

In the context of global change, developing mechanistic tools integrating the influence of environmental factors on toxicants bioaccumulation dynamics is required, as organisms will face unprecedented conditions. Mechanistic models based on the Dynamic Energy Budget (DEB) theory are relevant to predict individual contamination as a function of environmental temperature and food availability, quality and contamination. Moreover, this modelling framework allows including adverse effects (DEB-tox), often based on internal concentration. However, for emerging compounds like PerfluoroAlkyl Substances (PFASs), toxico-kinetics calibration is a challenge as their properties are not fully characterized yet and experiments are scarce. We here present the results of a research project aiming at investigating toxico-kinetics (TK) of PFASs in juvenile common sole from a contaminated nursery ground, the Gironde Estuary (France). In such a highly fluctuating environment, our goal was to extract information from the large variability observed in the measurements for several polychlorobiphenyls (PCBs) and PFASs. First, we designed environmental scenarios and prioritized environmental sources of inter-individual variability using a mechanistic model calibrated for CB153 thanks to experimental data. As CB153 is poorly biotransformable in fish, its bioaccumulation in juveniles mainly relies on the ingestion of contaminated food. Further, we considered this congener as an additional tracer of potential preys in the wild. Indeed, diet reconstruction from stomach contents and isotope data provided us with contrasted pictures. We predicted sole CB153 contamination for a range of environmental diet, food contamination and temperature scenarios. Comparing these predictions with *in situ* measurements, we were able to highlight the major influence of diet composition. The next step was to consider the other PCBs and PFASs with previously selected environmental scenarios. Discrepancies between model predictions and observations allowed us to formulate new modelling hypotheses taking into account the ability of soles to assimilate and metabolize these different compounds depending on their properties (e.g. hydrophobicity, spatial conformation, functional groups). This mechanistic approach prioritizing sources of variability provided new insights on the differential bioaccumulation between toxicant families in a key flatfish resource.

TU204

Investigating metabolic acceleration in dynamic energy budget models of copepods using the ecotoxicological model organism Nitocra spinipes

J. Koch, GhEnToxLab (Ghent University) / Applied Ecology and Environmental Biology; K. De Schamphelaere, Ghent University (UGent) / Applied Ecology and Environmental Biology

Copepods form an essential part of marine ecosystems and constitute a large portion of animal biomass on earth. Moreover, their small body size and short life cycle make them convenient test organisms in ecotoxicity studies. Beside acute toxicity, multiple works in the past focused on chronic life history effects of chemicals in copepods. Unfortunately, we usually lack a mechanistic explanation of observed effects which is required for realistic laboratory-to-field extrapolations. Models rooted in Dynamic Energy Budget (DEB) theory can help to evaluate sublethal toxicity data in terms of effects on the energy household of an animal. Although DEB models for new species are usually easily parameterised, the copepod life history shows distinct deviations from the 'standard DEB model' requiring further investigation. While some authors presume metabolic acceleration from birth until puberty, others suggest a von Bertalanffy growth curve which is truncated at the final moult. In this study we parameterised the two typified DEB models 'abp' (metabolic acceleration from birth to puberty) and 'sbp' (standard von Bertalanffy growth from birth to puberty) for the harpacticoid copepod *Nitocra spinipes* to investigate metabolic acceleration in copepods. As no high-quality data on length over time were available for *N. spinipes*, we performed a growth experiment over 28 days. Additional data from literature were used to aid the parameter estimation. Submodels for food (Holling's type II functional response) and temperature dependency (Arrhenius temperature correction) were calibrated on development time and reproduction data. While isomorphic growth is commonly assumed in DEB studies, it does not hold true for *N. spinipes* which grows more slender in the course of its development. Hence, we used the square root of the top view area as a length measure to scale with the cubic root of structural volume in length-to-volume conversions. Both models abp and sbp showed good fits to the given data. Overall, abp predicted the data slightly better compared to sbp with a mean relative error of 0.063 vs. 0.076 in sbp. However, we do not regard this difference clear enough to unequivocally confirm or reject metabolic acceleration in copepods. More detailed data on *N. spinipes* and other copepods are needed to reveal the most accurate model for the copepod life history. That said, both models are promising tools for the evaluation and extrapolation of toxicity data in *N. spinipes*.

TU205

Grey seal ecophysiology and environmental change

J. Desforges, Aarhus University (AU) / bioscience; G.M. Marques, University of Lisbon; K. Kauhala, Natural Resources Institute Finland Luke; K. Harding, University of Gothenburg Sweden

Marine mammals are considered as sentinel species for marine ecosystem health. In the Baltic, grey seals (Halichoerus grypus) can serve this purpose as they are top predators and have shown to respond to anthropogenic and environmental stressors over the past decades. These stressors can influence the physiology and health of grey seals, ultimately leading to individual and population level consequences. Acknowledging the need for mechanistic understandings of stressor effects, we have developed a full lifecycle bioenergetics model for Baltic grey seals using Dynamic Energy Budget (DEB) theory. We use the comprehensive information available in the literature on grey seal fetal development, lactation, growth and reproduction to parameterize and validate our model. Our model accurately predicted grey seal ontogeny and lifehistory traits, providing one of the first full descriptions of mammalian development in DEB. Recent reports have indicated that climate change effects on sea ice and food web dynamics have impacted grey seal condition (i.e. blubber thickness). We use our model to explore these relationships and confirm that grey seal body condition in the Baltic is vulnerable to change in food quality/quantity and can lead to down-stream consequences on reproductive success. The results offer new insights into physiology and ecology of Baltic grey seals with the potential to lead to novel approaches for the study of stress ecology and conservation of this species.

TU206

Evaluation of thermal stress on Daphnia magna using oxidative stress and life-history trait parameters

H. IM, J. Na, J. Jung, Korea University / Environmental Science and Ecological Engineering

Present study evaluated the effects of temperature (20 °C and 25 °C) on oxidative stress and life-history trait responses of *Daphnia magna* in short-term (5 days) and long-term (21 days) exposures. *D. magna* exposed to 25 °C exhibited continuous higher production of reactive oxygen species (ROS). In short term exposure, glutathione peroxidase (GPx) activity was significantly suppressed in elevated temperature. In contrast, daphnids showed significantly enhanced catalase (CAT) and glutathione peroxidase (GPx) activity under long-term exposure ($p < 0.05$). Lower lipid peroxidation (LPO) level at elevated temperature under prolonged exposure suggests that antioxidant enzymes successfully prevented ROS-mediated damage. In addition, exposing *D. magna* to elevated temperature significantly shortened time to first brood, brood size, and body length, but induced significantly higher male production ($p < 0.05$). Reduced body length at elevated temperature indicated that *D. magna* invested more energy into defense mechanisms rather than growth and reproduction to cope with the thermal stress. Moreover, a multi-generational study was performed to evaluate multigenerational effect of elevated temperature on *D. magna*.

TU207

Transport-protein metal binding links uptake biodynamics for predicting copper in tilapia

Y. Chen, Kaohsiung Medical University / Biomedical Science and Environmental Biology; W. Chen, Kaohsiung Medical University / Department of Biomedical Science and Environmental Biology; C. Liao, National Taiwan University / Department of Bioenvironmental Systems Engineering

Metal could bind to transport protein, then accumulate in the cellular and tissue. It points out that the metal ion accumulating in target subcellular compartment could reflect the metal toxicity. Copper (Cu) plays an essential role in cellular metabolism of aquatic organisms, but it would cause toxicity with excessive accumulation. The purpose of this study was to conduct the short-term exposure experiment to examine the Cu accumulation in tilapia, then combined with bioavailability and subcellular partitioning to estimate the Cu binding situation and mechanism of toxicity on gill. We developed a mathematical framework that quantified the Cu affinity and the amount of transport protein in different subcellular compartment. Results indicated that Cu accumulation in metabolically active pool (MAP) preferred to organelles than heat denatured protein, and Cu accumulation in metabolically detoxified pool (MDP) was metal rich granule. The estimated parameters of maximum Cu influx rate, total number of transport protein and affinity constant didn't have significant differences between MAP and MDP. However, the conditional stability constant of MDP 0.45 ± 0.005 ml μg^{-1} was

significant higher than that of MAP 0.269±0.018 ml μg^{-1} ($p < 0.001$), and the uptake rate constant of MDP 0.128±0.001 ml g^{-1} hr^{-1} was also significant greater than MAP 0.086±0.001 ml g^{-1} hr^{-1} ($p < 0.001$), it revealed that Cu was likely to bind on MDP in the low exposure concentration than that of MAP. This study concluded that Cu tend to accumulate in MDP, then may cause less toxicity to tilapia. Keywords: Copper; bioavailability; transport protein; subcellular partitioning

TU208

Relationships between subcellular metal partitioning and biomarkers of effects in white suckers (Catostomus commersonii) exposed to an environmental metal gradient

N. Urien, INRS-ETE / Centre Eau Terre Environnement; A. Urien, Université du Québec INRS / Centre Eau Terre Environnement; L. Ramilo, H. Sonnenberg, Ecological and Regulatory Solutions Inc; P.G. Campbell, P. Couture, Université du Québec, INRS / Centre Eau Terre Environnement

Discharges from metal mining operations may lead to metal accumulation and toxicity in aquatic species. Once metals enter cells, they can bind to sensitive components and cause deleterious effects. Nevertheless, metals can also be detoxified by binding to molecules designed to sequester them, limiting their toxicity. The objectives of this study were *(i)* to assess the subcellular distribution of metals and metalloids (Cd, Cu, Se and Zn) in livers of white suckers exposed to metal-mining effluents, and *(ii)* to investigate the links between the binding of specific metals to particular subcellular fractions and physiological effects. To this end, mature male and female fish were collected in three lakes downstream from a metal-mining effluent and one lake in a reference area. Subcellular partitioning among putative metal-sensitive fractions (MSF) and biologically detoxified fractions (BDM) in livers was determined after differential centrifugation and heat-denaturation steps. In parallel, a suite of biomarkers was investigated ranging from general indicators of energy accumulation to specific indicators of oxidative stress and metabolic or biosynthetic capacities. Total hepatic metal concentrations were significantly higher in exposed fish than in reference fish, with Cd (x6) and Se (x10) being accumulated the most. No differences between sexes were observed. Subcellular partitioning of metals was similar among areas but specific to metals; over 70% of the Cd burden was found in the heat-stable cytosolic proteins fraction, which includes metallothioneins. In contrast, the largest contributors to the total Se liver burden were the potentially metal-sensitive heat-denaturable proteins fraction (≈35%), and the organelles fraction (≈30%). These results suggest that Cd was well detoxified and regulated by white suckers, whereas the presence of relatively high Se concentrations in the MSF suggests that exposed fish were likely subject to stress. Principal component analysis showed that increasing [Se] in all of the fractions was strongly correlated with lower fish condition and associated with higher oxidative stress, suggesting a trade-off between growth and control of oxidative stress. Finally, this work will contribute to advancing our understanding of the toxic modes of action of metals in aquatic organisms and our capacity to monitor the risk for fish inhabiting metal-contaminated environments.

TU209

Development of an adverse outcome pathway for acetylcholiesterase inhibition in zebrafish (Danio rerio)

K.H. Watanabe, A. Mikhail, Arizona State University / School of Mathematical and Natural Sciences; K. Conrow, Arizona State University; N. Vinas, Mississippi State University / Engineer Research and Development Center

Acetylcholine is a neurotransmitter that is important for a broad range of processes in the body such as muscle activation. Acetylcholinesterase (AChE) is an enzyme that hydrolyzes acetylcholine in order to eliminate it from the body, and when AChE is inhibited acetylcholine levels increase. Excess acetylcholine at cholinergic synapses overstimulates muscarinic- and nicotinic-type receptors. These receptors are found in most organs of the body, thus multiple adverse outcomes may result. Moreover, a wide variety of chemicals including organophosphates, carbamates and some high nitrogen compounds, can inhibit AChE. Thus, the impact of AChE inhibition is large, yet relatively little research has been focused upon developing related adverse outcome pathways (AOPs) or a network for this molecular initiating event. This presentation focuses upon the construction of adverse outcome pathways that result from AChE inhibition in zebrafish (*Danio rerio*). We performed a comprehensive review of the literature to identify studies and datasets that could be used to construct an AChE AOP. Adverse outcomes include seizures, impairment of the retina architecture and behavioral changes. Preliminary AOPs for these outcomes will be presented with references to the studies that support the AOP, and identification of data needed for quantitative AOP development.

TU210

Development of a Novel Quantitative Adverse Outcome Pathway Predictive Model for Lung Cancer

T. Hill, US EPA NHEERL/ISTD/CB / ORD NHEERL Integrated System Toxicology Division; R. Conolly, US EPA RTP

Traditional methods for carcinogenicity testing are resource-intensive, retrospective, and time consuming. An increasing testing burden has generated interest in the adverse outcome pathway (AOP) concept as a tool to evaluate chemical safety in a more efficient, rapid and effective manner that better directs resource utilization. A central premise of the AOP concept is that pathway progression from the molecular initiating event (MIE) implies a definable "response-response" (R-R) relationship exists between each key event (KE) that drives the pathway towards the adverse outcome. Computational description of these R-R relationships in a quantitative AOP (qAOP) enables dose-response consideration of probabilities and uncertainty, as well as flagging of special at-risk populations or sentinel species. The qAOP also provides a platform to utilize early genomic and *in vitro* data streams for rapid, less resource-intensive hazard prediction, as well as the dose response and exposure duration that informs the level of risk. This poster describes a novel AOP/qAOP for lung cancer in the mouse from the MIE of CYP2F2-specific formation of reactive metabolites, advancing through KE for protein/nucleic acid adducts, diminished CC10 capacity and hyperplasia of CC10 deficient Club cells, and culminating in the adverse outcome of mixed-cell tumor formation in the airway. The AOP is independent of route of exposure and grounded in overlapping mechanistic events for naphthalene, styrene, ethyl benzene, isoniazid and fluensulfone in the mouse. The qAOP modeling is supported by defined mechanistic relationships and quantitative data (PB-PK, dose-response and time-course) from archival data in peer-reviewed literature. Findings will include evaluation of data supporting the cancer qAOP, suitability for characterization of R-R relationships, and identification of data gaps or additional research as required. This approach supports international efforts on use of quantitative effect thresholds for adversity predictions and incorporation of novel data streams into the cancer risk assessment process. *This abstract does not necessarily represent the views or policies of the U.S. EPA.*

TU211

A combined PBTK and qAOP-modeling approach to assess the impact of dioxin-like compound (DLC)-induced embryotoxicity on recruitment failure in European eels

M. Brinkmann, University of Saskatchewan / School of Environment and Sustainability and Toxicology Centre; M. Freese, J. Pohlmann, Thühnen-Institute for Fisheries Ecology; J. Doering, National Research Council at U.S. Environmental Protection Agency, Duluth; M. Damerau, L. Marohn, R. Hanel, Thühnen-Institute for Fisheries Ecology; M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre

The panmictic stock of the European eel (*Anguilla anguilla*) has seen a dramatic decline over the past several decades, and declines in recruitment as a result of maternally transferred contaminants has been proposed as one of several potential causes. In particular, dioxin-like chemicals (DLCs) have been identified as a class of chemicals of great concern for both European and American eels (*Anguilla rostrata*). DLCs bioaccumulate, are highly embryotoxic in many species of fish, and maternally transferred in artificially matured eels. However, to date researchers have been unable to locate reproducing adult eels or developing embryos in their natural spawning grounds in the Sargasso Sea. As a result, accurate embryotoxicity data to identify the potential causative chemicals are unavailable. Therefore, this study aimed to (a) parameterize a physiologically-based toxicokinetic (PBTK) model for European eels to account for the impact of changes in physiology that result from sexual maturation and migration on toxicokinetics, and (b) to couple this model with a quantitative adverse outcome pathway (qAOP) for activation of the aryl hydrocarbon receptor 2 (AHR2) of fishes to predict early life stage mortality of eels as a result of exposure to maternally transferred DLCs. The PBTK model was used to kinetically predict the redistribution of DLCs within the body of female eels during migration, and ultimately the concentration in gonads and eggs. A simple qAOP was described previously linking activation of species-specific AHR2 in an *in vitro* luciferase reporter gene assay using transfected COS-7 cells with embryo lethality across nine species of fishes exposed to DLCs. To this end, AHR2 was cloned from European eel and used to predict eel-specific relative potencies of five DLCs representing congeners measured at among the greatest concentrations in gonads of eels. Using this data, mortality of early life stages of eels was estimated based on the internal concentrations predicted by the PBTK model. Our integrated PBTK model and qAOP approach will ultimately shed light on the question whether early life stage mortality induced by exposure to DLCs has the potential to significantly contribute to the observed decline in recruitment of eels.

TU212

Salmonid pituitary cells as a test system for identifying endocrine disrupting compounds

L. Harding, University of Washington / Aquatic and Fishery Sciences; I.R. Schultz, NOAA NWFSC / Marine Science Laboratory; G. Young, Advisian WorleyParsons Group / Aquatic Sciences; P. Swanson, NOAA-NWFSC

The pituitary gland is a central regulator of reproduction, producing two gonadotropins, follicle-stimulating hormone (FSH) and luteinizing hormone (LH), which regulate gonadal development, sex steroid synthesis and gamete maturation. Despite its central role in regulating reproduction, there are limited data on impacts of endocrine disrupting chemicals (EDCs) on the pituitary gland. We have previously observed that waterborne exposure of previtellogenic coho salmon to 17α-ethynylestradiol (EE2) causes widespread effects on the pituitary transcriptome. Other in vivo studies with the selective serotonin reuptake inhibitor (SSRI) fluoxetine caused a decrease in FSH beta subunit (*fshb*) mRNA levels. These results motivated us to expand our studies by developing an in vitro test system using pituitary cells isolated from coho salmon and previtellogenic female rainbow trout. Preliminary studies were performed to optimize culture conditions and to establish the time course of *fshb* and LH β (*lhb*) subunit gene expression with and without addition of endogenous sex steroids (estrogen [E2] or 11-ketotestoerone). These initial studies suggested culturing with and without E2 was valuable as it could mimic the (+) feedback effects on LH that is observed from in vivo studies. After optimizing assay conditions, a suite of 12 contaminants and other hormones was evaluated for their effects on *fshb* and *lhb* gene expression. Each chemical was tested at 4-5 different concentrations up to solubility limitations in cell culture media. Results indicated more chemicals altered LH synthesis than FSH. The more potent chemicals were estrogens (EE2) and aromatizable androgens (testosterone), which induced *lhb*. The estrogen antagonist 4-OH-tamoxifen decreased the E2 stimulated expression of *lhb*. Among several SSRIs tested, the sertraline metabolite norsertraline was notable for both increasing *fshb* synthesis and decreasing the E2 stimulation of *lhb*. These results indicate that diverse types of chemicals can alter gonadotropin production in fish. Further, we have shown that pituitary cell culture is useful for screening chemicals with potential endocrine disrupting activity and can support quantitative adverse outcome pathway testing. Supported by EPA-STAR grant R835167.

TU213
SETAC Mechanistic Effect Models for Ecological Risk Assessment of Chemicals Interest Group
E. Zimmer, IBACON GmbH

Integrating life cycle approaches towards a sustainable circular economy (P)

TU214
Metal and mineral resources in LCIA - What's the problem?
R. Schulze, University of Leiden / CML; J. Guinee, University of Leiden / Institute of Environmental Sciences; R.A. Alvarenga, Z. Weng, J. Dewulf, Ghent University / Department of Sustainable Organic Chemistry and Technology; J. Drielsma, Euromines
There is currently a lack of consensus on how to assess impacts from abiotic resource use in life cycle impact assessment (LCIA). Unlike other environmental impact categories, abiotic resource use does not just have one single, explicitly agreed-upon, international management goal. The SUPRIM project focuses on impacts which occur directly from the use of abiotic resources such as minerals, metals, and natural materials. It concerns impacts associated with their availability or accessibility, but excludes impacts covered by other impact categories, such as toxic emissions or adverse working conditions. The current state-of-the-art LCIA for abiotic resources has been criticized by representatives of the metals & mining industry. The LCA community is developing new methods, which all focus on different issues associated with resource use. This lack of a broadly accepted method, likely attributable to the lack of a common perspective on resource use and a common understanding of the potential problem(s) related to the use of resources, was the starting point of SUPRIM. The aim of the project is to obtain an understanding of different stakeholders' views and concerns regarding potential issues associated with the use of resources. The gained insights are provided in the form of a structured overview of those views, and used as a basis for further method development. They are achieved by 'taking a step back' towards a structured discussion about potential problems with resource use, and different motivations behind resource management concepts. To guide the discussion towards a clear outcome, a framework was developed. It introduces distinctive criteria for the evaluation and/or formulation of perspectives and problems on resource use, which will enable a comparison of differences and overlaps between stakeholder views. The framework will be applied in a workshop with project external stakeholders from industry, policy and academia. The workshop outcome will be used to guide the further development of impact assessment from abiotic resource use in LCIA, such as a reduced future availability of the resources themselves, changes to their ability to provide functions, losses of certain desired properties in the environment or the technosphere, or an increased difficulty to access them. We aim to present both the framework developed for the formulation and evaluation of perspectives, and the outcome of its first application during the stakeholder workshop.

TU215
The relevance of the end-of-life stage for the environmental impact of batteries
J.F. Peters, Karlsruhe Institute of Technology KIT / Helmholtz Institute Ulm HIU; M.J. Baumann, Karlsruhe Institute of Technology KIT / Institute for Technology Assessment and Systems Analysis; C. Minke, Technische Universität Clausthal / Energy Research Center; M. Weil, Karlsruhe Institute of Technology KIT / Institute for Technology Assessment and Systems Analysis ITAS
Numerous LCA studies exist in the field of energy storage, especially batteries. However, the majority of these studies focus on the production and use phase, while the battery disposal or recycling is usually evaluated in separate studies that focus explicitly on this part of the life cycle. While of lower importance when comparing very similar batteries (e.g., different lithium-ion batteries (LIB)) with similar end-of-life (EoL) processes, this is increasingly relevant when comparing different electrochemical energy storage technologies. Thus, a thorough modelling of the EoL phase can be considered mandatory for a well-funded assessment. For evaluating this aspect we expand existing LCA studies on stationary batteries by a tentative modelling of their EoL processes (recycling) and compare the results. Three different battery technologies are considered for this purpose, an LFP-LTO battery (rack-mounted stationary LIB), a hybrid aqueous ion battery (AHIB) and a vanadium redox flow battery (VRFB), all with the same net storage capacity. The results show that considering the end-of-life stage actually does change the outcomes of the results significantly and that cradle-to-gate assessments are not appropriate for comparing very different battery technologies. Highly integrated batteries like the LIB have advantages under a cradle-to-gate perspective (higher energy density and thus lower material demand per provided capacity), while less integrated systems can have significant advantages when it comes to recyclability. The AHIB and VRFB are easy to dismantle and all major components can be recovered by mechanical dismantling on a macro-scale. The highly integrated LIB require complex processes and obtain a commingled fraction of micro-size particles that are difficult to separate and require significant process inputs while only recovering a fraction of the materials originally contained in the batteries. This can change the picture fundamentally towards an advantage of technologies easy to dismantle on macro-scale (AHIB and VRFB) in comparison with highly integrated cells (LIB). Thus, design for recyclability is highly important in terms of a future circular economy and might easily outweigh the possibly reduced energy density or lower performance.

TU216
Battery recycling efficiencies and their influence on the life cycle impacts of batteries
K. Boonen, A. Van der Linden, VITO
The EU Batteries Directive sets the minimum recycling efficiencies for waste batteries, as a percentage of their average weight. In the light of the circular economy, it may be argued that, in addition to the quantity, it is important to consider the specific materials that are recycled, the quality of the recycled fractions and their potential use. The Public Waste Agency of Flanders (OVAM) asked VITO to assess the effect of these factors. In this study, the environmental impact of the end of life is compared for different battery recycling routes. Furthermore, the impact of the rest of the life cycle of disposable and rechargeable batteries is calculated to put the impact of the end of life into perspective. A number of potential improvement options, such as a higher collection rate, higher functional recycling and a shift to different types of batteries, are evaluated.

TU218
New and Reconditioned Electrical and Electronic Equipment. How does change the environmental performance?
M. Pini, University of Modena and Reggio Emilia / Department of Sciences and Methods for Engineering; P. Neri, University of Modena and Reggio Emilia / Department of Sciences and Methods for Engineering DISMI; R. Gamberini, B. Rimini, F. Lolli, University of Modena and Reggio Emilia; A. Ferrari, University of Modena and Reggio Emilia / Department of Sciences and Methods for Engineering
The scope of this study, carried out within the LIFE12 ENV/IT001058 WEEENmodels, was to compare the environmental performance of the life cycle of new electrical and electronic equipment (EEE) and the reused one through the Life Cycle Assessment methodology. Both *attributional* and *consequential* LCI (Life Cycle Inventory) modelling have been implemented. A representative product has been considered for each WEEE group, assuming that it generates the same environmental damage of the other products belonging to the same category. In particular, the following representative products have been selected: refrigerator (R1), washing machine (R2), cathode ray tube (CRT) (R3), laptop (R4) and fluorescent lamp (R5). In addition, lower performance of reconditioned EEE has been taken into account. Different set of replaced components have been evaluated in order to understand which determines the best solution. Scenario A represents the set of replaced components, which damage more frequently; Scenario B is just an alternative set of replaced components. The environmental comparison between new and reused WEEE, adopting attributional LCI modelling, showed that Scenario B produces a damage decrease for all WEEE categories. Moving on the consequential LCI modelling, the environmental comparison highlighted for both scenarios a considerable damage reduction for the reused EEE respect the new one. Furthermore, for the reused R1, R2, R3 the analysis of results carried out environmental credits. This is due to the avoided burdens associated to the manufacturing of the new EEE, since the system boundaries have been enlarged until to considering the avoided production of the new product. Attributional and consequential LCI modelling performed different LCIA results. Following the methodological guidance for the identification of the most adequate LCI modelling framework presented by Laurent et al., 2014, it would recommend to adopt consequential LCI modelling. But we suggest to LCA practitioner to focus also the attention on the request of who commissioned the project, which often in the waste field are local administrations. Generally, they wants a snapshot of the real effects that waste management policies provoke on human health and environment. For this reason, *attributional* LCI modelling would be the proper LCI modelling to achieve this scope. Considering this LCI modelling the Scenario B determines the

best environmental performance.

TU219
The impact of European consumption of household appliances: insights from the LCA of efficiency measures and expected trends
F. Reale, EC JRC; V. Castellani, European. Commission - Joint Research Centre / Sustainable Resources, Bio-Economy; R. Hischier, EMPA / Technology and Society Lab; S. Sala, European Commission - Joint Research Centre / Bioeconomy unit
Household appliances are an important contributor to the overall impact generated by European citizens' consumption of products. In 2010 the energy-related products covered by the Ecodesign directive responsible for 53% of total EU-28 gross energy consumption in 2010. The aim of this study is to discuss main insights from the Life Cycle Assessment (LCA) of future scenarios of the European consumption of household appliances in the residential sector. The consumption is a Basket of Products (BoP) owned by an average European citizen. The BoP baseline consists of a process-based LCI model for a BoP that represents the most relevant household appliances in terms of energy consumption and market share: dishwasher, washing machine, drying machine, air conditioner, refrigerator, TV screen, computer, lighting, cooking appliances. A number of scenarios have been tested, covering the various life cycle stages including scenarios on the use phase, the waste collection, the electricity mix used. An overall scenario covering the design options for products energy efficiency and expected trends in purchase and user behavior has been calculated and compared with the baseline. The baseline hotspot analysis (with ILCD impact assessment method) confirmed the well-known relevance of the use phase of energy-related products, where the efficiency of products and consumer behaviour appear to be the two factors determining the BoP impact. Results of the scenarios assessed show for most of the categories a reduction of the overall impact compared to the baseline scenario. The reduction is more important for categories like e.g. GWP (due to the improved energy efficiency of products), IRP (thanks to the assumed "phasing out" of nuclear power plants in Europe) and AP (in this case, the reduction of the amount of coal-based electricity leads to reduced releases to the atmosphere of those substances contributing to AP). Due to the expected increase of the number of devices per person in the future, some of the impact categories – namely HTPc, FETP, LUC and FRD – show a higher potential impact in the scenarios than it is in the baseline. Obtained results show clearly that just heading for more efficient devices is a necessary, but not yet a sufficient condition towards more sustainability; we as a society have also to re-evaluate the way how we acquire (more and more) such devices and are spending more and more time behind them – here some limitations may would make sense.

TU220
Assessing economic and environmental effects of product replacement program using dynamic discrete choice model: As a case study of "home appliance eco-point system" in Japan
D. Nishijima, National Institute for Environmental Studies; S. Kagawa, Kyushu University; M. Oguchi, K. Nansai, National Institute for Environmental Studies
In evaluating environmental burdens from consumer durables, product lifetime is a key factor and most of the previous studies used average lifetime or lifetime distribution with a focus of engineering durability (Müller 2006; Kagawa *et al*., 2011; Nishijima, 2016). These product lifetime modelling techniques did not explicitly treat a relationship between product replacement and consumer's behavior. Whereas, the product replacement modelling techniques based on the economic maximum utility theory have been developed in economics and marketing fields (Schiraldi, 2011; Melnikov, 2013). It is beneficial to apply the economic product replacement models to environmental research. This study attempts to use the product replacement model for evaluating economic and environmental effect of polices for consumer durables. As a case study, we focused on air conditioners and analyzed the economic and environmental impacts of "Home appliance eco-point system" in Japan which was conducted during a period from May 2009 to March 2011 for encouraging consumers to replace their own products with new ones. Following the proceeding studies (Rust, 1987; Gordon, 2009;), we constructed the product replacement model of air conditioners in Japan by Bellman equation and a dynamic discrete choice model. We also estimated the logit parameters by the maximum likelihood estimation. We used the annual sales and replacement data of air conditioners during 1993 to 2015 (The Japan Refrigeration and Air Conditioning Industry Association; The Japan Electrical Manufactures' Association) and replacement and running cost data during the same period (Agency for Natural Resources and Energy of Japan; The Japan Refrigeration and Air Conditioning Industry Association). Using the estimated product replacement model, we analyzed the impact of "Home appliance eco-point system" on the CO_2 emissions and economic output by input-output framework. Through the results, we not only evaluated how effective the system was for reducing CO2 emissions and stimulating economy in Japan, but discuss how we could improve the system for obtaining more economic and environmental benefits.

TU221
Economic lifetime, hazard functions, and car inspection system
Y. NAKAMOTO, S. Kagawa, Kyushu University
Under the Paris Agreement adopted at COP21, Japan set itself a target of reducing its territorial greenhouse gas emissions by 26% (relative to the 2013 level) by 2030. To further reduce emissions in the transport sector, the government has set up both a technology policy and a demand policy, to try to improve the fuel economy of new vehicles and increase sales of next-generation motor vehicles as a proportion of new vehicle sales, respectively [Ministry of Land, Infrastructure, Transport and Tourism, 2017]. \n By assessing and estimating the economic lifetime of vehicles based on consumer behavior that maximizes utility level over time, we were able not only to specify the replacement purchase rate based on a dynamic discrete choice model but also to quantitatively analyze the environmental impact of changes in consumer behavior due to the adoption of policies because a motor vehicle inspection dummy, maintenance and repair costs, and new vehicle replacement purchase costs are explicitly included in the utility functions at the time of vehicle retention and replacement purchase. \n In this study, we used a DDC model to estimate car replacement purchase rates based on consumer behavior aimed at maximizing utility levels over time. By combining replacement purchase rates specified from source data with life-cycle CO2 emissions analysis, we demonstrated the impact of Japan's car inspection system on CO2 emissions derived from cars. The parameter estimate results obtained from our DDC model are robust, showing that car owners behave with a forward-looking perspective. In addition, it is clear that offering subsidies for car inspection costs can be expected to have a substantial effect on cutting CO2 emissions associated with the transport sector because it would dampen car replacement purchase behavior and thereby increase the average economic lifetime of cars. \n The results of this study show that revising Japan's car inspection system has the potential to cause a major turnaround in the replacement purchase behavior of the nation's car owners, thereby contributing to cutting CO_2 emissions. However, in practice, completely scrapping the current car inspection system would be very difficult. This is because, although abolishing inspections would relieve car owners of a painful cost burden, it might also put the safety of car operation at risk, due to the failure to detect problems that a car inspection would ordinarily detect.

TU222
Li-S batteries for electric vehicles, challenges for circular economy objectives
g. benveniste, C. Corchero, IREC; B. Amante, Universitat Politecnica de Catalunya UPC
The continuous and planned increase of the electrification in the transport sector is one of the main drivers of advances in energy storage for electric vehicle (EV) propulsion and present technological challenges to achieve the expected requirements. The implementation of the EVs on our roads remains a challenge and is below expectations foreseen. The elevated costs of the batteries and thus the EV cost, refrain the massive depletion of this technology. With the aim of reaching a field of 500 kilometers autonomy in the short term, it is necessary to investigate new materials and configurations of EV batteries. To this end, lithium-sulfur (Li-S) batteries are the closest battery technology capable of meeting these expectations. Although Li-S can overcome the technical issues, this solution still needs to demonstrate how the socio-economic-environmental barriers associated are solved, above all when considering their fitting in a circular economy society. There are no clear evidences of the environmental benefits due to the use of Li-S batteries as an alternative to Li-ion batteries. Moreover, there it is still unclear of how these batteries should be treated at their end of life with the aim of recovering the maximum amount of valuable materials. This study focuses on the methodological design to analyze the environmental and social aspects related to Li-S batteries using LCA perspective in a circular economy context. This research has the following objectives: 1)To evaluate their environmental profile; 2) To identify their possible use in a second life, once they cannot be used in an EV (e.g. their use in stationary applications); 3)To evaluate the associated environmental impacts and potential benefits due to material recovering using batteries recycling options. These objectives present a considerable number of challenges due to the lack of data in the Li-S data inventory collection, the uncertainties due to the feasibility of using them in second life and the lack of examples to analyze economic and environmental benefits of designing a customized recycling process. For this reason, the aspects covered by this study are extremely relevant in the frame of considering Li-S batteries technology as a suitable system within the objectives of a circular economy. This research is being carried out within HELIS Project. This project receives funding from the European Union's Horizon 2020 research and innovation program under Grant Agreement No 666221.

TU223
ATISOL C2C - Life cycle assessment as a tool for the ecodesing of a "vapour and air barrier membrane - insulator" system, in a cradle to cradle approach
S. Groslambert, University of Liège - Chemical Engineering / Dpt of Chemical Engineering - PEPs; M. Getlicherman, Derbigum; B. Colson, Sioen Felt & Filtration; I. De Vilder, Centexbel; A. Tilmans, Belgian Building Research Institute (BBRI); A. Léonard, Liège Université / Chemical Engineering - PEPs
The European directive on the energy efficiency of buildings requires the members to put on the market solutions for insulation of buildings that are simple, effective and lasting, but also respectful of the environment and of the users. To reduce energy losses and to guarantee the durability of the thermal insulation, it is necessary to have a vapor and air barrier on the warm side of the building, situated between the thermal insulation and the inside. Hence the passage of moisture from

the building is reduced, preventing condensation problems on the insulating material. Currently, the implementation of an insulation system combined with a vapor barrier presents three major problems: an important time for placing, a random durability in time (stability of tapes of junction, adherence to the existing walls, punching resistance), and finally a low disassembly and re-use level. The ATISOL C2C project aims to develop a complete solution (ecological insulation + renewable vapor barrier + coating), with the lowest environmental impact on its whole life cycle. The solution can be used in both new construction or during renovation. Compared to the state of the art, the solution that is developed is unique and innovative by its simplicity in terms of materials by integration of a vegetal self-adhesive binder to the spunbond reinforcement of the membrane, the latter being also made of renewable resources. The material is appropriate for application on the different wall coverings that can be found in a building. Due to the self-adhesive characteristics, the implementation is made easier in both common surfaces (walls, roofs and ceilings) and to the level of detail such as corners and junctions. In addition, the application of a clay finishing coating on the membrane completes the offer. The constructive system can be dismantled at the end-of-life of the building and the various elements are recovered and valued in a cradle-to-cradle perspective. A first step is already carried out: the Derbiskin©. The preliminary life cycle assessment results support the technical partners along the whole development and evolution of the membrane by pointing out the hotspots of the system, from the choice of the components of the vegetal binder or the spunbond reinforcement to the manufacturing process. This project is supported by the GreenWin Competition Clusters and subsidized by the Walloon Region (BE).

TU224

Life Cycle Assessment of Recycled Asphalt and Biomaterials for Road Pavements

A. Jimenez del Barco Carrion, The University of Nottingham; D. Lo Presti, The University of Nottingham / Nottingham Transportation Engineering Centre NTEC

Transportation is one of the main economy drivers in societies and roads are the most important mean of transport. To build and maintain roads is essential to ensure the efficiency and keep up the level of this service. However, these operations require a high consumption of non-renewable and raw materials (aggregates and petroleum-based materials) which is one of the major concerns nowadays in this field. To cope with this issue, the use of recycled materials in pavement engineering has become very popular in the last decades. In addition to the raw material saving, recycling reduces costs and save landfill space. In this regard, the increase of recycling rates of asphalt materials, aiming at 100%, is key to move towards the implementation of a sustainable circular economy in pavement engineering. However, the amount of recycled material that can be used in a pavement is limited due to some uncertainties related to its long-term performance. To cope with this issue, if high recycled asphalt amounts are to be used in asphalt mixtures, the recycled material has to be treated and new components have to be added in the asphalt mixture. These processes and new components may hide the advantages of using recycled materials from the environmental and economical point of view. Within the ERA-NET Plus Infravation 2014 Call, the project BioRePavation analysed three alternative biomaterials to be included in high-recycled asphalt content mixtures to help increase recycling rates in an European case study. A comparative full Life Cycle Assessment of the asphalt pavement was carried out for each alternative to determine whether the use of recycled asphalt mixture in high amounts including biomaterials still entails environmental advantages. From a preliminary analysis of the results, it is possible to affirm that using the asphalt mixes with high recycled asphalt content generally provided similar or lower carbon footprint than the asphalt mixes currently used in Europe. This type of studies are needed in order to encourage road authorities to use innovative technologies that can promote a circular economy.

TU225

Dynamic vs static LCA to explore the sustainability of industrial waste recycling

A. Di Maria, KU Leuven / MTM; A. Levasseur, École de technologie supérieure / Construction engineering; K. Van Acker, KU Leuven / Materials Engineering

LCA methodology is often used to promote the circular economy in the construction sector. However, LCA presents some methodological challenges when assessing the environmental impacts of building and construction materials. Construction materials can accumulate in buildings and infrastructures for several decades, with considerable stocks of materials along the life cycle. Due to the long life of construction materials, LCA should take into consideration also time related aspects. However, in the current LCA, any temporal information is lost, making most of the LCA methodologies better suited for retrospective assessment rather than forecasting purposes. To fill this gap, this study proposes a time-dependent LCIA on climate change, to assess the carbon footprint of two newly developed construction materials, produced through the recycling of industrial residues (stainless steel slag and industrial goethite). The results of the dynamic LCA are compared to the results of traditional static LCA, to see how the methodological development of dynamic LCA may have an influence on the final environmental evaluation for construction materials. Both dynamic and static LCA results shows that the recycling of industrial residues to produce new construction materials has the potential to mitigate the climate change impacts of construction blocks, by substituting traditional OPC concrete. Although the dynamic LCA did not result in a shift in the ranking between the three materials compared with static LCA, it provides a clearer picture on emission flows and their effect on climate change over time.

TU226

Pursuing the sustainable circular city - is environmental accounting supporting the transition?

A. Petit-Boix, S. Leipold, University of Freiburg / Chair of Societal Transition and Circular Economy

The transition from linear to circular economies is already in the international policy agenda and several actors are implementing this concept at different scales. In particular, cities are engaging in this process in their quest of turning into healthier, more sustainable environments, and they thus promote a number of circular initiatives. However, do these initiatives help to achieve the goals included in local sustainability agendas? Or are they less environmentally favorable than conventional, linear systems? Systematic environmental accounting might give an answer to these questions once decision-makers have access to practice-oriented studies. In this contribution, we seek to determine whether research has effectively quantified the environmental performance of the initiatives promoted in cities. To do so, we gathered the features of circular economy initiatives reported by a pool of cities to understand what they refer to when addressing circular economy. At the same time, we reviewed scientific literature that applied quantitative environmental tools to analyze case studies of circular economy practices. These tools included life cycle assessment, material flow analysis or input-output analysis. Our first results showed that there are many research gaps regarding the impacts of new strategies and a structured evaluation is needed. While research and practice are both interested in the implementation and evaluation of waste management practices, cities are engaged in a variety of initiatives that research has not explored yet, such as urban planning issues. This might put cities at a disadvantage if they are not able to select the most environmentally friendly initiatives that help them achieve their local sustainability goals while approaching circular economy.

TU227

Taking stock of a circular economy within planetary boundaries: A multi-scale analysis through consequential LCA

H. Helander, A. Petit-Boix, S. Leipold, University of Freiburg / Chair of Societal Transition and Circular Economy

Current institutional agendas are embracing the concept of “circular economy” (CE) in order to improve the sustainability of products and services and reduce the resource dependence. CE is applied through a broad range of strategies at various scales and needs to be comprehensively studied so as not to compromise the earth’s safe operating space. For this reason, consequential life cycle assessment (CLCA) provides suitable tools for understanding these changes. Our main goal is to create a methodological framework that enables the assessment of CE strategies across scales within the planetary boundaries. Two assessment levels will be considered, i.e., cities and products/sectors. The framework will be applied and tested through an analysis of bio-based products and sectors (i.e., wooden houses, cardboard packaging and food waste), as well as on European cities applying CE-related strategies. At the city level, CLCA will be combined with territorial LCA to provide information on the environmental variations associated with local CE strategies. Thus, we might be able to determine the impacts of production and consumption activities that meet the needs of a city before and after the application of CE strategies. At the product level, case studies will examine different CE strategies such as eco-design, recycling and cascade use of resources. To address how sustainability changes with scale, sector-wide scenarios for different measures and changes in market shares will be constructed through a CLCA approach. Opportunities and challenges for the specific sector and context will be identified to include side effects and to ensure assessing realistic pathways. In this way, the framework adapts to the specific requirements of each sector. To interpret the results in relation to the safe operating space, the planetary boundaries will provide reduction targets. This will be done by following existing proposals on the integration of LCA results into the planetary boundaries approach. By combining a bottom-up CLCA approach with a top-down planetary boundary framework, we provide a method that can take stock of “real” environmental sustainability progress, at different scales and thereby help companies, cities and countries to understand the environmental implications of CE strategies. This might enable the integration of environmentally friendly CE initiatives into a variety of sectors and scales.

TU228

Opportunities and threats in water treatment options as investigated by LCA

T. van den Brand, KWR Watercycle Research Institute / Water systems and technology; R. Hofman-Caris, KWR Watercycle Research Institute; A. Butkovskyi, Wageningen University WUR; B. Hofs, Evides Waterbedrijf; A. van Wezel, KWR Watercycle Research Institute / Chemical Water Quality and Health; S. Kools, KWR Watercycle Research Institute

In this research two LCA studies are presented as starting points in studies on water treatment processes. Case 1 on drinking water production and case 2 on industrial wastewater treatment. Both cases were aimed to unfold the potential of LCA as a tool to direct future research. We performed the work using Simapro 8.0 software, method Recipe Endpoint (E) and the Ecoinvent 3.0 database. First, in drinking water production, flocculants are used to remove particles, natural organic matter (NOM) and metals (like iron) from water. The sludge formed can be hydrolysed again to recover iron for production of new flocculants. Our LCA study showed that flocculants obtained from iron sludge after HCl dosage have a significant lower environmental impact than commercial available $FeCl_3$. Recovery of $FeCl_3$ flocculants from iron sludge to be applied in the drinking water purification or waste water treatment looks promising and this LCA study underlined that technical research into the quality of the flocculants is justified. A sensitivity analysis indicated that the iron content of the sludge is strongly determining the environmental impact; thus indicating that different types of iron sludge should be considered for further research. As second case study, we investigated water management in shale gas production, since hydraulic fracturing technologies require significant volumes of water for well development and produce high volumes of wastewater with highly variable composition. Different treatment options for waste water from shale gas production were compared in case 2. It is important to note that each process resulted in different effluent quality. Each process included a pre-treatment consisting of dissolved air flotation and biodegradation, followed by either discharge directly to seawater, or treatment with vapour compression distillation (VCD) with water discharge to surface water and injection of the brine back to the deep underground formation. The LCA study on of shale gas waste water treatment indicated that more detailed information on the concentrations of compounds in the waste water is required. A technical research into the efficiency of the VCD, to optimize compound removal from waste water, is recommended. In this study LCA has shown to be an effective tool to evaluate the direction of research within the water sector, evaluate possibilities for resource recovery and determine environmental impacts of processes.

TU229

Closing the loop in a territory: LCA approaches to boost resource recovery

M. Calvet, CETAQUA / MASE; M. Amores Barrero, CETaqua, Water Technology Centre; D. Marin, CETaqua, Water Technology Centre / Environment and Socioeconomics; M. Isasa, CETaqua Water Technology Centre / MASE; M. Termes, CETAQUA; M. Ruiz Mateo, CETaqua Water Technology Centre

The concept of Circular Economy is widely extended in political and business agendas and so is the concept of "Closing the loops". The idea that the value of materials and products should be maintained in the economy as long as possible and wastes minimised is understood and accepted. However, its implementation is bringing to the light questions as to which level to implement it (material, product, system, business and territory), which tools to use to decide on the most appropriate circular economy strategy to develop etc. Territories, understood as cities, municipalities and wider geographical areas, act as accumulators of resources that in the current linear model create negative externalities. However, these water, waste and energy flows if managed in a circular system could be valorised bringing massive opportunities to all territorial actors. This paper explores the application of the Circular Economy in two different case studies in Spain (San Feliu de LLobregat and Gavà in the Catalan Region) for which a methodology has been specifically created. The methodology is validated and its effectiveness demonstrated through the identification of more than 10 Circular Economy Opportunities in each case study. The role of LCA as a tool used in different stages of the Territorial Circular Model is explored i.e. at the data inventory gathering, resource flows analysis, assessment of the most appropriate circular economy strategy and the development of indicators to establish current levels of circularity and benchmarks. The need to develop a tool to assist in the data inventory gathering, data visualisation and material flows analysis to identify a greater number of circular economy opportunities is also highlighted. Conclusions of the research include the need to assess the identified opportunities from a technical, economic and social perspective. The importance of the development of an action plan to assist actors in the implementation of the preferred circular economy strategy is highlighted. Finally, the importance of monitoring of the assessed impacts of the strategy is emphasised.

TU230

Innovative method to optimize territorial organic waste resources

G.C. Vega, The Technical University of Denmark (DTU) / Management Engineering; J. Sohn, The Technical University of Denmark DTU / DTU Management Engineering; M. Birkved, Technical University of Denmark / QSA Dept of Management Engg

A truly environmentally sustainable bioeconomy requires integrative approaches for its design and implementation. A holistic assessment, where several steps are taken into account in order to arrive at thoughtful recommendations for future biotechnologies is proposed. The assessment incorporates common LCAs of biotechnologies with analysis of producer territories in order to provide site specific recommendations that take into consideration different geographical and feasibility constraints, the present and future energy grid, and production capabilities. The authors posit that a multi-criteria approach, such as this, can prevent unforseen burden shifting between environmental impacts while providing implementable decision support. Method: An LCA of various biotechnologies will be conducted with the aim to provide guidance for biorefinery ecodesign that would incorporate emerging biotechnologies and cascading products. Among these technologies, six pathways for anaerobic digestion (AD) and three different pathways to extract polyphenols will be assessed at the product level. At the territorial level a two-pronged approach will be used to achieve a representative data set for the territories, which will consist of material flow data from national and regional sources scaled down to the territory and individual producer data (primary when possible) scaled up to the territory level. A feedback loop will be established between the modules of biotechnology assessment and the foreground system at the level of the territory, in order to observe the effects of waste optimization on the territory. Results: The performance of the biotechnologies will very likely depend largely on energy consumption and the intended use of the new products viz. how the residual resources from wine production are used. At the territorial level the authors posit that local managerial practices, in terms of wine production will be greatly influencial for global warming, eutrophication and resource depletion potentials. Fertilizer inputs, both mineral and organic, and pesticide use will very likely differ from territory to territory and will impact the above mentioned categories as well as toxicity related impact categories. Another important aspect will be the energy consumption of the territories and the influence of future energy grid greening on the future impacts of the technologies proposed today.

TU231

Environmental Benefits of a Circular Economy: Connecting Waste Type and Geographic Proximity

R. Itten, R. Keller, M. Stucki, Zurich University of Applied Sciences / Institute of Natural Resource Sciences

The aim of a circular economy is to transform waste into resources. There is a plethora of waste and by-products that remain unused in the traditional linear industrial system. However, transformation from a linear to a circular system is challenging, limited by several constraints such as the availability of information on the specific composition of the waste, the availability in time and space, the quantity of waste as well as limited knowledge of the usability of such waste products. The goal of the SHAREBOX Horizon 2020 project is the development of a platform for the facilitation of synergies within the industry to enable a more circular flow of resources within the European processing industries. The SHAREBOX platform is a database of available waste and resources required by companies, enabling the transformation of waste to resources by matching supply and demand. The platform also serves as the first point of contact between different partners in a circular system. Furthermore, the platform enables the identification of new synergies overarching the different subsectors of the industries as well as optimal matching from the perspective of a circular economy. We analysed the implications of the transformation of different types of waste to resources when the industries are located in different geographic locations under consideration of the life cycle stage of transformation. Waste PET can be transported up to 10 000 km by lorry and still provide a net benefit regarding greenhouse gas emissions due to circular use. However, in case of concrete, the results are very different. A net benefit only occurs if the additional transport distance compared to primary concrete is less than 5 km. Transformation from linear systems to circular systems can substantially reduce total resource consumption as well as emissions of the whole value chain and therefore contribute to a greener economy. However, matching industries for transformations leading to the substitution of primary materials is still a major challenge. In addition, the environmental benefits of the reuse of resources is limited by the life cycle stage of the transformation as well as by additional transportation that may be required. The completeness of the scope will be crucial for the assessment and generalisations overarching different types of waste remain challenging.

TU232

Evaluation of nutrients and energy recovery technologies through Life Cycle approaches

M. Ruiz Mateo, CETaqua Water Technology Centre; M. Calvet, CETAQUA / MASE; S. Lopez, CETaqua Water Technology Centre / Santiation; M. Isasa, CETaqua Water Technology Centre / MASE; Y. Lorenzo-Toja, CETaqua, Water Technology Centre; D. Marin, CETaqua, Water Technology Centre / Environment and Socioeconomics

Conventional treatments for wastewater treatment are characterized by a high energy consumption, mainly attributed to the oxidation (removal) of carbon and nutrients. In the current economic and environmental context, there is a necessity to find solutions and provide strategies and technologies to be able to change the current concept of Waste Water Treatment Plants (WWTPs) from being energy consumers to resource recovery sites. In the meanwhile, a huge effort is done in the fertilizer industry to produce nitrogen fertilizers or to extract phosphorus from the finite reserves of phosphate rock, that moreover, are located out of Europe. The LIFE NECOVERY project aims at demonstrating, by means of a prototype, the feasibility of a new wastewater treatment approach, based on energy and nutrients recovery. The process tested in the project is based on an initial pre-concentration step that promotes the biosorption process maximizing the biogas production. The effluent of the pre-concentration unit is the influent of a nutrient recovery unit based on adsorption in zeolites. The LIFE ENRICH project (Enhanced Nitrogen and phosphorus Recovery from wastewater and Integration in the value CHain) goes a step further and aims at demonstrating the whole value chain for nutrient recovery in wastewater treatment plants and their valorisation in agriculture through different approaches boosting a model based on circular economy. This study intends to evaluate environmentally and economically the innovative processes tested in the LIFE NECOVERY and LIFE ENRICH projects by comparing them to conventional schemes of wastewater treatment. To do so, Life Cycle Assessment (LCA) has been the selected methodology to quantify the environmental burdens of the innovative schemes and the conventional plants where the prototypes are located: Vilanova WWTP and Murcia Este WWTP. Special focus has been put to impact on climate change, which is expected to be reduced thanks to the recovery of nutrients that could replace chemical fertilisers and due to the higher biogas production and its further valorisation. Life Cycle Costing (LCC) analysis has been undertaken in order to assess all relevant costs associated with the life cycle of both systems. The analysis includes cost incurred during construction phase (CAPEX and civil works) and operation and maintenance phase (OPEX costs e.g. energy, chemicals, transport) and is aimed to identify the most economic-friendly scheme.

TU233

Life Cycle Assessment of a novel process of polyhydroxyalkanoates production with waste and by-products from wine industry value chain

A. Novi, Università di Bologna / Centro Interdipartimentale di Ricerca per le Scienze Ambientali; L. Vogli, Università di Bologna / Centro Interdipartimentale di Ricerca Industriale su Energia e Ambiente; S. Righi, University of Bologna / Physics; S. Macrelli, R. Conti, Università di Bologna / Centro Interdipartimentale di Ricerca Industriale su Energia e Ambiente; C. Samorì, C. Torri, Università di Bologna / Dipartimento di Chimica Giacomo Ciamician; P. Galletti, Università di Bologna / Dipartimento di Chimica G Ciamician; E. Tagliavini, Università di Bologna / Dipartimento di Chimica G Ciamician Alma Mater Studiorum

EU wine production accounts for some 60 percent of worldwide output, with France and Italy being the largest wine producer countries in the world (Gaeta and Corsinovi, 2014). The wine industry influences the environment with the use of soil, water, energy, fertilizers and pesticides. In addition it produces liquid and solid organic waste that has to be managed in the proper manner in order to minimize environmental impacts. In recent years, some innovative technologies have been proposed for the valorization of winery waste and by-products (i.e. grape marcs, grape seeds, vinification lees, etc.) (Devesa-Rey et al., 2011). VALSOVIT is a research project funded by Emilia Romagna Region (Italy) which aims to valorize wine industry by-products. Its focus is the development of an integrated strategy for the transformation of waste from the whole oenological supply chain into high added-value products such as polymers, base chemicals, and molecules for the nutraceutical, cosmetic and agrochemical industries. In this framework, a novel experimental process for the valorization of wine lees and sewage sludge is carried out. These winery residues are subject to anaerobic acidogenic fermentation in order to produce volatile fatty acids (VFAs), which in turn are used to feed a mixed microbial community (MMC) able to accumulate polyhydroxyalkanoates (PHAs) granules as carbon and energy intracellular reserve. The last step consists of PHAs extraction using dimethyl carbonate (DMC). Life cycle assessment is applied to calculate and compare the environmental impacts related to the production of one kg of PHAs to those of an analogous amount of a fossil-based plastic (polypropylene, PP), and two bio-based plastics (bio-polypropylene, bio-PP, and polylactic acid, PLA). PHAs produced from wine industry residues show mixed results. Four different scenarios which vary in terms of feedstock used and pre-treatment process are analyzed. Results show that system setup including pyrolysis pre-treatment allows to reduce both energy demand and GHG emissions more efficiently than the setup without pyrolysis. The latter, on the other hand, is technologically simpler and therefore less effort and cost consuming than the one including pyrolysis. No significant differences between the two feedstock used are found in the results. Gaeta and Corsinovi, 2014. Economics, Governance, and Politics in the Wine Market. Palgrave Macmillan, US Devesa-Rey et al., 2011. Waste Management. 31:2327-2335

TU234

Environmental, social and economic challenges towards a bio-based economy: the STAR-ProBio project, Sustainability Transition Assessment and Research of Bio-based Products

P. Morone, Unitelma Sapienza University of Rome; S. Righi, University of Bologna / Physics; E. Merloni, University of Bologna; L. Summerton, University of York; L. Ladu, TechnischeUniversitaet Berlin; A. Koutinas, Agricultural university of Athens; S. Majer, DBFZ, Deutsches Biomasseforschungszentrum gemeinnützige GmbH / Bioenergy Systems; S. Ugarte, SQ consult; J. Golaszewski, UniwersytetWarminskoMazurski W Olsztynie; K. Waskiewicz, ChemProf; X. Bengoa, Quantis; F. Razza, NOVAMONT; H.V. Haraldsson, 12Swedish Environmental Protection Agency   Naturvardsverket; S. González-García, University of Santiago de Compostela CIF Q1518001A / Chemical Engineering; D. Fedrigo-Fazio, European Environmental Citizens Organisation For Standardisation; M. Grill, AgroVet GmbH

STAR-ProBio is a multi-actor collaborative Research and Innovation Action (RIA) coordinated by Unitelma Sapienza University and including 15 partners from 11 European countries. This project has received funding from the European Union's Horizon 2020 Research and innovation action under grant agreement No 727740. The overall objective of the project is to promote a more efficient and harmonized policy regulation framework for the market-pull of bio-based products. This will be achieved by developing a fit-for-purpose sustainability scheme, including standards, labels and certifications. An integral part of STAR-ProBio is the adoption of life-cycle methodologies to measure environmental, techno-economic and social impacts, and comprehensively assess the roll-out of bio-based products. The analysis of selected **case studies** on construction materials, bio-based polymers, and fine chemicals, will apply benchmarking against non-bio-based products. The project includes several activities, such as the identification of environmental, social and economic criteria to be considered in the development of a sustainability scheme, the development of an LCA approach for strategic and PEF-compliant policy decision support, the sustainability interpretation of end-of-life options taking into account the EU circular economy principles, the development of a methodology to compare techno-economic sustainability of bio-based products versus their fossil-based alternatives, the identification of consumers' sustainability preferences and expectations, the assessment of social and economic benefits of new sustainable value chains and the assessment of the status quo and description of existing approaches to quantify (direct and indirect) impacts of land use changes.

TU235

Integration of a Colombian bio-refinery from industrial palm oil waste into the circular economy

J. Torres, Universidad de la Salle / Grupo de Investigación en Gestión del Riesgo y Cambio Climático; I. Herrera, D. Garrain, A. Gamarra, CIEMAT / Energy Dpt Energy Systems Analysis Unit

Currently, the idea of a circular economy has an important role in the world political and business agendas about to decouple economic growth from resource constraints. Circular economy has not a single definition, nevertheless unlike the traditional linear take-make-consume-dispose approach, it searches to maximize the added value at each point in a product's life. In the Colombian context, palm cultivation is a major non-food agricultural commodity for the economy due to its market abroad. The volume of production places the sixth exporter of palm oil in the world. Furthermore, palm oil mills produce approximately 2 tons of concentrated solid wet biomass per ton of primary product commercialized (oil and kernel). Additionally, 0,7 cubic meters of liquid effluent per ton of fresh fruit bunches is also obtained. The aim of this research is to develop the circular economy approach in the Colombian palm oil industry, to account for the agriculture supplies and demands in a representative sample of the process chain. This study allows the characterization of the quantity of waste to be used in palm oil mill bio-refineries as a representative sample in order to identify potential risks. In addition, the work adds not only criteria for assessing the agricultural palm sector to establish indicators for a sustainable circular economy, but also methodologies based on Life Cycle Analysis to allow efficient management of resources, nutrients and agrochemicals in order to quantify the required amount to produce a given product. The knowledge of these parameters permits the identification of those elements that influence its magnitude, so that, different alternatives can be used to enable the sustainability of the oil palm industry. Finally, this research could contribute to develop knowledge for future decision makings towards the sustainability of resources and the optimization of processes carried out by palm-cultivation companies as part of their policy of environmental responsibility.

TU236

CRADLE-TO-GATE LIFE CYCLE ASSESSMENT OF BIOGAS PRODUCTION FROM PALM OIL MILL EFFLUENT

N. Abdul Aziz, M. Mohd Hanafiah, Universiti Kebangsaan Malaysia / Environmental Science

Exploring renewable energy sources is becoming increasingly important due to its low environmental impacts as compared to the consumption of non-renewable fossil fuel sources. Waste-derived biogas is one of the promising technologies that yields a renewable, sustainable, and green source of energy. In Malaysia, palm oil mill effluent (POME) can be a suitable feedstock for biogas production due to its abundant and high potential in energy generation. However, a comprehensive assessment need to be conducted to ensure the sustainability of POME-based biogas production. This study was conducted to evaluate cradle-to-gate life cycle environmental performance associated with the production of biogas by the anaerobic digestion of POME. The functional unit was defined as 1 tonne of POME used for biogas production and the system boundaries covered the plantation-processing mill-biogas plant stages. The life cycle assessment (LCA) was performed using ReCiPe 2016 environmental impact method and SimaPro 8.4 software. The present study demonstrates that the generation of electricity from biogas is advantageous comparing electricity production in conventional power plants. The results also able to identify hotspots in the life cycle of the biogas production where environmental performance of the system can be improved and environmental benefits can be achieved from the anaerobic digestion of POME with regard to the reduction of greenhouse gases emissions.

TU237

Challenges and open issues in assessing new technologies for circular economy solutions

P. Masoni, Ecoinnovazione srl / Sustainability Department; A. Zamagni,

Ecoinnovazione / LCA and Ecodesign Laboratory; D. Tonon, Ecoinnovazione srl
Circular Economy has become a concept quite known also within the public domain. It is a catchy term that puts together two words easily understood by everybody with a positive meaning. Moreover, it can be easily translated into simple rules/guidelines to follow to claim an improvement of our economy and its relation with the environment, such as: recycle, avoid dangerous substances, extend the life of the products. However, the reality is more complex, and the efforts to close the loops, i. e. reusing waste streams in other life cycles, does not come for free. For example, burden shifts from resource depletion to other environmental impacts are likely and common consequences. For this simple reason, life cycle assessment and life cycle sustainability analysis should be used to support the identification and understanding of the potential advantages of circular economy solutions, with the identification and management of the unavoidable trade-offs. In the case of innovative technologies developed to extract valuable substances from waste streams the complexities of the analysis are related to: scale-up from laboratory or pilot scale to full industrial scale; different possible industrial applications of the technology; a basket of diverse applications of the innovative semi-finished product/ingredient delivered by the new technology; diversity of the function of the technology; complex market of the substituted products, etc. This work presents and discusses how the above-mentioned challenges and open issues, with a focus on the diversity of the function of the technology, have been addressed in a specific case of an innovative technology to extract polyphenols from different waste streams. The presented example shows that the analysis can be rather complex due to the need of addressing different applications, identifying the benchmark and to the fact that the same technology can provide different functions according to the selected perspective. The oral presentation will detail how the main difficulties have been considered and addressed, such as the of scale-up, complex market of substituted products, different geographical location of the technology implementation. One key aspect is that the adopted perspective directly influences the results in terms of environmental preference of the innovative technology, and as such, the benefits of circularity solutions needs to be properly addressed and quantified, and are not inherently beneficial.

TU238

Circular economy: what does restaurant food waste generation data and consumers say?

R. Dagiliute, Vytautas Magnus University / Environmental Science Department; A. Musteikytė, Vytautas Magnus University

Around 88 million t. of food is annually wasted in the European Union. According to FAO (2013), 31–39% of food is wasted at consumption level in developed regions. This wastage has an enormous negative impact on the global economy and food availability and courses major environmental impacts. EU programme "Towards a circular economy: a zero-waste programme for Europe" (COM/2014/0398 final) aims to reduce by half food waste in EU by 2030. Roadmap to a Resource Efficient Europe (COM (2011) 571) aims to change consumption patterns and achieve 20% reduction in the food chain's resource inputs and halved disposal of edible food waste in the EU by 2020. As study (2008) on British households indicates, 61% of wasted food could be consumed if it would be better handled. Hence, changes in consumption patterns are in importance to reach those aims and reduce related impacts. This study analyses amounts of the food waste generated in a restaurant X (Vilnius, Lithuania) and explores consumers' attitude towards this problem. Catering business was closely monitored in terms of customers' flows and food waste generated. To find out consumers' opinion about this problem the guests of the restaurant were surveyed (174 in total). Research has shown that during the six months 14744 kilograms of food was thrown away in a restaurant. Amount of food waste was linked to the total number of customers during the selected timeframe. Most of food was discharged in December, and in the spring quantities of food wasted decreased mostly due to the seasonality. Weekend effect was also registered. Flour products composed the biggest share of all food waste. It was found that the restaurant consumers had low ecological consciousness - inadequate standpoint of consuming too much, poor knowledge about what impact food waste has on environment, recycling, and opportunities to waste less. 73% of those often eating at public caterers indicate often to leave some food uneaten. Only 5% always and 22% often ask to take-away left food. 43% of the rest indicate a shame as a reason to take food leftovers. Most often those were with higher education and higher incomes. Although respondents had no clue on their own negative impact to the environment, they all agree that much more information on solving problems like this is needed. Therefore, policies to encouraging food saving at home and public places should be promoted to deal with "food waste challenge" (2015).

TU239

Assessment of Carbon Footprint of a typical Spanish dietary pattern: The Atlantic diet

X. Esteve Llorens, Universidade de Santiago de Compostela / Chemical Engineering; M. Moreira, G. Feijoo, University of Santiago de Compostela / Chemical Engineering; J. Garrido, Universidad de Santiago de Compostela; S. González-García, University of Santiago de Compostela CIF Q1518001A / Chemical Engineering

Access to adequate nutrition is a basic human need that depends on numerous social, political and economic factors. Similarly, food patterns affect not only to food consumption but also its production, which cause health, social and environmental impacts. In particular, food chains that support diets are linked to environmental issues such as greenhouse gas (GHG) emissions, fossil energy requirements and land use. According to Garnett (2011) and Irz et al. (2016), 15-30% of total GHG emissions in developed countries are derived from food production, distribution and consumption. Therefore, environmental pressures from food systems are on the top of public health agendas, and sustainable food production and dietary patterns are considered of major interest. Consumption patterns vary significantly across Europe. In the southern countries, healthier diets richer in fruits and vegetables have been identified. In this sense, the traditional Atlantic diet is a common dietary pattern in Northern Portugal and Galicia (Northwest of Spain), culturally and climatic similar areas. The Atlantic diet is characterised by an abundant consumption of vegetables, fish and meat, mainly local and fresh products (seasonal food), cooked to maintain its characteristic flavour and taste. For this reason, it has been become a worldwide reference for a healthy diet. The main objective of this study was to quantify the carbon footprint of the Atlantic diet using a simplified Life Cycle Assessment (LCA) approach due to the lack of detailat certain stages of the life cycles of various foods. To do so, the production, transport and processing (when necessary) of the different food ingredients that constitute a typical daily menu was taken into consideration. According to the preliminary results, food production was the main responsible for contributions to the carbon footprint, mostly due to agricultural and livestock activities involved in the production of vegetables, fruit and meat. The findings from this study can be considered as a first step in an attempt to define a sustainable Atlantic diet. Moreover and in line with the literature (Pernollet et al., 2017), the use of a simplified LCA method reports accurate results at a lower demand of data collection than the full LCA. This research has been supported by a project granted by Xunta de Galicia (ED431F 2016/001). S.G-G. would like to express her gratitude to the Spanish Ministry of Economy and Competitiveness (RYC-2014-14984).

TU240

Assessing life-cycle impacts of the sharing economy: how to account for behavioural changes?

D.A. Chapman, KU Leuven / Department of Materials Engineering; K. Van Acker, KU Leuven / Materials Engineering; J. Eyckmans, KU Leuven / Faculty of Economics and Business

The sharing economy, facilitated by digital platforms, is expanding in to more and more areas of the economy and could help the transition to a more circular and sustainable economy. The *a priori* environmental benefits of sharing arise from providing wider access to existing goods, intensifying their use, resulting in fewer goods produced overall. Thus, in order to account for the impacts, goods must be assessed over their entire lifetime, particularly the manufacture and use phases. Life cycle assessment (LCA) is a tool that can be used to estimate such environmental impacts, but a comprehensive assessment should also include the various responses and behavioural changes of consumers to this new sharing marketplace. For example, sharing goods will affect the decisions and habits of consumers in many ways, with corresponding changes to market dynamics. This research consists of a review of studies into the sharing economy, and suggests how consequential LCA can be used to give a more detailed assessment of the environmental impacts. Particular attention is paid to how the behavioural changes in consumption should be accounted for in an LCA applied to the sharing economy.

Innovative techniques for enhancing and monitoring microbial activities for in situ remediation of contaminated sites (P)

TU241

Effects of plant growth and organic carbon addition on DDE degradation in soil

M. Cardoni, National Research Council of Italy / Water Research Institute; F. Mitton, University of Mar Del Plata; M. Di Lenola, National Research Council of Italy / Water Research Institute; L. Patrolecco, Water Research Institute-National Research Council / Water Research Institute; N. Ademollo, F. Spataro, National Research Council of Italy / Water Research Institute; K.S. Miglioranza, University of Mar Del Plata / Laboratorio de Ecotoxicología y Contaminación Ambiental, Instituto de Investigaciones Marinas y Costeras; M. Gonzales, University of Mar Del Plata; P. Grenni, National Research Council of Italy (CNR) / Water Research Institute; A. Barra Caracciolo, National Research Council / Water Research Institute

Although the use of DDT was banned in numerous Countries several years ago, owing to its high lipophilicity and persistence, this pesticide and its metabolites (*p´p´*-DDE and *p´p´*-DDD) are frequently found in the environment. Plant-assisted bioremediation can be a promising clean-up technology to contaminated soil remediation; it relies on the synergistic action between plant rhizosphere and microorganisms to remove toxic substances. In this work, *Solanum lycopersicum* together with dissolved organic carbon were added to DDE contaminated soil for bioremediation purposes in greenhouse microcosms. The experimental set was performed to assess the effectiveness on DDE biodegradation of tomato plant presence with and without the addition of two different DOC (with different humic substance composition). The pots were filled with contaminated soil (1 ppm of DDE) in presence/absence of tomato plants and watered with different kinds of DOC solutions; control soils (with/without plant and/or DDE) were also implemented. The plots were sampled after 40 days from DDE exposure. The effects of the different treatments on the natural microbial community and on DDE biodegradation ability were evaluated in terms of microbial abundance, viability, structure, dehydrogenase activity and DDE residual concentration. The results showed that the plant presence stimulated the overall soil microbial community activity but did not increase significantly the DDE biodegradation. The quality of the organic carbon in terms of fulvic and humic acids presence influenced differently both DDE degradation and microbial activity.

TU242

Soil microbial community associated to a poplar-assisted bioremediation study

A. Barra Caracciolo, National Research Council / Water Research Institute; V. Ancona, Water Research Institute - Italian National Research Council / Water Research Institute; P. Grenni, National Research Council of Italy (CNR) / Water Research Institute; M. Cardoni, M. Di Lenola, National Research Council of Italy / Water Research Institute; C. Campanale, Italian National Research Council; G. Garbini, Italian National Research Council / Water Research Institute; M. Lopez, Consejo Superior de Investigaciones Científicas; G. Aimola, V. Uricchio, Italian National Research Council / Water Research Institute

A poplar-assisted bioremediation strategy has been applying for four years to a historically polychlorinated biphenyls (PCBs) contaminated area in Southern Italy using the *Monviso* poplar clone. This clone was effective in promoting both a general decrease in contaminant occurrence and an increase in microbial activity in the chronically polluted area a little more than one year after planting. In fact, the synergistic interaction between poplar and soil microorganisms in rhizosphere promoted a reduction of overall PCBs concentration under the Italian legal limit (D.Lgs. 152/06) of 60 ng/g soil (Ancona *et al.*, 2018). A further sampling was performed four year later in order to assess the PCB residual concentrations at different depths and distance from poplar tree trunks inside the planting area. At the same time, microbial analyses were carried out to evaluate total microbial abundance, cell viability and dehydrogenase activity. Moreover, nucleic acids were extracted from soil. The hypervariable regions V4-V5 of the 16S rRNA gene were amplified and sequenced by MiSeq (Illumina). The structure of the microbial community in the planted and un-planted (control) soil was performed and compared and bacterial species involved in PCB degradation identified.

TU243

Plant-assisted bioremediation to recover multi-contaminated areas and provide biomass for renewable energy production

V. Ancona, Water Research Institute - Italian National Research Council / Water Research Institute; A. Barra Caracciolo, National Research Council / Water Research Institute; P. Grenni, National Research Council of Italy (CNR) / Water Research Institute; D. Borello, Università La Sapienza / Dipartimento di Ingegneria Meccanica e Aerospaziale (DIMA); M. Falconi, ISPRA Institute for Environmental Protection and Research / Soil Protection Department - Geological Survey of Italy; A. Basile, Regione Puglia / Servizio Rifiuti e bonifica; A. Massacci, Italian National Research Council / IBAF; V. Uricchio, Italian National Research Council / Water Research Institute

Phytoremediation is gaining popularity as a sustainable solution to contaminated soil remediation. In particular, plant-assisted bioremediation exploits synergistic action between plant roots and natural microorganisms (bacteria and fungi) to remove, transform or stabilize toxic substances in soil, sediment or water. Such remediation technology can be effectively applied to contaminated areas. It is based on the use of suitable plant species, selected to stimulate the biodegradation activity of rhizosphere microorganisms (*e.g.* through the production of radical exudates or oxygen release). The plant-assisted bioremediation is an environmental recovery strategy for areas affected by widespread and multiple contamination, ecologically and economically viable. At the same time, this technology can provide wood biomass that can be efficiently treated to produce renewable energy. Among these treatments, biomass gasification is a very efficient process to produce clean energy in the form of a fuel gas (syngas). Among plant species poplar has good energy production potential, can grow in different environments and it is among the fastest growing trees. Furthermore, poplar demonstrated the capability to absorb organic contaminants (i.e. heavy metals) from the soil in which it is cultivated. For these reasons, the potential use of poplar for syngas production has been investigating, using biomass collected from a plant assisted bioremediation area located in a multi-contaminated soil in Southern Italy. The implementation of these technologies is line with the sustainability criteria of the Renewable Energy Directive (EC 2009) and with those of the "circular economy", according to which by recovering energy from a material that would otherwise be a waste, taking care to separate any hazardous pollutants released during the process. An exhaustive Regulation, which establishes threshold limits of contaminants in the biomass and rules on how to manage it outside the remediation site, is necessary.

TU244

Microcosm experiment to assess the effectiveness of a Populus clone to enhance PCB biodegradation in a historically contaminated soil

L. Passatore, National Research Council / Institute of Agro-Environmental and Forest Biology (IBAF); A. Barra Caracciolo, National Research Council / Water Research Institute; M. Di Lenola, National Research Council of Italy / Water Research Institute; P. Grenni, National Research Council of Italy (CNR) / Water Research Institute; I. Nogues, National Research Council of Italy / Institute of Agro-Environmental and Forest Biology (IBAF); E. Guerriero, P. Benedetti, National Research Council / Institute of Atmospheric Pollution Research; A. Massacci, Italian National Research Council / IBAF

Greenhouse experiments have been performed to test the capacity of the *Populus* species (clone Monviso) to grow on a PCB historically contaminated soil and to improve soil quality in terms of contaminant transformation and autochthonous microbial community abundance and activity. The experimental set-up consisted of pots filled with the contaminated soil and poplar cuttings, under the following conditions: microbiologically active soil (TMA), previously sterilized soil (TS), microbiologically active soil in hypoxia (TMAA). Moreover, non-planted soil was used as control. PCB concentrations in soil samples and plant roots were analysed 6 months and 12 months after the start of the experiments. At the same time plant growth, biomass production and plant stress indicators (i.e. chlorophyll content, leaf fluorescence, antioxidant in plant tissues) were investigated together with cell abundance, diversity and viability of soil microorganisms under the different growing conditions. The overall results showed the capability of the clone Monviso to grow, transform and partially bioconcentrate PCBs in roots. The PCB transformations were initially higher in the microbiologically active soil; subsequently in line with a high microbial growth of the sterilized soil, the amount of indicator congeners found were similar between the two treatments. The anoxic treatment differed in terms of congeners detected, microbial community structure and activity and plant physiology stress indicators. However, the Monviso clone showed an unexpected capacity to produce biomass under flooding treatment. Consequently, the switching of aerobic and anaerobic conditions in rhizosphere can be a promising strategy to promote both the degradation of high/low chlorinated PCB congeners. This study was a preparatory experiment for a field experiment of plant-assisted bioremediation

TU245

Are PCB half-lives obtained in rhizoremedition experiments reliable? Pitfalls in experimental design and suggested guidelines for conducting the experiments

E. Terzaghi, University of Insubria (Como) / Department of Science and High Technology, Como; E. Zanardini, C. Morosini, University of Insubria / DSAT; G. Raspa, Sapienza University of Rome / Department of Chemical Engineering Materials and Environment; S. Borin, University of Milan / DeFENS; F. Mapelli, University of Milan; L. Vergani, University of Milan / Department of Food, Environmental and Nutritional Sciences; A. Di Guardo, University of Insubria / Department of Science and High Technology

In the last two decades bioremediation technologies have become ever more important as a sustainable alternative to traditional remediation techniques. In particular, there has been an increasing attention on rhizoremediation techniques, employing plant roots and their associated microorganisms to enhance the degradation of organic contaminants in soil. Many short-term laboratory/greenhouse experiments and long-term field trials have been conducted to investigate the most suitable plant species and environmental conditions that stimulate and favour microbial activities in the degradation of Polychlorinated Biphenyls (PCBs). Recently, an attempt to extrapolate rhizoremediation half-lives (rhizo-HLs) for the ten PCB families from these studies has been made (Terzaghi et al., 2018) providing important data for multimedia fate models that aim to predict the time needed to achieve regulatory thresholds in a PCB contaminated site where rhizoremediation techniques are applied and therefore to draw up its remediation plan. However, many of the studies available in the literature (more than the 80%) were not correctly set up to allow the calculation of PCB rhizo-HLs and could not be considered. In particular the main pitfalls in the experimental design referred to the type of chemicals (single congeners vs. mixture), contamination (spiked vs. aged) and experiment (greenhouse vs. field), the experimental time, the set-up of appropriate controls and replicates as well as the analytical and microbiological techniques adopted. The present work aims to 1) list and discuss the main pitfalls in the experimental design of previous and current rhizoremediation experiments and 2) propose guidance to perform appropriate experiments to obtain comparable, accurate and useful data for rhizo-HLs calculation. Moreover rhizo-HLs will be presented and compared with those obtained with other approaches.

TU246

Effect of Organic and Inorganic Fertilizers on the Bioremediation of Used Motor Oil Polluted Soil

P. Ferdinand, U.E. Ezeji, Federal University of Technology Owerri / Biotechnology

Three treatments (poultry manure (PM), Nitrogen Phosphorus Potassium fertilizer (NPK), and a combination of both) were used for bioremediation of soil spiked with used motor oil to determine the potential of these treatments in enhancing biodegradation of used motor oil in soil. The degree of biodegradation of the oil was studied for a period of 4 weeks under laboratory conditions. Hydrocarbon-utilizing bacteria counts were high in all the poultry manure-amended soil ranging between 9.0×10^6 and 30×10^6CFU/g compared to unamended control soil throughout the 4 weeks of study. Oil-contaminated soil amended with a combination of poultry manure and NPK fertilizer showed the highest reduction in total petroleum hydrocarbon with loss of 80% in the 4th week compared to other treatments. The results obtained demonstrated the potential of the treatments for oil bioremediation in the order: Poultry Manure and NPK > Poultry Manure > NPK.

TU247

Soil pollution and physico-chemical properties steer the bacterial community structure in the uneven highly polluted SIN Brescia-Caffaro site.

F. Mapelli, University of Milan- DeFENS / Department of Food, Environmental and Nutritional Sciences; L. Vergani, University of Milan / DeFENS; E. Terzaghi, University of Insubria (Como) / Department of Science and High Technology, Como; G. Raspa, Sapienza University of Rome / Department of Chemical Engineering Materials and Environment; E. Zanardini, C. Morosini, University of Insubria / DSAT; A. Di Guardo, University of Insubria / Department of Science and High Technology; S. Borin, University of Milan / DeFENS

Contaminants are strong ecological drivers steering the microbiome structure in polluted soils. Bioremediation relies on the residing microbial communities and their activity but can be limited by spatial heterogeneity of microbial populations, contaminants and soil chemistry. Studies aimed at identifying the drivers of microbiome selection are therefore pivotal to develop *in-situ* bioremediation technologies. In this perspective, the National Priority Site SIN Brescia-Caffaro (Italy) offers a fascinating natural laboratory due to its extremely high, old, mixed and uneven soil contamination by metals and organic pollutants, in particular polychlorinated biphenyls (PCBs). Aim of our study was to evaluate the spatial correlations between environmental factors (pollutant fingerprints and soil physico-chemical properties) and the soil microbial community structure. More than 120 soil samples were collected in three areas in the SIN-Caffaro along a tridimensional geostatistically conceived grid, and were analyzed to estimate the soil hydrolytic activity, the physico-chemical features and the concentration of metals and 79 PCB congeners. A cultivation-independent approach led to unravel the phylogenetic structure of the residing bacterial communities. By means of statistical analyses, we showed that significantly different bacterial communities were selected in the investigated areas within the SIN Brescia-Caffaro. Spatial distribution of bacterial populations within each site was significantly correlated with physico-chemical soil parameters and pollutant concentrations. Soil physico-chemical properties were also significantly correlated to the hydrolytic activity of the soil microbiome, a relevant indicator of soil quality and pollutant availability in historically contaminated sites. The results demonstrated that the soil properties and the contaminant profile in the SIN Brescia-Caffaro shaped the structure of the residing bacterial communities, leading to hypothesize that it drove the selection of populations able to degrade the contaminants. The detection in the SIN Brescia-Caffaro soils of the *bphA* gene, codifying for the biphenyl dioxygenase involved in the aerobic PCB degradation process, confirmed that they host an intrinsic natural attenuation potential, possibly exploitable for future remediation interventions. This study also highlighted the prospect of exploiting spatial patterns of bacterial diversity as proxies for monitoring polluted sites.

TU248

Laboratory-scale assessment of bioremediation of hydrocarbon-contaminated soil

F. Diana, University of Milano Bicocca; T. Stella, University of Milano-Bicocca / DISAT; M. Daghio, University of Milano - Bicocca / Department of Earth and Environmental Sciences; F. Pittino, University of Milano Bicocca; R. Ferrari, A. Francioli, HPC Italia s.r.l.; A. Franzetti, University of Milano - Bicocca / Department of Earth and Environmental Sciences

Recalcitrant hydrocarbons often persist in contaminated environments. Biological remediation strategies (bioremediation) are a widely used approach to remove hydrocarbons. This study focused on bioremediation of hydrocarbons contaminated soil from an industrial active site using biopiles. The site is contaminated by light and heavy hydrocarbons, the latter ones representing the most recalcitrant fraction. Bioremediation could be the best solution to recover this area considering both the size of the area and the economic/environmental costs of other technologies such as Dig&Dump. Biopiles will be built to treat the contaminated soil, air insufflation and nutrient addition will be considered to stimulate the aerobic biodegradation of hydrocarbons. In order to optimize this process, a lab-scale test was carried out and three different conditions were tested: natural attenuation (NA), addition of sawdust (SW) to soil to improve the soil structure and addition of compost as amendant (CO). Thirty-six bioreactors were set up (6 sampling points for each condition in duplicate) and incubated for 180 days. Air pumps were used to insufflate air into bioreactors with the exception of NA ones. Laboratory analyses were performed on soil and soil gas samples at the beginning of the experiment and 6 samplings were carried out during the incubation period. Chemical analyses (GC-FID) of total petroleum hydrocarbons (TPH) were performed to evaluate the degradation rates and microbiological/molecular analyses (Total Bacterial Count, Most Probable Number-MPN, High-throughput sequencing of the 16S rRNA gene and quantitative PCR) to assess the growth of bacteria potentially involved in the degradation process. The highest degradation rate was observed in CO bioreactors (first-order rate constant $K=0.180\ d^{-1}$) while lowest rates were observed in NA ($K=0.004\ d^{-1}$) and SW ($K=0.011\ d^{-1}$) in the first 60 days of incubation. However, a residual TPH concentration of » 900 ppm was reached in all bioreactors after 180 days starting from an initial concentration of 2660 ppm. The microbiological characterization suggested a selection of the bacterial community according to the chemical results. In this respect, MNP results showed a significant increase in the number of diesel-growing microorganisms in CO bioreactors. These data will be confirmed by qPCR of the catabolic gene *alk*B encoding for an enzyme potentially involved in the biodegradation of hydrocarbons (on-going analysis).

TU249

Influence of Surfactants and Mycobacterium vanbaalenii PYR-1 Bioaugmentation on 14C-Pyrene Mineralization and Microbial Community Structure in PAH-Contaminated Soils

D.C. Wolf, University of California-Riverside / Environmental Toxicology; J. Gan, University of California, Riverside / Department of Environmental Sciences

Polycyclic aromatic hydrocarbons (PAHs) are ubiquitous environmental contaminants that have potential mutagenic, carcinogenic, and teratogenic properties. Bioremediation has been recognized as a versatile approach to remediate PAH-contaminated soils. However, the biodegradability of PAHs is limited by their bioavailability to microorganisms in the soil porewater fraction. To expedite biodegradation, surfactants at the critical micelle concentration (CMC) has been added to enhance the bioavailability of PAHs. The aim of this work was to evaluate the effects of Brij-35 nonionic surfactant and rhamnolipid biosurfactant at three concentrations (0.1X, 1.0X, and 10X CMC) and the bioaugmentation of *Mycobacterium vanbaalenii* PYR-1 in PAH-contaminated soil using ^{14}C-pyrene as a model compound. Additionally, the bacterial community structure after the 50 day incubation was analyzed using 16S rRNA gene high-throughput sequencing and PAH-degrading genes were predicted using phylogenetic investigation of communities by reconstruction of unobserved states (PICRUSt). The addition of Brij-35 surfactant at all three concentrations resulted in increased PAH mineralization after 50 days incubation by indigenous microbe populations in the sandy loam and clay soil compared to the unamended and rhamnolipid-amended soil treatments. The bioaugmentation of *M. vanbaalenii* PYR-1 had an immediate impact on PAH mineralization in both soils, resulting in 60% mineralization after 10 days. The addition of rhamnolipid delayed PAH mineralization in both bioaugmented soil treatments in a dose-dependent manner. It appears that the rhamnolipid biosurfactant acted as a more favorable carbon source compared to ^{14}C-pyrene and was preferentially degraded. Similar PAH-degrading genera increased in relative abundance after PAH addition, especially *Bacillus* and *Sphingomonas*. Species richness and Shannon diversity decreased following the addition of ^{14}C-pyrene compared to the uncontaminated soil and the addition of rhamnolipid biosurfactant at 10X CMC in all soil treatments resulted in the lowest species richness and Shannon diversity. Using PICRUSt, PAH-degrading genes such as PAH dioxygenase subunits and aldehyde dehydrogenase were greatest in bioaugmented soil treatments compared to native soil treatments. Overall, the results of this study provide beneficial insights towards the abiotic and biotic processes as well as their complex interactions in the bioremediation of PAH-contaminated soils.

TU250

Italian field results of Emulsified Lecithin-based Substrate used as ERD treatment of Chlorinated Solvents in groundwater

A. Leombruni, M. Mueller, PeroxyChem LCC; F. Morlacchi, Centro Assistenza Ecologica

ELSMicroemulsion is a food-grade carbon that supports the treatment of a wide range of groundwater contaminants, including chlorinated solvents. ELS is the acronym for Emulsified Lecithin Substrate, a technology designed to create reducing conditions and to promote enhanced reductive dechlorination (*ERD*) reactions. In general, organic carbon addition in a saturated zone is well-known to promote conventional enzymatic reductive dechlorination reactions. This happens because carbon in the subsurface will support the growth of indigenous microbes in the groundwater environment. As bacteria feed on the soluble carbon, they consume dissolved oxygen and other electron acceptors, thereby reducing the redox potential in groundwater. As bacteria ferment the ELS, they release a variety of volatile fatty acids (VFAs) such as lactic, propionic and butyric, which diffuse from the site of fermentation into the groundwater plume and serve as electron donors for other bacteria, including dehalogenators. Lecithin itself is composed primarily of phospholipids, which have both hydrophilic and hydrophobic regions in their molecular structure. As a result, ELS tend to be stable emulsions, expectedly more stable than with only hydrophobic compounds. Further, phospholipids support remediation by providing essential nutrients (carbon, nitrogen, phosphorus) to bacteria. ELS Reagent was shown to effectively treat tetrachloroethylene (PCE) and its catabolites in the aquifer. The site is a former manufacturing facility in Italy impacted for more than 2,000 m^2 with PCE from a historical solvent release. The main contaminated area and the down gradient plume showed maximum PCE concentrations up to 5,000 ppb in the swallow aquifer. In 2016, the consultancy firm performed a field scale injection of ELS with a goal to reduce the PCE mass and its catabolites in the source area and the distributed plume and treat any residual

VOCs potentially migrating from beneath the former facility. A total of 4900 kg of ELS concentrate was emulsified and injected under pressure thought 51 fixed wells in the swallow contaminated aquifer. Subsequent field monitoring showed PCE and TCE below detection limits at all wells after 6 months. A 99.8% reduction of PCE and TCE was observed in the source and plume areas along with the reduction of the recognized catabolites, such as DCE or VC. Moreover, complete reductive dechlorination of 1,2-dichloropropane has also been observed in all the monitoring wells.

TU251

Cheese whey effects on microbial communities in contaminated groundwater of an urban area

D. Vlkova, Technical University of Liberec / Institute of Nanomaterials, Advanced Technologies and Innovation; I. Dolinova, Regional Hospital in Liberec / Centre of Clinical Biochemistry; S. Wacławek, A. Ševců, Technical University of Liberec / Institute for Nanomaterials Advanced Technology and Innovation

Chlorinated ethenes (CE) are the second most ubiquitous contaminants worldwide. Herein we describe an urban locality Novy Bydzov (Czech Republic) where groundwater pollution was identified in private wells in 2007. The source of CE was machinery, metal cutting, and chemical industry, now out of order. The improper handling of hazardous compounds (e.g. chlorinated hydrocarbons, mineral oils etc.) caused uncontrolled contamination of Quaternary aquifer which is about 4-5 meters thick, composed of sandy gravel and delimited by impermeable 400 meters thick Mesozoic strata. Application of different carbon sources (lactate, glycerol, cheese whey and polyhydroxybutyrate) on the CE-contaminated groundwater was previously tested in the bench-scale studies and based on these experiments, cheese whey was chosen for the *in situ* application. The effect of three consecutive cheese whey applications (first was in October 2017) on indigenous microbes was described using qPCR Due to the techniques after sampling time the DNA extraction was performed using a FastDNA Spin Kit for Soil according to manufactures' protocol. Extracted DNA was quantified using Qubit 2.0 fluorometer. Isolated samples were tested using qPCR method. An universal marker, 16S rDNA gene (total bacteria marker) was used as a control. Other monitored specific markers were focused on presence of *Dehalococcoides, Dehalobacter, Sulfurospirillium* and vinyl chloride (VC) reductases vcrA and bvcA. In addition denitrifying bacteria were monitored by *nirK* marker and sulfate reducing bacteria by *dsrA2* marker. All data are counted in relative values. Higher bacterial abundance was detected based on all tested markers after the first cheese whey application. This application will be repeated two more times. Generally, application was successful and bacterial biomass and specific markers for organohalide respiration increased and prevailed in higher concentrations. Moreover, higher bacterial abundance triggered efficient sequential dehalogenation of the CE contaminants. Specific markers are still being monitored in the treated groundwaters and will be discussed together with physico-chemical results.

TU252

The Influence of Nanoscale Zero-valent Iron (nZVI) in Combination with Various Organic Compounds (Modifiers) on Dehalorespiring Microflora

K. Markova, Technical University of Liberec / Institute for Nanomaterials, Advanced Technology and Innovation; D. Vlkova, Technical University of Liberec / Institute of Nanomaterials, Advanced Technologies and Innovation; I. Dolinova, Technical University of Liberec / Institute for Nanomaterials Advanced Technology and Innovation; J. Nosek, Technical University of Liberec

Among all the groundwater contaminants chlorinated ethylenes (CE) such as trichloroethylene (TCE) can be transformed by combination of abiotic and biotic methods under anaerobic conditions. Currently, nanoscale zero-valent iron (nZVI) is used for the treatment of chlorinated compounds via its strong reducing property. Biological reductive dechlorination of CE is contributed by dehalorespiration. The influence of nZVI in combination with carboxymethyl cellulose (CMC), molasses and detergent (anionic surfactant) on the specific dehalorespiring microflora was tested within this study. Groundwater contaminated with CE (1,2-cis-DCE and TCE) was collected from the chemical factory Spolchemie a.s. Batch tests with iron composite and various concentrations of CMC (0,25, 0,5 and 1 g/l), detergent (5, 10 and 20 g/l) and molasses (5, 10 and 20 g/l) were performed for periods ranging from 6 to 26 days. DNA was extracted after filtration of the tested water and used as a template for a real-time PCR amplification. 16S rDNA gene was used as a total bacterial community marker. Specific genes were used for detection of ongoing reductive dehalogenation (vcrA, bvcA, Dre DHC-RT and Dsb) and to monitor denitrifying and sulphate reducing bacteria (nirK and apsA). CMC bacteria protecting effect when nZVI is applied was observed. Positive effect was exhibited in total bacteria amount (16S rDNA), denitrifying (nirK) and sulphate reducing bacteria (apsA). CMC as the substrate for dehalorespiring bacteria was not confirmed. Detergent enhances nZVI subsurface migration parameters. Direct positive effect on bacterial populations only in denitrifying bacteria was observed. Detergent had even inhibiting influence on dehalorespiring bacteria. Molasses as carbon and electron source had positive effect on all studied groups of bacteria. Interestingly, in combination with nZVI molasses enhanced growth of dehalorespiring but not denitrifying and sulphate reducing bacteria. Molasses is suggested to serve as the substrate for fermentation which produces electrons utilised by dehalorespiration. Molasses as the substrate and nZVI with its pH buffering capacity presented the best conditions for dehalorespiring bacteria. The authors acknowledge the assistance provided by the project No. TF02000064 supported by TACR.

TU253

Mechanistic insight into microbial reductive dehalogenation

S. Zhang, UFZ   Helmholtz Centre for Environmental Research / Department of Ecological Chemistry; G. Schuurmann, Helmholtz centre for environmental research - UFZ / Department of Ecological Chemistry

Microbially mediated reductive dehalogenation provides a promising approach to remediate and detoxify halogenated aromatics. Despite extensive respective studies, the mechanistic understanding of the underlying chemical reactions is still limited. Interestingly, *Dehalococcoides mccartyi* strain CBDB1 and *Dehalobacter* strain 14DCB1 share a common substrate spectrum but yield different dehalogenation patterns, suggesting different sites of primary attack (aromatically bound halogen vs. H) by the nucleophile cob(I)alamin (vitamin B12).[1] The latter was unraveled through quantum chemical analyses of respective electronic structure characteristics. Building on these recent results, a perturbational molecular orbital (MO) approach has been developed for a more detailed analysis of the molecular initiating event triggering the reductive dehalogenation. Application to 93 aryl halides covering chlorinated benzenes, phenols, anilines, biphenyls, dibenzo-p-dioxins, and brominated benzenes reveals that the lowest symmetry-compatible σ* orbital located at the carbon-halogen bond mediates the dehalogenation step, and enables discriminating CBDB1-active from non-active substrates to 92%.[2] In the present communication, these findings are discussed including applications of the MO approach for predicting dehalogenation pathways and regioselectivity. Overall, our approach supports the view that the reductive aromatic dehalogenation proceeds through an inner-sphere electron transfer. [1] Zhang, S.; Wondrousch, D.; Cooper, M.; Zinder, S. H.; Schüürmann, G.; Adrian, L. 2017. Anaerobic Dehalogenation of Chloroanilines by *Dehalococcoides mccartyi* Strain CBDB1 and *Dehalobacter* Strain 14DCB1 via Different Pathways as Related to Molecular Electronic Structure. Environ. Sci. Technol. 51, (7), 3714–3724. [2] Zhang, S.; Adrian, L.; Schüürmann, G; submitted 2017.

TU254

Bacterial biosorption of PFOS from contaminated waters

M. Stylianou, Orebro University / The Life Science Centre-Biology; I. Ericson Jogsten, Orebro University / MTM Research centre; P. Olsson, Orebro University / The Life Science Center-Biology; J. Jass, Orebro University / SCHOOL OF SCIENCE AND TECHNOLOGY

Per- and polyfluorinated alkyl substances (PFASs) have been extensively used for commercial and industrial products since the mid-1900, and are still in use although they have been classified as bioaccumulative hazardous organic compounds (Stockholm convention 2009). Perfluorooctane sulfonate (PFOS) is highlighted as the most abundant PFAS reported to contaminate the environment, animals and humans. The most frequently applied method for PFOS remediation of water is by passing it through activated granular carbon filters. Currently, there are increasing efforts to find new strategies for equally efficient and cost-effective methods for PFOS remediation of contaminated waters. This study investigated the possibility of removing PFOS by microbial binding. We tested the binding capacity of live and dead *Escherichia coli* OP50 in different PFOS concentrations. The exposed bacterial pellets were subsequently analyzed for PFOS by UPLC-MS/MS. The deadbacteria were found to have high adsorption (286-3324 µg/g of bacterial pellet) whereas live *E. coli* cells showed 5 – 7 fold lower binding capacity (38-675 µg/g of bacterial pellet). Importantly, the data also revealed that dead bacteria have at least equal affinity for PFOS isomers as the linear compound; which defines the applicability of PFOS bioremediation with dead bacteria as a promising alternative approach. We propose that microbial binding of PFOS can be applied as a novel, less costly technique for PFOS environmental elimination.

TU255

Hexavalent chromium reduction in a biocathodic microbial electrolysis cell

G. Beretta, Politecnico di Milano / Civil and Environmental Engineering; A. Mastorgio, E. Sezenna, S. Sabrina, Politecnico di Milano

Groundwater is the environmental matrix most frequently affected by anthropogenic hexavalent chromium contamination. Due to its cancerogenicity, Cr(VI) has to be removed, hopefully using environmental-friendly and economically sustainable remediation technologies. To overcome the limits of the currently applied bioremediation technologies, an alternative strategy is the use of BioElectrochemical Systems (BESs) to stimulate bioreduction of Cr(VI). BESs include a set of technologies based on biological reactors where an electrode (anode) can function as the final electron acceptor for the oxidation of organic compounds; then electrons flow through the circuit and reach the cathode that acts as the electron donor for the bioreduction of oxidized species. In the present study, we have assessed if Cr(VI) can act as an efficient terminal electron acceptor for an anaerobic biocathode in a Microbial Electrolysis Cell (MEC). The cathode was first inserted into the cathodic compartment of a dual-chamber Microbial Fuel Cell, and inoculated with autotrophic colture origenate from anaerobic digester sludge. After 30 days of acclimation, the electrode was transferred into the cathodic chamber to work at -300 mV (vs. SHE) as the biocathode in a Cr(VI)-reducing MEC. An abiotic control and an open circuit (OC) control were also operated in parallel. Hexavalent chromium dissolved concentration was analyzed at the initial, during the experiment and final time by spectrophotometric method, while the dissolved total chromium was analyzed by ICP-MS. During the whole test, the current intensity was monitored. At the end of the experiment, the microbial characterization of the communities enriched on the biocathode and in the cathodic solution was performed by 16S rRNA gene sequencing. The acclimation phase in the MFC allowed the formation of an electroactive biofilm on the electrode. A decrease in Cr(VI) concentration was observed at the end of the tests, both in the polarized reactor and in the OC reactor. However, the BES ensured higher removal efficiency than the pure chemical process. In addition, higher current values were measured in the BES compared to the abiotic control, thanks to the biofilm interaction with the electrode. The results from microbial characterization showed that the bacterial community on the surface of the electrode was affected by the cathodic polarization, and it was different from the biomass on graphite in the open circuit system.

TU256

Enhancing Reductive Dechlorination Combined with In-Situ Chemical Reduction for the Remediation of a Heavy Contaminated Chlorinated Solvents Source Zone in South of Italy

f. arjmand, I. Bona, L. Moretti, M. Cremonesi, CH2M Hill

The present site comprises an urban site where a historical Chlorinated Compounds-CHC (mostly PCE) contamination has been released in aquifer before eighties, and characterized by a long-term monitoring activity. Contamination is present in shallow aquifer and was higher than 10 mg/L. The efficiency of the remediation is currently about 99.9%, removed more than 300 Kg PCE. The site characterization integrated with a MIP investigation to identify the plume. The plume has been addressed into four areas then a combination of In-Situ Enhanced Reductive Dehalogenation and In-Situ Chemical Reduction was selected to secure contaminant removal due to biodegradation, approaching the electron donors for PCE. This combination allows to have a reducing ambient due to producing hydrogen which helps groundwater to reach an anaerobic environment which is favorable for the microorganisms to degrade the PCE into the end product, ethylene. The first injection applied in a pilot scale (Phase1) to calibrate the injection for the site conditions. Based on the successful results of this phase, the full-scale planned for phase two and applied in two steps. First step covered the northern part of the plume (area A) in the upgradient and main source zone (area B) which is the most contaminated area. In Area B also the vadose zone has been treated. After a year (step 2), the injection took place in area C near to the site boundary and in area D downgradient of the site. Due to PCE bioremediation we have production of daughter products to prevent the accumulation of these by product an air sparging and soil vapor extraction plants have been installed in the site boundary to remove them from the soil vapor and aquifer. During the ERD we have observed methane production because of methanogenesis reaction, CH2M has decided to install a biofiltration plant, to prevent any dangers for the residential areas nearby. The challenge this complex geology has been solved by using fixed injection points with non-return valves corresponding to the depth of treatment in each aquifer. This allowed for accurate and tailored dosage application of the product without any risk of cross-contamination. Due to the rapid effect of injection, it has been possible to observe very good reduction rates within only few months from the application. PCE, has already shown reduction of three orders of magnitude and in some points, we reached the target, with daughter compounds appearing without accumulation.

TU257

Bioelectrochemical sulfide scavenging from hydrocarbon contaminated marine sediments

M. Daghio, University of Milano - Bicocca / Department of Earth and Environmental Sciences; E. Vaiopoulou, Ghent University / Center for Microbial Ecology and Technology (cmet); C. Perri, University of Milano-Bicocca / Department of Earth and Environmental Sciences; M. Zoeter Vanpoucke, Ghent University / Center for Microbial Ecology and Technology (cmet); A. Sherry, Newcastle University / School of Civil Engineering & Geosciences; C. Cruz Viggi, National Research Council / Water Research Institute (IRSA); I.M. Head, Newcastle University / School of Civil Engineering & Geosciences; A. Franzetti, University of Milano - Bicocca / Department of Earth and Environmental Sciences; F. Aulenta, National Research Council / Water Research Institute (IRSA); K. Rabaey, Ghent University / Center for Microbial Ecology and Technology (cmet)

Thermodynamically favorable electron acceptors (e.g. oxygen) are quickly consumed in hydrocarbon contaminated marine sediments. In this environment the biodegradation of contaminants often occurs via anaerobic pathways. Due to the high abundance of sulfate in marine water, hydrocarbons are often degraded by sulfate reduction and toxic sulfide is produced. Several studies have showed the possibility to couple anaerobic hydrocarbons degradation to current production in Bioelectrochemical Systems (BES), in which an electrode (anode) can be used as a solid electron acceptor by several groups of microorganisms. Anodic sulfide oxidation is another important process that can scavenge sulfide from the environment through biological and abiotic processes. The aim of this work was to assess if the bioelectrochemial stimulation is an effective strategy to promote both hydrocarbons and sulfide removal from marine contaminated sediments. A reactor (POL) was built by connecting a bioelectrochemical cell to three holders containing artificially contaminated sediment. The anode (polarized at 0 mV *vs* Ag/AgCl) was made by a graphite plate and the cathode was made using a stainless steel mesh. Weathered North Sea crude oil was used to contaminate the sediment. Artificial marine water was continuously recirculated into the system (flow rate 0.69 L/day). An abiotic control (ABI) and an open circuit control (OC, disconnected electrodes) were also set up. Total petroleum hydrocarbons (TPH) in the sediment, sulfur species, and current production were monitored over time. Samples of the sediment and of the anodic biofilm were collected to characterize the microbial communities by high-throughput sequencing of the 16S rRNA gene. TPH removal was observed in all the tested conditions. Contaminants removal was linked to current production up to around 5 mA (POL) and negligible current was observed in ABI. Sulfate reduction was also observed indicating the involvement of the sulfur cycle in the process. Members of the families *Desulfuromonadaceae* and *Prolixibacteraceae* dominated the anodic community.

TU258

Freshwater sediment enrichments to improve MFCs performance for in situ remediation application: a phylogenetic microbial characterization

C. Armato, University of Torino / Department of Public Health and Pediatrics; D. Ahmed, Istituto Italiano di Tecnologia / Centre for Sustainable Future Technologies (CSFT@PoliTo); D. Traversi, University of Torino / Department of Public Health and Pediatrics; V. Margaria, M. Quaglio, Istituto Italiano di Tecnologia / Center for Sustainable Futures@Polito; G. Gilli, University of Torino / Department of Public Health and Pediatrics; G. Saracco, Istituto Italiano di Tecnologia / Centre for Sustainable Future Technologies (CSFT@PoliTo); T. Schilirò, University of Torino / Department of Public Health and Pediatrics

One of the possible application for Microbial Fuel Cell (MFCs) is the in situ remediation of contaminated sites. MFCs operation links the removal of pollutants from contaminated sites to the production of current by means of the activity of electrochemically active microorganisms (EAMs), able to degrade substrate producing a flow of electrons. EAMs have potential applications in bioenergy production, green chemical synthesis, bioremediation, bio-corrosion mitigation, and biosensor development. The aim of this work was to investigate the effect of two enrichments, a general (Gen) and a ferric citrate (FeC) ones, to increase the percentage of EAM in order to improve the MFCs performances. A freshwater sediment (Fw) sample was chosen as inoculum source. The effect of the enrichment procedures was compared in term of both electrochemical performance and biological characterization. The microbial community was subjected to three sequential enrichments and then used as inoculum for the MFCs. Anodic potential and voltage were continuously monitored. DGGE, sequencing and rt-qPCR techniques were used to investigate the EAM community. Moreover microbial α-diversity was calculated. The enrichment effect was evaluated both for the precultures and for the three components of MFCs (planktonic, biofilm and rod). Results showed that the MFC inoculated by Gen enrichment preculture had better performance than the FeC one (shorter start-up time, lower anode potential, higher current and power density). The main source of variability resulted to be the kind of enrichment, both in the preculture and in the MFCs. Proteobacteria, Bacteroidetes e Firmcutes resulted as the main Phyla in our samples. *Geobacteraceae spp.* and *Pseudomonas spp.* decreased more during the FeC enrichments and their DNA concentration was higher in the Gen-MFCs and FeC-MFCs, respectively. Microbial population enriched with FeC showed a lower Shannon diversity index, both in the preculture and at the MFCs level ($p< 0.05$). Enrichment with FeC decrease the relative abundance of EAM and the microbial diversity. Previous studies show the need of a heterogeneous community dominated by EAM to improve the remove of contaminants and to increase the performance of the MFCs. The present work indicates that Gen enrichment promoting the development of a self-balancing community seems to be a preferential approach to be implemented in *in situ* application.

TU259

Integration of molecular and isotopic analyses to investigate the potential of aerobic biodegradation at a site contaminated by Monochlorobenzene

I. Pietrini, Politecnico di Milano / DICA; F. de Ferra, G. Carpani, Research Center for Non Conventional Energy - ENI; L. Zaninetta, Syndial / Environmental Business Services; M. Marchesi, Politecnico di Milano; L. Alberti, Politecnico di Milano / Department of Civil and Environmental Engineering; T. Stella, University of Milano-Bicocca / DISAT; A. Franzetti, University of Milano - Bicocca / Department of Earth and Environmental Sciences

Bacterial communities associated with contaminated sites represent a great opportunity for environmental bioremediation considering that bacteria are able to use a wide number of chemical compounds as a source of carbon and energy. The use of an integrated approach based on different methodologies to gather more information about site-specific potential for bioremediation is gaining a wider acceptance from public authorities. The main objective of our work was to define quantitative indicators to assess the intrinsic degradation potential of a monochlorobenzene (MCB)-contaminated aquifer by the use of a “toolbox” based on isotopic and molecular biology analyses. Microcosms with groundwater collected from a MCB-contaminated site were set up under aerobic and anaerobic conditions to simulate both natural attenuation and biostimulated degradation processes. Enrichment factors for ^{13}C were determined by Compound Specific Isotope Analysis (CSIA). High-throughput sequencing (Illumina) and Ion Torrent analysis and quantitative PCR were performed to gain insights into the structure of the microbial community and to identify functional biomarkers. The investigation of the potential anaerobic degradation pathways is not shown because the obtained data are not consistent. On the other hand, during the aerobic degradation MCB was completely depleted upon addition of nutrients and CSIA results confirmed negligible C isotope fractionation under oxidative conditions. The catabolic *tod*C gene, encoding for toluene dioxygenase, and *Pseudomonas* were identified as molecular and taxonomic markers, respectively. Recently, analyses of the identified molecular and taxonomic markers for the aerobic degradation of MCB were also applied to another area contaminated by MCB, and nearby the first site, to establish whether an aerobic approach for site reclamation from MCB would be successful in the extended area.

TU260

Isotopic and Molecular Biology fingerprinting of a complex contaminated industrial area

T. Stella, University of Milano-Bicocca / DISAT; I. Pietrini, Politecnico di Milano; F. de Ferra, G. Carpani, Research Center for Non Conventional Energy - ENI; M. Marchesi, Politecnico di Milano; L. Alberti, Politecnico di Milano / Department of Civil and Environmental Engineering; A. Franzetti, University of Milano - Bicocca / Department of Earth and Environmental Sciences

The management and the remediation of large contaminated areas with multiple pollutant sources and different environmental conditions represent a big challenge to site owners. Therefore, a detailed chemical, isotopic and microbiological site characterization (fingerprinting) is crucial to evaluate, at first, the intrinsic remediation of the contaminated area (natural attenuation) and, then the potential of enhancing specific biodegradation processes (biostimulation). This study aimed at gathering chemical, isotopic and molecular biology data from a contaminated industrial area to quantify the complex mixtures of contaminants, to provide information about the sources of contamination and to assess the presence of potential degraders and, thus to enhance the on-going biodegradation processes. Contaminated groundwater was collected from 19 piezometers in a restricted area of the site. Chemical analyses of chlorinated ethenes, 1,1,2-trichloroethane (1,1,2-TCA), 1,2-dichloroethane (1,2-DCA), benzene, toluene, xylene isomers, ethylbenzene and chlorinated benzenes were performed following the standard protocols. Compound-Specific Carbon Isotope Analysis (C-CSIA) were carried out to define the isotopic signatures of 1,1,2-TCA, 1,2-DCA and chlorinated solvents (PCE, TCE, 1,2-*cis*-DCE, VC). The structure of the microbial community was determined by Illumina High Throughput Sequencing, whereas its functional profile was assessed by quantitative PCR of key genes encoding for enzymes involved in specific metabolisms. Trichloroethylene and 1,2-dichloroethane (1,2-DCA) were found in most of the water samples at high concentration as well as 1,2-*cis*-DCE. Illumina sequencing data showed a great bacterial diversity probably due to contamination heterogeneity. However, species belonging to *Burkholderiales* and *Rhodocyclales* orders were predominant in 1,2-DCA and VC-contaminated groundwater, respectively. The functional characterization based on the quantification of catabolic genes enconding for reductive dehalogenases (*PceA, TceA, VcrA, BvcA*) and oxidative enzymes (*etnC, etnE*) will be accomplished (on-going analysis) as well as isotopic analyses.

TU261

Microbial ecology and ecosystem services: a key role for biotechnological applications

G. Lembo, ENEA CR / Department of Ecological and Biological Sciences; A. Signorini, ENEA-Italian Agency for New Technologies / Energy and Sustainable Development; V. Mazzurco Miritana, S. Rosa, A. Agostini, V. Pignatelli, ENEA-Italian Agency for New Technologies / Energy and Sustainable Development, Laboratory of Biomass and Biotechnology for Energy; M. Fenice, University of Tuscia / Department of Ecological and Biological Sciences; G. Massini, ENEA-Italian Agency for New Technologies / Energy and Sustainable Development, Laboratory of Biomass and Biotechnology for Energy

Microorganisms are the invisible component of terrestrial bioma, they contribute decisively to the ecosystem functionality, providing goods and services. Their play a crucial role in biogeochemical cycles function.They determinant for the water, air and soil quality. Despite microorganisms are of micrometric size (1µ--1mm), their activities impact on a planetary scale. They are ubiquitous and show remarkable metabolic versatility. They are able to thrive even in extreme environments. Very often different strains of microorganisms perform their metabolic activities in close relationship and/or have co-evolved mutual dependence for performing complex processes where members of the food chain depend on the previous ones for their substrates. Human kind is largely relaying on microorganism for its survival; they provide fundamental ecosystem services and perform complex biochemical activities to degrade residues and transform food. The scientific community is increasingly exploring the potentiality offered by functional microbial biodiversity to improve the human wellness and sustainability. Currently, a much interest is addressed towards biotechnological techniques that supply clean and affordable renewable energy sources exploiting the activities of microbial communities. This is the case of the anaerobic digestion (AD) process, through which, in the absence of oxygen, the complex organic matter is transformed into gaseous products, such as CH_4, H_2 and CO_2. Although the engineering and technological aspects of the AD have been thoroughly studied, the microbial community is still managed as a 'black box', since most of the AD plants lack microbiological planning and monitoring. On the other hand, interactions between the microbial components have an indisputable impact on the combined performance of the bioprocess as a whole. Disruptions in the AD process are often related to a poor understanding of the ecology of the microorganisms responsible for the associated biochemical reactions. In this work, insights about microbial community dynamics, investigated with innovative molecular techniques, are presented in order to improve the understanding of the linkages between natural and biotechnological ecosystems, and, by applying microbial ecology principles, improve the environmental engineering practices.

TU262

Evaluation of bioremediation potential in groundwater using newly-developed software

M. Nechanická, Technical University of Liberec / Institute for Nanomaterials Advanced Technology and Innovation; D. Vlkova, Technical University of Liberec / Institute of Nanomaterials, Advanced Technologies and Innovation; I. Dolinova, Technical University of Liberec / Institute for Nanomaterials Advanced Technology and Innovation; M. Marchesi, Politecnico di Milano

Bioremediation is one of economic and effective environmental techniques being applied for the removal of different contaminants from the groundwater. To achieve a complete overview on bioremediation processes, knowledge about molecular-genetic, physicochemical, and chemical characteristics of the groundwater as well as geological parameters of the site is needed. The aim of this work is, therefore, to develop a user-friendly software allowing assessment of bioremediation process of chlorinated ethenes, even to unprofessional users. The software enables an interpretation of input data, resulting in evaluation of the potential for natural bioremediation at the contaminated sites. Suitability of conditions for bioremediation is simultaneously evaluated in this software. Moreover, data from one sampling round are only used. To ensure widespread user availability, the program was created in Microsoft Excel. Actual data from the Novy Bydzov site were used to verify and demonstrate program's functionality in this work.

Anthropogenic and natural sources of environmental contaminants highlight the impacts of opposing and conflicting regulations (P)

TU263

REMEDIATION OF AQUATIC ECOSYSTEMS: ADSORPTION OF PHOSPHORUS BY SAWDUST

G. Pantano, T.C. Souza, P.S. Fadini, A.A. Mozeto, Federal University of Sao Carlos

Introduction: Despite the scenario of excess phosphorus in some aquatic environments, there has been concern with the depletion of phosphate rock deposits, which can affect global food security. A possible solution to this contrast is the use of sawdust to remove the excess phosphorous from eutrophic systems, for further use as fertilizer. The aim of this work was to study the phosphorus adsorption using sawdust as organic adsorbent. **Methods:** This study was carried out with water and sediment samples from an eutrophic ecosystem, Barra Bonita reservoir, located in Barra Bonita city, São Paulo State, Brazil. The microcosm experiments were conducted in 5 L glass jars, that were filled with sediment and water from the reservoir. Among the 36 assembled microcosms, 18 were used as controls and 18 were used as treatment (with bags containing sawdust). The dissolved oxygen, iron, and orthophosphate were determined in interstitial water and water from the jar's water column. Emerging contaminants and adsorbed phosphorus (P) were determined in sawdust. **Results and discussion:** Dissolved oxygen values in the control microcosms were significantly higher ($p < 0.05$) in comparison to the treatment microcosm, as a consequence of the organic matter oxidation present inside the bags. The lowest concentration of Fe(II) found in the water column of the control microcosms causes the oxidation of the superficial sediment and this oxidized layer was responsible for the reduction of internal flow of P. In the interstitial water the Fe(II) concentration is much higher than in the water column due to the large amount of Fe present in the sediment. The concentration of orthophosphate in the water column varied during the experiment, in the treatment microcosm the decrease was indicative of phosphorus adsorption. It was observed that the adsorption of phosphorus on sawdust began after 57 days. The maximum adsorption was at 214 days (41.4 µg P g^{-1} sawdust). The adsorption of atenolol and caffeine was not observed in sawdust. The concentrations of carbamazepine, diclofenac, paracetamol, ibuprofen, naproxen, propranolol, triclosan, estrone, 17-estradiol and 17- ethinylestradiol are lower than the limit of quantification (LOQ). **Conclusion:** Sawdust is considered a biosorbent, of easy access and low cost, to use in the remediation of eutrophic environments. The possibility of phosphorus recovery is important to ensure water and global food security. **Acknowledgments:** FAPESP (2016/00490-6)

TU264

Formation potential of trifluoroacetate and its estimation by means of the TOP assay

J. Janda, DVGW Water Technology Center / Analysis and Water Quality; K. Nödler, TZW DVGW-Technologiezentrum Wasser / Analysis and Water Quality department; F. Lange, DVGW Water Technology Center / Analysis and Water Quality; C. Zwiener, Environmental Analytical Chemistry, Center for Applied Geoscience, University of Tuebingen / Geosciences; H. Brauch, DVGW Water Technology Center / Analysis and Water Quality

Trifluoroacetic acid is the perfluorinated carboxylic acid with the shortest-possible chain length and thus a small, persistent molecule. Due to its high acidity ($pK_a <$ 0.23) it occurs in its anionic form (trifluoroacetate, TFA) in the aquatic environment and is considered as highly mobile. Photochemical conversion of volatile refrigerants (e. g. 1,1,1,2-tetrafluoroethane) is the most frequentliy discussed anthropogenic source of TFA. However, its formation in the environment has also been shown for other substances containing trifluoromethyl moieties. Hence, a large number of active substances in modern crop protection agents, pharmaceuticals or industrial chemicals have to be regarded as potential TFA precursors. In the present study, the TFA formation potential of environmentally relevant substances was determined using the so-called total oxidizable precursor assay (TOP assay). In order to analyze the resulting concentrations of TFA, a method for quantitative extraction of the analyte from the higly alkaline and saline digestion solutions was developed. The subsequent measurement was performed using ion chromatography coupled to tandem mass spectrometric detection (IC-MS/MS). The oxidative transformation of 10 precursors (pesticides: flufenacet, fluopicolide, fluopyram, flurtamone and tembotrione; pharmaceuticals: fluoxetine and sitagliptin; industry chemicals: 4:2 FTSA and 6:2 FTSA) led to substance-specific molar yields between 7.1% (6:2 FTSA) and 96 % (sitagliptin). It is known from previous studies that TFA can be formed during waste water treatment. Therefore the formation potential of samples from six treatment plants (WWTPs) was investigated. As expected, more TFA was formed after oxidation of the influents (up to 180 % increase relative to the concentrations without oxidative treatment) than of the effluents (between insignificant and 140 %). Interestingly, one WWTP exhibited a strong (biological) formation of TFA, which could be confirmed quantitatively using the TOP assay.

TU265

A Challenge for pesticide regulators: The example of 1,2,4-triazol in groundwater - Overview of regulatory strategies in Germany, Denmark and France

B. Smith, BVL / Department for plant protection products; W. Tüting, BVL; A. Gimsing, The Danish Environmental Protection Agency / Pesticides and Gentechnology; A. Boivin, ANSES; A. Gathmann, BVL / Department for plant protection products

The substance 1,2,4-triazol is a known metabolite of several fungicidal active substances used in plant protection products. Modelled groundwater concentrations of pesticide uses of each active substance are below the limit value of 0.1 µg/L. In plant protection regulation, this limit value has to be applied for 1,2,4-triazol due its toxicological relevance according to the regulation (EC) 1107/2009. Exceedance of this trigger has been questioned considering that several fungicidal active substances forming 1,2,4-triazol may be applied consecutively. In addition, plant protection products are not the single source of 1,2,4-triazol. It can also originate from biocide uses, nitrification inhibitor uses as additive to fertilizers, or even be formed naturally in forest soils. Consequently leaching of 1,2,4-trizol from these different sources might lead to exceedance of the limit value for groundwater in agricultural catchments. Therefore, the competent authorities of Germany, Denmark and France have initiated monitoring programs and incidence reporting. If entries above 0.1 µg/L in groundwater are found and entry via pesticide application is considered likely, authorities may have to decide on further risk management actions. Possible mitigation measures are discussed and the difficulties in dealing with different sources and regulatory frameworks are highlighted.

TU266

PPPs on the basis of natural compounds: nature challenges analytics

M. Andre, F. Stahl, C. Jansen, SGS Institut Fresenius GmbH

For many plant protection products (PPP) using natural compounds as an active ingredient, considerable background levels are frequently observed in untreated control material. These contaminations originate from both, natural and anthropogenic sources, making the method development and residue analysis more challenging. There are different routes for natural background concentrations resulting from natural sources as amongst which are: physicochemical degradation from higher compounds (e.g. degradation of triglycerides to fatty acids), microbiological activity or the use of a plant product as active ingredients (e.g. rapeseed oil). Besides the natural occurrence of the active ingredient or parts of it, anthropogenic routes of contaminations are also diverse: some active ingredients of PPPs were used in industrial production processes (e.g. short-chained fatty acids as softener for plastic materials), other compounds are incorporated in materials used for solvent production. Both may lead to high background levels. Both routes, the anthropogenic as well as the natural, can lead to background level contaminations of the active ingredients, making it hard or in some cases impossible to find contaminant-free control material and/or to determine these active ingredients at low concentration levels. Furthermore, natural compounds used as active ingredients in PPPs or their derivatives are of low molecular weight and thus leading to fragments < 100 Da in LC-MS/MS analysis. These are more difficult to analyse as the signals of these mass transitions are often disturbed.

Persistence & Biodegradation Assessment (P)

TU267

Implication of microbial adaptation for the persistency of emerging pollutants

B.A. Poursat, University of Amsterdam/IBED Institute / Institut for biodiversity and ecosystem dynamics; M. Braster, VU University Amsterdam; R. Helmus, University of Amsterdam / IBED; R.J. van Spanning, VU University Amsterdam; P. de Voogt, University of Amsterdam / IBED; J. Parsons, University of Amsterdam / IBED-ELD

Regulatory determination of the persistency of organic chemicals is mostly done using OECD ready biodegradability tests (RBTs). RBTs, however, suffer from several problems that lead to a high variability of the results and, hence, to difficulties in their interpretation. The origin and history of the inocula is one of the major causes of that variability. Nowadays, it is evident that results of RBTs change over time as microbial populations apparently adapt within years to metabolise previously persistent chemicals. Several studies also show an improvement of the biodegradation rates even after a short period of pre-exposure to the tested chemical. As such, there is a need to assess the influence of this process on RBTs. We, therefore, used chemostat systems to expose activated sludge microbial communities to 3 different chemicals, 4-chloroaniline, N-methylpiperazine and metformin. Two of these chemicals are considered as emerging contaminants and are persistent according to RBTs. The biodegradation capacity of the activated sludge and of the exposed inocula was assessed in batch culture using the OECD 310 guideline for testing of chemicals. Different phases of biodegradation were measured following CO_2production (OECD 310) and the compound and product concentration by LC-MS/MS. Community changes in the chemostats were determined by 16s rRNA sequencing. The results of these experiments show enhanced biodegradation capacity for N-methylpiperazine after pre-exposure to this molecule. Moreover, microbial communities exposed to metformin were able to degrade this molecule and its known persistent transformation product, guanylurea, which is considered as persistent in fresh water. These preliminary results show that microbial communities can adapt to degrade a molecule that was initialy persistent.These results are a first step to understand adaptation mechanisms and their implication for the persistency of organic compounds of emerging concern.This project aims at correlating microbial adaptation and biodegradation performance in time. The ultimate aim is to design more robust and realistic RBTs using adapted inocula.

TU268

Prioritization of organic compounds based on their persistence in dissolved phase

L. FUSTER, EPOC, University of Bordeaux / EPOC UMR 5805; M. Dévier, University of Bordeaux / EPOC / LPTC UMR 5805 CNRS; P. Pardon, K. Le Menach, UMR CNRS EPOC Universite Bordeaux / EPOC UMR 5805; P. Mazellier, University of Bordeaux / EPOC UMR 5805; H. Budzinski, University of Bordeaux

When considering the large number of chemicals potentially present in the environment, the scientific community seeks to establish a pertinent list of priority compounds. Assessing the persistence of chemicals such as pharmaceuticals or polar pesticides represent a need in order to realize a better prioritization of compounds of concern**.** Persistence in dissolved phase is widely dependent on intrinsic properties of compounds but also on environmental conditions such as temperature, salinity, or presence of microorganisms. Estuaries are characterized by gradient and high variability of salinity and suspended solids (SS). The environmental risk assessment may need to be adapted to dynamic conditions such as those prevailing in transitional areas. This study focuses on the persistence of polar pesticides and pharmaceuticals into turbidity maximum zone of the macrotidal Seine estuary. Laboratory batch experiments simulating mixing conditions of the discharge of wastewater into estuarine water were performed. The influence of SS concentration, salinity and abiotic control was assessed on a selection of 60 polar pesticides and 51 pharmaceuticals. In order to compare relative compound persistence in dissolved phase, a persistence index based on the half-lives of the compounds was calculated. Briefly, marks depend on half-life values of each condition, and the average mark gives the persistence index. Risk quotient is calculated with measured environmental concentrations of each compound in the Seine estuary. Of the 111 monitored compounds, 33 were quantified at the initial time. Only 3 exhibited a persistent behavior (e.g. atrazine) while biotic degradation was shown to be the main attenuation process for 15 molecules (e.g. diuron). Degradation was enhanced by increasing concentrations of suspended solids. Because half-lives of compounds presented important variations between all experimental conditions, valuable prioritization was complex to achieve in such conditions and consequently in transitional zones. A persistence index and measured concentration in the Seine estuary were used together and allowed a categorization of compounds into 4 levels of environmental concern. Moreover, non-targeted analysis highlighted the formation of 794 compounds during 21 days of incubation, in high concentration level of SS condition. In order to improve risk assessment, formation of transformation products should have to be considered and included in prioritization schemes.

TU269

OECD 308 tests to explore differences in persistence of pharmaceuticals and microbial diversity between two rivers

C. Coll Mora, Stockholm University / Environmental Sciences and Analytical Chemistry; Z. Li, Stockholm University / ACES; R. Bier, S. Langenheder, Uppsala University / Department of Ecology and Genetics/Limnology; A. Sobek, Stockholm University / Department of Environmental Science and Analytical Chemistry ACES

Persistent pharmaceuticals in aquatic ecosystems are of particular societal concern and the OECD 308 guideline is often used to obtain the biodegradation half-lives required for risk assessment. The environmental relevance of OECD 308 has been criticized in recent years regarding the difficulty to interpret multiphasic processes (biotic and abiotic) and the lack of compartment-specific half-lives for water or sediment as an outcome. In particular, biodegradation processes in the sediment can vary according to the microbial communities, which may be impacted by the settings chosen for the test (i.e. sediment-water ratio, aerobic-anaerobic conditions and initial concentration levels). In this study, we have investigated the differences in biodegradation of a mixture of 9 pharmaceuticals (acetaminophen, caffeine, carbamazepine, diclofenac, fluconazole, metformin, oxazepam, tramadol, and venlafaxine) using water and sediment collected from rivers Fyris and Gründlach, before and after the discharge of a wastewater treatment plant (WWTP). Bottle incubations were set following the OECD 308 guideline, spiked with pharmaceuticals and incubated for 40 days at 16°C in the dark with daily aeration. Water samples were taken at 10 time points and analyzed in UHPLC-MS/MS. The microbial community composition in the sediment was analyzed with Illumina sequencing of bacterial 16SrRNA to provide more insight into the biodegradation potential in the different treatments. The dissipation half-lives obtained for diclofenac, oxazepam, tramadol, and venlafaxine are significantly different ($p < 0.001$) between rivers and between locations. Additionally, the half-lives of non-sterile treatments are significantly shorter than sterile ($p < 0.01$) for all compounds except carbamazepine, indicating that dissipation reflects a combination of biodegradation and sorption mechanisms. Furthermore, compound concentrations remained constant in river water treatments, with only caffeine and acetaminophen showing signs of degradation. Dissipation of pharmaceuticals in the sediment compartment is more relevant than in water for the 2 rivers. Although sorption cannot be completely excluded, the experiment results indicate that the biotic processes could be divergent between rivers, but also between sediments taken up- and downstream the WWTP. An analysis of the microbial diversity in sediment and water of each river will complement the differences in dissipation rates observed for the treatments.

TU270

Compartment-Specific Screening Tools - Development and Application to Assess Potential Persistence of Organic Compounds in Water, Sediment and Soil

T. Junker, ECT Oekotoxikologie GmbH; A. Coors, ECT Oekotoxicologie GmbH; G. Schuurmann, Helmholtz centre for environmental research - UFZ / Department of Ecological Chemistry

The persistence assessment under REACH ideally relies on compartment-specific degradation half-lives that are generally derived from aerobic laboratory simulation studies for surface, aquatic sediments or soil. Albeit these data are given top priority, they are not available for most of the compounds since simulation tests are time-consuming and expensive, and they are required only for compounds with a production volume of 100 or more tons per year. Thus, screening information (e.g. results from ready biodegradability tests (RBTs) or quantitative structure-activity relationship (QSAR) models) are used in the absence of simulation test data to decide whether a substance is considered as “not persistent” or “potentially persistent” according to the screening criteria. However, RBTs only exist for the water compartment and QSAR models were mainly developed based on qualitative experimental biodegradation data related to water-only test systems. Therefore, half-lives for soil and sediment are calculated by multiplying the half-life in water by constant factors. However, this extrapolation is questionable due to different conditions in the compartments, which might affect biodegradation in connection with physico-chemical properties of the compounds. Consequently, there is a lack of experimental quantitative biodegradation data for soil and sediment at the screening test level. In the present work compartment-specific screening tools for water-sediment (Water-Sediment Screening Tool, WSST) and soil (Soil Screening Tool, SST) were developed based on the existing MITI test system (OECD TG 301C). The test systems MITI, WSST and SST were applied to determine degradation data for a set of fifteen test compounds. The results demonstrate that the WSST and the SST are suitable to determine sound and reliable biodegradation data including biodegradation kinetics for compounds on the screening test level. Furthermore, the results were used within the scope of a Compartment-Specific Persistence Screening (CSPS). The results of the CSPS were in good agreement with the REACH standard persistence screening, resulting in slightly more conservative but reasonable results. Thus, the data can be applied to identify potentially persistent compounds in the regulatory context. Beyond that, the results can be used as input parameters for multimedia fate models to assess the environmental fate of the compound, e.g. overall persistence (P_{ov}).

TU271

Persistency assessment of pesticides in Denmark

A. Gimsing, The Danish Environmental Protection Agency / Pesticides and Gentechnology; A. Aagaard, S. Marcher, The Danish Environmental Protection Agency / Pesticides and Biocides; V. M�ller, The Danish Environmental Protection Agency

Persistent active substances can affect the environment over long periods of time, as such substances can be distributed and accumulated within and outside the areas where they are used. Persistent substances constitute a long-term and difficult-to-quantify risk of spreading in the environment and affect organisms. Persistent substances can also cause effects on and lead to residues in subsequent crops. This also applies to the metabolites of an active substance. Therefore active substances with a DT50 above 180 days cannot be approved in Denmark. The persistency evaluation is based on an assessment of available reliable half-lives from both laboratory and field studies. All half-lives should be normalised to 20 °C and pF2. Assessment of persistency should not be based on average or percentiles of the data. Instead data are assessed by considering the soil types used and focusing on soil types representative for Danish conditions. If in general these soils have a DT50 above 180 days, products with such active substances cannot be authorized for outdoor uses in Denmark. If only some of the soil DT50 values are above 180 days, an ad hoc assessment is performed to decide if these findings constitute the major part of data and if it is likely that DT50 for Danish soils is above 180 days under field conditions relevant to the intended use. The persistency evaluation should be performed for both the active substance and metabolites. However, metabolites which fulfill certain criteria are considered to be of no concern regarding persistence.

TU272

Influence of Winter Conditions on Fungicide Persistence in North American Golf Course Turfgrass

P. Koch, University of Wisconsin - Madison / Molecular and Environmental Toxicology Center

Fungicides are routinely applied to golf course turfgrass prior to winter in temperate climates around the world to protect the plants against psychrophilic plant pathogenic fungi. The persistence of these fungicides in the varying environmental conditions present during winter is poorly understood despite important implications for human and environmental health as well as disease control on the turf. A 3-year field study was initiated at the University of Wisconsin - Madison (Wisconsin, USA) in 2015 to determine which environmental conditions most influenced the persistence of the fungicides propiconazole and chlorothalonil. Foliar concentration of both fungicides was measured using liquid chromatography-mass spectrometry, and a bioassay was conducted in a controlled environment chamber using the psychrophilic plant pathogenic fungus *Microdochium nivale* to determine the date when disease protection was lost. Fungicides were applied once on 20 Nov 2015 and again on 5 Dec 2016 and 10-cm diameter turfgrass cores were collected biweekly from the experimental area throughout each winter. Both winters experienced above-average temperatures in December with frequent rainfall events, and the concentrations of both fungicides in the turfgrass leaf tissue fell dramatically within the first 28 days after application. In addition, a corresponding increase in *M. nivale*-disease symptoms in the controlled environment chamber was observed as fungicide concentration decreased. These results suggest that fungicides do not persist in winter conditions following rainfall events, though it remains unclear whether they persist for prolonged periods of time on frozen turf and under prolonged snow cover.

TU273

Biodegradability of novel graft copolymer with levan and polystyrene

B. Lončarević, Institute of Chemistry, Technology and Metallyrgy, University of Belgrade / Department for Chemistry; M. Lješević, Institute of Chemistry, Technology and Metallurgy, University of Belgrade; G. Gojgić-Cvijović, D. Jakovljević, Institute of Chemistry Technology Metallurgy University of Belgrade / Department of Chemistry; V. Nikolić, Innovation Center Faculty of Chemistry University of Belgrade; M. Vrvić, V. Beškoski, Faculty of Chemistry University of Belgrade / Department of Biochemistry

The significant increase in plastics productions caused waste management problems which is particularly relevant for polystyrene plastic as the most dominant packaging material. Therefore, investigations of new biodegradable polymers are increasing. Graft copolymerization is important technique for physical and chemical modification of polymers. The microbial levan is biocompatible, biodegradable, renewable and eco-friendly fructose based polymer. It can be produced from sucrose by wide range of microorganisms using levansucrase enzyme. In the present study graft copolymer with microbial levan and polystyrene was synthetized, characterized and its biodegradable potential was investigated.

Levan was isolated after fermentation of *Bacillus licheniformis* strain. Syntheses of copolymer were performed by the free radical reaction using potassium persulfate as initiator. Verification of the synthesis was recorded by ^{13}C NMR Bruker AVANCE III 500 spectrometer. Biodegradation potential in aerobic conditions of obtained copolymer was investigated using Micro-Oxymax respirometer (Columbus Instruments, Ohio). O_2 consumption of samples mixed with soil was measured in period of 28 days. The ^{13}C NMR spectrum of copolymer showed signals corresponded to both components. Consumption of O_2 was higher in copolymer sample (705.0 L) compared to control (350.9 L) and polystyrene (499.5 L) after 673 h. The formation of levan and polystyrene graft copolymer was confirmed by ^{13}C NMR analysis. Results after 28 days in aerobic biodegradation in soil shows that obtained novel copolymer has biodegradation potential, however additional tests for biodegradation are needed.

TU274

Aerobic degradation of styrenated phenol in soil: influence of the temperature and of the characteristics of the soils

M. Enrici, SOLVAY / HSE - PRA PS; P. Chagnon, SOLVAY / Research and Innovation

The persistence of chemicals is assessed through their kinetic of degradation in the environment. Several simulation tests are available to evaluate the half-life of the chemicals in different environmental compartments. The half-life is then compared to the Annex XIII criteria of REACH to decide if the substance is be considered as Persistent (P) or very Persistent (vP). Nevertheless, the interpretation of those tests is complex because several parameters may influence the kinetic of degradation despite a standardization of the methods In the present project, the rate and route of transformation of a styrenated phenol compound was investigated in four different soils and at two temperatures: 12°C and 20°C under aerobic conditions. Statistical analysis is performed to assess the influence of the temperature on several endpoints: mineralization rate of the parent compound, kinetic of degradation of the parent compound, kinetic of degradation of the main metabolites, formation of non-extractable residues. In addition, the influence of the characteristics of the soil is assessed to evaluate the representativeness of those tests for the evaluation of the actual fate and behavior of such chemical in the environment.

TU275

Comparison of kinetics and products of degradation determined for the toluenediamine substances in the OECD-standardized ready biodegradability and sediment simulation tests

C.R. Boegi, BASF SE / FEP/PA; C. Gaertner, Fraunhofer Institute for Molecular Biology and Applied Ecology IME; H. Schwarz, BASF SE / RB/TC; R.J. West, International Isocyanates Institute, Inc. / Toxicology and Environmental Research Consulting

The OECD ready biodegradability tests (RBT) are designed to approximate the rate/extent to which substances are degraded in the environment, such as is more precisely measured in the OECD simulation tests. This work compares results obtained from both test types for degradation of the toluenediamine (TDA) substances. Degradation of [14C]-2,4- and 2,6-TDA was studied according to the OECD Guideline Nos. 301B and 308, wherein their disappearance, formation of degradation products, and evolution of $^{14}CO_2$ were measured from initial doses of 0.5 mg/L. The 301B test used an inoculum collected from a domestic sewage treatment plant, while the 308 test used water/sediment collected from two diverse tributaries of the Rhine River. Disappearance of TDAs in the RBT followed pseudo-first-order kinetics, and half-lives for the 2,4- and 2,6-TDAs were approximately 43 and 17 d, respectively. For 2,4-TDA, evolution of $^{14}CO_2$ was equivalent to 4 and 7% of the applied radioactivity (AR) after 28 and 63 d, respectively, while that from 2,6- TDA was 12 and 24% of AR, respectively. The TDAs were removed by >90% in the RBT, with the balance of AR associated with the biosolids. In the 308 test, the TDAs were rapidly transformed from their fully-dissolved state in water to a non-extractable residue (NER) in the sediments beneath. After the first 11 d, radioactivity in the river water was reduced to < 10% of AR, several transiently-formed degradation products were detected (tentatively identified by high resolution LC-MS), and NER accounted for ≥ 79% of AR for both TDAs in both river systems. Disappearance of TDAs was fitted to a dual-first-order-in-parallel kinetic model, with 50% depletion times (DT_{50}) of approximately 0.4 – 1.0 d and 0.7 – 1.2 d determined for the 2,4- and 2,6- isomers, respectively, in both river systems. Yields of $^{14}CO_2$ were ≤ 10.6% of AR for the 2,4-isomer and ≤ 8.3% of AR for the 2,4-isomer in both river systems after 100 d. In all cases, < 1.5% of AR could be freed from the sediment using vigorous solvent extractions. The results of both test types show that the TDAs are not persistent in the environment, and are transformed by concurrent biodegradation and abiotic reactions. While the RBT gave a reasonably conservative approximation of the DT_{50} times and $^{14}CO_2$ yields in aerobic surface water/sediment systems, it did not give a realistic representation of the fate mechanisms which result in formation of NER with natural organic matter in the environment.

TU276

Evidence for Anaerobic Microbiodegradation of PCBs and PBDEs in Sediment cores from an e-Waste Site, South China

B. Mai, Guangzhou Institute of Geochemistry; C. Huang, Guangzhou Institute of Geochemistry, Chinese Academy of Sciences; Y. Zeng, Guangzhou Institute of Geochemistry

Biodegradation of polychlorinated biphenyls (PCBs) and polybrominated diphenyl ethers (PBDEs) is an important transformation and detoxification route in the environment. To better understand the in-situ microbial degradation of PCB and PBDE in anaerobic sediment, three sediment cores from an e-waste dismantling site, South China, were sampled (named #1, #2, and #3, respectively). Positive matrix factorization model (PMF), compound specific isotope analysis (CSIA), and microorganism analysis were used to trace the in-situ biodegradation of these pollutants. High levels of PCB (44-67800ng/g, dw) and PBDE (62-792000ng/g, dw) were found in all the samples and the concentrations were general increased from bottom to the top layers. PMF analysis indicated that the technical mixtures are the dominant PCB and PBDE input and dehalogenation takes place in the sediment cores, especially for PBDEs. This conclusion was supported by the microorganism analysis, substantial *Dehalococcoidetes* were found in the sediment cores. The range of the relative abundance of *Dehalococcoidetes* for three sediment cores (#1, #2, #3) were 1.50-9.01%, 1.47-5.24%, and 0.20-2.55%, respectively, which were significantly correlated with the ratios of factor 2 (biodegradation source) /∑PBDEs (with the p values of 0.02, 0.05 and 0.01, respectively). As for CSIA analysis, only the stable carbon rations ($\delta^{13}C$) of BDE 28, BDE47, BDE85, and BDE99 in the top 20cm of the #3 sediment cores were obtained. An increase in the $\delta^{13}C$ values for BDE 28 and a slightly decrease in the $\delta^{13}C$ values for BDE 85 were found with the increase of the depth sediment cores, indicating a potential biotransformation of these compounds in the cores. No significant differences in the $\delta^{13}C$ values of BDE 47 and BDE 99 were observed in the sediment cores, possibly due to the complicated fate of these compounds, such as BDE47 and BDE99 being both reactants and products during the debromination processes.

TU277

Transformation and degradation mechanisms of flame retardant triphenyl phosphate in aquatic environment

Y. Choi, Gwangju Institute of Science and Technology; S. Kim, Gwangju Institute of Science and Technology / School of Earth Science and Environmental Engineering

Organophosphate flame retardants (OPFRs) in aquatic environment are concerned because they are latest alternative chemicals of brominated flame retardants. Among OPFRs, triphenyl phosphate (TPHP) shows high consumption volumes, as well as high concentration in water. TPHP caused toxic effects especially in aquatic organisms but research of biotransformation products is insufficient. Kinetic studies of TPHP and transformation products are important to understand the effects on environmental organisms. To identify the biotransformation products of TPHP, *daphnia magna* was investigated due to standard test species in aquatic environment. TPHP was exposed to individual *daphnia magna* and each samples were separated by biota and remaining medium. *Daphnia magna* were homogenized and remaining medium were extracted with solid phase extraction. Samples were analyzed using liquid chromatography-tandem mass spectrometry (LC/ESI-MS/MS) for quantitation and ultra-high-performance liquid chromatography-electrospray high-resolution tandem mass spectrometry (UHPLC-HRMS/MS) for qualification. Two major biotransformation products were detected in the study based on phase I & II biotransformation mechanisms. Diphenyl phosphate (DPHP), product of phase I reaction, was identified for biotransformation products both biota and medium. Among phase II reaction, sulfonyl triphenyl phosphate was verified; intermediate metabolites were not significantly detected due to brief retention times. Parent compound (TPHP) and hydrolysis products (DPHP) were calculated by degradation ratios relative to control. Significant tendency were observed between TPHP and DPHP; as TPHP showed decreased, degradation product (DPHP) ratios increased. In conclusion, hydrolysis and sulfation were major mechanisms for biotransformation products of TPHP in environment. As a result, the risk to aqueous organisms must be estimated in order to develop regulations for organophosphate flame retardants in aquatic system.

TU278

Photolytic and biological degradation of silicon organic compounds

E. Grabitz, Leuphana University Lueneburg; O. Olsson, Leuphana University of Lueneburg / Institute for Sustainable and Environmental Chemistry; N. Mitzel, University Bielefeld / Inorganic and Structural Chemistry; K. Kuemmerer, Leuphana University Lueneburg / Institute of Sustainable and Environmental Chemistry

This study provides new data on the degradability and persistence of a selected group of partially new silicon organic compounds. Polysiloxanes are an important group of industrial chemicals, which are frequently produced in high amounts. They are widely used in industry, personal care products and agriculture. In general, siloxanes occur ubiquitous in the environment in different concentrations (e.g. in water from ng to mg per L). Since these polysiloxanes are only cleavable by chemicals, potential substitutes, which are better degradable in the environment, are urgently needed. Therefore, a partially new synthesised homogenous group of silicon organic compounds (p-$MeOC_6H_4SiMe_3$, o-$MeOC_6H_4SiMe_3$,(p-$MeOC_6H_4)_2SiMe_2$, p-$Me_2NC_6H_4SiMe_3$, o-$Me_2NC_6H_4SiMe_3$) with higher water solubility was investigated to provide new and reliable data on photolytic and biological degradation of these substances in aqueous solutions. Each compound was irradiated with artificial sunlight (xenon lamp, 300–800 nm, SUN-test CPS+). During the irradiation time of 8 hours, samples were taken every two hours and analysed directly with HPLC-UV/vis. An adapted closed bottle test (OECD 301 D) was used to investigate ready biodegradability. The degradation rate was determined by measuring the depletion of diluted oxygen during a period of 28 days. It was found that artificial sunlight degrades the compounds to a certain degree. After 6 hours, 99 % of the substance p-$Me_2NC_6H_4SiMe_3$ was primary eliminated. During the test, generation of more polar transformation products was observed. The other substances were eliminated between 15 and 65 % with treatment of sunlight during a time period of 8 hours. No ready biodegradability could be observed for these five substances. The results from the biodegradation test confirm the data available in literature and in our own database on siloxanes. Increased water solubility of the newly synthesised silicon organic compounds did not result in an increased biodegradability in water.

TU279

Biodegradation of adsorbed oil pollutants: Research on a model system

J. Milic, Institute of Chemistry, Technology & Metallurgy / Department of Chemistry; M. Ilic, IChTM / Department of Chemistry; B. Lončarević, Institute of Chemistry, Technology and Metallyrgy, University of Belgrade / Department for Chemistry; T. Solevic Knudsen, IChTM / Department for Chemistry; J. Avdalovic, Institute of Chemistry, Technology & Metallurgy, University of Belgrade / Department of Chemistry; N. Lugonja, Institute of Chemistry, Technology & Metallurgy, University of Belgrade / Centre of Chemistry

Environmental pollution by various types of oil has been, and continues to be a specific and serious problem. Investigations and development of new techniques are required, as well as improvements to known ones. Sorbent materials are attractive because they collect the oil and separate it from the oil spill site by absorption. The addition of sorbents to oil spill areas facilitates a change from liquid to semi-solid phase and once this change is achieved, the removal of the oil by removal of the absorbent structure then becomes much easier. At this stage, pollutants are separated and concentrated, unlike the environment conditions where pollutants could spread to very low concentration when it is challenging for apply bioremediation techniques. In this study we tested adsorption and degradation of crude oil, diesel oil and mazut as s model substrates. Two types of natural sorbents were used: organozeolite and bentonite. Petroleum pollutants sorption was investigated in the batch tank. Sorption was conducted with sorbents (1g/100 mL) placed in Erlenmeyer flask (500 ml) with 100.0 ml of tap water and oil pollutant (0.6 ml). Sample was then shaked in laboratory shaker for 24 h at 20 °C. Supernatants and sorbents were separated by decantation. Biodegradation ability of adsorbed pollutants has been tested by microorganisms isolated from oil contaminated site, and O_2 consumption and CO_2 production was measured in period of 5 days by Micro-Oxymax respirometer. Adsorbed total petroleum hydrocarbons were determined after adsorption and respiration experiments by GC and gravimetric analysis. Obtained results showed highest biodegradation potential with bentonite/diesel (BED) model and lowest biodegradation potential with organozeolite/mazut (OZM) model, with cells consummation of 80913,53 µl and 5834,53 µl of O_2 within 115 hours, respectively. The production of CO_2 by cells in BED model was more than twofold higher that by OZM model. As well, BED model obtained highest TPH decomposition at the end of experiments. This results indicate that bioremediation process can be successfully used on adsorbed pollutants, where added value is recycling of sorbent material; but further investigation are needed to determine the best sorbent for each of potential pollutants from environment. **Acknowledgements** This work was supported by Ministry of Education, Science and Technological Development of the Republic of Serbia, Project No: III 43004.

TU280

Applying high-resolution mass spectrometry to evaluate chemical persistence in un-spiked natural waters

Z. Li, Stockholm University / ACES; M. McLachlan, Stockholm University / Environmental Science and Analytical Chemistry (ACES)

Microbial degradation (biodegradation) is an important mechanism for removal of organic contaminants in natural systems. For many compounds, biodegradability is a fundamental determinant of the environmental fate of the compound. The OECD 309 guideline ("Aerobic Mineralization in Surface Water") is one of the most important tests for providing kinetic biodegradation data in surface waters for use in persistence assessment and risk assessment. The OECD 309 simulation test measures biodegradation in aerobic natural waters that have been spiked with test chemicals and incubated in the laboratory. However, these experimental conditions do not accurately simulate natural aquatic environments, where a variety of microbial organisms are exposed to chemicals with much lower concentrations. As a result, legitimate questions have been raised about the ability of such standard biodegradability tests to predict persistence of compounds in natural systems. This study was designed to test the hypothesis that biodegradation is significantly different in a spiked system than in a natural system. OECD 309 experiments were carried out with and without spiking. Water from Lake Norra Bergundasjön in southern Sweden was used, a recipient for wastewater treatment plant effluent with a freshwater dilution factor of ~4. A mixture of 16 test compounds comprising a range of biodegradability was used for the spiked systems. Four sets of experimental conditions were used (all in duplicate), i.e., spiked lake water, un-spiked late water, spiked artificial lake water (inflowing lake water mixed with the effluent (80:20, v/v) from the wastewater treatment plant which discharges into the lake), and un-spiked artificial lake water. Incubation conditions followed the standard OECD 309 protocol, lasting for a period of 60 days at 20 °C in the dark. Triplicated water samples were collected at 11 time points. After adding a mixture of internal standards, the sample aliquots were filtered and analyzed with UHPLC-Orbitrap-MS/MS using direct injection. Data was processed using both the target approach and the non-target approach, where the implementation of liquid chromatography coupled to high-resolution mass spectrometry allows for screening of organic contaminants in aquatic systems. The biodegradation kinetics (half-lives) of the detected compounds in the spiked and un-spiked waters will be compared and contrasted to evaluate the hypothesis.

TU281

A Ultimately Transformed Organic Carbon (UTOC) approach to assess biodegradability of complex chemicals

M. Cregut, F. Brillet, University of Nantes / GEPEA CNRS UMR CBAC Laboratory; C. Sweetlove, lOREAL SA / Research and Innovation; J. Chenèble, LOreal Research / Research and Innovation; J. Lharidon, LOréal Research & Innovation / Life Sciences Direction; T. Gerald, University of Nantes / Microbiology

According to the United Nations (UN), a substance is the "chemical elements and their compounds in the natural state or obtained by any production process". This definition has evolved according to different acts of regulation. Another category of substances is UVCB: Unknown or Variable composition, Complex reaction products or Biological materials" such as crude oils or vegetal extracts. In addition, there are "mixtures or solutions composed of two or more substances in which they do not react". The assessment of complex mixture biodegradability can be limited by technical issues and/or difficulties to rule on ready or inherent biodegradability. This work is composed of three different studies to introduce and improve Ultimately Transformed Organic Carbon (UTOC) as a quantification tool for biodegradation. The UTOC includes the inorganic carbon resulting from respiration and the carbon assimilated by microorganisms. This measurement strategy was initially compared to a DOC DIE-AWAY test (i.e. OECD 301A) and then successively tested and validated with non-soluble chemicals, pure and in an emulsion mixture (consisting in a virtual cosmetic formula). The UTOC approach highlighted the beneficial effects of an emulsion on the biodegradation of these substances. Prior to assess a natural complex mixture, pass levels to differentiate unequivocally ready and inherent biodegradability were determined. This latter step aimed to reinforce safety in the assessment for substances of unknown composition. Based on the principle of reducing the probability of persistent parent products or generation of toxic by-products during biodegradation, the UTOC approach was reinforced with ecotoxicological tests using a weight of evidence approach for a moderate % of biodegradation. Finally, the methodology was assayed and validated by an investigation of the biodegradability and ecotoxicology assessment of a vegetal extract. It appears that coupling ecotoxicological tests with UTOC concept has been shown to provide a robust safety assessment approach, and further research should focus on more complex substances (viscous or solid, absorbable, volatile). The advantages of UTOC are clear; it appears as an appropriate method to quantify the initial raw material converted to an inert product by the action of microorganisms to determine the ready biodegradability of an unknown substance such as a vegetal extract.

TU282

Development of a multi-sensors device to assess the biodegradation of chemicals

M. Cregut, University of Nantes / GEPEA CNRS UMR CBAC Laboratory; v. le cunff, L. Catherinot, E. Calzolai, Y. Pichot, TRONICO; C. Sweetlove, lOREAL SA / Research and Innovation; E. Grangé, S. Jouanneau, University of Nantes / GEPEA CNRS UMR CBAC Laboratory; M. Durand, University of Nantes / UMR CNRS GEPEA CEBAC Laboratory; J. Chenèble, LOreal Research / Research and Innovation; A. Lahmar, University of Nantes / GEPEA CNRS UMR CBAC Laboratory; J. Lharidon, LOréal Research & Innovation / Life Sciences Direction; T. Gerald, University of Nantes / Microbiology

Most of the methods used to evaluate biodegradation have been defined for almost 50 years. According to the fact that annually, hundreds of new chemicals require a biodegradability assessment; the development of new metrological solutions needs to be investigated. Indeed, few measurement systems, enabling an automated biodegradation assessment are available, apart from those based on manometric or oxygen consumption measurements, which present certain limitations to assess complex or volatile chemicals. To increase the reliability of the assessment, notably for volatile and complex chemicals, our objective was to develop a multiparametric platform disposing of its own measuring methodology. A research project was therefore conducted to develop this methodology while integrating automation of measurements to tackle another major challenge in biodegradation assessment. To reach this technological bottleneck, a validation of different technologies of sensors has been performed to assess their reliability and accuracy in operating conditions. This first step is crucial prior to establish a carbon balance analysis, using several

modeling steps involving the use of different parameters such as O_2, CO_2, pH, T°C, Pressure and Biomass. These technological investigations will be used to create an unique automated device enabling the evaluation of biodegradation of a chemical whatever its physicochemical characteristics.

TU283
Investigations on key parameters of an innovative biodegradation test based on cell proliferation
S. Rey, Firmenich / Biotechnology; B. Özel Duygan, University of Lausanne / Fundamental microbiology; S. Leocata, L. Baroux, P. Merle, Firmenich; J. van der Meer, University of Lausanne / Department of Fundamental Microbiology; M. Seyfried, Firmenich
Standard OECD screening tests for biodegradation are useful tools for determining the potential of chemicals to undergo decomposition and mineralization in the environment. Most of these are carried out at high test compound concentration and are based on simple readouts such as CO_2 formation or oxygen consumption, and usually neglect biomass formation. Our research attempts to fill a gap in the knowledge on bacterial physiology in tests conducted at more environmentally relevant concentrations of industrial chemicals compared to existing OECD 301 series tests. Recently the feasibility of performing cell counting by flow cytometry was demonstrated for mixed cultures from WWTP sludge and lake water. The present poster reports on key parameters of this new biodegradation test method, notably effects of initial cell and test compound concentration and test duration on the test outcome, as well as on yield determination and on data analysis of flow cytometric cell counting. As test compounds, selected reference chemicals were chosen from the ECETOC MCC/007 report list suggested for method development for readily and non-biodegradable compounds. Alongside cell counting, several test compounds were analyzed in parallel for CO_2 and disappearance of parent compound, and comparison with results from standard screening tests will be presented.

TU284
Challenges and Solutions of Ready Biodegradation Study with Difficult Substances
T. Suzuki, Kao Corporation, S.A. / Product Safety & Regulations; T. Sasa, D. Tomiyama, Kao Corporation / Safety Science Research; M. Yamane, Kao Corporation / Safety Science; O. Morita, Kao Corporation / Safety Science Research
Ready biodegradation studies (OECD TG 301) are required for registrations and the development of chemicals for various applications. Current test systems work well for many substances, but some substances, called “difficult substances” sometimes face problems with these test systems. Here challenges and solutions in ready biodegradation studies are presented with water insoluble or/and volatile substances, as examples of “difficult substances”. A hydrocarbon, 15-methylhentriacontane, is insoluble in water and tends to stay on the water surface. An initial ready biodegradation study indicated that it was not ready biodegradable. It was hypothesized that this was because the microorganisms could not access the test substance well on the surface water. To overcome this challenge, the test substance was wrapped in a nylon sheet so that it could stay in the water for access by the microorganisms. With this test system, the biodegradability was remarkably improved and it was regarded as ready biodegradable. This method and result were accepted by the Japanese authority. An aroma chemical, 3,5,5-trimethylhexanoic acid, is insoluble in water and tends to stay on the water surface and volatilize. An initial ready biodegradation study indicated that it was not ready biodegradable. It was hypothesized that this was because the test substance disappeared from the test system by volatilization. To elucidate this hypothesis, the concentration of the test substance and a possible hydrolyzed metabolite were monitored by chemical analysis in the water phase. The result indicated that the test substance volatized within 48 hours. This result strongly suggested that the low biodegradation result was due to the rapid volatilization and disappearance of the test substance from the test system. The possible solutions to this challenge will be discussed in the presentation.

TU285
Influence of inoculum origin and adaptation on biodegradation of emerging contaminants
B.A. Poursat, University of Amsterdam/IBED Institute / Institut for biodiversity and ecosystem dynamics; J. Dalmijn, University of Amsterdam / IBED; M. Braster, VU University Amsterdam; R. Helmus, University of Amsterdam / IBED; R.J. van Spanning, VU University Amsterdam; P. de Voogt, University of Amsterdam / IBED; J. Parsons, University of Amsterdam / IBED-ELD
Assesment of microbial biodegradation is a key parameters for estimating the environmental risk of new organic chemicals. Commonly used tests for the assesment of ready biodegradability (RBTs) have been designed as simple and inexpensive methods to identify chemicals that are not expected to be environmentally persistent in most of the ecosystems. However, RBTs suffer from several problems that lead to a high variability of the results and, hence, to difficulties in their interpretation. These tests are low throughput, space consuming and poorly reproducible. Moreover, the origin of the inoculum is also a cause of variability in RBTs results. Pre-exposure of the inoculum to the tested chemical prior to any test has been proposed as a method to improve biodegradability testing. Pre-exposure could allow a better persistency prediction of chemicals present at low levels in wastewater or of newly produced chemicals by including the natural adaptation ability of microbial communities. Therefore, in order to assess the influence of the inoculum origin and of pre-exposure on RBTs, we compare the biodegradation capacity of activated sludge from different Dutch wastewater treatment plants before and after pre-exposure to five different chemicals. Carbamazepine, diclofenac and metformin are commonly detected pharmaceuticals in wastewater, while 4-chloroaniline and N-methylpiperazine are industrial chemicals with erratic behaviour in RBTs. In this research, an effort is made to miniaturize the standard OECD 310 procedure. As this test requires large volume vessels, it is difficult to perform large scale tests with multiple inocula and compounds using this guideline. Hence, sealed bottles and 96 well plates are used for the incubation and elimination is measured by following the CO_2 production (OECD 310) and the compound concentration by LC-MS/MS. The results of these experiments are expected to show differences between responses of inocula that are not pre-exposed. After pre-exposure we expect to reduce the risks of errors of test results and to enhance biodegradation. Different responses between the different inocula will give valuable information about the future environmental fate of the tested compound. Finally, this knowledge will develop more accurate ready biodegradation testing and lead to a more comprehensive environmental risk assessment of persistent chemicals.

TU286
Investigations on the role of adaptation in OECD biodegradation screening tests
F. Miffon, C. Dick, Firmenich; K. van Ginkel, AkzoNobel; M. Seyfried, Firmenich
Until recently, adaptation was recognized by the European Chemicals Agency as one of the options in so-called enhanced ready biodegradation tests (RBTs) to provide proof of non-persistence of a test chemical. Since June 2017 (time of publication of the latest ‘Guidance on Information Requirements and Chemical Safety Assessment Chapter R.7b: Endpoint specific guidance’), adaptation has been explicitly excluded. This decision was presumably based on the concern of a perceived lack of capacity for adaptation in the natural environment but failed to provide scientific justification to generally put into question the environmental representativeness of lab results obtained from lower tier tests. As shown previously, positive results from enhanced RBTs are useful in persistence assessments and help avoid false negatives, usually resulting from the stringency of this type of tests. Based on previous results obtained from enhanced RBTs conducted after adaptation, we selected a structural family of compounds for which solid and reproducible effects were seen after adaptation with different inocula. One of the compounds was studied in more detail regarding possible mechanisms of adaptation. The outcome of this study will form the basis for further investigations on the environmental representativeness of positive results obtained from enhanced RBTs with adapted inocula.

TU287
Use of Chemical Analysis to Enhance Interpretation of Biodegradability Tests: A Case Study with Two Gas-to-Liquid (GtL) Products
J. Dawick, G. Whale, C. Hughes, Shell Health / Risk Science Team
The Oslo Paris (OSPAR) Commission, which oversees the OSPAR Convention, currently implements the harmonised mandatory control system (HMCS) for use and reduction of discharges of chemicals in the exploration and production of oil and gas offshore in the North East Atlantic. Chemical suppliers must submit a Harmonised Offshore Chemical Notification Format (HOCNF) to the implementing OSPAR member state authority to certify use of their products offshore. Marine biodegradability screening tests (BST’s) are an intrinsic part of offshore chemical control schemes and the HOCNF registration process. However, the lack of robustness of the marine biodegradation methods has been highlighted in a series of ECETOC workshops, [1] particularly when these are used to assess complex, volatile and poorly water-soluble substances (e.g. petroleum products).We have found that the inclusion of abiotic controls and chemical analysis for total petroleum hydrocarbons (TPH) in freshwater BST’s demonstrates that disappearance of test substances from the test system is often far greater than is suggested by use of standard ultimate biodegradability metrics. [2] Here we present results from two marine BST tests (OECD 306 and BODIS) conducted on two synthetic hydrocarbon Gas-to-Liquid (GtL) products. In addition to measuring ultimate biodegradation (by oxygen uptake) in these tests, additional analytical techniques have been incorporated to enhance the interpretation of results. Extraction and analysis of test samples using gas-chromatography has been conducted to i) confirm substance dosing and ii) understand the extent of abiotic losses in the test system. In addition, two-dimensional gas chromatography (GCxGC) was employed to characterise hydrocarbons present in test samples, which were compared back to the composition of the original test substances. An overview of the results and our recommendations on how marine biodegradability tests can be improved and interpreted will be provided. **REFERENCES** [1] ECETOC Workshop Report No.34 – Improvement of the OECD 306 Screening Test. Published September 2017. Available online via: http://www.ecetoc.org/publication/workshop-report-no-34-improvement-oecd-306-screening-test/ [2] Hughes. C., Whale. G., Mead, C. (2015). Investigation into the

biodegradability of various hydrocarbon solvents. SETAC Europe annual meeting 2015, Barcelona, Spain.

TU288

Organising an international ring test to improve the marine biodegradation screening test

A. Ott, T. Martin, Newcastle University / School of Engineering; G. Whale, Shell Health / Risk Science Team; J. Snape, AstraZeneca UK Ltd. / AstraZeneca Global Environment; B. Rowles, Cefas Lowestoft Laboratory; R. Davenport, Newcastle University / School of Engineering

A series of international standardised tests have been approved by the OECD to measure the relative biodegradability of substances. Among these tests, biodegradation screening tests (BSTs) form the first tiers of assessment, offering relatively simple and cheap characterisations of biodegradability. Most parameters in these BSTs are highly prescribed and conservative, but the microbial inoculum is the least controlled parameter. The resulting high levels of variation have been recognised as a limitation since the introduction of these tests up to today and are especially reported for the marine BST OECD 306. BSTs were designed over two decades ago and are not, in their current form, effective as screens for persistence. In recent years, regulatory emphasis has shifted from identifying chemicals that are rapidly biodegradable to identifying chemicals that are potentially persistent in the environment. Technical guidance documents, which have been prepared under the European chemicals regulation system known as REACH, have suggested several improvements to effectively assess persistence with BSTs. Within their nature, these modifications and enhancements also address a number of the commonly discussed reasons for high variability and poor reliability of BSTs. The Cefic LRi ECO11 project investigated and validated several enhancements for the marine BST. It was possible to represent the bacterial diversity in the sampled environment better by increasing the microbial inoculum to environmentally relevant concentrations through tangential flow filtration. This standardisation led to a significant decrease in variability between replicates in comparison to the standard marine BST. By extending the test duration beyond the persistence half-life threshold, previously reported elongated biodegradation lag phases for chemicals in surface water were recognised. A ring test project was conducted from 2016-18 to validate these intra-laboratory findings from Cefic LRI ECO11 in 13 other testing facilities within Europe, North America and Japan. The test protocol, developed by academia, industry, CROs and regulatory bodies, together with information on the organisation and conduction of the ring test will be presented.

TU289

Tissue-specific accumulation of triphenyltin compounds in marine fishes in sub-tropical Hong Kong

R.C. Sham, K.K. Ho, The University of Hong Kong / The Swire Institute of Marine Science and School of Biological Sciences; K.M. Leung, The University of Hong Kong / The Swire Institute of Marine Science

The extensive use of organotin (OT) compounds in antifouling paints and other industrial uses (e.g. as fungicides, wood preservatives, and antibacterial textiles) have resulted in massive release of these compounds into urbanized coastal marine environments. Even though the International Maritime Organization (IMO) of the United Nations enacted a global prohibition on the usage of organotin-based antifouling agents on hulls of sea-going vessels in September 2008, Hong Kong had not adopted any regulatory legislation to restrict the production, usage and release of these compounds until early 2017. High concentrations of these compounds, especially triphenyltin (TPT), are still being detected in coastal marine environments of Hong Kong and Shenzhen, China. Organisms inhabiting these areas are particularly susceptible because they can bioaccumulate TPT through direct contact with contaminated seawater and sediment, and through dietary uptake. Nonetheless, a comprehensive tissue-specific accumulation profile of TPT compounds in marine fishes is still lacking, and such information will help reveal their toxicokinetics and identify targeted organs of accumulating these contaminants. This study was, therefore, designed to investigate the distribution pattern of TPT in the bodies of four marine fish species, namely *Collichthys lucidus*, *Cynoglossus bilineatus*, *Johnius belangerii*, and *Johnius heterolepis*. For each species, 15 tissue types ($n = 4$) were extracted for quantification of TPT concentrations and its degradation products (i.e., di- and mono-phenyltin) using gas chromatography mass-spectrometry. We found that the accumulation tendency of TPT was highly tissue-dependent. Highest concentrations of TPT were consistently found in livers, whereas scales and swim bladders contained the least amount of TPT. Mass-balance model showed that muscles (dorsal and ventral) generally contributed to 50% of the total body burden of TPT in these fishes on a wet-weight basis. Regression analysis suggested that TPT concentration of the whole organism could be predicted using its concentration in dorsal muscles ($p < 0.05$, $r^2 = 0.973$), which indicated that dorsal muscles can actually represent the contamination in the whole organism on dry-weight basis. Our findings from profiling the distribution pattern of TPT compounds would help identify potential TPT-induced organ-specific toxic effects in fishes, and investigate the potential of bio-magnification of TPT in marine food webs.

TU290

POPs in the terrestrial environment of Schirmacher Hills, Antarctica: A preliminary study and implications for PCB degradation kinetics

s. katakam, IIT Hyderabad; A. Qureshi, IIT Hyderabad / Civil Engineering; P. Chakraborty, SRM University; A.K. Tiwari, NCAOR / Polar environment

We present preliminary results on the occurrence of Polychlorinated biphenyls (PCBs) and Organochlorine pesticides (OCPs) in the terrestrial environment [moss, and water] of Schirmacher Hills, Dronning Maud Land, Antarctica. α-HCH concentrations (4.48 ng/g dw) were comparable to other locations while *p,p′*-DDE (31.2 ng/g dw) concentrations are higher than those observed by 5 times or more. Out of the tested 28 polychlorinated biphenyl (PCB) congeners, only 6 PCBs were detected. Σ_{28}PCBs in both moss (122 ± 115 ng/g dw, $n = 5$] and water (30 ng/L and 165 ng/L, $n = 2$) are higher by up to 10 times compared to other studies around the continent. Heavier congeners (hexa through nona) in both moss and water samples constitute 52–100% of Σ_{28}PCBs. This suggests that some localized sources of PCBs may still exist in the Schirmacher Hills region. It is possible that the old research stations, or tourism base, in the area may be one of those sources. While the observed congener distribution can be expained using congener distributions in known commercial PCB mixtures (Aroclors, Sovol and Clophen) in some samples, a post-depostition transformation in snow/ice (in glaciers) is required to explain the remaining observations. Box modeling exercise to reproduce congener distribution in our environmental samples suggests that degradation half-lives of PCB congeners in snow should differ by at least 20 times between penta-/hexa-/hepta- (1750 h) and octa- (35000 h) congeners, as against the uniform value of one year (8760 h) suggested previously. Different half-lives could be possible, either if the rate of actual photoreductive-dechlorination process is different for congeners other than the two congeners which were experimentally determined previously or due to occurrence of hydroxylation reactions in snow that have been shown to be responsible for a more efficient degradation of lighter congener (PCB-7). It is also possible that the higher intensity of solar radiation in Antarctica is driving a faster degradation reaction, albeit for the lighter congeners only. Overall, PCB congener distribution in some samples can be explained by a direct contribution from sources, while for others a post-deposition degradation step is required to reproduce the observations. More studies are required to identify and constrain the PCB sources in the Schirmacher Hills, and PCB congener degradation kinetics in snow.

TU291

Degradation of crop protection products in Brazilian soils

N. Baudin, S. Marshall, Syngenta Product Safety / Product Metabolism and Analytical Science; G. Bending, University of Warwick / School of Life Sciences; I. Bramke, M. Garrod, Syngenta Product Safety / Product Metabolism and Analytical Science; C. Mckillican, Syngenta Crop Protection LLC / Product Metabolism and Analytical Science

Before CPPs become commercially available they are subjected to rigorous testing according to strict regulatory guidelines, including understanding the fate of these compounds in the soil environment. The global use of CPPs requires an understanding of their behaviour in a range of soils, including those from both temperate and tropical regions. Moreover, results from previous CPPs fate studies have shown fundamental differences in Brazilian soils compared to temperate soils. The aim of my project is to determine the major physico-chemical and biological properties controlling the degradation of pesticides in Brazilian soils. A set of 4 different soils, prescribed for regulatory testing to encompass the typical range of properties, such as pH, organic matter, clay content and cation exchange capacity (CEC), including an on crop version and a pristine version of these soils, was used in my study. My first experiment focussed on the rate of degradation and mobility of the fungicide thiabendazole in four different Brazilian soils and one temperate soil. Thiabendazole exhibited slow degradation due to it adsorbing onto the soil solid surface, thus being unavailable to microorganisms in the soil pore water. Thiabendazole half-lives (DT_{50}s) and distribution coefficients (K_{OC}s) were higher in some Brazilian soils compared to the temperate soil due differences in their physico-chemical properties. Further pesticides will be tested to determine which key physicochemical and biological properties are the driving force for a compounds fate in tropical soils.

TU292

Study of the Degradation of Bisphenol A by the basidiomycete fungus Trametes versicolor, via HPLC-DAD

C.E. Gracio, V. Bianchi, P.G. Silva, A.C. Montini, E.C. Lima, C.L. da Silva, UFABC / CCNH

Bisphenol A (BPA) is a compound widely used in plastificants such as polycarbonates and resins. Its use has been increasing in the last years and researches point that it may be detected in the environment in great concentrations. Moreover, this substance is classified as a pollutant of emerging concern because of its persistence in the environmental systems and its uncertain damages to both human and animal health. Some studyes connect the exposition to this compound with cancer and other diseases.In this work, it was evaluated the ability of the fungus specie *Trametes versicolor* (Institute of Botany of São Paulo) in degrading BPA by growing the mycelium in a enriched liquid medium and adding a Sigma brand pattern to it. After that, 2mL of sample were periodically purchased and analyzed in an Agilent 1220 Series HPLC with DAD detector. 87.78% of removal was the average efficiency of the degradation; slightly smaller than other species that our group has investigated in previous works, such as *Trametes villosa*. In future works, the activity of the enzymes like Laccases and Manganese-Dependent Peroxidase should be taken into consideration and evaluated, as well as to compare the degradation with an abiotic system containing the BPA pattern – in order to evaluate its persistence in the culture medium.

TU293

Soil dissipation of paraffin oils: Improvement of the microbial degradation and impact on soil dissipation.

P. Adrian, A. Barret, CEHTRA SAS; G. Destrycker, CEHTRA; P. Lemaire, TOTAL Fluides

The study was conducted according to OECD 307 and the active substance applied onto soils according to Good Agricultural Practices in EU. Four soils were freshly collected from sites in Germany and handled per the International Standards Organisation Standard ISO/DISS 10381-6 Part 6 and Good Laboratory Practices. Following incubation the soil samples are extracted and further analysed by GC/MS i.e. monitoring of one ion (m/z) for the internal standard (66 m/z) and one ion for the test substance (57 m/z). Satisfactory method performance was achieved at each degradation interval, as verified by recovery efficiency testing. The test substance, Paraffin Oil CAS (72623-86-0) as produced by TOTAL Fluides, dissipated to below 50% of the original concentration over a 17-day period after the initial treatment (DAT) in all four soils evaluated in the study. From 17 DAT through 122 DAT the concentration of Paraffin Oil CAS (72623-86-0) as produced by TOTAL Fluides continued to decrease to 80-90% dissipated in all four soils evaluated. From 122 DAT through 300 DAT there was not any substantial dissipation of Paraffin Oil CAS (72623-86-0) as produced by TOTAL Fluides in any of the four soils occurring. Two treated soil samples were then treated with dextrose, ammonium nitrate and monopotassium phosphate in order to improve the remaining bioactivity. The remaining residues of the applied substance was then followed for two weeks. The results showed that for one soil a complete dissipation of the remaining residues occurred although in these time of incubation a plateau was observed prior treatment. A methodology is therefore proposed to demonstrate for certain chemicals that the degradation is of biological origin and to integrate these results in the proposed end points.

TU294

Leaching of PAHs from Coal Mining Heap Samples from the Saarland

T. Schiedek, Applied Geosciences / Applied Geoscience

After 250 years coal mining stopped 2012 in the Saarland, Germany. Ca. 80 mining heaps remained (up to 100 m tall). Heaps contain a significant amount of natural coal, well known as a source of polycyclic aromatic hydrocarbons (PAHs). PAHs are pollutants with high persistence, toxic impact on organisms. This study aims at quantifying the amount of PAHs which could be potentially and under almost real conditions leached from heap sediments. Samples (top 10 cm) from heaps of Duhamel, Göttelborn, Lydia, Reden, Viktoria and 2 coal samples, were extracted and used inbatch experiments. Leaching experiments with an automatic extraction unit (Dionex300) were executed, using acetone (potential leaching) and water at different temperatures (40°C and 80°C, "real" leaching). Additionally, batch experiments were conducted to investigate leaching under near-equilibrium conditions (10 days). The 16 EPA-PAHs and four additional PAHs (1-methylnaphthalene, 2-methylnaphthalene, benzo[e]pyrene and perylene) were analysed by gas chromatography with mass detection. Additionally total organic carbon (TOC) and physico-chemical parameters (pH and TDS) were analysed. The heap samples contained a potential concentration in the range of 0,01 - 36 mg/kg. The highest value of 36 mg/kg was found in the heap Lydia (most abandoned PAH was naphthalene). In general, light PAHs (mass lower 202 AMU) were found in concentrations up to 40 times higher than heavy PAHs. Coal samples showed 4-times higher PAH concentrations (most abandoned light PAHs) than sediment samples. However, the water extractions showed only light PAHs. The batch experiments (3 samples per heap, 1 coal) showed only light PAHs in the water phase (concentrations from 0,1 – 0,5 µg/L), with 2-methylnaphthalene (0,5 µg/L) in the coal sample. The highest concentration of total PAHs of a heap was found at Lydia, ca. 6 times higher than the lowest concentration found in the heap Victoria. Potential light PAH concentration in sediments (acetone extraction) were ca. 3 orders of magnitude higher than water extractions at 40°C and 80°C or in batch experiments. The extract at 80°C showed 20 times higher concentrations than at 40°C for the lighter PAHs. TOC content was found to be above 60% in coal samples (with 90% OC). Sediment samples showed TOC values in the range of 2% - 8%. Light PAHs from heaps have been found to be mobile, but maybe immediately sorbed by natural TOC. However, dust emissions may pose a potential risk from heaps.

When ecotoxicology meets trophic ecology (P)

TU295

Will detoxification processes developed by marine mammals still be efficient in the future?

P. Mendez, Observatoire Pelagis; J. Spitz, Observatoire Pelagis Université de La Rochelle/CNRS; F. Caurant, Université de La Rochelle / LIENSs

In marine mammals, food is the main route of entry for contaminants. Their concentrations can largely vary among prey species, for that reason differences in bioaccumulation will arise from differences in predator diet. Among all the contaminant that marine organisms faced, metallic trace elements (MTE) are natural substances that have been present on the earth since its formation. MTE can be divided in essential and non-essential in function of their biological role in the organisms. Low concentrations of essential elements can lead to deficiency effects. On the contrary, excess of non-essential elements [i.e. cadmium (Cd), mercury (Hg) and lead (Pb)] can induce toxic effects. However, their long-term presence in to the environment has allowed to marine mammals and other marine organisms to developed mechanisms to mitigate the potential toxic effects of these non-essential elements. The best known detoxification process is the demethylation of Methyl-Hg by Selenium (Se) forming granules of tiemmanite (Hg:Se) in their liver. Today, anthropogenic activities induced a continuous increase of Hg concentrations in the environment, altering prey availability as well as the composition of prey communities. Such changes could affect the ability of marine mammals to control the negative impacts of their exposure to non-essential elements. Here, we investigated the temporal trends of Hg and Cd in liver and kidneys (main storage tissues) of 183 individuals of the smallest cetacean species in the North Atlantic: the harbour porpoise (*Phocoena phocena*). Both elements showed a significant increase ($p < 0.05$) of concentrations between 1999 and 2013. Notably, we highlighted a strong increase of the number of individuals exhibiting extreme values among the range of measured concentrations. In parallel, we analysed essential trace elements in 78 forage species (i.e. jellyfish, crustaceans, cephalopods and cartilaginous and bony fish) to assess their quality for predators. Results showed broad differences of their essential element composition. In particular, selenium concentrations can largely vary between species suggesting different Se exposure among marine mammal species depending on their diet, which means that some of them could be less protected against Hg toxicity. Thus, changes in prey quality could have cumulative effects in cetaceans (increase of toxic elements and deficiency in essential ones) impacting the efficiency of detoxification processes in the future.

TU296

Impact of biofilm growth on mercury accumulation in Daphnia magna

s. issa, Norwegian University of Science and Technology; T.M. Ciesielski, Norwegian University of Science and Technology / Department of Biology; S. Einum, Norwegian University of Science and Technology / Centre for Biodiversity Dynamics CBD; . Mikkelsen, Norwegian University of Science and Technology / Department of Chemistry; V. Jaspers, Norwegian University of Science & Technology / Biology

A largely neglected issue in lab-based toxicity testing of pollutants is the potential for other components in the biotic community to influence the effects of toxins on focal species. For example, *Daphnia* are commonly used as a model organism for such studies to infer toxicity thresholds of aquatic organisms. Nevertheless, such tests are usually conducted in highly standardized conditions and with a minimum of naturally occurring biofilm. Although this allows for standardization, it may hinder ecological relevance. Biofilm commonly grows in culture medium and serves as additional food for *Daphnia*. It can aslo accumulate mercury (Hg), a pollutant of high international concern because of its long-range transport across the globe and its various toxic properties. As such, biofilm can play a central role in the transfer of Hg to higher trophic levels in freshwater ecosystems. By taking this into account, we can better predict effects of Hg in these ecosystems. Therefore, we conducted an experiment where single *Daphnia magna* clones were exposed at 20ÂC to 0.2 Ђg/L and 2 ɋg/L Hg ($HgCl_2$) in the presence and absence of biofilm. Our objective was to test for a significant effect of Hg accumulation in biofilm on Hg accumulation in daphnids. Results showed no significant effect of biofilm on Hg uptake in *Daphnia*. However, biofilm served as an additional source of selenium (Se) to daphnids, thereby increasing Se/Hg molar ratios in the animals. Thus, biofilm played a central role in the transfer of Se through the freshwater food web and in decreasing the risk from Hg toxicity in *Daphnia*.

TU298

Multiple stressor effects on resource quality for consumers: a case study with phototrophic biofilm exposed to phosphorus and ionic silver

M. Danger, K. Sanchez-Thirion, LIEC; C. Crenier, LIEC Université de Lorraine CNRS; A. Bec, Université Clermont Auvergne; J. Ferriol, ECOLAB UMR CNRS UPS INPT; A. González, Universidad de Las Palmas de Gran Canaria; F. Perrière, Université Clermont Auvergne; L. Ten-Hage, ECOLAB UMR CNRS UPS INPT; V. Felten, LIEC / LIEC CNRS UMR; J. Leflaive, ECOLAB UMR CNRS UPS INPT

Autotrophic biofilms are fundamental biological compartments of many aquatic ecosystems, representing notably a major resource for many invertebrate consumers. To date, most studies have tried to understand the impacts of stressors on microbial communities or on functional processes taking place into the biofilm mat. Far less studies investigated the indirect effects of stressors on upper trophic levels through alterations of the quality of biofilms. We investigated, through a laboratory study, the single and combined effects of phosphorus (P) availability and silver contamination on the elemental (C:N:P ratios) and biochemical (fatty acid profiles) compositions of a diatom-dominated biofilm. We hypothesized that 1) P would enhance the elemental quality while 2) P and silver, through the replacement of diatoms by more tolerant primary producer species, would reduce the biochemical quality of biofilms for their consumers. The quality of biofilms for consumers was assessed for a common crustacean species, *Gammarus fossarum*, by measuring organisms survival and growth rates. Results mainly showed that species replacement induced by both stressors affected biofilm fatty acid compositions, and that P immobilization permitted to achieve low C:P biofilms, whatever the level of silver contamination. Gammarids growth and survival were not significantly impacted by the ingestion of silver-contaminated resource. On the contrary, we found a significant positive relationship between the biofilm P-content and gammarids growth, while biofilm fatty acid contents were unrelated to this parameter. This study underlines the large indirect consequences stressors could play on basal resources quality for consumers, and, in turn, on the whole food web.

TU299

Soil pollution induced changes in leaf litter chemical composition and in detritivore physiology and activity.

A. Le Navenant, LIEC Université de Lorraine CNRS; E. Billoir, Université de Lorraine, CNRS UMR 7360; A. Cébron, LIEC CNRS UMR Université de Lorraine; S. Coq, CEFE, CNRS, Montpellier; V. Felten, LIEC / LIEC CNRS UMR; J. Nahmani, CEFE, CNRS, Montpellier; F. Maunoury-Danger, LIEC - Université de Lorraine - CNRS

In terrestrial ecosystems contaminated with high metal contents (brownfield), recent studies showed that, surprisingly, leaf litter decomposition process could be maintained despite deep changes in bacteria to fungal abundance ratio and invertebrate detritivore community structures. To disentangle the potential mechanisms leading to this pattern, we evaluated chemical characteristics of birch litter (*Betula pendula*) produced on 10 sites along a metallic contamination gradient to assess the effects of the contrasted litter characteristics on microbial colonization and litter consumption by, the diplopod *Glomeris marginata*, used as a model detritivore. Our results reported an impact of soil contamination on leaf litter chemical composition, leading in turn to significant impacts on diplopod physiology (in particular with an increasing oxidative stress when diplopod were exposed to contaminated litters). However, pollution mediated changes in leaf litter chemistry had no significant impacts on microbial litter colonization (bacteria:fungi ratio) and litter consumption by detritivore, confirming the high resilience of litter decomposition process to soil metallic contamination.

TU300

Decomposition rates and feeding activity of soil fauna in relation with stages of plant colonization in mine soils of a Mediterranean area

A. Peñalver Alcalá, J. Alvarez-Rogel, M. Tercero Gómez, Escuela Técnica Superior de Ingeniería Agronómica. Universidad Politécnica de Cartagena / Ciencia y Tecnología Agraria; M. González-Alcaraz, Department of Biology & CESAM - University of Aveiro / Biology & CESAM

Organic matter decomposition (tea bag index) and feeding activity of mesofauna (Bait Lamina) were studied in an abandoned mine tailings area. Six environments were studied: A) Within the mine tailings: 1. Bare soils (S); 2. Small groups of *Pinus halepensis* trees (2-5) ≤≈2.5 m high, growing scattered (P); 3. Isolated *P. halepensis* trees >≈4 m high with shrubs and herbs under the canopy (P+MS); 4. Dense patches with several *P. halepensis* trees (>5) >≈4 m high and shrubs and herbs under the canopy (DP+MS). B) Outside the mine tailings: 5. Polluted forest with *P. halepensis* trees > 5 m high and shrubs and herbs under the canopy (PF); 6. Control forest not contaminated with *P. halepensis* trees > 5 m high and shrubs and herbs under the canopy (CF). Roiboos and green-tea bags were buried in each environment for a total of 110 days. Tea bags were regularly collected from each environment, to calculate mass remaining, decomposition index and organic carbon and nitrogen of the remaining material. In each plot, two groups of 5 baited sticks were vertically inserted. The number of holes partially and fully empty after 20 days was recorded to calculate the % of holes fed upon. After ≈50 days, the percentages of mass remaining in the tea bags were: - DP+MS, P+MS and S: green tea ≈50-55%, roibos tea≈90%; -PF, CF and P: green tea ≈80-85%, roibos tea ≈96%. These percentages were maintained until the day 110. The lower decomposition in CF and PF can be related with more abundance of resources in forest soils outside of the tailings which could induce to microorganisms to use other sources of nutrients different from tea material. However, in more stressed environments, such as the soils within the tailings, a source of organic matter easily decomposable, mainly the green tea, could stimulate microbial activity by a priming effect. Besides, within the tailings, the decomposition in S (the most unfavorable environment "a priori") could be favored by the high soil temperature (average ≈28 ºC), as a consequence of the lack of vegetation, while in other environments the shading by plants maintained the temperature between ≈23 and ≈25 ºC. Feeding activity was (% of holes fed upon): CF =42%, P =39%, S= 31%, P+MS =21%, AF =8%, DP+MS =7%. The high % of holes fed in bare soils (S) could be related to the scarcity of resources in this environment which stimulated the consumption of the bait.

TU301

Effects of mineral supplements on lead exposure in free-ranging herbivores

J. Pareja Carrera, IREC-UCLM / IREC-UCLM; M. Martinez-Haro, IREC-Instituto de Investigación en Recursos Cinegéticos / Department of Life Sciences; J. Rodr�guez-Estival, University of Castilla-La Mancha / IREC-UCLM; J.E. Smits, University of Calgary / Ecosystem and Public Health; M. Durkalec, National Veterinary Research Institute / Department of pharmacology and toxicology; R. Mateo, IREC-CSIC- UCLM / Grupo de Toxicología de Fauna Silvestre

Lead (Pb) mining has contributed to the extensive release of Pb into natural environments for centuries. In former mining districts, now on the Iberian Peninsula mainly livestock and hunting estates, Pb persisting in the soils and vegetation of affected areas may constitute an environmental and health risk. Since Pb is an important toxic metal for both animals and people, there is a need to explore how to prevent or reduce exposure. Therefore, we studied the effect of commercial mineral supplements on Pb bioaccessibility through laboratory and field approaches. One aim was to prevent or reduce absorption in herbivore inhabiting mining areas and thus reduce the possible exposure route to people. In our previous work, we identified one mineral supplement rich in calcium (Ca) and phosphorus (P) that specifically altered Pb solubility and absorption in a digestive tract simulation model. Here, we go one step further to evaluate the effects of that commercial mineral supplement on Pb absorption and on immune status in goats from an old mining area. Two groups of goats from two plots with similar soil Pb concentrations were selected. One group was supplemented with the commercial mineral salt for 20 days, whereas the other one served as control. Then, the Pb exposure was evaluated in blood, milk and feces, and the phytohaemagglutinin (PHA)-skin test was used to evaluate T-cell-mediated immunocompetence. Results showed that all goats responded to the PHA, but no significant difference was detected between groups. Blood and milk samples were collected the same days that the PHA-skin test was conducted. Blood Pb levels in supplemented goats were slightly lower than in the non-supplemented goats (4.6 µg/dl vs 5.3 µg/dl). Milk Pb levels in non-supplemented goats were 2-fold higher than in supplemented animals (0.012 vs. 0.006 µg/g wet weight). Based on our study, supplements enriched with Ca and P appear to decrease the Pb burden in the animals, likely through competition in intestinal absorption with the nutrient, Ca, preferentially absorbed over the toxic metal, Pb, and possibly through reduced geophagia by the animal. These supplements could serve as a cost-effective measure to reduce Pb exposure of livestock and wildlife in areas contaminated with Pb. An assumed additional advantage is reduced the risk of Pb exposure through milk consumption by the local human population.

TU302

Analysis of anticoagulant rodenticides, neonicotinoids and fipronil in liver of predatory birds

D. Schenke, Julius Kühn-Institut - Federal Research Centre for Cultivated Plants / Institute for Ecological Chemistry, Plant Analysis and Stored Product Protection; A. Geduhn, Federal Environment Agency / Section IV 1.4 Health Pests and their Control; A. Esther, Julius Kuehn-Institute - Federal Research Centre for Cultivated Plants / Institute for Plant Protection in Horticulture and Forests, Vertebrate Research

Pesticides in predatory birds have been drawing much attention worldwide in regard to species declining and protection. Pesticides are used for pest management of animal species such as commensal rodents and sap-sucking insects. However, pesticides can lead to secondary poisoning, when predators take up pesticide residues from primarily exposed target or non-target species. The analysis focused on anticoagulant rodenticides, neonicotinoids and fipronil which were regularly applied in the years 2011 to 2013. We obtained liver samples of 89 avian predators from this period, which were collected from veterinary institutions or private persons from 26 administrative districts in Germany. Avians were found dead or were euthanized shortly after admission to the veterinarian. Defrosted liver samples were spiked with surrogates and homogenized in a mixture of methanol and water (2:1/v/v) and cleaned up by solid supported liquid extraction with a diatomaceous earth column (Geduhn et al., 2014, DOI 10.1016/j.scitotenv.2014.07.0490048-9697). Quantification of the analytes was performed by LC-ESI-MS/MS with a calibration from 0.1 to 100 ng/ml ($r^2 > 0.99$) and a signal to noise ratios > 6:1 for the lowest concentration level. The neonicotinoids imidacloprid with the metabolites 5-OH-IMD and IMD-olefine, thiamethoxam and clothianidin with TZMU and TZNG were not found in the predators although expected especially in case of insect-consuming species such as little owl (*Athene noctua*). Similarly, we detected no residues of the phenylpyrazole fipronil, which has a higher bioaccumulation potential and the metabolites F-sulfone, F-sulfide and F-carboxamide. One to four substances of the rodenticides chlorophacinone, difenacoum, bromadiolone, brodifacoum, flocoumafen and difethialone were found in 30% of the liver samples, originated from 14 different districts. Brodifacoum was detected in more than 70% of these samples. No sample contained coumatetralyl and warfarin. Residues occurred more often in avian predators specialized on rodents than in generalists; e.g. 44% of the 26 liver samples from common bussards (*Buteo buteo*) contained residues. The portion was with 80% even higher for red kite (*Milvus milvus*) but only 5 samples of this species were examined. A residues distribution pattern will be presented but more samples are necessary for final statements.

TU303

Trophic Magnification of POPs including PFCs Within A Terrestrial Food-Web of An Avian Top Predator, the Cooper's Hawk (Accipiter Cooperii)

K. Fremlin, SFU / Department of Biological Sciences; J.E. Elliott, Environment Canada / Science Technology Branch; F. Gobas, Simon Fraser University / Resource & Environmental Management

Protocols to assess bioaccumulation of POPS within terrestrial systems are far less developed compared to aquatic systems. Presently, regulatory agencies in Canada, the USA, and the EU use only bioaccumulation information for fish to assess the bioaccumulation potential of chemicals. However, recent studies have shown that some chemicals that are not bioaccumulative in aquatic food-webs do biomagnify in terrestrial food-webs. To better understand the bioaccumulation behaviour of chemicals in terrestrial food-webs, we aim to produce a food-web model to assess the biomagnification of POPs in an apex avian predator, the Cooper's hawk. Over 100 samples were collected from various trophic levels of the food-web including hawk eggs, songbirds, invertebrates, and berries. All samples were analyzed for a number of contaminants listed as priorities for monitoring by the Chemical Management Plan of the Canadian federal government. Stable isotope analysis of $\delta^{13}C$ and $\delta^{15}N$ signatures of hawks, songbirds, invertebrates, and berries was used to estimate the trophic position of each organism. Legacy POP concentrations were expressed in terms of lipid equivalent concentrations to account for variability in the fractions of lipid and non-lipid organic matter measured in each sample. PFC concentrations were expressed in terms of protein equivalent concentrations to account for the fraction of protein within each sample, which was estimated as the product of the percent of nitrogen measured in each sample and a nitrogen:protein conversion factor. Censored regression by maximum likelihood estimation was used to assess the relationship between the natural logarithm of each lipid or protein equivalent concentration and trophic position. Trophic magnification factors (TMFs) were determined as the antilog of the regression slope. TMFs of legacy POPs ranged from 0.61 to 38.40, indicating that most legacy POPs are biomagnifying in this terrestrial food-web. TMFs of PFCs ranged from 11.8 to 544.6, indicating that PFCs are also biomagnifying in this terrestrial system and potentially at higher magnitudes than legacy POPs. Overall, terrestrial TMF values for legacy POPs were comparable to or higher than TMF values determined for several aquatic systems; whereas, terrestrial TMF values for the PFCs were considerably higher than TMF values found in aquatic systems.

TU304

Seasonal dynamics of zooplankton community, trophodynamics and Hg across a gradient from a DOM rich river to a marine system

S. Schultze, University of Oslo; A. Ruus, NIVA / NIVA; K. Borga, Department of Biosciences, University of Oslo / Department of Biosciences; T. Andersen, D.O. Hessen, University of Oslo / Department of Biosciences; H. Veiteberg Braaten, O. Kaste, NIVA Norwegian Institute for Water Research; A. Poste, Norwegian Institute for Water Research

Recent increases in terrestrially derived dissolved organic matter (tDOM) in freshwater ("browning") and marine systems ("coastal darkening") have been noted in several studies in boreal areas. This leads to the question if high riverine input of terrestrial derived material will affect the food web dynamics of recipient brackish and marine ecosystems. The presence of tDOM may affect light attenuation, primary production, visual responses, nutrient availability and bacterial abundance - all factors that may affect food webs in different manners. Terrestrial inputs can also directly and indirectly influence inputs, bioavailability and food web uptake of contaminants such as mercury (Hg). While several studies exist on effects of browning on productivity and community composition of freshwater systems as well as bioaccumulation of contaminants, there is considerably less known about OM and Hg dynamics at the freshwater-marine interface. To address these issues we characterized physicochemical conditions, lower food web structure and Hg dynamics along a river- fjord continuum in southern Norway. Comprehensive water (surface and deep water) and zooplankton samples were collected on five occasions in 2015/2016. Physical-chemical parameters and nutrient concentrations were measured alongside data on chlorophyll a, bacterial as well as viral abundance and zooplankton composition. Methylmercury concentrations in zooplankton were analysed and trophic position and food origin was established with the help of stable isotope measurement (δ13C and δ15N). Conservative mixing, reflected in the salinity, was the major structuring force for physicochemical conditions in the system. DOC, TOC, total Hg and silicate concentrations reflected physical mixing thus showing a clear decline from freshwater to marine. Overall bacterial abundances were higher in freshwater than marine dominated systems and show clear seasonal patterns. Nutrients reflected both physical mixing patterns as well as biological (bloom) processes. Salinity and seasonality were major structuring forces for the zooplankton community, and zooplankton from the more freshwater influenced inner fjord appeared to have higher dietary reliance on terrestrial carbon sources than zooplankton from the outer fjord. We also found higher Hg concentrations in zooplankton from the more freshwater influenced sites, highlighting the importance of riverine Hg inputs for contamination of coastal biota.

TU305

Spatial comparison of contamination and biomagnification profiles of triphenyltin compounds in sub-tropical marine environments of Hong Kong

R.C. Sham, K.K. Ho, The University of Hong Kong / The Swire Institute of Marine Science and School of Biological Sciences; X. Wang, Xiamen University / Department of Environmental Science and Technology; K.M. Leung, The University of Hong Kong / The Swire Institute of Marine Science

Biomagnification of lipophilic organic contaminants is one of the major pathways to accumulate xenobiotic substances in marine organisms. Interestingly, the magnitude of biomagnification is not necessarily consistent in organisms across the marine food chain. Triphenyltin (TPT) compounds, which is moderately lipophilic ($log\ K_{OW}$ ~3.5), are commonly used in antifouling paints on sea-going ship hulls and submerged mariculture facilities in Hong Kong, Mainland China, and Taiwan. Studies have suggested that TPT can be biomagnified along the lower part of the trophic food chain (i.e., among primary producers, invertebrates, and fishes), while their magnification potential has remained unclear among the higher trophic organisms, such as larger fishes, dolphins, and seabirds. To date, only two studies have investigated whether the biomagnification of TPT occurs in marine organisms at higher trophic levels; however, their findings were contradictory. Therefore, we aimed to evaluate the biomagnification potential of TPT in high-trophic organisms across a spatial gradient from the more-contaminated western waters to the less-contaminated southern waters of Hong Kong. We have divided the western and southern waters into four sites, namely inner estuary (WI), outer estuary (WO), south of Lantau Island (SL), and southeast of Hong Kong Island (SE). Environmental (seawater and sediments) and biota samples (including molluscs, crustaceans, fishes and marine mammals) collected from the respective sites were analysed using gas-chromatography mass-spectrometry to examine the concentrations of six organotin compounds (i.e., mono-, di- and tri-butyltin; mono-, di- and tri-phenyltin). Preliminary results showed that seawater samples from WO had the highest concentrations of TPT ($F_{3,12} = 21.28$, $p < 0.05$) and the results from biota samples indicated a concentration gradient from the western to southern waters (WI > WO > SL > SE). The above findings were consistent with our hypothesis that the western waters are more polluted than the southern waters due to the influx and polluted freshwater from the Pearl River. Our forthcoming results on whether TPT can be biomagnified in the higher trophic organisms will further shed light on its biomagnification potential at the higher food chain with consideration of its lipophilicity and octanol-water partition coefficient (*log Kow*).

TU306

Comparative trophodynamics of polychlorinated biphenyls and chlorinated paraffins in an urban river

P. Labadie, UMR CNRS EPOC Universite Bordeaux / UMR 5805 EPOC; A. Goutte, EPHE / UMR METIS; K. Maciejewski, UMR EPOC; C. Simonnet-Laprade, UMR5805 EPOC; K. Le Menach, UMR CNRS EPOC Universite Bordeaux / EPOC UMR 5805; F. Alliot, EPHE / UMR Metis; R. Santos, Hepia, University of Applied Sciences Western Switzerland / Ecology and Engineering of Aquatic systems research group; H. Budzinski, University of Bordeaux

Trophic magnification factors (TMFs) have been extensively used to asses the biomagnification potential of organohalogens in numerous aquatic and terrestrial ecosystems. While the trophodynamics of legacy persistent organic pollutants is relatively well known, that of more emerging halogenated contaminants remain scarcely documented. This is particularly the case for short and medium-chain chlorinated paraffins (SCCPs and MCCPs, respectively), which quantitative analysis remain challenging. In the present study, we aimed at investigating the biomagnification of these compounds in the trophic web of an urban river heavily impacted by urban inputs: the Orge river (near Paris, France). In addition, a comparative study was performed, using polychorobiphenyls (PCBs) as benchmark chemicals (i.e. positive control for biomagnification). Abiotic and biotic samples, ranging from primary producers to piscivorous fish (n=45), were collected in this systems and analysed for PCBs, SCCPs and MCCPs. Stable isotopes of nitrogen were used to estimate trophic levels and to compute TMFs using a Linear Mixed-Effects Model (*lmec*) accounting for the difference of samples between taxa. Our results show the expected biomagnification of the targeted PCB congeners (i.e. TMF >1), thereby validating both the sampling strategy and the data treatment. SCCPs exhibited TMFs in the range 0.4 –2.0 and the extent of biomagnification was directly related to structural features such as alkyl chain length and chlorine content. Conversly, MCCPs almost consistently displayed TMFs < 1, likely as a consequence of their higher biotranformation rates compared to SCCPs. Such results provide additional data for the risk assessment of chlorinated paraffins.

TU307

Copper and mercury effective body residues in freshwater macroinvertebrates as related to benthic community metrics from a mining river basin.

I. Moreno, L. Méndez-Fernández, University of the Basque country UPVEHU / Zoology and Animal Cellular Biology; M. Martínez-Madrid, University of the Basque Country UPV EHU / Genetics, Physical Anthropology and Animal Physiology; N. Costas, I. Pardo, University of Vigo / Ecology and Animal Biology; P. Rodriguez, University of Basque Country / Zoology and Animal Cell Biology

Copper and mercury body residues in 10 macroinvertebrate taxa were used to model the alterations in benthic community metrics due to metal bioaccumulation in mining areas of the Nalón River basin (Spain). The studied taxa are potentially useful as water quality biomonitors and cover different functional feeding styles. This is part of a larger study in North Spain that aims to develop biota quality standards of several heavy metals, to contribute to water quality management to take forward the conservation of macroinvertebrate communities. The specific objectives of the study were: first, to model the relationships between 4 macroinvertebrate community metrics (number of families and abundance of EPT and PT), one multimetric (METI) and a predictive model (NORTI), using the Cu and Hg body residues as predictor variables; second, to assess Cu and Hg toxicity to benthic macroinvertebrate communities through the estimation of effective body residues (ER); and third, to investigate the taxa-specific differences in metal ERs in relation to their feeding styles. The ERs were estimated for each taxon and metal from the best non-linear models, selected using Akaike's Information Criteria, and compared with the 90th percentiles (P90) of the data distribution in the reference sites of the study area, considered an approach to threshold (=no-effect) concentrations. In most cases, dose-response models were fitted for Cu, but only in few instances for Hg. Results showed that Cu-ER_{50} and Cu-ER_{25} in 4 taxa (Baetidae, Hydropsychidae, Ephemerellidae and Microdrili oligochaetes) were usually less than 2 times above the P90, calculated for the same taxa. These ERs in other 3 taxa (Heptageniidae, Ephemeridae, Rhyacophilidae) were mostly within the range of 2.1 to 5.0 times the P90. The largest ratios were found in ER_{50} for Lumbricidae and Perlidae, which reached5 to 12 times the P90 values. In the case of Hg, the predator taxa (Rhyacophilidae and Perlidae) and some of their potential preys, e.g. mayflies and simulids, showed ERs that were typically within the range of 1 to 3 times their respective P90s.

TU308

Trophic transfer of Cadmium nitrate in a simplified marine food chain: experimental feeding rate of gelatinous zooplankton Aurelia sp. and Sanderia malayensis on crustacean Artemia sp.

E. Costa, C. Gambardella, V. Piazza, CNR ISMAR; S. Lavorano, Costa Edutainment spa Aquario di Genova; M. Faimali, F. Garaventa, CNR ISMAR

Trophic interactions are a crucial vector of contaminant transfer in both aquatic and terrestrial ecosystems. In the marine environment, Cnidarian jellyfish are known to play an important role in food webs as major predators of metazooplankton and as prey of apex predators, influencing the microbial loop, through direct and indirect effects, besides regulating the marine biogeochemical fluxes. In this study, the potential contaminant transfer was investigated in simplified marine food chains. The nauplii of the brine shrimp *Artemia sp.* and the ephyrae of *Aurelia* sp. and *S. malayensis* were selected as primary and secondary consumers, respectively. Cadmium nitrate was selected as toxicant. Performed experiments consisted in feeding ephyrae, every 24 hours for 5 days, with nauplii of *Artemia sp.* previously exposed, for 6 hours, to different concentrations (0.1-0.5-1-2-4 mg/L) of Cadmium nitrate; this range was selected through preliminary trials aimed to define the cadmium LC_{50} value for crustacean larvae. At the end of feeding experiment (5 days), the effect of Cadmium nitrate treated crustaceans on ephyrae jellyfish was investigated by the "ingestion rate method", the "predatory performance" and biometrics and bioenergetics parameters (Disch diameter, ash-free dry weight_AFDW and gross growth efficiency_GGE). In addition, 24 hours after each feeding treatment, two ecotoxicological end-points were evaluated on jellyfish ephyrae: Immobilization and Frequency of pulsation (number of pulsations/min). Results showed a 100% of feeding rate and predatory performance in both control and treated jellyfish (*A. aurita* and *S. malayensis*). Cadmium nitrate treated *Artemia* nauplii, once ingested, caused in ephyrae a decrease of Disch diameter and AFDW and also an inhibition of GGE% (*Aurelia* sp.EC_{50} : 3.82 mg/L). As regards ecotoxicological assays, immobilization was never affected (effect < 50%), while frequency of pulsations showed a significant decrease after each feeding treatment. These findings suggest a contaminant transfer from crustacean nauplii to ephyrae able to induce sublethal effects.

TU309

Tissular injuries in Crassostrea virginica as evidence of the trophic transference of copper and cadmium via Chlorella sp.

F. Mares-Guzman, Universidad Autonoma Metropolitana Iztapalapa / Hidrobiology; G. Barrera Escorcia, Universidad Autonoma Metropolitana Iztapalapa / Hidrobiologia; X. Guzman-Garcia, Universidad Autonoma Metropolitana Iztapalapa / Hydrobiology

Essential metals are found in organisms in small quantities carrying out\n biological functions. In contrast non-essential metals do not have a known biological\n function. When metals are incorporated by the organisms they can cause damage and their presence indicates contamination. Several human activities contribute to the increasing load of both essential and non-essential metals in the aquatic\n environment. Microalgae such as *Chlorella* sp., are the primary link in the\n trophic chain. By being in direct contact with the environment, they can\n incorporate contaminants by absorption or adsorption. If these algae\n accumulate contaminants, such as metals, the organisms that feed on them like\n the oyster *Crassostrea virginica* can in turn incorporate them through filtration, which may have negative consequences. The objective of this work \n was to evaluate the effects derived from the trophic transfer of copper and\n cadmium from *Chlorella* sp. to *C. virginica*. Microalgae were\n cultured for 110 h at a sublethal dose of copper and cadmium (0.1 mg / l). A concentration of 30 X 10 ^{6} cells was given to C. virginica\n for 21 days. The evolution of histopathological lesions in C. virginica was\n evaluated in days 0, 5, 10, 15 and 20 of the assay. The analysis\n performed in the digestive gland revealed diverse lesions ranging from the\n loss of cilia and covering membranes, to the increase in the light of the digestive gland tubules, as well as the presence of various\n inflammatory processes. Other organs such as the gills, presented\n inflammation and injuries that compromise the body's physiological processes\n such as feeding and breathing. These damages were evident after the first 96\n hours of exposure to the contaminated food. However, lesions associated\n with cadmiun exposure, a non-essential metal, in more than 50% of organisms\n could be observed on day 10 and those associated with more than 50% \n animals in cooper exposure were deferred to day 15. The presence of *Chlorella*\n sp. in the digestive tract made possible to associate the injuries with\n trophic metal poisoning, and the prevalence of lesions with metal and exposure time.

TU310

Can microplastics save us? Effects of microplastic particles and particle-bound trace contaminants in an artificial aquatic food web

L. Hanslik, COS University of Heidelberg / Aquatic Ecology and Toxicology; A. Batel, University of Heidelberg / Aquatic Ecology and Toxicology; T. Braunbeck, University of Heidelberg / Centre for Organismal Studies

Since 2015, studies dealing with toxic effects of microplastics in freshwater ecosystems come into focus. Still, little is known about vertebrate models as final consumers in food web experiments. The present study investigates the transfer of pristine microplastic particles and a model polycyclic aromatic hydrocarbon, along an artificial food chain with *Artemia* spec. nauplii and zebrafish (*Danio rerio*). Therefore, cryogenically grinded microplastic particles, made of polystyrene (

TU311

Toxicokinetics links predator-prey dynamics to assess zero-valent iron nanoparticles bioaccumulation in a Caenorhabditis elegans-Escherchia coli ecosystem

Y. Yang, National Taiwan University / Bioenvironmental Systems Engineering; Y. Lin, National Health Research Institutes / National Institute of Environmental Health Sciences; H. Lin, National Taiwan University; C. CHEN, National Taiwan University / Bioenvironmental Systems Engineering; C. Liao, National Taiwan University / Department of Bioenvironmental Systems Engineering

BACKGROUND: Zero-valent iron (Fe^0) nanoparticles (NPs) are one of the most paramount NPs applied in environmental remediation that the potential impacts on the ecological dynamics and soil ecosystem health are of great concern. **OBJECTIVE:** The primary objective of this study was to simulate dynamic models linking biokinetic and consumer-resource dynamics in the *Caenorhabditis elegans* (*C. elegans*)–*Escherichia coli* (*E. coli*) OP50ecosystem. **METHODS:** The biokinetic parameters, uptake and depuration rate constants of bacteria and worms were obtained from toxicokinetic experiments and related published literature. Biomass dynamics of bacteria and worms were estimated by employing the Lotka-Volterra model. Dynamics of Fe^0NPs accumulations, bioconcentration factors (BCFs), biomagnification factors (BMFs) were simulated based on the consumer-resource dynamics. A sensitivity analysis was also performed to characterize the influence of consumer-resource-related physiological parameters. **RESULTS:** Results showed that biomass of worms increased steadily from 22.25–51.61 g L^{-1}, whereas the biomass of bacteria decreased rapidly from 17.17–2.29 g L^{-1} and attained a steady-state after 2 h of the simulation in the scenario of 100 mg L^{-1} Fe^0NPs exposure. We also observed that internal concentrations of Fe^0NPs were estimated to be 67 and 1768.85 μg L^{-1} in worms and bacteria, respectively. In addition, the BCF of bacteria was found to be 17.69, close to the experimental results. Moreover, the BMFs of worms were maintained to be consistently smaller than 1 during 24 h exposure. Results also indicated that internal concentrations of Fe^0NPs in worms were mainly influenced by biomass conversion rate for bacteria ingested by worms, whereas parameter of death of worms had the smallest effect on worm internal concentrations. **CONCLUSIONS:** Model application to toxicokinetic results confirms the hypothesis that the consumer-resource dynamics are effectively associated with Fe^0NPs accumulations in bacteria and worms that the bioaccumulation kinetics and consumer-resource dynamics are likely to be dominated by the same physiological parameters.

Use of Effect Based Methods in the context of the national and european legislative framework for the protection of aquatic ecosystems (P)

TU312

INTEREST OF IN VITRO BIOASSAYS (YES/YAS) FOR THE SCREENING OF ENDOCRINE DISRUPTION IN SURFACE WATERS OF WALLONIA (BELGIUM)

C. Chalon, ISSeP; Y. Marneffe, Inst. Scientific de Service Public / Ecotoxicology Department; A. Claessens, C. Frippiat, ISSeP; K. Nott, SWDE; V. Brahy, F. Delloye, SPW-DGO3-DEE

This study is part of the BIODIEN project. This project aimed at conducting, for the first time, a screening campaign of endocrine disruptors (ED) in waters of Wallonia (groundwater, surface water and wastewater). Almost 200 substances were screened, including hormone estrone, alkylphenols, phthalates, chlorophenols, perfluorates, PBDEs, PCBs, HAPs and pesticides. In parallel with analytical methods, YES and YAS bioassays were conducted in order to quantify estrogenic and androgenic activities in surface waters. Antagonist activities were also evaluated. Over 71 river samples (concerning 24 river sampling points from the regional monitoring network), estrogenic activity was detected and quantified in 53 samples and could reach levels up to 11.7 ng E2eq/l (mean: 2.1±1.6 ng E2eq/l). Androgenic activity was never detected. On the other hand, estrogenic and androgenic antagonist activities were detected in 42 % and 55 % of the samples, respectively. When the estrogenic activity is compared to the EU-Watch List EQS for E2 (0.4 ng.l), 60 % of the samples exceed this value. The estrogenic activity was compared to the chemical results. A good correlation was found with the estrone concentration but also with other ED (e.g. bisphenol A, perfluorates). This study is, in a way, the first attempt in Wallonia to follow the *recommendations for the use of effect-based methods (EBM) for monitoring of estrogens in surface waters* emanating from the Science to Policy Interface (SPI) Estrogen monitoring project. These recommendations were presented at the last EU-WG chemicals held in October 2017 and this would possibly lead to the introduction of EBM in regulatory monitoring under the Water Framework Directive (WFD), especially for estrogens.

TU313

Ecotoxicological tools to assess the impact pollution of tributaries to the Alqueva Reservoir (Southern Portugal)

P. Palma, Instituto Politécnico de Beja / Department of Tecnologies and Applied Sciences; S. Fialho, A. Lima, Instituto Politécnico de Beja; A. Penha, H. Novais, Instituto de Ciências da Terra; S. Monllor, Institute of Environmental Assessment and Water Research IDAEA CSIC / Department of Environmental Chemistry; N. Guillem-Argiles, Institute of Environmental Assessment and Water Research IDAEACSIC / Institute of Environmental Assessment and Water Research IDAEA; M. López de Alda, Institute of Environmental Assessment and Water Research IDAEA-CSIC / Department of Environmental Chemistry; P. Alvarenga, LEAF Centro de Investigação em Agronomia, Alimentos, Ambiente e Paisagem, Instituto Superior de Agronomia, Universidade de Lisboa; M. Morais, R. Salgado, Instituto de Ciências da Terra

Degradation of surface waters and biodiversity loss at different spatial and temporal scales occurs through multiple stressors whose effects are difficult to separate and identify. Efficient management of water bodies depends on the development and selection of robust, sensitive and easily applicable tools that allow prioritizing the pressures and stressors that act in a basin, and mitigate their effects. The Alqueva reservoir constitutes the most important water supply source in southern Portugal, a semi-arid region with high levels of water scarcity and where agriculture is one of the main activities. The aim of the present study was to assess the use of an ecotoxicological tool-box in tributaries of the Alqueva reservoir for detecting chemical alterations that may influence the water quality of the reservoir. Water samples were collected along 2017 at four tributaries of Alqueva (streams of Zebro, Álamos, Amieira and Lucefécit) and analyzed for: (i) physical chemical support elements (pH, temperature, dissolved oxygen, conductivity, chloride, total phosphorus, Kjeldahl nitrogen, ammonium, nitrite, nitrate, BOD, COD), (ii) hazardous substances (pesticides), and (iii) ecotoxicological endpoints, using bioindicators representing different trophic levels (*Vibrio fischeri*, *Thamnocephalus platyurus*, *Daphnia magna)*. In general, Zebro and Lucefécit presented concentrations of BOD (Zebro: 4.0-35.5 mg $L^{-1;}$ Lucefécit: 2.3-7.5 mg L^{-1}) and total phosphorus (Zebro: 0.18-6.23 mg $L^{-1;}$ Lucefécit: 0.02-1.92 mg L^{-1}) that compromise the support of biological life, with regard to nutrient and oxygenation conditions. As regards pesticides, the concentrations detected were low, being bentazone the compound quantified at highest levels. Lucefécit was the tributary that presented higher concentrations of pesticides (with values of bentazone of 1.94 µg L^{-1}), probably due to the intensive agriculture crops around it. Concerning to ecotoxicological analysis, the results highlighted the toxicity in sublethal parameters (reproduction, feed inhibition or growth inhibition) induced, mainly, by samples from Zebro and Lucefécit streams. So, the results from the ecotoxicological tool-box allowed identifying the streams that promote a higher chemical impact to the reservoir, which is essential to delineate management actions to improve the water quality of the reservoir.

TU314

Effects based tools for use in conjunction with passive samplers

R.J. Brown, O.L. Tran, wca consulting; G. Whale, Shell Health / Risk Science Team; M.J. Spence, CONCAWE; D. Leverett, wca

As part of an ongoing review of the Water Framework Directive (WFD), the European Commission (EC) is considering "effects based tools" (EBTs) for use as an alternative to, or in combination with, the monitoring of individual substance concentrations. There are several ways in which EBTs may be integrated into environmental monitoring but one approach, which integrates chemical exposure over time, is to conduct toxicity profiling on extracts from passive samplers deployed in surface waters. The aim of this project was to provide a critical evaluation of available EBTs that could be used in conjunction with passive samplers, and propose a list of assays for use in monitoring surface waters associated with the oil and gas industry. A list of possible EBTs was compiled based on recent published reviews on this topic. These assays were then broadly screened based on commercial availability, general validation maturity, previous application to environmental samples, and suitability for use with passive sampler extracts to derive a short list of 22 assays for more detailed consideration. The short-listed assays included novel whole organism bioassays (or surrogates), and *in vitro* or bacterial assays for endpoints based on endocrine disruption (oestrogen, androgen and thyroid), genotoxicity, oxidative stress, and metabolism of polyaromatic hydrocarbons (PAHs). Commonly used whole-organism assays (e.g. acute invertebrate, algae and fish tests) were not subject to this screening, since they are already well proven and no detailed evaluation was required, however, they were considered as part of the final recommendations. The shortlisted EBTs were then subject to a detailed review, based on the published scientific literature, to identify relevant information with respect to their performance, interpretation, and application. The EBTs were compared using the information identified in the literature reviews and an initial suite of thirteen bioassays were recommended for the monitoring of surface waters associated with refinery effluents using passive sampler extracts. This recommended suite of EBTs should be considered a starting point for use in the monitoring of waters receiving refinery effluents, to be further developed based on experience in using the assays for this purpose, and incorporating new relevant bioassays once they have achieved a sufficient level of validation maturity. This review will be published as a Concawe report in 2018.

TU315

Innovative ecotoxicological monitoring strategies for the protection of aquatic ecosystems and the implementation of the Water Framework Directive (WFD)

W. Cristiano, Institute for Environmental Research, RWTH Aachen / Department of Ecosystem Analysis; H. Hollert, RWTH Aachen University / Institute for Environmental Research; C. Di Paolo, RWTH Aachen University / Ecosystem Analysis (ESA); I. Lacchetti, Istituto Superiore di Sanità / Environment and Health; L. Mancini, Italian Institute of Health (ISS) / Department of Environment and Health; M. Carere, Italian Institute of Health ISS

The Water Framework Directive (WFD, 2000/60/EC) regulates the European water policy and addresses the EU member states to achieve good qualitative and quantitative status of all water bodies. Despite the efforts to reduce the release of chemicals into the aquatic environments, pollution is still widespread across Europe, and new emerging substances should be assessed and managed. The general goal of this project (realised in the framework of the '*Torno subito*' Lazio Operational Programme European Social Funding 2014-2020) is to select and define innovative methods to assess the toxicity due to the exposure to different pollutants, especially the emerging substances and respective mixtures, with a focus on aquatic ecosystems and human health. This goal has been achieved, in a first step, by making a literature review on the priority and emerging substances widespread in the aquatic environment, to investigate their effects on the development of zebrafish (*Danio rerio*) embryos. Then, a few toxic substances that are relevant for our goals have been selected and analysed through the fish embryo acute toxicity test (FET) and other assays; particular attention has been given to the sub-lethal effects. Afterwards, environmental samples from different aquatic systems in Italy will be analysed to detect the chemicals present in these sites as well as their toxic effects. In order to reach a better comprehension of the effects of such substances on the ecosystems, bioassays with other organisms from different trophic levels (bacteria algae, daphnids) will also be performed. The study will ultimately aim to provide recommendations for the implementation and the update of the monitoring strategies of the WFD, as well as to enhance the current EU activity on Effect-Based Methods.

TU316

Chemical and Ecotoxicological Monitoring of a marine coastal area in the Central Italy

M. Carere, Italian Institute of Health ISS; I. Lacchetti, Istituto Superiore di Sanità / Environment and Health; R. Scenati, L. Lucentini, M. Beccaloni, Italian Institute of Health ISS / Environment and Health; P.M. Gucci, Italian Institute of Health ISS / Environmental and Health; F. Falleni, Italian Institute of Health ISS / Environment and Health; C. Gavasso, Lega Navale; M. Cicero, Italian Institute of Health ISS; G. Marsili, Osservatorio Ambientale di Civitavecchia; L. Musmeci, Italian Institute of Health ISS / Environment and Health; V. Frau, L. Zingari, Consorzio Ambientale di Civitavecchia; E. Veschetti, Italian Institute of Health ISS

A monitoring campaign has been performed in Central Italy with the aim to characterize the chemical quality status of the coastal marine area in order to detect the possible impact of the emissions of a Coal fired power station and other sources of pollution in proximity of the city of Civitavecchia. The sampling has been carried out in two different seasons of the year along the marine coastal area and in a transitional surface waterbody (Saline di Tarquinia). The analysis has been performed in the water column and in the first 20 centimeters of the sediments. The chemical substances analyzed included several priority substances of the WFD (water framework directive) and other chemical substances: Metals, Dioxins, PCB, PAH, Naphtalene. The Ecotoxicological assays have been performed with the use of algae (*Phaeodactylum tricornutum)* and crustaceans (*Artemia franciscana* and *Tigriopus fulvus*). The results have showed a diffuse light exceedance of the sediment environmental quality standards of the Italian legislation for some metals (e.g. arsenic, lead, chromium, mercury) and naphthalene; the data of the water column are in general below the environmental quality standards, but Uranium has been detected in surface water samples at concentrations above the available PNEC (predicted no effect concentration). Ecotoxicological effects have been detected with the algae and could be related to the substances detected (e.g. heavy metals) or other substances released in the area (transitional waterbody). In general the results show a situation in which the quality of the sediments is not in a good status, although the level of concentrations should not cause a high risk for the aquatic ecosystems; the chemical contaminants can derive from different sources of pollution (industrial, urban, agricultural, atmospheric deposition) of the area and management measures should be reinforced in order to achieve the good quality status required by the water framework directive. The presence of Uranium in the marine coastal area should be further investigated to understand the possible role of the Coal fired power station.

TU317

USE OF DIAGNOSTIC STRAINS OF THE SALMONELLA/MICROSOME ASSAY FOR THE IDENTIFICATION OF MUTAGENIC PROFILES IN WATER SAMPLES AND SUSPENDED PARTICULATE MATTER

D.A. Morales, State University of Campinas / Faculty of Technology; J. Rossetto Martins Zwarg, School of Technology, UNICAMP; R. Massei, Helmholtz Centre for Environmental Research   UFZ; T. Schulze, M. Krauss, W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; G. Umbuzeiro, School of Technology, UNICAMP / LAEG

The Salmonella/microsome mutagenicity assay uses the genetically modified Salmonella *enterica* serovar Typhimurium bacterium, and the exploration of these different genetic characteristics allows the detection of various classes of mutagenic compounds. The objective of this study was to use different strains for the identification of mutagenic profiles and hinting at the class of compounds responsible for the detected mutagenicity. The research was performed with organic extracts of surface water and suspended particulate matter (SPM) collected in 13 different sites along the Danube River during the Joint Danube Survey 3 (JDS3). The extracts were evaluated with the Salmonella/microsome microsuspension assay with and without metabolic acrivation (S9). The strains used were: TA98, YG1041, TA1538 and YG5185 with specific genetic characteristics for the detection of different compounds. A total of 69% surface water and 92% SPM samples were positive for at least one strain/condition. The applied methodology compared the profiles obtained at different sites along the Danube River. In this way, it was possible to identify different sources/types of compounds or mixtures that are causing mutagenicity. We conclude that aromatic amines and nitro compounds for surface water and also polycyclic aromatic hydrocarbons for suspended particulate matter appear to be responsible for mutagenicity at some of the sites, because the obtained profile were similarly representative compounds of these classes. Other sites indicate the presence of other types of compounds or mixtures that need to be investigated. Non-target chemical analysis is currently carried out to test if it is possible to correlate the observed profiles with individual compounds or groups of chemicals. **ACKNOWLEDGMENTS** The authors thank FAPESP Project 2013/16956-6. José Ricardo R. M. Zwarg thanks FAPESP Project 2015/11399-7 for the IC scholarship. Daniel A. Morales thanks CAPES for the PhD scholarship. The SOLUTIONS project has received funding from the European Unions Seventh Framework Programme for Research, Technological Development and Demonstration under grant agreement no. 603437.

TU318

NTA meets EDA: A practical example

J. Kuinke, V. Hinnenkamp, P. Balsaa, A. Simon, IWW Rheinisch-Westfälisches Institut für Wasserforschung gGmbH; T.C. Schmidt, University of DuisburgEssen

Organic micropollutants play an important role in the assessment of water bodies that are used for drinking water production. On one hand, micropollutants pass through the wastewater system in sewage treatment plants and subsequently in surface water. On the other hand, there are direct discharges from industry and in addition, there are diffuse sources from agriculture, or from households, buildings and settlements. As major pollution events (i.e. PFC in the river Ruhr) show, water suppliers must always expect to find new critical substances that could pose a potential health risk in drinking water, especially when using surface water. For this reason, it is necessary to initiate a proactive screening of contaminants and their potential effects. Continuous monitoring by high-resolution mass spectrometry also makes it possible to analyze water pollution retrospectively, gain knowledge about temporal dynamics and discharge patterns and thus identify the source of the contamination more frequently. In this context, a fast, robust and routine method for the determination of organic micropollutants is needed. This project is an approach to analyze organic micropollutants in water samples with a combination of non-target-analysis (NTA) and effect-directed-analysis (EDA). Samples were taken regularly over a period of one year in order to obtain an annual progression of the water pollution. A LC IMS QTOF system was used to carry out the NTA. Different endpoints were analyzed for the EDA: cytotoxicity (MTT assay), endocrine activities (ER-CALUX and AR-CALUX) as well as genotoxicity (p53-CALUX, umuC assay and Ames assay). Due to the investigation of raw water samples, no significant biological effect of the individual samples was to be expected. The focus of this project was therefore on the identification of seasonal exposure patterns of the micropollutant load. The coupling of NTA with a test strategy for toxicological effects forms an innovative approach with potential for preventive product quality assurance of the water supplier.

TU319

Imposex levels in gastropods from the Northern Adriatic Sea (Italy): a proposal of classification according to the Water Framework Directive

F. Cacciatore, ISPRA-Institute for Environmental Protection and Research / Mitigation and Prevention of Impacts; R. Boscolo Brusà, C. Antonini, M. Formalewicz, ISPRA - Institute for Environmental Protection and Research; M. Marin, University of Padua; A. Bonometto, M. Gabellini, ISPRA - Institute for Environmental Protection and Research

Butyltins (BTs) - i.e. *mono*- (MBT), di- (DBT) and tributyltin (TBT) - are synthetic compounds worldwide used in industrial and agricultural applications giving rise to contamination of aquatic environments. Organotins, which include BTs, were banned on antifouling paints, the main route to aquatic pollution, by Reg. 782/2003/EC. Presently, due to their persistence, toxicity and bioaccumulative properties, TBT compounds are included among the priority hazardous substances according to the European Water Framework Directive (WFD) and its daughter Directive 2008/105/EC. Imposex, the superimposition of male sexual characteristics on females of gonochoristic gastropods, is the most studied effect of TBT exposure and it is generally recognized as a specific response to organic tin compounds. For this reason, under the implementation of the WFD, imposex measurements have been indicated as a useful tool to link chemical and ecological status assessment. In this context, the aim of this study was to propose a classification of imposex according to WFD using two species of gastropods collected in the Northern Adriatic Sea and in the Venice Lagoon: *Nassarius nitidus* (Jeffreys, 1867) and *Hexaplex trunculus* (Linnaeus, 1758). The first species, less sensitive to BT pollution, can be found in the inner parts of the lagoon, whereas the latter, more sensitive, occurs only near the lagoon inlets or in the sea. To define Ecological Quality Ratio (EQR) class boundaries within WFD, the relationship between the ecological impact caused by BT pollution and the reproductive capacity of the gastropod populations was assessed. This preliminary attempt showed that most of the sites were in Bad ecological status before the ban and have reached mostly a Moderate status at present, with very few sites in Good or High status. A comparison between the two species was also performed showing advantages coming from the combined use of both species to cope with the ecological quality assessment in a wider study area.

TU320

Lessons Learned from Sibro Dam and River Restoration in Sweden

E. Hällqvist, C. Becker, P. Bönlökke Adamsen, P. Gliveson, A. Sahlén, Ramboll

Aquatic ecosystems in the European Union are under pressure from growing demand for\nsufficient quantities of good quality water for human use. The Water Framework Directive\naims to protect and improve water quality and aquatic ecology; environmental quality\nstandards (EQS) have been established as legal tools with which to set requirements for\nmember states. In Sweden, all major surface waters are classified according to the current\nstatus of the water designated by authorities in the respective water district. The ecological\nstatus of surface water comprises three different types of quality factors according to the\nframework - biological, physicochemical and hydro-morphological. The latter defines\nconnectivity and biodiversity in the ecosystem, since many aquatic organisms are\ndependent on the ability to migrate during their life cycle. Water power represents a large\nfraction (almost 50%) of electricity production within the country, and a large proportion of\nSwedish rivers are affected hydro-morphologically. At present, there are 11,000 active and\nabandoned dams in Swedish rivers, and 1,800 are hydroelectric power plants. All of these\ndams impact the ecological connectivity of rivers and have a negative impact on\nbiodiversity. In Sweden, a common national strategy is to increase the use of hydropower\nplants as an alternative to reliance on fossil fuels. In the same time water power is the\ngreates individual cause of physical impacts in lakes and streams. The challenge at this\nearly stage of Sweden’s national energy strategy is to identify technologies and\nmanagement practices that promote hydroelectric power with minimal long-term adverse\necological impacts. To illustrate the challenges, this paper summarizes work conducted\nover the past 2 years to manage the future of the Sibro Dam located in southern-central\nSweden. The project was initiated after previous dam repair work involving the diversion of\na large reach of the Sibro River resulted in serious consequences for nationally protected\nindigenous mussels and other aquatic life. The responsible municipality is obligated to\nimprove ecological connectivity at Sibro Dam and regulation of Lake Båven. The planning\nwork included preparation of an environmental impact assessment (EIA), detailed\nengineering design for fish passage, engagement with local communities and\ncommunications between the municipality of Nyköping and Sweden’s federal court.\nKeywords\nFish passage; Sweden; ecological connectivity; environmental impact assessment

Behavioural Ecotoxicology: Unravelling behavioural responses to chemical contaminants in the environment (P)

TU321

Impacts of methylmercury on growth, respiration and swimming in larvae of a marine forage fish

x. ye, Stony Brook University / School of Marine and Atmospheric Sciences; N. Fisher, SoMAS @ Stony Brook University / SoMAS

Marine and estuarine fish accumulate methylmercury (MeHg) to elevated concentrations, often higher than in freshwater systems. Because MeHg is a neurotoxin, it is plausible that high tissue concentrations could affect behavior in marine fish which in turn could affect their populations in contaminated waters. Here we examined sublethal effects of MeHg to a marine forage fish at the larval stage, the Sheepshead minnow *Cyprinodon variegatus*. Because the bioavailability of MeHg from different food types may lead to different MeHg internal distributions and toxic effects, we compared artificial and natural diets with varying MeHg concentrations. Artificial (commercial fish flakes containing methylmercury) or natural diets (zooplankton containing MeHg, obtained from MeHg-contaminated phytoplankton) were prepared; MeHg concentrations ranged from zero (controls) to as high as 7.8 ppm. The larvae were fed control and MeHg-contaminated diets from an age of 7 days until 5 weeks when they reached juvenile stage. Growth rates, respiration rates, and swimming activity were tested. Results indicate that MeHg-rich diets—either artificial or natural foods--have no significant impact on fish growth rates under any treatment. However swimming activity (swimming speed, acceleration, active time and swimming distance) was impaired after 3 weeks exposure to natural diets containing 7.8 ppm but not 2.5 ppm; artificial diets containing MeHg up to 4.8 ppm had no discernible effect on swimming. In addition, MeHg as low as 4.8 ppm had a small but significant impact on the respiration rates of these fish. The data suggest that mortality and growth are unlikely to be affected by these MeHg concentrations, but swimming activity may be reduced at elevated MeHg concentrations, and this could influence the success of populations in the wild through impairment of predation or avoidance of predators.

TU322

Comparability of Zebrafish Embryo Behavioral Assays: A Need for Standardization of Experimental Factors

A.O. Ogungbemi, Helmholtz centre for environmental research - UFZ / Bioanalytical Ecotoxicology; D. Leuthold, Helmholtz Centre for Environmental Research - UFZ GmbH / Bioanalytical Ecotoxicology; S. Scholz, Helmholtz Centre for Environmental Research / Department Bioanalytical Ecotoxicology; E. Küster, Helmholtz Centre for Environmental Research, Dept.Bioanalytical Ecotoxicology / Department Bioanalytical Ecotoxicology

In recent times, zebrafish embryos have gained wide acceptance as an alternative test model for drug development and toxicity testing. In particular, the behavioral response of zebrafish embryos is a useful endpoint to detect neurotoxic and neuroactive chemicals. Consequently, several behavioral test methods have been developed including photomotor response test (PMR), locomotor response test (LMR), spontaneous tail contraction test (STC) and touch evoked response test (TER). Although these methods are distinct in their application, most of their experimental parameters lack consistency in protocols such as exposure time, imaging time, age of exposure, endpoint parameter, statistical analysis etc. Therefore, there is a need to standardize these methods in order to enable comparability of test results, as well as, to ensure accurate prediction of chemical activity in zebrafish. To initiate this standardization process, we embarked on a meta-analysis of existing behavioral assays to ask these questions: 1.) Are there consistencies in hypo/hyper behavioral activity of zebrafish embryos when different assays are used? 2.) Despite lack of standardization, is it possible to aggregate the data from different assays to give useful behavioral activity? 3.) Is it possible to determine which experimental parameters are most influential for the behavioral assays? Based on the meta-analysis, we conclude that, results from different behavioral assays (LMR, PMR and STC) are consistent with the predicted activity of a chemical. Even though, effect concentrations vary to some extent among the considered behavioral assays, most of the variability could be explained by the most influential parameters including: exposure time, age at exposure and concentration range. These results can be useful to identify the most important experimental factors in an effort to standardize behavioral assays for toxicity testing.

TU323

Effects of 17α-ethynylestradiol (EE2) on social behaviors of the false clown anemonefish (Amphiprion ocellaris)

T. Chen, National Museum of Marine Biology and Aquarium / Department of Biology; C. Lu, National Dong Hwa University / Institute of Marine Biology; C. Chen, National Taiwan University / Institute of Environmental Health

The synthetic estrogen 17α-ethinylestradiol (EE2) is extensively used in oral contraceptive pills, medication, cosmetics, and personal care products. It is also widely used in livestock and aquaculture. EE2 enters aquatic ecosystems via wastewater discharges and effluents of sewage treatment plants. EE2 is commonly detected in wastewater effluents and surface waters including coastal water. Although coastal regions are often impacted by sewage discharges, no study has been done to address the effect of environmental estrogens such as EE2 in coral reef fish. Agonistic behavior is crucial for maintaining social hierarchy in many coral reef fish. Endocrine disrupting contaminants such as EE2 may interfere fish social structure via disrupting their agonistic behavior. In this study, we aimed to use the false clown anemonefish (*Amphiprion ocellaris*) as an experimental model to characterize endocrine disrupting effects of EE2 in coral reef fish, with an emphasis on social behaviors. For the exposure experiment, the fish were randomly distributed to separate tanks to form small colonies consisting of three individuals and were exposed to an environmental concentration of EE2 (30 ng/L) for 4 weeks. During this period, social behaviors including agonistic behavior, submissive response, and shelter utilization were videotaped and quantitatively analyzed once a week. Our results show that growth and survival were significantly affected by social hierarchy rather than the EE2 treatment. Social hierarchy was not altered, but social behaviors of the middle-ranked fish were significantly affected by EE2, suggesting that EE2 may cause different impact in different ranks.

TU324

Impacts of environmentally realistic antidepressant exposure on reproductive behaviour and sperm traits in fish

J.M. Martin, M. Saaristo, Monash University / School of Biological Sciences; M.G. Bertram, Monash University / Biological Sciences; S. Hannington, J. Tanner, Monash University / School of Biological Sciences, Monash University, Victoria, Australia; M. O'Bryan, Monash University / The Development and Stem Cells Program of Monash Biomedicine Discovery Institute and the Department of Anatomy and Developmental Biology, Monash University, Victoria, Australia; B.B. Wong, Monash University / School of Biological Sciences

Pharmaceutical contaminants are increasingly being detected in ecosystems worldwide. Indeed, more than 1 in 10 currently manufactured pharmaceuticals have been found in the environment. One pharmaceutical pollutant of environmental concern is the antidepressant fluoxetine, which has repeatedly been reported in aquatic ecosystems. Worryingly, the primary target molecule of fluoxetine is conserved across a wide range of non-target species-. As a result, by directly acting on the central nervous system and neuroendocrine pathways, fluoxetine can affect a range of ecologically important behavioural and physiological processes in wildlife. Despite this, the effects of environmentally relevant fluoxetine exposure on processes of sexual selection in aquatic biota remain uncertain. This is concerning as sexual selection processes directly influence mating outcomes and so are fundamental to individual fitness, as well as the viability of populations and species. To address this knowledge gap, we investigated the impact of 30-day exposure two environmentally realistic levels of fluoxetine (average measured concentrations: 30 and 380 ng/L) on a range of reproductive behaviours, as well as sperm quality, in the eastern mosquitofish (*Gambusia holbrooki*), a promiscuous freshwater fish with internal fertilisation. We focussed on these traits because reproductive behaviour and sperm quality are both crucial fitness determinants, and are known to be vulnerable to disruption by other chemical pollutant classes. We found that fluoxetine exposure impacts reproductive behaviour in fish at field-detected concentrations, altering both association time and copulatory behaviour carried out by males towards females. Fluoxetine exposure, however, did not significantly impact sperm quality measures (i.e. performance and viability). In combination, our results indicate that fluoxetine exposure can alter reproductive behaviours with direct bearing on fitness in fish and, further, highlight the need for ecotoxicological testing using sub-lethal exposure concentrations and ecologically important behavioural endpoints.

TU325

Determining the effects of antidepressants on multiple behaviours in a marine and freshwater amphipod

S.A. Kohler, University of Portsmouth / Animal Physiological Ecology; A. Ford, University of Portsmouth / Biological Sciences; M.O. Parker, University of Portsmouth

Behavioural assays have been gaining recognition as a viable endpoint in ecotoxicology as they provide a link between biochemical and ecological effects of environmental contaminants. Psychotropic drugs are designed to modulate behaviours in humans, and preclinical studies have demonstrated that these compounds can also alter behaviours in aquatic vertebrates. The effects of behavioural modulating drugs have been tested from a pharmacological discipline using anxiety-like behaviours including thigmotaxis (wall hugging) and scototaxis (light avoidance) on mice and zebrafish, using well-defined behavioural assays. These pharmacological methods have been translated to ecotoxicological studies on vertebrates but comparatively few have been done on invertebrate species. This ongoing study aims to translate these techniques to model crustaceans for the purpose of assessment of environmental risk using the antidepressant fluoxetine as a model compound. Specimens of the marine amphipod, *Echinogammarus marinus* and the freshwater amphipod *Gammarus pulex* were exposed to environmentally relevant concentrations of fluoxetine from 0.001-1 μg/L during 1 day, 1 week, and 2 week exposures. Activity was measured as swimming velocity and choice experiments were used to determine phototactic and thigmotactic response. Both *E. marinus* and *G. pulex* showed alterations in activity at concentrations as low as 1ng/L and as soon as 1 day compared to controls (P=< 0.05). Significant differences were observed in thigmoactic and phototactic behaviours both between treatments and with length of exposure. Results indicate that fluoxetine can have an effect on some amphipod behaviours at environmentally relevant concentrations. These results may have implications for future study design of these types of experiments and aid the development of high-throughput analysis on common laboratory invertebrate species.

TU326

Inter-species variability in the behaviour of a marine and freshwater amphipod

S.A. Kohler, University of Portsmouth / Animal Physiological Ecology; A. Ford, University of Portsmouth / Biological Sciences; M.O. Parker, University of Portsmouth

Invertebrate species are used in standard ecotoxicology testing to assess environmental effects of contaminants. However standardised invertebrate models are limited to relatively few species. Behavioural ecotoxicology is expanding with techniques and endpoints used in pharmacology being translated to other vertebrate and invertebrate species for use in ecotoxicology. Despite this, data on the control behaviours of model organisms such as crustaceans, and the inter-species variability in behaviours are currently under-studied. The aims of this study were to provide control data on a range of behaviours for use in ecotoxicological testing, using amphipods as model organisms. Behaviours commonly associated with anxiety in pharmacological studies including activity, phototaxis (light/dark preference) and thigmotaxis (wall hugging) were assessed in the marine amphipod *Echinogammarus marinus* and the freshwater amphipod *Gammarus pulex* using choice assays. Both organisms exhibited negative phototactic and positive thigmotactic behaviours (P=< 0.001 respectively) however, differences in sensitivity to these assays were observed between species. *E. marinus* showed a significantly greater sensitivity to the phototaxis assay than *G. pulex* (P=< 0.001), while the reverse was found for the thigmotaxis assay (P=< 0.001). Swimming velocity was used as a measure of activity. Significant differences were observed in swimming behaviours between species when exposed to a light stimulus (P=< 0.001) which may be attributed to differences in life histories between the two species. The results of this study provide evidence of phototactic and thigmotactic behaviours in two model crustacean species and describes two behavioural assays with potential for use in behavioural ecotoxicology. In this study we demonstrate that closely related species are capable of very different behavioural responses. The inter-species variability in sensitivity to behavioural assays found between the two amphipods in this study highlights the importance of control data on your model species for behavioural studies.

TU327

Physiological basis of individual tolerance to the benzodiazepine oxazepam in zebrafish (Danio rerio)

L. Vossen, Uppsala University / Department of Neuroscience; J. Fick, Umea University / Department of Chemistry; T. Brodin, Umea University / Department of Ecology and Environmental Science; S. Winberg, Uppsala University / Department of Neuroscience

Pharmaceuticals are common contaminants in aquatic ecosystems. Among the most prescribed pharmaceuticals globally are the benzodiazepines (e.g. Valium), a class of psychoactive drugs used to treat anxiety and induce sedation. Benzodiazepines are persistent in the environment, and their target, the GABA-A receptor, is evolutionarily conserved throughout the vertebrates. Behavioural changes have been described for juvenile Eurasian perch (*Perca fluviatilis*) and Fathead minnows (*Pimephales promelas*) at environmental concentrations. We recently found that also wild-caught zebrafish (*Danio rerio*) show reduced fear responses after 7 days of exposure to the benzodiazepine oxazepam (1, 10 or 100 microgram per Liter). Intriguingly, fear responses were partially restored after 28 days of exposure. Here we analyse the physiological and genetic basis of this tolerance to oxazepam, including peak cortisol levels in response to a stressor, concentrations of monoamine neurotransmitters in brain as well as mRNA expression of brain $GABA_A$ receptor subunits and mRNA expression of liver enzymes involved in the metabolism of oxazepam. We then correlate these measures of physiological and genetic tolerance with the individual's behavioural tolerance. The results will shed light on the potential for inter-individual variation in oxazepam tolerance to mitigate the effects of benzodiazepine pollution.

TU328

Reversible behavioural alterations in burbot, Lota lota, from exposure to environmentally relevant levels of oxazepam

J. Sundin, Norwegian University of Science and Technology / Department of Neuroscience; F. Jutfelt, Norwegian University of Science and Technology / Department of Biology; J. Fick, Umea University / Department of Chemistry; M. Thorlacius, Marine and Freshwater Institute; T. Brodin, Umea University / Department of Ecology and Environmental Science

Benzodiazepines are frequently detected in the environment. They persist in wastewater effluent and can be found at high concentrations in treated effluent. Furthermore, several benzodiazepines are resistant to photodegradation, enabling them to persist in the environment. Benzodiazepines are designed to alter human behaviour by binding to GABA-receptors, which are found in a wide range of animals including all vertebrates. We investigated the effect of the benzodiazepine oxazepam on behaviour using the burbot, *Lota lota*. We found that high levels of oxazepam affected swimming activity, diurnal as well as nocturnal, while the environmentally relevant level had no detectable effect. There was also an effect on boldness, with fish exposed to high levels of oxazepam spending more time hiding than the control- and low level fish. Interestingly, the effects of high oxazepam were no longer detectible when the fish were tested again after being kept in water without drugs for five days. Our results suggest that effects of pharmaceuticals may be reversible, if the exposure duration is relatively short and the animal have the possibility to move to uncontaminated water.

TU329

Behavioural endpoints and biochemical biomarkers as tools to investigate effects of citalopram in brown trout (Salmo trutta f. fario)

M. Ziegler, University of Tubingen / Animal Physiological Ecology; S. Tisler, University of Tuebingen / Environmental Analytical Chemistry; L. Reinelt, University of Tubingen / Animal Physiological Ecology; R. Triebskorn, University of Tuebingen / Animal Physiological Ecology

Citalopram (CIT) is a selective serotonin reuptake inhibitor (SSRI) which is commonly used as an anti-depressant drug. It binds to proteins responsible for the serotonin reuptake from the synaptic cleft and thereby inhibits the reuptake of serotonin into the pre-synapse. Due to high consumption rates and moderate elimination during wastewater treatment, CIT is one of the most abundant SSRIs in surface water. Several studies showed that environmentally relevant concentrations of 1 µg/L may affect aquatic organisms. The aim of this study is to investigate effects of CIT in different life stages of brown trout (*Salmo trutta* f. *fario*) with focus on development, behaviour and individual health. Both, eggs of fish in the eyed ova stage and 8 months old juveniles were chronically exposed to four concentrations of CIT (1, 10, 100 and 1000 µg/L) in a in a semi-static three-block design accompanied by a control exposure. The larvae were exposed for 5 month at 7°C and 11°C, the experiments with juvenile fish were conducted for 4 weeks at 7°C. To investigate the effects of CIT on the embryonic development, mortality, hatching rate, and heartbeat rate were recorded. During the exposure, also behavioural endpoints were observed. Besides, several biomarkers indicative for fish health were investigated, such as cortisol-level, acetylcholinesterase activity, hsp70-level and the histological condition of the liver. After exposure to 1000 µg/L CIT, length and weight of both larvae and juveniles were significantly reduced. Furthermore both stages showed an enhanced swimming activity and an increased swim up in the aquaria. In an artificial swimming measurement device (small aquaria with a diameter of 17 cm), videos were recorded for 20 minutes with the aim to quantify changes in the swimming behaviour. Due to the settings of the cameras, the aquaria were strongly illuminated and the water was not ventilated during the recording. This stress situation resulted in a high activity of fish except for those exposed to 1000 µg/L CIT. These moved significantly less with a lower velocity than the control fish. The results of both experiments make evident that 1000 µg/L CIT affects both larvae and juvenile brown trout, on one hand by making them more agile in the aquaria, but also by depressing stress-induced flurry swimming. The study is embedded in the Effect-Net (effect network in water research) Project which is funded by the Wassernetzwerk Baden-Württemberg.

TU330

Assessing the direct and indirect effects of chemical contaminants on the behaviour, ecology and evolution of wildlife: A conceptual framework

K. Arnold, University of York / Environment; M. Saaristo, Monash University / School of Biological Sciences; T. Brodin, Umea University / Department of Ecology and Environmental Science

Chemical contaminants, e.g. metals, pharmaceuticals, pesticides, are changing ecosystems via effects on wildlife. Most studies examine a limited range of endpoints, species and/or contaminants under laboratory conditions. Recent work explicitly based in ecological realism, however, reveals that chemical contaminants have direct and indirect effects at multiple levels of organisation by affecting behavioural responses of animals. Given that behaviour reflects multiple physiological changes at low contaminant concentrations, and links individual- to population-level processes, it provides a sensitive tool for holistically assessing contaminant impacts. Here, we develop a conceptual framework that integrates direct and indirect effects of chemical contaminants on behaviour, under environmentally relevant concentrations and natural contexts. Within our framework, we show how the consequences of contaminants can extend beyond individuals. We use altered predator-prey interactions to demonstrate cascading contaminant effects through communities, exerting both positive and negative effects on distinct populations. Moreover, contaminants can be potent evolutionary forces selecting directly for compensatory behaviours or indirectly on downstream behaviours, via selection on resistance genes. To help implement our framework, we supply tools to design ecologically realistic experiments and risk-assessments. Although predicting effects of contaminants is complex, existing knowledge in ecology and evolution needs to be applied to this global environmental challenge.

TU331

Scent and sensibility: EE2 disrupts male mate choice in fish

M. Saaristo, C.P. Johnstone, Monash University / School of Biological Sciences; K. Xu, University of Alberta / Department of Renewable Resources; M. Allinson, The University of Melbourne / School of Chemistry; B.B. Wong, Monash University / School of Biological Sciences

Among the handful of studies that have studied the behavioural effects of endocrine disrupting chemicals (EDCs), only a few have attempted to disentangle the mechanisms underlying behavioural changes, such as mate choice. In fish, for example, ecological studies have shown that males base their mate choice on multiple cues and both visual and chemical cues play an important role in choosing the most suitable mate. Therefore, it is crucial to understand if and how EDCs affect mate choice cues (e.g. visual and chemical cues), and further, if one cue is affected disproportionately. Accordingly, the aim of this study was to investigate the impacts of a 28-day exposure to 17α-ethinyl estradiol EE2 (measured concentration 12ng/L) - a synthetic estrogen used in the contraceptive pill and a widespread contaminant of aquatic systems - on visual and chemical communication in the guppy. To examine the impact of EE2 on male mate choice, we ran a standard choice assay, which was conducted in two parts to disentangle visual cues from chemical cues. First, we allowed a single male (either control or EE2) to court two size-matched females (one control and one EE2-female). In this visual cue experiment, the male was only able to see the females, but not to smell them. Second, we introduced chemical cues (control and EE2-female) to the trial tank paired randomly with the females. We found that there was no significant effect of EE2-treatment on total time males spent associating with the females, when given only visual cues. There was, however, a significant effect on courtship 'sigmoid' display with both control and EE2-exposed males spending more time performing sigmoid displays for control females compared to EE2-exposed females. When males were presented with both visual and chemical female cues, males (control and EE2) entered the association zone more frequently, if EE2-exposed female was paired with an EE2-chemical cue. In contrast, sigmoid display showed a reverse pattern, with males preferring EE2-exposed females that were paired with control chemical cues. Not only does our study uncover a previously unknown behavioural impact of EE2-exposure on chemical cues, but also raises the possibility that EE2-exposure may have mixed and complex effects on mate attractiveness. Our results underscore the importance of studying multiple mate choice cues simultaneously, and highlights the possible ecological implications of altered chemical communication for exposed wildlife.

TU332

Effects of tributyltin on the eyes, swimming, feeding and growth of newborn guppies Poecilia vivipara

D.V. Paulo, C.F. Mariz Jr, M.K. Alves, R.M. Barata, UFPE Universidade Federal de Pernambuco / Department of Zoology; R.N. Alves, UFPE Universidade Federal de Pernambuco / Zoology; P.S. Carvalho, UFPE - Universidade Federal de Pernambuco / Zoología

Although the use of the antifouling contaminant tributyltin (TBT) has been banned since 2008 by the International Maritime Organization, it still persists in coastal environments due to its remobilization from contaminated sediments and also as a result of illegal use, including tropical regions along the Brazilian Atlantic Coast. *Poecilia vivipara* is a promising model for tropical estuarine fish ecotoxicological studies, and we focus here on its feasibility to address fish early life stage toxicity caused by TBT. Newborn *Poecilia vivipara* fish at six days after birth (dab) obtained from a laboratory breeding stock were exposed for 96h to waterborne tributyltin at 0.1; 1.0; 4.5; 7 and 9 µg TBT L^{-1}, plus controls and solvent controls. After exposure, we evaluated swimming speeds and trajectories of the fish, counter-current swimming resistance, ability to capture Artemia nauplii, growth in weight and length, and histology of the eyes and retina. Macroscopic analysis of the eyes showed a darkening of the iris region after exposure to 4.5; 7 and 9 µg TBT L^{-1}. Histopathological analysis of the retinal pigment epithelium (RPE) indicated a hyperpigmentation of the pigment epithelium villi and basal region in TBT exposed fish. In addition to these alterations, RPE invaginations, photoreceptor degeneration, iris epithelial cell atrophy and iris melanin condensation were observed. After exposure to 7 µg TBT L^{-1}, swimming speed, swimming resistance, daily capture of Artemia nauplii and growth in weight were reduced by 85%, 60%, 33.6% and 56% relative to controls, respectively. The histopathological changes detected in the retina and iris may have reduced the fish visual exploration and prey detection capacity, which together with the detected effects in swimming endpoints might have led to a deficiency in prey capture and growth. These changes can reduce the chances of exposed fish to recruit to the adult population.

TU333

Chemosensory behavioral reactions of zebrafish larvae to environmental contaminants

S.K. Könemann, C.M. vom Berg, Eawag Swiss Federal Institute of Aquatic Science and Technology / Environmental Toxicology

Background: Selecting an appropriate behavioral response to a potential rewarding or threatening stimulus is critical for the survival of an animal. Therefore, organisms possess an innate ability to react to threatening and rewarding situations they are naturally exposed to. However, the ability to naturally respond to environmental cues might be severely affected by anthropogenic activity and behavioral outcomes become unpredictable. In particular, neuroactive and psychoactive substances in the aquatic environment released by wastewater treatment plants or agricultural run-offs might potentially change the perception and interpretation of natural cues by aquatic organisms, especially fish. **Aim:** We are investigating whether environmental contaminants lead to attractive or aversive responses in fish, and are additionally interested in the neuronal mechanism underlying the observed behavioral response. We aim to better understand how environmental contaminants change natural behavioral responses of fish in order to better predict their impact on the ecosystem. **Methods:** We are using zebrafish larvae as a model organism, because they are amenable for behavioral analysis and mechanistic dissection of complex processes. Larvae are exposed to a point source of test chemical at different concentrations and the behavior is tracked with an automated video recording system. Various parameters such as the larva's space use, locomotor activity and velocity are evaluated. Active neuronal regions are detected by staining the larvae for an endogenous activity indicator (pERK) after the behavioral assessment. **Results:** For Nicotine we found an attractive response at 1 uM, expressed by an increased dwell time in the nicotine containing zone. Higher concentrations (10 uM), on the other hand, appear to be clearly aversive, and larvae tried to escape the dish. Attractive and aversive responses have been reported to be attributed to differential activity levels in the Habenula with according activation or inhibition of the reward center in the teleost brain. We are investigating whether neonicotinoids (Imidacloprid, Thiacloprid) and psychoactive pharmaceuticals (Citalopram, Lamotrigine, Oxazepam) found in European waters trigger similar behavioral patterns. **Outlook:** We will dissect which chemosensory system and higher brain areas are involved in the behavioral reactions to environmental chemicals. This will advance our understanding of the impact of chemicals on fish behavior.

TU334

Urban sewage effluents into an alpine stream: are information on behavioural effects on Daphnia magna suitable to protect alpine cold adapted species?

V. Di Nica, University of Milan - Bicocca (VAT IT12621570154) / Department of Earth and Environmental Sciences; V. Lencioni, F. Bellamoli, MUSE-Museo delle Scienze Trento / Dept. of Invertebrate Zoology and Hydrobiology; T. Pescatore, Water Research Institute Italian National Research Council IRSA-CNR / Dept. of Earth and Environmental Sciences; C. Ferrario, University of Milano Bicocca; S. Villa, EMA European Medicines Agengy

Even if identified as pristine, mountain freshwater ecosystems could be threatened by chemical pollutants through the discharge of effluents from wastewater treatment plants (WWTPs). Recently, residues of pharmaceuticals and personal care products have been detected in different alpine rivers downstream of WWTPs. Acute toxicity tests performed with *Daphnia magna* are among the most internationally used bioassays for monitoring the toxicity of effluents. However, acute toxicity tests do not take into consideration endpoints that may provide early warning signals about the health of the exposed populations, such as behavioural changes. Altered behavioural signals could be induced at sublethal concentrations which are significantly lower than the corresponding L(E)C50. In this study, we compared the sensitivity as mortality and swimming of *Daphnia magna*, and *Diamesa cinerella* gr.larvae, a chironomid (Diptera Chironomidae) common in cold freshwaters in the Alps, often associated to pristine environments. Both organisms were exposed for 24 and 48 hrs to different dilutions of effluents collected from a WWTP located at the Tonale Pass locality, in Trentino (1799 m a.s.l., NE Italy). The aim was to verify if *D. magna* could be employed in biomonitoring programs for WWTPs located in Alpine areas as surrogate of cold freshwater best adapted species. Mortality rate and behavioural responses (as swimming, analysed with two video tracking systems: LoliTrack Systems and ImageJ/wrMTrck) were compared. No mortality or change in behaviour was observed in the two organisms under exposure to undiluted samples. Exposure to serial dilutions of the effluent caused mortality only in *D. magna* (15% of mortality after 24 hrs at 1:1000 dilutions; 15% and 20% of mortality after 48 hrs at dilutions of 1:100 and 1: 1000, respectively). For the behavioural investigations, exposure to dilutions of the treated effluent induced significant alterations of swimming parameters in both organisms (e.g., the time spent in activity in *D. magna*; the average speed of movement and the cumulative distance travelled in both) at both the exposure times. Overall, these findings emphasised a higher sensitivity of *D. magna* than *D. cinerella* gr. to treated effluents. Accordingly, *D. magna* might be proposed as model organism to test the toxicity of WWTP effluents in alpine streams.

TU335

Do silver and titanium dioxide nanoparticles influence the fish kairomone induced anti-predator defence in Daphnia magna?

A. Beasley, University of Siegen; S. Hartmann, University of Siegen, Institute of Biology / Department of Chemistry and Biology; K. Witte, University of Siegen / Department of Chemistry and Biology

Daphnia possess a trait of phenotypic plasticity, whereby kairomones from fish induce growth and the formation of structures such as a spine or helmet. The resulting increase in body size, allows the daphnids to defend themselves from the predators in their natural environment. As the common link between green algae and fish in the food chain, daphnia are considered a key component in the freshwater system. Their ability to grow adequate defensive structures, is therefore necessary, to prevent an ecological imbalance in the freshwater environment. Ag and TiO_2 manufactured nanomaterials (MNMs) are widely used in the commercial industry because of their unique properties. Silver is known for its antimicrobial properties and is therefore used in soaps and bandages as well as clothing and washing machines. Titanium on the other hand is used in products such as sunscreens, paint and toothpaste because of the bright white pigment it contains. Due to their small size, nanoparticles are not being effectively removed from wastewater treatment plants and end up in freshwater systems such as rivers and streams. Filter feeders, like the cladoceran *Daphnia*, take up these nanoparticles and are therefore of particular scientific interest, to establish what impact the MNMs are having on the freshwater cycle and food-chain. In our study, we investigate the effects of Ag (NM300K) and TiO_2 (NM105) MNMs on the predator defence response; by chronically exposing *Daphnia magna* to fish kairomones and a range of nanoparticle concentrations. This experiment was conducted in accordance with OECD guideline No. 211 over a course of 21 days. For statistical analysis, we measured the body length, body width and tail spine length after each moult and counted the reproductive success of the individual daphnids. An SEM picture is taken of each daphnid at the end of the experiment, to observe and compare the microscopic details of the anti-predator defences in the treated and untreated daphnia.

TU336

Behavioral and Physiological Responses of Daphnia magna to Fluoxetine and Propranolol Exposure

M.E. Nielsen, P. Roslev, Aalborg University / Biology and Environmental Science

Fluoxetine and propranolol are neuroactive human pharmaceuticals that occur as pollutants in surface waters. The potential effect of such pharmaceuticals on aquatic organisms including invertebrates has raised some concern but many adverse effects are not well characterized. In this study, 6 behavioral and physiological parameters in the freshwater Cladoceran *Daphnia magna* were compared for their responses to fluoxetine and propranolol exposure: mobility (dichotomous response), active swimming time, swimming distance, swimming velocity, swimming acceleration speed, and survival in the absence of food (starvation-survival). Changes in swimming behavior of *D. magna* were quantified by video tracking of single organisms followed by image analyses. Active swimming time and swimming distance appeared to be more responsive behavioral endpoints than swimming velocity and swimming acceleration. The EC50s for fluoxetine and propranolol determined from swimming time and swimming distance were comparable (1-2 mg/L). At low sublethal exposure concentrations (µg/L), nonmonotonic responses in swimming behavior were observed in *D. magna*. Behavior profiling estimated from multiple behavioral parameters showed that fluoxetine and propranolol stimulate swimming activity at 1-10 µg/L. EC50 values for fluoxetine and propranolol estimated from survival time in the absence of food (starvation-survival) were much lower than EC50 values estimated from changes in swimming behavior. Starvation-survival is strongly affected by energy metabolism and we suggest that this parameter can be a potential sensitive endpoint for determining adverse effects of pharmaceutical to *D. magna*. Combining behavioral and physiological responses to high and very low exposure concentrations should be considered in models predicting adverse effects of pharmaceuticals to non-target organisms.

TU337

How toxic is a non-toxic nanomaterial: Behaviour as an indicator of effect in Danio rerio (zebrafish) exposed to nanogold

T. Botha, North-West University / School of Biological Science; S. Brand, North-West University; V. Wepener, North-West University - School of Biological Sciences / School of Biological Sciences

Gold nanoparticles are used as drug delivery vectors based on the assumption that they have a low toxicity. Literature has however showed conflicting results over the last few years. This study aimed at investigating the toxicological effects of nanogold (nAu) over a range of indicators from sub cellular to whole organism level. Gene regulation, changes in oxidative stress biomarkers and swimming performance were assessed in *Danio rerio* (zebrafish) following exposures to nAu. Adult zebrafish were exposed to nAu for 96 hours, swimming performance was measured post exposure. Liver tissue was collected for DNA microarray and Real Time Polymerase Chain Reactions (RT-PCR) analyses to determine changes in gene expression (catalase, superoxide dismutase and metallothioneins). Whole body samples were stored in respective buffers for protein biomarker analysis (catalase, superoxide dismutase, acetylcholine esterase, malondialdehyde, cellular energy allocation and metallothionein). Swimming behaviour was assessed in 1.1 L Tecniplast™ tanks for a period of six hours and videos were analysed using Noldus EthoVision software. The critical swimming speed was performed in a Loligo® swimming tunnel, briefly fish were acclimatized within the chamber for one hour and swam at a starting speed of 2 bl/s with a 0.5 bl/s speed interval, fish were swam until they were unable to keep up with the increasing water flow. The DNA microarray revealed that 20 mg/L was the least related to the control group. At 20 mg/L there was a significant increase in gene expression for all genes analysed but protein biomarkers showed no significant response. The behaviour results showed significant changes in distance moved, swimming speed, acceleration bouts, zone alterations and time spent within the top zone- responses which are seen in fish responding to toxicological stress. The exposed fish has a significantly lower critical swimming speed when compared to the control. Since swimming performance and social interaction during swimming is essential to life whole organism behaviour shows a toxicological response to nAu which is in agreement with genetic responses seen.

TU338

The effects of silver and silver nanoparticles via different routes of exposure on behaviour in marine amphipods

M. Vannuci-Silva, UNICAMP / Institute of Biology; S.A. Kohler, University of Portsmouth / Animal Physiological Ecology; G. Umbuzeiro, School of Technology, UNICAMP / LAEG; A. Ford, University of Portsmouth / Biological Sciences

Behavioural responses are an important endpoint because they provide a link between biochemical and ecological effects of environmental contaminants. Silver is increasingly being used in nanomaterials and, consequently, being released into the environment in different forms. The behavioural consequences of metal exposure in crustaceans are scarce, especially in marine organisms. Therefore, the aim of this work was to evaluate the effects on the swimming behaviour of the marine amphipod *Echinogammarus marinus* after exposure to silver, in its salt (AgCl and $AgNO_3$) and nanoparticle (E. marinus (n=20 per treatment) were exposed individually. The exposure via water was performed with Ag (from $AgNO_3$) at 0; 5; 25 and 100 µg L^{-1} for 96 hours. In the exposure via food, the animals were fed on alternate days with control food or food impregnated with AgCl or AgNP (approximately 200 mg kg^{-1}) during 7, 14 and 28 days. The movements of the amphipods were tracked using a DanioVision™ system with EthoVision®XT software for behavioural analysis under 3 minutes dark/ 3 minutes light cycle. Differences in velocity of swimming, response to light and thigmotaxis were evaluated. In the Ag exposure via water, light significantly increased maximum velocity for all treatments ($p \leq 0.05$) indicating an escape-related behaviour; excepting at 100 ug L^{-1}, where the maximum velocity had no difference between lights off and on (p=0.110), showing that silver at this concentration had a significant effect on the response to light; no significant effects were observed in frequency in centre zone for all treatments (p>0.05), although, cumulative duration in centre zone was significantly different for 25 ug L^{-1} treatment (p=0.048), where animals spent more time in the centre zone when was dark. Results from the exposure via food are currently undergoing analysis. The current results indicate that silver has effects on swimming and response to light behaviours in *E. marinus*, indicating that exposed animals in the environment could be more vulnerable to predation. *Acknowledgement:* The authors thank São Paulo Research Foundation FAPESP 2016/19635-4 for the financial support. We also thank Professor Dr Theodore Henry from Heriot-Watt University for providing the food (control, AgCl and AgNP) used in the feeding exposure.

TU339

Developing methods to determine aquatic invertebrate behavioural endpoints for regulatory ecotoxicology studies

A.C. Brooks, Cambridge Environmental Assessments / Department of Enviromental Science; A. Lawrence, Cambridge Environmental Assessments / Regulatory Ecotoxicology; M. Hackett, Cambridge Environmental Assessments / Cambridge Environmental Assesssments; M. Allen, F. Joyce, F. Pickering, Cambridge Environmental Assessments; H.S. Schuster, Cambridge Environmental Assessments (CEA) / Aquatic Ecotoxicology

Under current plant protection product (PPP) regulation, Tier I aquatic ecotoxicology studies measure mortality (acute) or reproduction (chronic). For invertebrates, these parameters are used to address the current protection goals of maintaining populations. Agreed guidelines for reproduction studies for aquatic invertebrates are currently available using *Daphnia* or *Chironomus*; however, for some substances, other taxa/species may potentially be more sensitive e.g. Ephemeroptera, Plectoptera, Trichoptera (EPT) species. Currently, there are no agreed methods for measuring reproduction endpoints for non-standard test species, such as EPT species. EPT species often live as larvae for a prolonged period and have an aerial adult stage, and thus assessing reproductive endpoints from a full life cycle in the laboratory is not easily achieved. Therefore, a proxy measure for reproductive effects is desirable. Although behavioural endpoints (e.g. predator response, locomotion, feeding activity) do not directly relate to the protection goals of maintaining populations, they may still have a useful role in regulatory risk assessment. For example, for organophosphates (OPs) with steep dose-response curves and thus a narrow exposure window between acute and chronic effects, significant differences on reproduction can be due to mortality of adults rather than true reproductive effects; therefore, designing specific reproduction studies for e.g. EPT species may not be necessary for such substances. Instead, if a risk assessment were undertaken using acute and behavioural endpoints and acceptable risks were concluded, then it would be unlikely for effects at the population level to arise. Here we will share our experiences of developing methods to measure such behavioural endpoints in one standard (*Daphnia magna*) and two non-standard (EPT: mayfly, caddis) test species that are suitable for use in regulatory toxicity tests, integrating the regulatory needs with the practicalities of ecotoxicology testing.

TU340

The effects of sublethal doses of pollutants on crop pest, Spodoptera littoralis

D. SIAUSSAT, Institute of Ecology and Environmental Sciences / Institute of Ecology and Environmental Sciences

Pesticides have long been used as the main solution to limit agricultural pests but their widespread use resulted in chronic or diffuse environmental pollutions, development of insect resistances and biodiversity reduction. The effects of low residual doses of these chemical products on organisms that affect both targeted species (crop pests) but also beneficial insects became a major concern, particularly because low doses of pesticides can induce various effects. In addition to the negative effects, some studies highlighted unexpected positive - also called hormetic - effects on insects, leading to surges in pest population growth at greater rate than what would have been observed without pesticide application. The present study aimed to examine the effects of sublethal doses of various representative products of large pesticide families used against a major pest insect, the cotton leafworm Spodoptera littoralis, and known to present a residual activity and persistence in the environment. Using an integrated approach from genes to behavior, we studied the impact on the peripheral olfactory system and the sexual or feeding behavior of our crop pest model following application of sublethal doses of deltamethrin, methomyl and chlorpyrifos. Whereas sublethal doses of methomyl appeared to disrupt the feeding behavior of larvae, we demonstrated a hormetic response of males to sublethal dose of deltamethrin. We completed our study by molecular (qPCR), biochemical (proteomic, AChE activity and metabolic) and electrophysiological approaches in order to decrypt the involved mechanism in pesticide response as well as in the behavioral disruption.

TU341
The effect of copper nanoparticles on olfaction in rainbow trout (Oncorhynchus mykiss)
P. Razmara, University of Lethbridge / Department of Biological Sciences; E. Mohaddes, Universitty of Lethbridge; E. Lari, G.G. Pyle, University of Lethbridge / Biological Sciences
Fish rely on olfaction for their survival, growth, and reproduction. Impairment of olfactory function can pose a threat to fish survival on the small scale and population loss on the larger scale. Metal contaminants (e.g. copper) can impair fish olfaction. Although the copper ion (Cu^{2+}) has drawn the most attention in olfactory toxicology, the impact of copper nanoparticles (CuNPs) on fish olfactory systems has not been well determined. The objective of this study was to investigate time-dependent effects of CuNPs and Cu^{2+} on olfactory acuity and olfactory–mediated behaviours of rainbow trout. To establish CuNPs or Cu^{2+} induced olfactory-impairment thresholds, inhibitory concentration (IC) curves were determined. Fish were exposed to a geometric dilution series of CuNPs or Cu^{2+} for 24 hours, and fish olfactory acuity was measured using electro-olfactography (EOG). Afterwards, fish were exposed to CuNPs or Cu^{2+} at concentrations known to impair olfaction by 50% (322 and 6.8 µg/L for CuNPs and Cu^{2+}, respectively) for a 24 h or 96 h exposure period. The response of fish to a social cue (taurocholic acid) was studied using EOG and a choice maze behavioural assay. After the behavioural experiment, fish olfactory rosettes were dissected to investigate if there was any DNA fragmentation as a marker of apoptosis that might be induced by CuNPs or Cu^{2+}. Results of EOG revealed that while a 96 h exposure to CuNPs caused a significantly greater impairment of fish olfactory function relative to a 24 h exposure to the same concentration, fish olfactory acuity partially recovered after 96 h under continuous Cu^{2+} exposure. Behavioural responses of rainbow trout to the social cue supported the results of neurophysiological experiments. Although fish exposed to control water or Cu^{2+} for 96 h had an avoidance response to an alarm cue, those exposed to the CuNPs did not respond to the alarm cue. Results of DNA fragmentation indicated apoptosis was not the mechanism of olfactory toxicity for CuNPs or Cu^{2+} in the exposed fish. In summary, over the same exposure periods, CuNPs caused progressive deterioration of olfactory acuity, whereas at least a partial olfactory recovery was documented for continuous Cu^{2+} exposure. However, the mechanism of olfactory toxicity caused by CuNPs is not clear and needs further investigation.

Informed substitution of hazardous chemicals for circular economy: science and practice (P)

TU342
Perfluoroalkyl acids concentrations in liquid wastes: a survey campaign and implications for waste disposal
M. Peruzzo, Eurolab; S. Valsecchi, Water Research Institute - Italian National Research Council IRSA-CNR; A. Scalco, Eurolab; S. Polesello, Water Research Institute- CNR / Water Research Institute
A survey campaign has been carried out to determine the concentrations of twelve perfluoroalkyl acids (PFAA) in liquid wastes, before the disposal in dumpsite or incinerator. The common characteristic of these wastes was that they were classified as “wastes without dangerous substances” and could be disposed without specific treatments. Waste samples (about 120) came from solid waste treatment plants, drinking water and isolated sewage treatment plants, landfill leachate, solid waste storage facilities, car washing, septic tanks, laundry sludge and wastes from various industrial plants such as paper, food, wood, furniture, glass and pharmaceutical industries. The large spectrum of activities allows us to get a panorama of the diffusion of PFAA in the economic sectors and to evaluate the impact of the different sources. The percentage of samples which presented total PFAA concentrations greater than 1 µg/L was 65%. The maximum concentration measured was 900 µg/L. As regards the single congeners, the percentage of positive samples (i.e. > 0.05 µg/L) ranged from 5% to 37.5% for the different compounds. It is very interested to note that PFOA and PFOS were found only in 5% of the samples, while the highest findings were for PFBA (57%) and PFBS (37.5%), highlighting the increasing diffusion of short chain PFAAs respect to the already restricted C8-PFAAs. It is also interesting to note that one of the samples with the highest concentrations was a found in the pharmaceutical industry, and it was an aqueous washing solution of water liquors. The overall survey underlines the need for a more accurate characterisation of wastes and the risk of transferring PFAA pollution from production sites to disposal sites, which can be located also in no-impacted areas.

TU343
Regenerated Textile raw materials: chemical contamination for LCA
A. Franchi, Buzzi Laboratorio Analisi
It’s essential, for every actor involved in the supply chain of a textile company, to increase awareness that a regenerated material requires proper and specific evaluation standards. These should ensure compliance with private protocols and mandatory laws and also ensure a proper control of levels of contamination. CID (Italian Consortium for Detox Implementation) with the support of local actors (chemical laboratories) made a study concerning chemical contamination of regenerated materials in order to propose a PRSL (Product Restricted Substance List) for regenerated and recycled textile materials. The adoption of a PRSL for regenerated textiles would guarantee the safe re-use of these materials as an alternative to their disposal. This case study takes into account regenerated wollen textiles (high wool content > 70%) derived by post-consume materials (knitted apparel, apparel made up by carded woven and combed woven), pre-consume materials (combed and carded woven, knitted fabrics, spinning and twisting trimmings), and regenerated cotton–type materials (derived from denim recycling). Operational plan involved quantitative and qualitative assessment concerning regenerated wollen-type raw material used by carded spinning companies in Prato textile district, sampling (more than 100 wollen-type regenerated raw material selected by origin and type and more than 40 cotton-type regenerated materials) and chemical analysis (made by Buzzi Lab) of some priority groups of concerned substances: APEOS (Ethoxylated Alkylphenols), Aromatic amines from azo-colorants, Chlorophenols, PFC (per-fluorinated compounds), Allergenic and Carcinogenic Dyes, Heavy metals from artificial perspiration solution Results was that about 150 sample were analysed and chemical contamination were found for aromatic amines (16% of total samples), APEOS (100% of samples), Chlorophenols (26 % of total samples), PFC (62% of total samples) heavy metals (82% of total samples), Allergenic and carcinogenic dyes (6% of total samples). Data analysis permits to establish a PRSL (Product Restricted Subtance List) protocol for regenerated raw materials with the aim to have an unique PRSL available for brands, manufacturing companies and every actor involved in the textile supply chain. The PRSL adpotion could improve the recycling of textile materials as an alternative to their disposal

TU344
Challenges for a comparative risk assessment among conventional hazardous substances and alternatives for textile finishing. Two case studies: flame retardants and durable water and oil repellents.
N. Fuentes, J. Damásio, V. Gonzalez-Andres, M. Diez-Ortiz, G. Janer, Leitat Technological Center
Some of the chemicals used in textile finishing processes are known to be toxic, persistent and bioaccumulative. Indeed, some of them (e.g., PFOA and DecaBDE) are listed in the Authorisation list and will be restricted after the sunset date. Alternative products are currently proposed by chemical companies for textile applications. Here we present two case studies (FLAREX and MIDWOR projects) focusing on the substitution of hazardous substances used as Flame Retardants (FRs) and Durable Water and Oil Repellents (DWORs). FRs are added to fabrics to inhibit the combustion process, and typically are products with a chemistry based on halogenates. DWORs are added to fabrics to repel water, oil and dirt, and typically are products with a chemistry based on long chain fluorocarbon polymers. These projects aim to support industry in the selection of alternatives. Alternative finishing additives available on the market were selected for laboratory validation of technical performance and industrial demonstration. In addition, a comparative risk assessment of conventional and these alternative formulations should be provided to ensure the reduction of environmental and human health impacts. One of the main challenges for a comparative risk assessment of these products is that the active substances in most of them are polymers and therefore are not subject to registration under REACH regulation. The potential human health and environmental impacts of these materials are related to their content or possible release of monomers (both with limited information). Moreover, the commercial formulations offered by chemical companies do not provide detailed composition. In fact, in the commercial products evaluated the chemical identity of the active substance was not reported in the safety data sheet. This is due to the lack of obligation to report ingredients that are not triggering the hazardous classification of the mixture. Under this scenario, we propose to base the comparative risk assessment on the toxicological profile of the chemical family of corresponding monomers (based on the information supplied by the providers), and the operational conditions necessary for the application of each of the products (assuming that the risk mitigation measures will not change within an industrial setting). The results will support industry to select functional and safer alternatives.

TU345
Substitution of firefighting foams containing per- and polyfluorinated alkyl

substances (PFASs)

A. Biegel-Engler, German Environment Agency - UBA / Chemicals; L. Vierke, C. Staude, German Environment Agency / Chemicals

Per-and polyfluorinated alkyl substances (PFASs) are heat resistant and show a low friction resistance. Because of these properties PFASs are for example widely used in aqueous film forming firefighting foams (AFFF). PFASs are not degradable in the environment. Thus, each release of AFFF into the environment causes a contamination. Long chain PFASs (such as PFOS and PFOA) need years to leach from top soil layers into the groundwater or into surface water. Short chain PFASs however reach ground water resources much faster due to their mobility in soil. Those contaminations already caused closed drinking water wells. Remediation is costly and long lasting. Although fluorine free foams are available and used at several European airports many firefighters hesitate to use them instead of AFFF. Restriction and authorization are regulatory measures under REACH which can be used to minimise releases of PFASs into the environment. An international regulation via the Stockholm Convention is possible as well. In addition the dialogue with stakeholders can lead to voluntary actions and may be an alternative measure to reduce environmental releases. Scientists and manufacturers need to be encouraged to develop environmentally friendly firefighting agents without fluorinated chemicals. Moreover, scientists, authorities and NGOs need to bring together knowledge about the new substances, such as analytical methods, and information on their fate and behaviour in the environment. This presentations provides an overview on regulatory actions regarding PFASs in the EU and further ideas how to substitute firefighting foams containing PFASs.

TU346

The Paradigm of Substitution - expand your view

M. Zimmer, ZVO e.V.; M. Metzner, Fraunhofer Gesellschaft

Many people mention substitution as the most promising option for risk reduction in the use of SVHCs. But it has to be considered that technical solutions are embedded into complex structure-effect relationships along of equally complexity. Many different properties and outcomes have to be considered. Otherwise a thorough assessment of the applicability of an assumes alternative will fail. The surface treatment sector as a major cross-sectional community of service providers has long term experience with innovation and substitution suggested by different players for many different reasons – risk reduction being one of them. In particular the SMEs have constantly been confronted with lots of different ideas and approaches. Hence they have deep insight into unexpected side effects and regrettable outcomes. And they are able to give indicators for real promising and applicable approaches to substitution. The authors will present some significant examples of substitution attempts – and they discuss arguments why they might be considered successful – or not.

TU347

A pilot case on how Socio-Economic Evidence can inform Risk Management decision making to assess Substitution versus Recycling for non-ferrous metals slags in safe use applications

H. Waeterschoot, M. vander Straeten, Eurometaux

The implementation of the European Circular Economy policy leads to more recycling, including closing the loop on substances. This policy combined with the increasing complexity of articles leads to increasing amounts of hazardous substances and impurities being available for recycling or reuse. Recycling processes in the metals sector produce besides pure substances for safe reuse, also final slags that collect some of the impurities that cannot be recovered at economic conditions. In parallel, the human health and environmental effects data generated by EU REACH and CLP Regulations lead to increasing hazard identifications and harmonised hazard classifications. Hazard endpoints of Very High Concern like CMR (carcinogens, mutagens and reproductive toxicity) or respiratory sensitisation may trigger substitution-based Risk Management Measures but also reduce the reuse in safe applications for “precautionary reasons”. In such cases, socio-economic evidence may be helpful to assess costs and benefits from a broader perspective, including Circular Economy and carbon footprint considerations.\n A pilot study conducted at a non-ferrous plant specialised in the recycling of complex end-of-life articles and materials allowed to evaluate this impact and develop a tool for assessors to check how and to what extent a change in a relevant hazard classification could impact the reuse capacity of final slags. The tool allows metal companies to assess their situation in respect to Substitution (materials loss or disposal) versus Reuse in safe applications.\n

Developments in the ecological and human health risk assessment of biopesticides: microorganisms, semiochemicals and botanicals (P)

TU348

Ecotoxicity of the hydrolate byproduct of three biopesticides on the unicellular green algae Chlamydomonas reinhardtii

D. Ballestero, J. Val, E. Langa, San Jorge University; E. Navarro, CSIC - Spanish National Research Council / Dept. Recursos marins renovables; M. Pino, San Jorge University / Facultad ciencias de la salud; A.M. Mainar, Universidad de Zaragoza

Synthetic pesticides have been widely used in intensive production systems throughout most of the 20th century. However in the last decades, environmental and human health concerns demand safer substances, so research on biopesticides has been increased. Although a large number of studies have been published focusing on the biological activity of biopesticides on target organisms, studies regarding toxicological effects on non-target organisms, are scarce. The BIOCROP Project (Biopesticide development by chemical and biotechnological tools) has allowed the screening of several plants extracts for bio-activity against a selected set of crop pests and arthropod vectors. Some of these compounds have showed its effective value as biopesticides. The extracts will be optimized by means of traditional and supercritical CO_2 technologies, as well as microbiological transformations. In the extraction process the organic and the aqueous fraction (hydrolate) have been separated. Both of them showed active compounds, being capable to act as biopesticides. In order to exclude a negative effect on the environment, these products should be studied on non-target organisms. **The aim of this study was to measure the acute ecotoxicity of hydrolates obtained of the semi industrial vapor-pressure essential oil extraction of three selected aromatic plant species; a domesticated *Artemisia absinthium* (Teruel, Spain), *Dittrichia graveolens* (Ciudad Real, Spain), and an experimentally pre-domesticated *Lavandula luisieri* (Toledo, Spain) using the algae *Chlamydomonas reinhardtii* as aquatic model organism.** Results indicate that all of three extracts having biopesticide activity are likely to cause toxic effects on the photosynthesis of *Chlamydomonas reinhardtii*, being *Lavandula luisieri* the most toxic compound followed by *Artemisia absinthium* with a very similar toxicity,and *Dittrichia graveolens* presentin the lower toxicity. These results allow for a better understanding of the safety that natural crop protectants can play in the aquatic environments. Acknowledgements: *We thank J. Burillo and J. Navarro for his generous cession of the extracts used in this study and the financial support of* MINECO-FEDER (CTQ2015-64049-C3-2-R)

TU349

Ecotoxicological evaluation of the hydrolate byproduct of Satureja montana on Daphnia magna and Vibrio fischeri

E. Oliva, Universidad San Jorge; E. Terrado, San Jorge University; J. Navarro, Centro de Investigación y Tecnología Agroalimentaria de Aragón (CITA); M. Pino, San Jorge University / Facultad ciencias de la salud; A.M. Mainar, Universidad de Zaragoza; D. Ballestero, San Jorge University

The increasing demand of natural bioproducts for cosmetic use, food or phytotherapy is based on the awareness about adverse effects on health and the environment. In particular, this work is focused on the plant *Satureja montana (Lamiaceae)*, which has demonstrated a wide range of applications due to its important antioxidant and antimicrobial activity. Furthermore, *Satureja* species have a sweet flavor and simple cultivation characteristics. The main components, thymol and carvacrol (oxygenated monoterpens), are supposed to be responsible for these biological activities. Although there are a substantial number of studies where *Satureja* speciesare evaluated for their biological and pharmacological activities, as well as its chemical characterization, limited data are available on ecotoxicological characterization. Consequently, the aim of this study is to evaluate the acute ecotoxicity of the hydrolate obtained from *Satureja Montana (Ejea, Aragón)* by the traditional method of steam distillation on two organisms widely used as indicators of ectotoxicity: the freshwater crustacean *Daphnia magna* and the marine bacterium *Vibrio fischeri*. Both tests are standardized for the purpose of determining the toxicity expressed as EC_{50}. Our results indicate that *the hydrolate of S. Montana* are likely to cause toxic effects on *D. Magna and V. Fisheri* but only at high dilutions (*LC50 values in the range of* 0,5% in both cases). These studies allow us to know the possible environmental effect that these promising plant extracts can cause as a source of cosmetic and pharmaceutical applications with the aim of ensuring more environment-friendly processes and products. *Acknowledgements: We thank J. Burillo, for his generous cession of the extracts used in this study and the financial support of MINECO-FEDER (CTQ2015-64049-C3-2-R)*

TU350

THE IMPACT OF THE HYDROLATE BYPRODUCT OF THREE BIOPESTICIDES ON THE SOIL ENVIRONMENT

M. Pino, San Jorge University / Facultad ciencias de la salud; D. Ballestero, J. Val, San Jorge University; E. Sánchez, Colegio internacional Anfora; E. Navarro, CSIC - Spanish National Research Council / Dept. Recursos marins renovables; J. Urieta, Universidad de Zaragoza

The extended use of synthetic pesticides has resulted, during the last century, in the pollution of the agricultural soil environments. As an alternative to these products, environmentaly friendly biopesticides are, nowadays, being developed. Although biological activity of biopesticides on target organisms is well known, studies focusing on the effects on soil non-target organisms are scarce. The BIOCROP Project (Biopesticide development by chemical and biotechnological tools) focus on the production and optimization of plant/fungal/agriwaste-based crop protectants via cultivation techniques, biotransformation, selective extraction and separations by traditional and supercritical CO2 technologies. In the traditional extraction process the organic and the aqueous fraction (hydrolate) have been separated. Both of them showed active compounds capable to act as biopesticides. In order to exclude a negative effect on the environment, these products were tested on soil non-target organisms (microbial community and earthworms). Soil microbial communities from an ecological farming crop have been exposed to three hydrolates, obtained by semi industrial vapor-pressure essential oil extraction, from three aromatic plant species: *Artemisia absinthium, Dittrichia graveolens and Lavandula luisieri.* The effects on the microbial community has been assessed using the community-level physiological profile –CLPP-. This method relies on the ability of the microbial community for degrading different carbon sources present in Biolog Ecoplates®). The acute toxicity of hydrolates was also tested by *Eisenia foetida* bioassay. Results indicate that hydrolates caused acute adverse effects in E. foetida, in particular *D. graveolens and A. luisieri* (LC50 in the range of dilution of 10-2). All three biopesticides provoked changes in the soil microbial ability to degrade different carbon sources compared to control. These results allow for a better understanding of the impacts of natural crop protectants in the soil environment as a pest management alternative. Acknowledgements: We thank J. Burillo and J. Navarro for his generous cession of the extracts used in this study and the financial support of MINECO-FEDER (CTQ2015-64049-C3-2-R)

TU351

Acute toxicity of emulsifiable concentrate of Alpinia galangal essential oil against Cyprinus carpio

H. Kim, K. Kim, H. Jeon, Y. Choi, Y. Kim, S. Lee, Kyungpook National University

Essential oils have exhibited their fumigational and topical toxicities on insect pests and they are developed as safe biopesticides. However, their use may be caused potent toxic effects to non-target organisms in the environment. It needs to be determined their negative effects on non-target organisms in the environment. *Alpinia galangal* essential oil (AGEO) has been considered to control the outbreak insect pest, *Ricina* sp. in South Korea. Acute toxicities of AGEO against *Cyprinus carpio* were assessed to understand its possible toxic effect on the representative aquatic organism, *Cyprinus carpio* in a static condition followed by OECD guideline 203 in 5 L beakers. As AGEOs were formulated for emulsifiable concentrate (EC) as an active ingredient, they were mixed with ethanol and tergitol in a ratio of 5:4:1. Tergitol is a surfactant and it did not show any toxic effect on the fish, so that it was used for the formulation of emulsifiable concentrate for AGEO. Three different AGEOs were prepared according to their extraction methods as steam distillation, solvent extraction and supercritical fluid extraction. After the EC formulation was prepared, they were ready to expose to *C. carpio* to determine LC50 values. All emulsifiable concentrates of three different AGEO showed no mortality on the tested fish during 96-hour incubation. Therefore, 48-h LC_{50} values for the VFEO were under the toxicity criteria of level 3 for the pesticide to *C. carpio* standardized by Korea Rural Development Administration. Based on these results, AGEO can be considered to use as a natural insecticide.

TU352

Chronic toxicity of emulsifiable concentrate of cinnamon essential oils against Cyprinus carpio

H. Jeon, K. Kim, H. Kim, Y. Kim, Y. Choi, S. Lee, Kyungpook National University

Recently, many researchers have developed natural insecticides to control insect pests using plant essential oils (EOs) due to their eco-friendly safe properties. Cinnamon EO is one of important EOs to be a potent candidate and is formulated as an emulsifiable concentrate (E.C.). As its use is recognized as safe, it needs to be determined its negative effect on the environment using bioindicators. To evaluate the negative effect on the ecosystem, chronic effects of cinnamon EOEC against *Cyprinus carpio* was determined in a static condition for 40 days. When cinnamon EOs are used as an active ingredient, they are mixed with ethanol as solvent and tergitol as surfactants. To select an appropriate surfactants, 8 different types of surfactants (Tween 80, Sodium dodecyl sulfate (SDS), Nondient, Triton X-100, Sodium dodecyl Benzene Sulfonate (SDBS), Koliphor, Tergitol and Mixture of SDBS and Nondient) were tested for the formulation and tergitol showed the lowest toxicity to the fish in an acute toxicity test. With the result of the acute toxicity of cinnamon EOEC, chronic toxicities of cinnamon EOEC was determined at the 5 different concentrations for 40 days. Each concentration was triplicate exposed to 10 of *C. carpio* adults. The treated five concentrations were 0.08, 0.16, 0.64, 2.56, and 5.12 ppm and the mean survival rate was 8.48 ± 0.47. The survival rate of the control group was 9.33 ± 0.58 and the positive control containing ethanol and tergitol was 8.00 ± 1.00. The mean temperature and pH of the test water was 24.06 ± 0.58 °C and 7.51 ± 0.03, respectively. The mean of dissolved oxygen of the test water was 7.29 ± 0.07 mg/L and the mean of hardness was 82.14 ± 2.04 mg/L. After the complete exposure, the mean of length of alive fishes was 3.00 ± 0.17 cm and the weight was determined as 0.37 ± 0.17 g. With these results, cinnamon EOEC may be considered as safe, natural insecticides for the environment.

TU353

Thiosemicarbazone scaffold for the design of antifungal and antiaflatoxigenic agents: evaluation of ligands and related metal complexes

s. montalbano, uniersity of parma / Department of Chemistry, Life Sciences and Environmental Sustainability; f. bisceglie, d. rogolino, University of Parma / Department of Chemistry, Life Sciences and Environmental Sustainability; M. Study Group, University of Torino, Brescia, Pisa, Perugia and Salento / Dep of Medical and Surgical Specialties Radiological Sciences and Public Health; f. degola, University of Parma / Department of Chemistry, Life Sciences and Environmental Sustainability; D. Feretti, University of Brescia Italy / Department of Medical and Surgical Specialties Radiological Science and Public Health; g. pelosi, University of Parma; m. pioli, f. restivo, m. carcelli, g. spadola, University of Parma / Department of Chemistry, Life Sciences and Environmental Sustainability; C. Zani, University of Brescia Italy / Department of Medical and Surgical Specialties Radiological Science and Public Health; i. zerbini, University of Brescia / Department of Medical and Surgical Specialties, Radiological Sciences and Public Health; a. buschini, University of Parma / Department of Chemistry, Life Sciences and Environmental Sustainability

Food safety is the safeguarding and protection of food from anything harmful affecting consumer health and is an extremely important issue facing the world. Food hazards can be divided into physical, chemical and biological. Examples of biological hazard are mycotoxins, that are toxic secondary metabolites produced by many species of filamentous fungi. Generally, mycotoxins represent a significant threat to human health as they can be carcinogenic, neurotoxic and toxic to endocrine or immune system. In particular, aflatoxins are a class of mycotoxin produced principally by two species of *Aspergillus*, *A. flavus* and *A. parasiticus*. Aflatoxins are found in various cereals, oil seeds, spices and nuts as a result of a fungal contamination that can occur in the field, during harvest, transport and storage. IARC has classified aflatoxins in Group 1 as carcinogenic agents to humans. The most dominant and potent aflatoxin is aflatoxin B1 and several studies indicate that high exposure to AFB1 can cause chronic toxicity and increases the incidence of hepatocellular carcinoma. A lot of methods can be applied to eliminate these toxins from food and guarantee the food safety and health concerns of consumers. Our research aims to develop new typologies of inhibitors of *Aspergillus* proliferation and of aflatoxins production, harmless to the environment and to human health. We have evaluated the biological activity of several thiosemicarbazone ligands starting from molecules of natural origin, like vanillin, perillaldehyde and their derivatives. In order to improve the biological activity, metal complexes were then synthesised. These molecules once synthesized and characterized, were initially tested to determine their antifungal and antiaflatoxingenic activity towards *A. flavus*. These compounds showed different efficacy in reducing fungal growth and mycotoxin accumulation. The most active compounds were used to perform cyto- and geno-toxicity tests on healthy human cells, particularly on human cell lines deriving from the districts that can be exposed to xenobiotics. Furthermore, we performed toxic and genotoxic assays on bacteria and plant cells. In conclusion, this approach allows us to study the antifungal and antiaflatoxin activity of several thiosemicarbazones and to determine the potential risk for environment and human health with a view to use these compounds in field. Financial support: Fondazione Cariplo-Project N. 2014-0555, http://aflatox.unibs.it/

Understanding human and environmental exposure to chemicals in urban systems (P)

TU354

Electronic products are related with household exposures in Canadian residents

M.L. Diamond, C. Yang, University of Toronto / Department of Earth Sciences; L. Jantunen, Environment and Climate Change Canada; D. Tsirlin, Cancer Care Ontario / Population Health and Prevention, Prevention and Cancer Control; L. Latifovic, Cancer Care Ontario; S. Harris, Cancer Care Ontario, University of Toronto / Population Health and Prevention, Prevention and Cancer Control

Key Words: electronic products, hand wipes, household exposure, FRs and plasticizers Novel flame retardants (NFRs), polybrominated diphenyl ethers (PBDEs), organophosphate esters (OPEs), and phthalates esters (PAEs) have wide applications as flame retardants (FRs) or/and plasticizers in consumer products, building materials, and industrial uses. Their wide-spread use has led to population-wide exposures (Carignan et al. 2017; Hammel et al., 2016; Hoffman et al. 2015). Some of these exposures have been related to adverse health effects (e.g., Carignan et al. 2017). Therefore. information on major exposure sources is needed to reduce exposures and ultimately prevent adverse health outcomes. Here we report on a household exposure study of Canadian women by determining levels of selected FRs and plasticizers in paired household air and dust, hand surface wipes of participants, as well as wipes of principle household electronic devices, including their cell phones. PAEs had the highest overall concentrations followed by OPEs by approximately one order of magnitude, and NBFRs and PBDEs (three orders of magnitude less than PAEs). Multiple compounds were found in wipes of individual electronic products suggesting either their usage in many products or migration into the surface polymer of these products from other sources indoors. Statistical analysis showed that OPE profiles on a participant's hands most frequently resembled the profiles found in that person's hand-held electronic products, notably that person's cell phone. Correlations for all compound classes were also found between compounds in large, stationary electronic devices (e.g., TVs) and the room's air and dust. However, the above correlations were not found for any OPEs between cell phones and air and dust, and were found for only two PAEs. Our results indicate wipes of cell phones were a stronger predictor of compounds on participant's hands, while large and stationary electronic products related more to household environment contamination of selected FRs and plasticizers. Our results indicate that participants are exposed to various FRs and plasticizers through their daily household environment. The Canadian adults' external exposure of hands to FRs and plasticizers were related to the levels in their household electronic products, particularly handheld devices such as cell phones. Handheld devices could contribute to human exposure through direct contact during use while large and stationary electronic products could be important sources and sinks in household environment.

TU355

Modelling diffuse emissions and fate of engineered nanoparticles used in outdoor paints to urban surface waters at high spatial and temporal resolution

M.D. Nuñez, University of York / Environment; A. Praetorius, University of Vienna / Department of Environmental Geosciences; A. Boxall, University of York / Environment Department

The expansion of the nanotechnology sector is leading to an increased use of products containing engineered nanoparticles (ENPs) in outdoor urban environments. Outdoor materials, such as construction materials, paints and coatings, are subject to weathering and ageing processes and will consequently lead to emissions of ENPs to the surrounding environment over time. Data on measured environmental exposure concentrations are still lacking for ENPs. Until analytical and monitoring techniques for ENPs in environmental matrices become available, modelling tools are the best approach to estimate exposure levels. Furthermore, models can analyze a wide range of potential scenarios and predict possible future trends of urban exposure which cannot be achieved by monitoring. In this study we present the results obtained from the application of a new modelling approach that combines an emission and a fate model for ENPs with high spatial and temporal resolution for an urban environment. The model was applied to the study of titanium dioxide (TiO_2) ENP emissions when incorporated in outdoor paints in the city of York (UK). The model emission calculations are based on locally collected data on outdoor paint usage (outdoor paint application ratios and frequency of application for York) and information about the sewage network connectivity around the city. Reliable and official sources of information, such as Yorkshire Water and York City Council, and surface water characteristics acquired from an extensive and local monitoring campaign performed in the rivers Ouse and Foss, helped to parametrize the river fate model. Using the model, the transport and fate of TiO_2 ENPs in the rivers circulating within the city (the Ouse and the Foss) could be studied and spatially resolved results obtained. The identification of hot spots of emissions within the city and the study of ENP transport and fate are accomplished by this approach.

TU356

Occurrence and human exposure of parabens, triclosan and triclocarban in personal care products from Korea

S. Mok, Hanyang University / Marine Sciences and Convergent Technology; J. Lim, Hanyang University; M. Lim, Seoul National University / Environmental Health Sciences; J. Park, Seoul National University / Environmental Health Science; K. Lee, Seoul National University, Graduate School of Public Health; H. Moon, Hanyang University / Marine Sciences and Convergent Technology

Parabens (*p*-hydroxybenzoic acid esters), triclosan (TCS) and triclocarban (TCC) have been extensively used in various cosmetics and personal care products (CPCPs) as preservatives due to their antimicrobial activities. However, little is known about the occurrence and exposure levels of parabens, TCS and TCC associated with the consumption of CPCPs in our daily life. In this study, ten parabens and their metabolites, TCS and TCC were measured in 243 CPCPs, which comprised of leave-on products (n=157), rinse-off products (n=59) and baby care products (n=31), collected from Korean market during 2016-2017, using liquid chromatography-tandem mass spectrometry (LC-MS/MS).Among ten parabens, methyl paraben (MeP) showed the highest detection rate (57%), followed by propyl paraben (PrP, 49%) and butyl paraben (BuP, 41%). TCC had only 20% of detection rate and TCS was rarely detected in the samples. Total concentration of parabens widely varied with ranging from < LOQ to 10200 µg/g. Concentrations of TCC and TCS ranged from < LOQ to 340 ng/g and < LOQ to 14.0 ng/g, respectively. Higher concentrations of parabens (> 1000 µg/g) were found at skin cares, sunscreen, face cleanser, eyeliners, body/hand lotions and lipstick. The daily exposure levels of parabens associated with the consumption of CPCPs were calculated using exposure factors, obtained from questionnaire-based survey and previous studies, and concentrations measured in our study. The mean daily exposure levels of parabens were 16.2 and 0.14 µg/kg body weight/day for mothers and their infants of Korea, respectively. Among CPCPs, some leave-on products such as skin cares, body/hand lotions, and sunscreens were the major contributors (> 80%) to total exposure levels of total parabens.

TU357

Characteristics of exposure factors for consumer products in Korean infant and caregivers pair

K. Lee, Seoul National University, Graduate School of Public Health; M. Lim, Seoul National University / Environmental Health Sciences; J. Park, Seoul National University / Environmental Health Science

Concerns about potential health risks of chemicals in consumer products like cosmetic, personal care products, food containers are growing. Especially infant and children are more vulnerable to chemical exposure compared to adults. Since exposure to these chemicals could be determined by consumer products usage pattern, an accurate measurement of the usage patterns of consumer products is important for realistic exposure assessment. The aims of this study were to determine exposure factors of consumer products for child and mother and analyze the relationship between consumer exposures of caregiver and infant. We determined the exposure factors of 12 kinds of cosmetics (3 basic cosmetics, 1 UV protection products, 3 hair products, 3 body products and 2 cleansing products) for adults and 10 kinds of consumer products (2 cosmetics, 3 oral supplies and 7 household products) for children and 11 kinds of food containers for household. Survey was conducted on 505 mother-infant pairs from Oct. to Dec. 2015 in Seoul metropolitan area by a structured questionnaire. The number of subjects were determined by proportionate quota sampling based on the population composition ratio in children's sex and age distribution from 0 to 4. All cosmetics investigated in this study were used on a daily basis and usage rates ranged from 52.1% to 98.0%, except 9.9% for hair styling product and 7.7% for deodorant. The frequency of food intake by food containers ranged 2.52 to 17.39 times a month. The use of children's oral supplies varies according to the age of the child. There were a significant difference in the mother's usage rates of lotion, hair products and vinyl package food by age of children. These exposure factor characteristics could be useful input data for exposure and risk assessment for chemical regulation.

TU358

Analysis of metabolites of organophosphate and pyrethroid pesticides in urine from Italian children

N. Bravo, CSIC-IDAEA / Department of Environmental Chemistry; J. Grimalt, Institute of Environmental Assessment and Water Research IDAEA CSIC / Department of Environmental Chemistry; B. Bocca, G. Calamandrei, A. Alimonti, Istituto Superiore di Sanità

Organophosphate (OP) and pyrethroid (PYR) pesticides are commonly used in agriculture, domestic environments and gardening. They eliminate insects because of their strong potential to disrupt the brain and nervous system of these organisms. Unfortunately, this neurotoxic effect is not selective enough as to avoid potential damage to other non-target species, including humans. OP and PYR pesticide exposure has been related to several human health effects, including respiratory, digestive, reproductive and neurological problems, among others. Children are more vulnerable than adults to environmental pollutant exposure because their organs and metabolism are still under development. Thus, their detoxification mechanisms are not yet mature. Once in the human body, OP and PYR pesticides are typically metabolized and excreted in urine within 4-48 hours after exposure, depending on the compound. Organophosphates are metabolized into dialkyl phosphates (DAPs) and specific compounds, such as 3,5,6-trichloro-2-pyridinol (TCPY, metabolite of chlorpyriphos), 4-nitrophenol (PNP, metabolite of parathion), malathion dicarboxylic acid (MDA, metabolite of malathion), 3-chloro-4-methyl-7-hydroxycoumarin (CMHC, metabolite of coumaphos), 2-isopropyl-6-methyl-4-pyrimidiol (IMPY, metabolite of dialzinon) and 2-diethylamino-6-methyl pyrimidin-4-ol (DEAMPY, metabolite of pirimiphos). Concerning pyrethroids, most compounds such as permethrin, cypermethrin, deltamethrin, allethrin, resmethrin and fenvalerate are metabolized into one single compound, 3-phenoxybenzoic acid (3-PBA). On the other hand, cyfluthrin is metabolized into 4-fluoro-3-phenoxybenzoic acid (4-F-3-PBA). Therefore, 3-PBA and 4-F-3-PBA can be used as a biomarkers of the most common PYR pesticides. The determination of the above mentioned compounds was performed using isotope dilution solid phase extraction UPLC-MS/MS. Human urine samples (n=199) from Italian children at 7 years were analyzed. Neuropsychological and psychomotor development of the children was assessed at 18-40 months by using the BAYLEY scales. In addition, neuropsychological development and intelligence were assessed at 7 years by using the NEPSY-II and the WISC, respectively. The compounds detected the most were DEAMPY (98%), PNP (97%), 3-PBA (91%) and TCPY (87%). The metabolite showing the highest concentration was DEAMPY with a median of 3.0 ng/mL followed by PNP, 3-PBA and TCPY with medians of 1.3, 0.56 and 0.47 ng/ml, respectively.

TU359

PAH levels in parturient and newborns from Aveiro region, Portugal.

M. Monteiro, Aveiro University / Biology; M. Fraga, Biology Department CESAM Aveiro University; C. Gravato, Faculdade Ciências da Universidade de Lisboa / department of Biology & CESAM; C.J. Silva, University of Aveiro / Biology Department & CESAM; A.L. Machado, University of Aveiro / CESAM Department of Biology; A.M. Soares, University of Aveiro / department of Biology & CESAM; S. Loureiro, Universidade de Aveiro / Biology

Environmental exposure to humans may be critical in some residential and working areas, and therefore biomarkers can and should be used as early warning tools to depict exposure and evaluate effects. Polycyclic aromatic hydrocarbons (PAHs) are a group of priority chemicals to be studied and monitored as they are considered carcinogenic and teratogenic. In the present study we aimed to monitor human fetal exposure to PAHs by measuring concentrations of naphthalene, phenanthrene, pyrene and BaP equivalents in placenta, umbilical cord and mother's blood (plasma and blood cells) of 49 parturient from Aveiro region, Portugal. Information organized in questionnaire forms, tissues and organs were collected following the parturient consent. Levels of PAHs equivalents were measured by a fluorescence methodology and were correlated with exposure to tobacco smoke as well as with other information regarding mother's lifestyle (e.g. urban or rural residential area, exposure to vehicles exhaust). In general, the studied group presented higher PAHs levels in the placenta and lower PAHs levels in the umbilical cord blood. The low molecular weight PAHs (naphthalene and phenantrene) measured in placenta presented higher levels than high molecular weight PAHs (pyrene and benzo[a]pyrene). Moreover, increased levels of naphthalene and phenanthrene equivalents were associated with exposure to vehicle exhaust, while higher levels of benzo[a]pyrene were associated with exposure to tobacco smoke at work. The highest naphthalene, pyrene and BaP equivalents levels were found in homogenized placenta of mothers who smoked in the third trimester of pregnancy. No significant correlations were found between PAHs levels and anthropometric data of newborns, but in general, higher PAHs levels were found in newborns groups with lower weight, head circumference, and length. Maternal-infant biomonitoring can be a major asset in evaluating environmental exposure to contaminants, which can also provide high value information for preventive medicine.

TU360

A modelling framework to link aggregate exposure pathways with internal exposures and potential bioactivity

J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology; J. Westgate, L. Hughes, ARC Arnot Research & Consulting; L. Li, University of Toronto at Scarborough / Department of Environmental Sciences; X. Zhang, University of Toronto Scarborough / School of Engineering and Applied Sciences; B. Givehchi, J.M. Armitage, University of Toronto - Scarborough / Physical and Environmental Sciences

The aggregate exposure pathway (AEP) model is a conceptual framework to help align chemical exposure information including (i) production, use and release, (ii) chemical fate and concentrations in various multimedia (urban and rural environments, biota), (iii) external exposures (e.g., contact rates), and (iv) internal exposures (e.g., blood concentrations) for human and ecological receptors. Some exposure models include elements of the AEP framework and are useful tools for organizing data, quantifying chemical concentrations throughout the source-to-exposure continuum and identifying research needs to address uncertainty in chemical evaluations. We present an overview of the Risk Assessment IDentification And Ranking-Indoor and Consumer Exposure (RAIDAR-ICE) modelling framework. RAIDAR-ICE includes direct and indirect near-field exposures and can include far-field exposures for aggregate human exposure assessment. The RAIDAR-ICE model is parameterized in this case study for about 200 organic chemicals comprising a broad range of chemical properties representative of commerical chemicals to demonstrate model applications for exposure and risk-based prioritization. Based on assumed emissions to air in the indoor environment, intake fractions (used for ranking exposure potential) range from 0.0018 to 0.37 emphasising the relatively high potential for human exposures to chemicals in used in indoor environments primarily due to their proximity to emission sources. Following the same exposure scenario, unit emission rate based whole body concentrations (exposure potential) range from 3×10^{-13} to 5×10^{-8} mmol/kg. The differences in ranking chemicals for exposure based on either external (intake fraction) or internal (concentration) exposure metrics are substantial due to chemical-specific differences in toxicokinetics. In absence of well-defined chemical use information, the model calculated critical emission rate can be used to gauge potential risks and provide guidance for proposed new chemical use. Using in vitro bioactivity data from the ToxCast program as an assumed "effect threshold", the critical emission rates of the case study chemicals span approximately 8 orders of magnitude. Including estimates of actual chemical use rates allows for the calculation of risk (bioactivity)-based estimates; the results of which span 10 orders of magnitude. Recommendations for addressing uncertainty in the model and its required input parameters are presented.

TU361

ENVIRONMENTAL IMPACT OF LEAD MINING ON THE BIO-ECOSYSTEM IN ISHIAGU TOWN OF EBONYI STATE IN SOUTH-EASTERN NIGERIA

S. Anika, UNIVERSITY OF NIGERIA, NSUKKA; V. Ahur, Federal University of Agriculture Makurdi / Department of Veterinary Physiology, Pharmacology and Biochemistry; P. Onyeyili, Federal University of Agriculture Makurdi / Department of Physiology Pharmacology and Biochemistry

Lead is a soft, dense and ductile metal found naturally in the environment and accounting for 0.0016% of the earth's crust. However, due to its ubiquitous nature, it is used in several industrial processes which can result to severe environmental pollution which can pass across food chains to animals and man. In 2010, about 400 deaths especially among children in Bukkuyum and Anka LGA of Zamfara State, Nigeria, due to chronic lead toxicity were reported by Medecins Sans Frontieres (MSF, Holland) to the health authorities (UNEP/OCHA 2010). The cause of the high mortality was acute and chronic lead poisoning as a result of massive environmental contamination from artisanal mining and processing of gold in Pb-rich ore by poor headsmen and farmers. This outbreak was reported as the worst in modern history (UNEP/OCHA 2010). Open-pit mining of lead in the Ishiagu Region of Ebonyi State since 1965 has exposed large volumes of marcasites, pyrites and tailings contaminating the environment and food chain pathways. The research was designed to investigate the environmental impact of lead mining on the bio-ecosystem of Ishiagu town and environs. Soil, water, grasses/plants, food, fish and quarry dust were collected between March and May 2017, processed and analysed for lead concentrations. All water samples exceeded WHO recomended safety limits for lead. Soil and food samples contained values. Sampling of Ivo River, the main communal water source showed links to upstream pollution as the river passes through lead miing fields. The result showed the negative impact of lead mining in Ishiagu and the neeed for regulatory agencies/government to take measures to avert consequences of lead poisoning in human beings.

TU362

Evaluation of potential risk of rare earth element contamination from leachate originating from electronic waste disposal

M. Makombe, Scientific Services, Cape Town / Department of Chemistry; C. Van der Horst, University of The Western Cape / SensorLab Department of Chemistry; V.S. Somerset, CPUT / Chemistry

Rare Earth Elements (REEs) form critical elements required in technological accessories. Their presence in electronic waste gives leads to environmental pollution. Therefore, there is a constant necessity for accurate data and reliable fast analytical methods. This review discusses and compares the methods given for the determination of rare earth elements and heavy metals in electronic waste and other associated environments. Classical analytical methods such as mineral acid dissolution and alkaline fusion are commonly employed. Various instrumental techniques that have been used recently includes electrothermal vaporiser or laser ablation accessory connected to ICP-MS or ICP-OES, X-ray Fluorescence and electro-analytical techniques. These methods of analysis including their accuracy, limit of detection (LOD) and limit of quantification (LOQ) are compared and discussed. This study further investigates the preparation, dissolution methods and instrumental techniques with a focus on rare earth elements in electronic waste and its disposal into the environment. **Key words**: Rare earth elements; electronic waste; fusion optimisation; spectroscopy; wastewater

TU363

A stonework snail as a new biomonitor of metal contamination in the urban environment

E. Rota, B. Braccino, R. Dei, University of Siena / Department of Physical Sciences, Earth and Environment; N. Bianchi, University of Siena / Department of Physical, Earth and Environmental Sciences; S. Ancora, University of Siena / Physical sciences, Earth and environment; R. Bargagli, University of Siena / Department of Physical Sciences, Earth and Environment

Papillifera papillaris (O.F. Müller) is a small pulmonate gastropod commonly dwelling on stone walls and monuments in Italian and Mediterranean urban environments. This widespread, low-vagile and omnivorous organism, which barely interacts directly with soil and inhales fine particles, is a promising indicator of metal deposition and bioavailability in urban environments. In fact, other organisms widely used as biomonitors of urban pollution, i.e. mosses, lichens and vascular plants, accumulate particles of soil and rock dust, making it difficult to recognize the element contribution from atmospheric deposition and the metal bioavailability to consumers. By analysing the chemical composition of the shells, soft tissues and faeces of snails collected from vegetated walls, at roadside and control sites, in three small towns in Tuscany (Central Italy), we found that the soft tissues of *P. papillaris* (purged of the gut contents) showed the highest Cd, Cu and Zn concentrations at the most trafficked sites. Data from faeces suggested that this species ingests large amounts of inorganic particles scraped from the stone surfaces, as indicated by the very high Al, Cr, Fe, Mn, and Pb concentrations. Most lithophilic elements and Pb are scarcely absorbed in the snail digestive tract and soft tissues mainly accumulate Cd and essential elements such as Cu, Zn and Mn. Although the chemical composition of the shells was characterized by low concentrations of all analyzed elements, the samples collected at the most trafficked sites had significantly higher Cu, Fe, and Zn concentrations. This bioaccumulation in *P. papillaris* shells likely remains after death, potentially providing a historical record of the snail exposure to metals over lifetime.

TU364

Metals Distribution in Urban Garden Soils in Greater Victoria, BC, Canada

M. Dodd, Royal Roads University / School of Environment & Sustainability

This study was conducted to determine heavy metal distribution in surface soils in residential and community garden soils in Greater Victoria, BC, Canada Over 500 soil samples were collected ffrom 190 residential and community gardens and analyzed using a portable XRF. A subset of the samples were analyzed by ICP-MS. A comparison of the XRF metal concentrations to to the ICP-MS data indicated that the XRF was a suitable technique for the rapid analysis of the large number of samples collected. Elevated concentrations of metals including Pb, Cu, Cr and Zn, were found in some garden soils. Based on homeowner interviews, historical maps and archival reviews, Pb contamination was primary attributed to the use of leaded paints and housing maintenance practices. Potential sources of the other metal contaminants included the use of wood preservatives, septic fields, automotive repair and old orchards. An interactive map of metal distribution based on the data obtained was developed and made available to the public. Metal bioavailability was assessed using an in vitro bioaccessibility assay and the data used to assess the risk associated with soil ingestion. The estimated daily intake was determined for each element incorporating metal bioaccessibility data. Using the median concentrations, the calculated EDI values were well below the respective tolerable daily intake suggesting that the risk associated with ingestion of metal contaminants were minimal. Dandelion samples were also collecetd and analyzed as surrogate plants to determine potential metal uptake. Metal bioaccumulation factors and translocation factors for the dandelion samples also suggested that the potential for the uptake of the metals studied was low. However there were isolated gardens with elevated Pb concentrations which were identified as being of concern. Recommendations for limiting Pb exposure in these gardens were provided to the homeowners.

TU365

Soil quality analysis, a lever for identifying sources of trace elements and managing urban allotments for urban agriculture production

M. Legras, C. Dubois, L. Castel, UniLaSalle Campus Rouen / Aghyle Unit

Projects seeking to produce home-consumption vegetables in urban areas and, more generally, to set up agricultural production inside cities, are increasingly numerous. Agglomerations and local public authorities also need references on thresholds of contamination in trace element and their transfer into plants. At European level, soil quality regulations are not homogeneous; in France, the legislation is mainly based on the contents in vegetables or fruits, the link with the soil never made. Some studies have highlighted the potential risk of metal contamination of vegetables grown in urban areas and the lack of site-specific risk assessments. However, experimental trials are still lacking on the potential of using urban soil as a good substrate for producing vegetables for domestic consumption. We assessed the quality of the soil on a site in the Rouen agglomeration (Normandy, France) for three uses: pasture, a forest recreational area and market gardening area. However, the city raises questions about the future management of this last area already cultivated for many years. We have analyzed the main physicochemical characteristics of soils, the trace elements (Pb, Cu, Ni, Zn, Cd, Hg) in certain vegetables and fruits and in soils, as well as the history of agricultural practices since the sixties and the topography of the site. Transfer coefficients in consumption vegetables have also been quantified. Our results showed that the zones had a physical and chemical heterogeneity due to the effects of the different cultivation techniques used, the urbanization and the topography of the site. Some metals contents are often above the recommended limits, and soil conditions (pH = 8) significantly reduce the mobility of metals. The concentration of Pb in some of the cultivated area samples was above limits, which makes gardening practices unsuitable for the area. Our results demonstrate that site-specific studies are needed before planning urban cropping areas, and educating urban gardeners about sustainable cropping techniques is a priority for safe feeding.

TU366

Vertical movement of PCBs in agricultural soils impacted by an historical contaminated site: using SoilPlus model to predict discharge, dynamics of movement in soil, and rhizoremediation potential

A. Di Guardo, University of Insubria / Department of Science and High Technology; G. Raspa, Sapienza University of Rome / Department of Chemical Engineering Materials and Environment; E. Terzaghi, University of Insubria (Como) / Department of Science and High Technology, Como; S. Borin, University of Milan / DeFENS; F. Mapelli, University of Milan; E. Zanardini, C. Morosini, University of Insubria / DSAT; S. Armiraglio, Municipality of Brescia / Museum of Natural Sciences; V.M. Sale, S. Anelli, P. Nastasio, ERSAF

Among the national priority polluted sites, the SIN Brescia Caffaro is located in a mid size city Brescia, (200,000 inhabitants) in northern Italy. The site derived from the activity of the former Caffaro s.p.a., a chemical factory among the largest former polychlorinated biphenyls (PCBs) producer in Europe, which produced such chemicals for more than 50 years up to mid 80'. About 100 Ha of agricultural areas were contaminated by a mix of Persistent Organic Pollutants (POPs, mostly PCBs, dioxins, furans) and heavy metals (Hg, As) in variable concentrations, often exceeding the safety values. Contamination mostly resulted because of runoff irrigation with contaminated waters. PCBs were measured in three different former agricultural areas and in three different points per area, in vertical cores up to 1 m depth. The resulting samples were representative of 0-10, 10-20, 20-30, 30-40, 40-60, 60-80, 80-100 cm. The results of concentration measurements with depth (for about 80 PCB congeners) confirmed a general tendency of PCBs to be confined to the upper 40-60 cm (depending on the congener). For example, in field A, PCB 28 ranged from 150 to 250 µg/kg in the top 30 cm to about 0.6 µg/kg at 1 m depth; PCB 209 (peculiar of Caffaro production) ranged from 15000 to 13000 µg/kg in the top 30 cm, descending to about 13 µg/kg at 1 m depth. A gradient was also observed along the runoff water flow direction. These concentrations of PCBs were then compared to those obtained by the SoilPlus model (a multilayered dynamic multimedia fugacity model) and used to predict discharge amounts and conditions regulating vertical movement. The objective was to reconstruct soil concentration profile during the historical contamination to predict discharged amount, potential for additional vertical movement, and conditions regulating chemical bioavailability for future PCB rhizoremediation.

TU367

Metals and metalloids in inhalable fractions of urban road dust

C.L. Wiseman, University of Toronto / School of the Environment; J. Nui, C. Levesque, P.E. Rasmussen, Health Canada

Road dust is highly enriched with metals and metalloids such as Cu, Sb and Zn, due to road surface attrition and wear of automotive components. Despite the importance of road dust as a source of inhalable particles (< 10 µm), little has been published regarding elemental enrichment patterns in particle sizes relevant to inhalation exposures. The goal of this study is to evaluate the contribution of road dust to airborne particulate matter, focussing on metals and metalloids in the inhalable particle size range. Road dust samples were collected from a variety of street types in 2015-2016 in collaboration with the City of Toronto, representing a total road length of about 840 km. Two types of samples were generated by the regenerative-air sweepers: the bulk hopper debris and finer dust box samples. The 50th percentile particle size diameter of the dust box samples was determined by laser analysis to be 9.4 µm, which represents the inhalable fraction. A total of 64 samples (32 inhalable and 32 bulk samples) were subjected to a 4-acid digestion (HF, $HClO_4$, HNO_3 and HCl) followed by multi-element determination using Inductively-Coupled Plasma Mass Spectrometry (ICP-MS). Results showed that the inhalable fractions of road dust were enriched with metals and metalloids relative to the bulk debris, including Cd (0.55 vs. 0.25 µg/g), Zn (649 vs. 252 µg/g), Sb (8.2 vs. 2.2 µg/g) and Pb (80 vs. 54 µg/g). The enrichment of elements of known toxicity in the inhalable fraction is of particular concern, given the bioaccessibility of this particle size range. Available data on the total weight of road dust collected by the City of Toronto each year, combined with the elemental concentrations of the road dust determined in the present study, provides the means to calculate annual flux estimates. For example, Pb loadings in the inhalable fraction alone are estimated to range between 70 kg/yr and 141 kg/yr, which is a significant source relative to the city-wide total of 513 kg/yr atmospheric Pb emissions reported by industries and municipal facilities in the National Pollutant Release Inventory. This example demonstrates the importance of non-exhaust forms of traffic emissions which have not been included in emission inventories to date. Next steps will involve the collection of whole, unfractionated road dust samples from a variety of road types for particle size distribution analyses to refine elemental loadings in each size fraction.

TU368

Sequential extraction and particle size distribution of Cd, Cu, Pb and Zn in street dust of Belgrade (Serbia)

T. Djordjevic, Faculty of Chemistry, University of Belgrade; N. Zaric, Innovation Center of the Faculty of Technology and Metallurgy, University of Belgrade; T. Solevic Knudsen, IChTM / Department for Chemistry

The aim of this study was to investigate the differences in distribution of Cd, Cu, Pb, and Zn in mobile phases extracted from different size-fractions in street dust particles from Belgrade, the Capital of Serbia. The metals investigated were chosen as the most significant pollutants according the European Environment Agency. The street dust samples were collected in summer 2016 at three different locations. The samples were fractioned into three sizes with diameters of: < 63 µm, 63 – 250 µm, and 250 – 500 µm. From different size fractions the metals were isolated into three fractions using a modified sequential extraction procedure after Tessier (Tessier at al., 1979): adsorptive and ion-exchangeable phase (using ammonium acetate); moderately reductive phase (using ammonium oxalate and oxalic acid); and organic sulphide phase (using hydrogen peroxide acidified with nitric acid). These fractions were analysed by inductively coupled plasma optical emission spectrometry (ICP-OES) using an ICP iCap6500Duo-Thermo Scientific instrument. The results showed that the concentrations of the metals (based on the sum of these three fractions) were in the following order: Zn > Cu > Pb > Cd. Comparison with the Serbian national Regulations demonstrated that the concentrations of Cu and Zn in these three fractions were higher than the Maximum allowed values at some locations only, indicating serious contamination with these metals at some locations. The phase partitioning study revealed that Zn and Cu were bounded mainly in the second phase. Pb and Cd were predominantly associated with the second phase. However, Pb was in one sample predominantly associated with the third phase while Cd was in one sample predominantly associated with the first phase. Detailed analysis of distribution of metals in different size fractions did not indicate any patterns suggesting a different origin of these metals at different locations. References: Regulations about allowed quantities of dangerous and harmful matters in soil and irrigating waters and methods about their analysis, Official Gazette of the Republic of Serbia, No. 23/94 (in Serbian) Sequential extraction procedure for the speciation of particulate trace metals. A. Tessier, P. G. C. Campbell, and M. Bisson. Analytical Chemistry, 1979, 51 (7), pp 844–851

TU369

"New" OPEs: isopropylated, tert-butylated and di-tert-butylated Triarylphosphate Isomers in E-waste, House, Car and NIST SRM Dust

L. Jantunen, Environment and Climate Change Canada; T.F. Rodgers, University of Toronto / Chemical Engineering and Applied Chemistry; V.H. Arrandale, Cancer Care Ontario; S. Bernstein, Environment and Climate Change Canada; J.

Guo, Indiana University - Bloomington / SPEA; S. Harris, Cancer Care Ontario, University of Toronto / Population Health and Prevention, Prevention and Cancer Control; P. Kurt-Karakus, Bursa Technical University / Environmental Engineering Department; L. Melymuk, Masaryk University / RECETOX Research Centre for Toxic Compounds in the Environment; L.V. Nguyen, University of Toronto - Scarborough / Department of Physical and Environmental Sciences; M.E. Shoeib, Environment Canada / Atmospheric Science and Technology Directorate; T. Shoeib, American University of Cairo; W. Stubbings, Indiana University - Bloomington / School of Public and Environmental Affairs; C. Turgut, Adnan Menderes University / Environmental Toxicology and Biotechnology; M. Venier, Indiana University / SPEA; G.M. Webster, Simon Fraser University / Faculty of Health Sciences; C. Yang, M.L. Diamond, University of Toronto / Department of Earth Sciences

Organophosphate esters (OPEs) have been measured at relatively high concentrations globally in remote to urban locations with highest levels in indoor environments. Studies of OPEs often comment that these are "new" compounds, citing their use as replacements of some now-restricted brominated flame retardants. Here, "even newer" organophosphate esters were investigated in this study of dust samples from several locations with varying site types. The sample locations and types tested here include dust from Canada (houses from Vancouver and Toronto), Turkey (homes and offices from Istanbul), and Egypt (homes, offices and cars from Cairo), dust from an e-waste facility in Toronto, Canada, and NIST house dust (SRM 2583-2585 from American homes). The new OPEs investigated include several isopropylated triarylphosphates (ITP) and tert-butylated triarylphosphates (TBTP) and a di-tert-butylated triarylphosphate and di-tert-butylated triarylphosphite. These compounds are used as flame retardants, but are also used in hydraulic fluids and as plasticizers. ITP is primarily used in foam and is a component of Firemaster 550 whereas TBTP is in Firemaster 600. Preliminary results indicate ITPs and TBTP levels are found in the ng/g range in these dust samples but at levels lower than the typically analyzed OPEs compounds such as tris(2-butoxyethyl)phosphate (TBEP), tris(2-chloroethyl)phosphate (TCEP) and tris(chloropropyl)phosphate (TCPP). Even though these are new to us and only recently included on some national regulation lists, these compounds are found in NIST SRM dusts that were collected in the mid-1990s thus have been high production volume chemicals for many decades. These compounds are of concern because they have long range transport potential as one ITP isomer was sought and found in arctic water and sediment. A few studies have shown that humans are exposed ITPs due to detection in wristbands and metabolites in urine. These compounds are also of concern due to their reported toxicity especially related to neurological effects. \n

TU370
OXIDATIVE POTENTIAL OF PARTICULATE MATTER COLLECTED AT INDUSTRIAL AND URBAN SITES
g. simonetti, E. Conte, Sapienza University of Rome; c. perrino, CNR Institute of Atmospheric Pollution Research / Institute of Atmospheric Pollution Research; L. Massimi, Sapienza University of Rome / Environmental Biology; S. Canepari, Sapienza University of Rome / Chemistry
The scientific world is still questioning about the effects of airborne particulate matter (PM) concentration and composition on human health, and different scientific approaches have been evaluated in order to gain information about it. The measurement of the oxidative potential (OP) is generally considered as a predictive index of PM ability to generate reactive oxygen species (ROS) in biological organisms, and different acellular assays are currently used in literature for its determination. In this work we applied three of the most used OP assays (dithiothreitol - DTT, acid ascorbic – AA, and 2′,7′-dichlorofluorescin –DCFH; Fang et al., 2016, Huang et al., 2016) to $PM_{2.5}$/PM_{10} samples and to size-segregated samples collected by a 10-stage impactor. Samplings were performed at an industrial site near Ferrara (Po Valley; Italy) and at a traffic urban site in Rome (Italy). All the samples were also analysed for anion, cations, macro- and micro-elements, total organic content, elemental carbon and water-soluble organic carbon, in order to identify the relationships between OP values and PM chemical composition and dimension. Despite the very different composition of PM in the two monitored areas, OP values were scarcely dependent on the sampling site: species whose concentration is very different in the two areas, such as secondary inorganic ions, seem thus to play a negligible role in the ROS generation. Each assay showed a different sensitivity towards the oxidant species: the DTT method was more sensitive to organic substances, while the AA method was more sensitive towards dusts rich in metals and metalloids. Furthermore, the DTT assay evidenced a greater affinity with particles in the fine mode, while AA responded mainly to particles in the *course* fraction. DCFH results appear to be driven by a competition between several factors, some increasing the response and some suppressing it. Fang, T. et al.: Oxidative Potential of Ambient Water-Soluble PM2.5 in the Southeastern United States: Contrasts in Sources and Health Associations between Ascorbic Acid (AA) and Dithiothreitol (DTT) Assays. *Atmos. Chem. Phys.* **2016** 16, 3865–79. 10.5194/acp-16-3865-2016 Huang, W. et al: Optimization of the Measurement of Particle-Bound Reactive Oxygen Species with 2',7'-dichlorofluorescin (DCFH), *Water Air Soil Pollut.* **2016**, 227, 164. 10.1007/s11270-016-2860-9.

TU371
Chromatographic determination of the pathway of nevirapine in wastewater at a wastewater treatment plant
S. Nzube, Cape Peninsula University of Technology / Department of Chemistry; C. Van der Horst, University of The Western Cape / SensorLab Department of Chemistry; V.S. Somerset, CPUT / Chemistry
Emerging pollutants (EPs) are defined as substances that have been detected, which is not included in the routine monitoring programmes at the EU level, and whose fate, behaviour and ecotoxicity effects are not well understood. Pharmaceuticals used in the treatment of HIV, known as anti-retrovirals, are becoming prevalent and there is a need to quantify these pollutants and minimise any adverse affects to aquatic and human health. Nevirapine (NVP) is commonly used in the anti-retroviral treatment of HIV infection. It is known as a non-nucleoside reverse transcriptase inhibitor of the dipyridodiazepinone class, commonly used to minimize viral resistance. This study reports the isolation and chromatographic characterisation of NVP in wastewater samples, with concentrations ranging between 250 to 500 ng/L. Further evaluation of the impact of NVP on the aquatic ecosystem was also considered and reported in this study. **Keywords:** Anti-retrovirals; Emerging pollutants; Chromatography; Wastewater

TU372
Leucomethylene blue: a selective photometric reagent for chlorine dioxide analysis in water
R. Devesa, Aigues de Barcelona / Chemistry. Laboratory; F. Estrany, Universitat Politecnica de Catalunya UPC; A. Garbayo, Agbar, Barcelona Water Company; X. Aldazabal, Polytechnic University of Catalonia
Drinking water supply companies have traditionally focused their efforts on providing a product with health guarantees, a safe and clean water. Disinfection has been the main purpose of the water treatment. A broad range of disinfectants and technologies are available. Chlorine dioxide has been increasingly used because of its ability to avoid the formation of trihalomethans (THMs), the most common and well-known disinfection by-products. N,N-Diethyl-p-phenylenediamine (DPD) method has been without any doubt the most common procedure for the analysis of free and combined chlorine (chloramines) and also, chlorine dioxide. While this method easily permits the differentiation between free and combined chlorine, the selectivity of the analysis of chlorine dioxide when chlorine is present has recently been questioned. As a result, this procedure has been removed from Standard Methods (American Water Works Association) and qualified as "reserved" method. Given this circumstance and the need of having a selective method for chlorine dioxide, several UV-VIS spectrophotometric methods have been evaluated by our group (1). Here, the results using leucomethylene blue are presented. This chromophore agent is obtained by reduction of methylene blue and its use for the analysis of chlorine dioxide is scarcely described in the literature. Our experiments show that it presents a good analytical performance, and what is more important, measurements are not interfered by elemental chlorine. The method showed a good accuracy with real water samples (relative error below 14 % for chlorine dioxide concentrations between o and 1.5 mg/L. This reagent has revealed to be the best option among the different compounds that we have used – amaranth, lissamine green, and choro phenol methyl red-. The only drawback of the procedure is the interference by high concentrations of sulphate. Ongoing work is taking place to avoid it by previous precipitation of the interferent or liquid extraction of the dye with an organic solvent once the colour has developed. (1) P. López et al. Chemical and sensory analysis of chlorine dioxide in drinking water. Part II. SETAC Europe 26th Annual Meeting. May 2016. Nantes (France).

TU373
Fate and effects of triclosan in subtropical freshwater benthic microcosms
F. Peng, Wageningen UR; P. van den Brink, Alterra/Wageningen UR / Aquatic Ecology and Water Quality Management Group b Alterra; G. Ying, Guangzhou institute of Geochemistry Chinese Academy of Sciences; H. Selck, Roskilde University / Dept Science and Environment; D.T. Salvito, Research Institute for Fragrance Materials (RIFM) / Dept of Environmental Science; N. Diepens, Wageningen University
Triclosan (TCS) is one of the top 10 most commonly detected organic pollutants in the aquatic environment. However, little is known about the toxicity and bioaccumulation of sediment-associated TCS. We examined the effect of sediment-associated TCS on the snail, *Viviparidae Bellamya*, and the worm, *Limnodrilus hoffmeisteri*, and assessed worm bioaccumulation during a 28 days exposure period in microcosms mimicking a subtropical freshwater sediment system. The results showed that TCS was detected in worm tissue with biota sediment accumulation factor (BSAF) values ranging between 0.67-6.3, suggesting that TCS could be accumulated in worms. The results of mass balance assessment showed that, during the experiment period, TCS amount in the microcosm was reduced 3.4% to 11.4% and 3.5% to 10.9% in the systems with and without macroinvertebrates, respectively. Based on the experimental conditions used, we conclude that sediment-associated TCS (8 µg/g dry weight (dw)) is unlikely to affect, at least in the short term, survival and growth of snails and worms in sediments, with no observed effect concentrations (NOECs) of 8 µg/g dw for both

organisms.

TU374
Joint Annual Meeting of the International Society of Exposure Science and the International Society for Environmental Epidemiology (ISES-ISEE 2018)
M.L. Diamond, University of Toronto / Department of Earth Sciences

Challenges in setting, meeting and measuring specific protection goals for plant protection products (P)

TU375
French Phytopharmacovigilance: a national scheme for monitoring the adverse effects of plant protection products
F. Botta, ANSES / DER; F. Eymery, T. Quintaine, M. Hulin, J. Rety, O. Yamada, M. Merlo, ANSES
Phytopharmacovigilance is the latest complement to ANSES's existing missions concerning the assessment of the risks associated with plant protection products before marketing, and the issuing and withdrawal of marketing authorizations. It is also fully in line with the third component of the Ecophyto plan. This national plan, established for the first time in 2008, was recently renewed. It aims to reduce pesticide uses in accordance with the requirements of Directive 2009/128, establishing a framework for Community action to achieve the sustainable use of pesticides. A specific scheme for funding these studies is planned through a tax on sales of plant protection products payable by the marketing authorisation holders. To meet this objective, phytopharmacovigilance relies on three fundamental and complementary methods of data collection and knowledge production: a network of surveillance or vigilance bodies, collection of spontaneous reports and *ad hoc* studies on the adverse effects of plant protection products. These studies are financed by PPV to meet three different needs: 1) when the information provided by the surveillance and vigilance bodies is seen to warrant clarification, 2) to investigate spontaneous reports or 3) to collect new data / information. To identify the adverse effects of plant protection products on biodiversity and ecosystems, Phytopharmacovigilance is based on the systematic and regular collection of information produced by the existing surveillance and vigilance bodies, covering risks and impacts on wildlife, crops, fauna, flora, air, water and soil. In the full article (2 pages word/pdf), a detailed list of studies and network working on "Pesticides impacts on biodiversity" and "Monitoring of pesticides (water, air, etc..)" is described.

TU376
Measuring and Modeling Aluminium Bioavailability and Toxicity to Aquatic Organisms
W.J. Adams, Red Cap Consulting; P. Rodriguez, PHR Consulting; B.A. Stubblefield, Oregon State University / Environmental and Molecular Toxicology; A.S. Cardwell, Oregon State University / Faculty Research Assistant; D.K. DeForest, Windward Environmental LLC; R. Gensemer, GEI Consultants / Ecological Division; E. Nordheim, European Aluminum
The ability to accurately predict the aquatic toxicity of aluminium (Al) in natural surface waters has eluded scientists for the past several decades. In 1988, the US Environmental Protection Agency (USEPA) released nationally-recommended ambient water quality criteria for Al of 750 and 87 µg/L as acute and chronic criteria, respectively. However, these applied only to waters with a pH between 6.5 and 9, and the chronic toxicity database was limited. Therefore, in 2009 we assembled a team of scientists to help expand this database and identify a means for measuring and predicting the toxicity of Al to aquatic organisms as a function of water chemistry. A series of chronic toxicity tests were performed, as part of this effort, with several freshwater species. The species were selected to meet requirements for the EU REACH dossier, USEPA water quality criteria or European Water Framework guidelines for environmental quality standards. To develop bioavailability models, multiple tests with a green alga (*Pseudokirchneriella subcapitata*), a cladoceran (*Ceriodaphnia dubia*), and a fish (*Pimephales promelas*) were performed across a range of DOC, hardness and pH conditions. These latter data were included in the development of a biotic ligand model (BLM) for the prediction of toxicity as a function of water chemistry. The toxicity data sets were also used to develop a multi-linear regression (MLR) model to provide a simplified means to predict toxicity as a function of DOC, hardness, and pH. Due to its complex environmental chemistry, measurement of the "toxic" form of Al in natural waters cannot be performed using the conventional "total" or "dissolved" analytical approaches. Studies have recently been completed which allow for the measurement of "bioavailable" Al in natural waters where suspended solids are present and contribute to measurements of total Al, but are non-toxic. The presentation will focus on modeling Al toxicity and measuring Al in natural waters.

TU377
Modelling impacts of chemicals on ecosystem services
N. Galic, Syngenta Crop Protection, LLC / Environmental Safety; V. Forbes, University of Minnesota / Ecology, Evolution & Behavior; C. Salice, Towson University / Environmental Science & Science Dept.; P. Thorbek, Syngenta / Environmetal Safety
Protection goals for the ecological risk assessment of chemicals are increasingly being framed in terms of ecosystem service delivery [1]. However, the type of data collected to assess risk is generally at the level of individual organisms or simplified multi-species systems. Currently, extrapolation from what is measured to what we want to protect uses overly simplistic approaches, such as risk quotients or toxicity-exposure-ratios. Ecological models provide a more mechanistic way of connecting these disparate levels and allow for integration of other relevant information as well as feedbacks across levels of organization [2]. Here we present output from the National Institute of Mathematical and Biological Synthesis (NIMBioS) working group (www.nimbios.org/workinggroups/WG_o2e). The two case studies provide a demonstration of a recently developed framework that allows quantitative and mechanistic linkages of the effects of chemicals and other stressors from impacts on individual organisms to the delivery of ecosystem services [3]. The first case study applies an individual-based model (IBM) [4] to quantify impacts of potential endocrine disrupting chemicals on services provided by trout populations in a mountain stream in Colorado, USA. The second case study uses an aquatic ecosystem model [5] to evaluate impacts of an insecticide on multiple ecosystem services delivered by a lake ecosystem, modified to represent a reservoir in Iowa, USA. The first case study is an example where managing for provision of the service GCT population provides will differ depending on the level of EE2 in the system. If EE2 concentrations are low, then management would need to focus on controlling BT populations. For high concentrations of EE2, management first needs to focus on reducing EE2, followed by control of BT. The second case study is an example where game fish species responded differently to exposure to the insecticide, as a result of interspecific interactions, and the economic valuation of this service needs to take into account with angler preferences. The service of water clarity for recreational activities was valued using threshold-based estimations of days fit for recreation. We provided concrete examples of how ecological modeling can be used to quantify impacts on ecosystem services from data gathered in standard testing. We discuss challenges and ways forward.

TU378
Sulphur: conflicting protection goals
G. Brouwer, Delphy / team fruitteelt; F.M. Bakker, Eurofins-Mitox
Sulphur is a key fungicide in biological fruit production. Following a recent risk assessment, the use of sulphur in The Netherlands has been rigorously limited. Current registrations allow for two applications, which is incompatible with disease control in biological top fruit production. The regulatory decision was technically correct and based on considerations for non-target arthropods, as risk for the egg parasitoid *Trichogramma* could not be excluded. Under current European regulations *Trichogramma* is one of the sensitive indicator species selected for higher tier testing. As this is a natural enemy of several Lepidopteran pest species, the indicator is clearly linked to the specific protection goal of preserving natural pest control potential. However, in the absence of alternatives, at a broader level this specific objective frustrates the current system of biological production as a whole. The specific protection goal must clearly be balanced against the broader objective of preserving biological production. To understand the importance of egg parasitoids such as *Trichogramma* in Dutch orchards, the Dutch Fruit Growers Organisation have investigated the control potential of egg parasitoids in 8 orchards (4 biological, 2 conventional and 2 untreated), using parasitization rate as a functional endpoint. The investigations show that parasitization rates are extremely low (< 1% of bait cards and < 0.005% of the host eggs showed parasitization), suggesting a minor role of *Trichogramma* and other egg parasitoids in these systems in The Netherlands, at least at the time of the study (late summer 2017). Conventional orchards showed no parasitization and regular (i.e. having sulphur treatments) and untreated orchards did not show differences. These results show that specific protection goals may have country specific weight and need to be considered and balanced against potential negative impacts of eventual protective measures, such as in this case jeopardizing biological top fruit production.

The Need for Resilience in Environmental Impact Assessment (P)

TU379
Recovery in environmental risk assessments at the European Food Safety Authority (EFSA)
T. Brock, Alterra, Wageningen University and Research Centre / Environmental Risk Assessment Team; F. Bigler, Retired; G. Frampton, University of Southapton; C. Hogstrand, Kings College London / Division of Diabetes and Nutritional Sciences; R. Luttik, Retired; F. Martin-Laurent, INRA Dijon; C.J. Topping, Aarhus University / Department of Bioscience; W. Van der Werf, Wageningen University; A. Rortais, European Food Safety Authority
The European Food Safety Authority (EFSA) performs environmental risk assessments for single potential stressors such as plant protection products, genetically modified organisms and feed additives and for invasive alien species that are harmful for plant health. In 2015-2016 a Working Group of the Scientific Committee of EFSA (the authors of this abstract) explored how ecological recovery is covered under current single-stressor Environmental Risk Assessment (ERA)

schemes at EFSA and how recovery could be assessed considering the complexity of the environment. An important aim of these activities was to promote a dialogue between different panels of EFSA and risk assessors and risk managers responsible for the food and feed chains. Another important aim was to provide risk assessors with a conceptual framework to address ecological recovery in ERAs for any assessed products, and invasive alien species that are harmful for plant health. This framework proposes an integrative approach based on well-defined specific protection goals, scientific knowledge derived by means of experimentation, modelling and monitoring, and the selection of focal taxa, communities, processes and landscapes to develop environmental scenarios to allow the assessment of recovery of organisms and ecological processes at relevant spatial and temporal scales. Due to the complexity of ecological systems and the need to evaluate effects and recovery of ecological attributes in spatial and temporal dimensions, a systems approach is required. The systems approach allows the integration of the various species, environmental factors, scales, and stressor-related responses necessary to address the context dependency in ecological recovery. The presentation will highlight the most important conclusions, challenges and recommendations to appropriately address ecological recovery in ERA for potential stressors that fall under the remit of EFSA EFSA Scientific Committee, 2016. Recovery in environmental risk assessments at EFSA. EFSA Journal 2-016; 14(2):4313. 85 pp

TU380

Habitat Equivalency Analysis for a Restoration Resilience Model of the Rio Doce Basin

P.N. Booth, Ramboll Environ / Ecological Sciences; E. Singer, F. Gomes, R. Arantes, Ramboll Environ / Sao Paulo; R. Wenning, Ramboll Environ

A Resilience Model was prepared to support environmental, economic, and social restoration of the Rio Dôce Basin after the Fundão Dam failure in Mariana, Minas Gerais State, Brazil. A Habitat Equivalency Analysis (HEA) was applied to quantify lost ecosystem services resulting from the disaster and was intended as a means of scaling the total terrestrial habitat restoration required. The HEA is being refined through a process of stakeholder engagement to determine the valued environmental components (VECs); and thus the valued ecosystem services that should drive the resilience restoration goals for each reach of the Rio Dôce River. HEA is a well- accepted spatially and temporally explicit method for integrating multiple complex and difficult to measure environmental variables into a few metrics to determine overall losses and gains in ecosystem services resulting from impacts or restoration actions. The HEA method is adaptable to any ecosystem and its flexibility allows for variability in the valuation of ecosystem services between communities and cultures. This paper focuses on the development and application of HEA within the context of the Resilience Model, and how selection of VECs as indicators to guide the focus, scale, and location of restoration is aimed at developing an overall ecosystem restoration program that is at once cost-effective and results in a more resilient Rio Dôce basin.

TU381

Using risk and recovery information in environmental cost-benefit analysis for determining appropriate risk management actions at major industrial facilities

A.E. Bartram, Ramboll Environ / Product Safety, Ecology and Sediment Management; S. Deacon, Ramboll Environment & Health Limited

Operators of chemical manufacturing plants and fuel storage depots are required to undertake site safety assessments, with specific emphasis on the control of major environmental accident hazards (SEVESO Directive 2012/18/EU). Many facilities are in close proximity to water bodies, the coast, and or protected conservation areas. Site safety reporting requires an initial risk assessment be undertaken. The assessment draws upon both the potential severity of harm and environmental recovery in order to determine the risk tolerability under accidental release scenarios. Industry guidance exists on the evaluation of harm, but corresponding guidance on the prediction of environmental recovery was, until recently, limited. On behalf of the Energy Institute, Ramboll Environ developed a guide for risk assessors to determine the environmental recovery duration following major accidents of releases of SEVESO substances. Published October 2017, the guide provides a step-wise framework to identify an appropriate recovery duration based on the chemical(s) of interest and types of habitats on and around a site. It includes consideration of key factors affecting recovery and the fate and effects of SEVESO III chemicals, such as PBT substances with the potential for longer-term impacts/delayed recovery. If an assessor determines that risks are intolerable, then the regulator requires facilities to consider appropriate investment in order that risks are as low as reasonably practicable. Ramboll Environ has pioneered a methodology based on ecosystem services concepts to evaluate the costs and benefits of potential risk management options. The method is used to evaluate the risk in terms of potential ‘damage avoided’ by putting risks into a socio-economic context. Case study examples will be provided where a range of infrastructure upgrade options at fuel storage depots are compared. Environmental Cost Benefit Analysis is used to determine if the upgrade would be grossly disproportionate to the benefit that the upgrade would provide, i.e. the damage avoided. The studies incorporate site-specific baseline ecology, other receptors and ecosystem services provided to society. The poster will also reflect upon lessons learned by both regulators and industry in the method development. References: Energy Institute (2017). Guide to predicting environmental recovery durations from major accidents. Supporting guide to the Environmental risk tolerability for COMAH establishments guideline

TU382

Addressing Resilience in Ecosystem Services Assessment

K. Mustajärvi, Ramboll E&H / Ecological Services; R. Wenning, Ramboll Environ / Ecology & Sediment Management; E. Bizzotto, H.R. Diogo, Ramboll / Ecological Services

An ecosystem services approach to landscape and nature restoration planning and damage assessment should fully account for all aspects of the environment and the human well-being derived from protection, enhancement and repair to natural resources caused by natural or human-caused events. With respect to damage assessment, conceiving of a resource as a part of an ecosystem that supplies valuable goods and services to people provides a basis for measuring changes and for valuation of those goods and services. With respect to landscape and nature restoration planning, an ecosystem services approach can lead to innovative ideas for ecological infrastructure in cities and promote ecological rehabilitation that may be ecologically and socially desirable and also economically advantageous. Resilience, however, is a key consideration that, to date, has been inadequately considered in ecosystem services assessment work. Considerations of resilience are especially important in ecosystems, because increasing resilience can reduce the risk that highly valued goods and services will cross critical thresholds and irreversibly degrade or change. Resilience also plays an important role in maintaining conditions that will sustain the provision of ecosystem services; a resilient ecosystem will stabilize and recover quickly to a prior or new baseline condition such that the flow of goods and services can be assured. This paper discusses three technical challenges that must be overcome for incorporating resilience in ecosystem services planning work. First, baseline conditions must be established for the goods and services produced by the ecosystem prior to altering the environment or repairing the damage caused by natural or human-caused disasters. Associated with this challenge is the added complication of shifting baselines in the context of climate change, which generates considerable uncertainties for projecting future recovery of services. The second challenge relates to the establishing the relative values of different ecosystem services, and projected changes to pricing and value by society in the future. The third challenge is the limited ability of current ecosystem models to provide defensible projections of the complex and intertwined social-ecological relationships defining a future sustainable flow of goods and services.

TU383

Use of cost modelling techniques to manage environmental subsurface risks, liabilities and uncertainties in Spain

P. Wouters, M. Ferreira, I. Harper, Ramboll Environ / Environment and Health; . Salinas, Red ElÃctrica de EspaЂa

Companies owning large portfolios of properties are often faced with a high degree of uncertainty in relation to the subsurface conditions of their sites. This makes it difficult, if not almost impossible, to develop and implement a rational and cost-effective strategy to manage their regulatory obligations and financial liabilities. A customised probabilistic risk model was developed to facilitate the management of environmental and reputational risks at a portfolio of over 500 industrial sites in Spain. The sites are equipped with industrial equipment that contains large amount of oils. The equipment differs significantly in age, size, design and make, and is equipped with non-standardised spill containment systems. The sites are distributed throughout the Spanish mainland and in the Baleares and Canarias islands, and their environmental and social settings show a large variability. Site investigations were known to have been performed at only 5% of the sites. Initially an Environmental Risk Assessment Model was developed using technical, regulatory, social and environmental data. The compiled and consolidated data were entered into a calculation model developed in Microsoft Office Excel™.Monte Carlo simulation was used to manage the potential range of scenarios that could be associated with particular assumptions in the model. OracleЂ_s Crystal Ball™ add-on to Excel was used to assign probability distributions to such uncertain model inputs. Probabilistic inputs were considered as risk scenario triggers for specific events at specific sites, such as: the likelihood of historic contamination being detected; a new contamination event being generated either on-site or off-site; a subsoil investigation being triggered; soil remediation being required; active or passive groundwater remediation being required; and implementation of a groundwater monitoring programme being required. Probabilistic inputs were also applied to the various cost scenarios that might be triggered. The model generated an environmental risk ranking, expressed in purely financial terms. Ten high risk and 23 moderately-high risk sites were identified and an environmental action plan focussing on these highest priority sites was prepared. This allowed the portfolio owner to direct financial and human resources required for site investigation, remediation and preventive maintenance to those sites which could give rise to the highest financial and reputational liabilities.

TU384

Quality stakeholder involvement for resilience in environmental risk

assessment

Y. Tomkiv, Norwegian University of Life Sciences (NMBU) / Faculty of Environmental Sciences and Natural Resource Management; B. Wynne, Norwegian University of Life Sciences / Centre for Environmental Radioactivity CERAD CoE; D. Oughton, Norwegian Public Roads Administration / Norwegian University of Life Sciences / Centre for Environmental Radioactivity (CERAD CoE)

There is a global call for sustainability and systemic approaches in environmental risk assessment that consider economic and societal aspects of risk, in addition to the environmental aspect. Early and effective stakeholder involvement plays fundamental role in these considerations. Stakeholder involvement is actively utilized in both policy and research, moreover, it is widely recognized that stakeholders will increasingly influence future consideration of environmental risk and assessment, including decision-making. However, it is important to ensure the quality of the stakeholder involvement activities in order to support democratically legitimate and robust processes. However, the existing systems of criteria tend to be focused on a rather narrow evaluation of the method applied. We argue that there is need for evaluation that goes beyond a simple assessment of the methodology and addresses the wider context that the stakeholder involvement activity is held in. This paper uses two stakeholder engagement events: one from the field of emergency preparedness and one from nanoremediation, to explore the applicability of existing evaluation criteria for a quality assessment of stakeholder or broader public involvement. We use criteria developed by Rowe and Frewer (2000), namely, representativeness, independence, influence, transparency and early involvement. The results of our evaluation showed that current criteria focus too much on the acceptance of the outcomes rather than the process itself and, as such, are not sufficient for assessing the quality of a stakeholder engagement. We will present proposals for extended criteria that address the limitations and highlight the principles for a more democratic stakeholder involvement. Rowe, G., & Frewer, L. J. (2000). Public participation methods: A framework for evaluation. Science Technology & Human Values, 25(1), 3-29.

TU385

Assessment and Management of Radiation Risks following a Nuclear Accident: The Shamisen Project Recommendations

D. Oughton, Norwegian Public Roads Administration / Norwegian University of Life Sciences / Centre for Environmental Radioactivity (CERAD CoE); E. Cardis, ISIGlobal; T. Schneider, CEPN; Y. Tomkiv, Norwegian University of Life Sciences / Centre for Environmental Radioactivity CERAD CoE

The Fukushima Daiichi accident in 2011 represents a poignant reminder of the complex interplay between environment, society and economics. Contamination of both terrestrial and marine ecosystems had wide reaching impacts for the affected populations. While the strict control of foodstuffs ensured that the radiological impacts on human health were minimal, the economic and societal consequences have been enormous. The loss of livelihood from bans on fishing and farming have hit farming and fishing communities, exacerbating the already existing concern for recruitment of younger generations to family businesses. The return of evacuees to decontaminated areas has been low, particularly for families with young children, leading to demographic changes in societies. Other social and cultural impacts arise from lack of access to beaches, places of heritage and festivals. The economic consequences from food bans go beyond thathe loss of sales, market value decreased in all products from the area due to loss in consumer trust (20% decrease compared to the rest of Japan). Strategies for radiation risk management are often at odds with the actual needs of the affected populations, and if not carried out properly can cause more harm than good. Recognising this, the EU SHAMISEN project has published a set of recommendations to improve radiation risk mangement after a nuclear accident. Experience suggested that existing recommendations had a technical focus, with less attention paid to social, ethical, psychological issues and that the information tended to be directed towards the decisions made by experts rather than for support of affected populations. This paper presents the main conclusions and recommendations of the SHAMISEN project. The 28 recommendations promote a management strategy that targets the overall well-being of populations, that addresses not only radiation effects, but also aims to alleviate psychosocial impacts and strengthen stakeholder engagement. \n \n

TU386

SETAC Ecosystem Services Interest Goup

S.E. Apitz, SEA Environmental Decisions Ltd

Air Pollution, Biomonitoring and Human Health (P)

TU387

Assessment of Indoor Radon Concentration and Trace Metals Composition in University Building Microenvironments

M. Mohamed Ali, Universiti Kebangsaan Malaysia; M. Mohd Hanafiah, Universiti Kebangsaan Malaysia / Environmental Science; M. Khan, Universiti Kebangsaan Malaysia / Centre for Tropical Climate Change System

This study measures the indoor particulate matter (PM_{10}) composition and the equilibrium equivalent radon (EEC_{Rn}) concentration in two university buildings with different ventilation systems. A low volume sampler using Teflon filter paper was used to collect the PM_{10} samples and inductively coupled plasma mass spectrometry was used to determine the concentration of heavy metals. The concentration of indoor radon was measured using a radon detector model DOSEman PRO. The potential human health damage due to the inhalation of carcinogenic and non-carcinogenic trace elements was also determined based on the US EPA standard. The equilibrium factor and the annual effective dose on the lung cancer risks of each occupant were calculated and a correlation of the radon concentration was made with the annual inhalation dose of the occupants at the indoor stations. The results showed PM_{10} concentrations recorded in Building 1 and Building 2 ranged between 19.1 to 237 µg m^{-3} and 23.4 to 159 µg m^{-3}, respectively. In Buildings 1 and 2, the principal component analysis and multiple linear regression showed that the main source of pollutants in PM_{10} were from the crustal source (20%) and combustion (21%), respectively. The effective lifetime carcinogenic risks (ELCR) in Buildings 1 and 2 were 1.90E-3 and 1.65E-4, respectively. The hazard quotient (HQ) represents the non-carcinogenic risk, with 7.73 and 6.46 in Building 1 and Building 2, respectively. The average equilibrium equivalent radon measured in Building 1 and Building 2 was 2.33 ± 0.99 and 3.17 ± 1.74 Bqm^{-3}, respectively. The average annual inhalation doses recorded at Buildings 1 and 2 were 0.014 ± 0.005 mSv y^{-1} and 0.020 ± 0.013 mSv y^{-1}, respectively. For trace metals, the ECLR and HQ values were found to exceed the permissible limits suggested by US EPA, whereas the values of equilibrium equivalent radon concentration were still below the standard recommended by ICRP.

TU388

Paradigm for PM2.5 Chemical and Biological Characterization: Paired Home and Personal PM2.5 Samples in Kheri, India

C. Roper, A. Perez, Oregon State University / Department of Environmental and Molecular Toxicology; P. Hystad, Oregon State University / College of Public Health and Human Sciences; S. Simonich, Oregon State University / Depts of Chemistry and Environmental Molecular Toxicology; R.L. Tanguay, Oregon State University / Sinnhuber Aquatic Research Laboratory and the Environmental Molecular Toxicology

The global public health impact from household fine particulate matter ($PM_{2.5}$) is extremely large however, there is a limited understanding of health effects associations with specific $PM_{2.5}$ chemical constituents as well as the underlying mechanisms of these adverse health effects. These research gaps can be addressed through use of a high-throughput screening platform to quickly gain biological response data. A subset of homes in Kheri, India that participated in the Prospective Urban and Rural Epidemiological (PURE)-AIR pilot study were selected to identify differences in chemical and biological measurements of household $PM_{2.5}$. In 6 households, personal air monitors collecting $PM_{2.5}$ were worn by female participants and paired with stationary monitors, resulting in personal (n=6) and home (n=6) $PM_{2.5}$ filters for each household. $PM_{2.5}$ was removed from filters via sonication in methanol. Aliquots of individual filter samples were removed for oxidative potential assessment. Remaining $PM_{2.5}$ samples of the same collection method were then pooled (n=6/group) and the soluble fraction of $PM_{2.5}$ from DMSO extraction was prepared for developmental toxicity testing performed in zebrafish (n=32/treatment) starting at 6 hours post fertilization (hpf). Aliquots of the pooled samples were used for chemical analysis (polycyclic aromatic hydrocarbons (PAHs, n=120), elements (n=20)) and oxidative potential assessment with methods identical to those used for individual filters. Significant differences were observed in oxidative potential between personal and home $PM_{2.5}$ for both individual and pooled samples. Significant mortality in zebrafish was observed starting at 24 hpf in personal $PM_{2.5}$ samples and by 120 hpf in home $PM_{2.5}$ compared to blank filter controls. Chemical analysis is underway to allow for correlations to be investigated between these biological responses and chemical constituents. This research is the first study to use paired home and personal $PM_{2.5}$ samples with chemical, oxidative potential, and developmental toxicity data, identifying the differences in these measurements between household and personal $PM_{2.5}$. Importantly, it outlines procedures for large-scale analysis of the PURE-AIR study which includes planned $PM_{2.5}$ measurements in 4,000 homes and will ultimately allow for correlation of human health effects with chemical and biological data to identify improved health metrics for $PM_{2.5}$ exposures.

TU389

Toxicity of airborne particulate matter as a factor to choose the most convenient school

F. Sánchez Soberón, Universitat Rovira i Virgili / Chemical Engineering; N. Serra, Universitat Rovira i Virgili / Laboratory of Toxicology and Environmental Health; V. Linares, University Rovira i Virgili / Laboratory of Toxicology and Environmental Health; M. Bellés, Universitat Rovira i Virgili / Laboratory of Toxicology and Environmental Health; M. Schuhmacher, Rovira i Virgili University / Departament d Enginyeria Química

One of the critical decisions parents have to face is choosing a good school for their kids. Proximity, high ratio of teachers to students, and access to convenient educative infrastructures are the driving factors determining school's choice. However it is used to assume that environmental air inside schools is safe enough.

Among the different air pollutants found in schools, $PM_{2.5}$ (airborne particulate matter smaller than 2.5 µm; also referred as "fine PM") is considered as the most injurious one. Since this pollutant is potentially very harmful, toxicity of $PM_{2.5}$ on lung cells has been widely studied. However, most of the publications on this topic are focused on studying $PM_{2.5}$ effects on human alveolar cells for short periods of time after applying doses far higher than environmental levels. To surpass this gap, we propose the present study. On it, we collected two fractions of fine PM ($PM_{2.5-1.0}$ and $PM_{1-0.25}$) inside classrooms of schools located under the influence of three different environments: urban, petrochemical, and chemical. Subsequently, we exposed human alveolar epithelial cells (A549) to IC_5 doses obtained from a previous study. Then, apoptosis and release of 17 cytokines from the cells was studied after five different exposure times: 0, 6, 24, 48, and 72 hours. Preliminary results showed no differences in cytokines release among the two PM sizes or three sampling sites. However, differences aroused when comparing the levels of seven cytokines (MCP-1, TNF-α, INF-g, G-CSF, IL-6, IL-7, and IL-8) versus exposure times. These differences became significant after 24-48 h from exposure, and increased till reaching the maximum value after 72 hours. Results from this study will be useful not only to better understand the way of action of $PM_{2.5}$, but also to schools managers and parents.

TU390

Acute Impacts of Extreme Hot Temperature Exposure on Emergency Room

Y. Lan, C. Chang, C. Chung, China Medical University

Abstract The purpose of this study was to assess the effects of extremely high air temperatures on hospital emergency room visits (ER) related to alcohol addiction and other mental illnesses in Taiwan. A time series study was conducted using health and climatic data from 2000 to 2010 in Taiwan. A national health insurance database, temperature database, and air quality surveillance database were used for this study. Relative risks (RRs) for increases in emergency room (ER) visits were estimated for alcohol addiction and other mental illnesses after exposure to extremely hot temperatures (the 99th percentile) while using the 50th percentile of the daily mean temperature as reference. Poisson regression models using a distributed lag non-linear model (DLNM) were used. We adjusted for the effects of humidity and outdoor air pollutants. We found an association between alcohol addiction and other mental illnesses and mean daily temperature at 23.6? on ER visits. The association was strongest within 0–7 days after exposure to hot temperatures. Increases (RR 1.02, 95% CI 1.01–1.04) in major depressive disorder (MDD) ER visits was observed over a cumulative period of 7 days after exposure to high ambient temperature (99th percentile vs. 50th percentile). The opposite association was reported for alcohol addiction (RR 0.99 , 95% CI 0.98–0.99). No significant associations with anxiety, dementia, and delirium were estimated. Our findings suggest that extreme temperatures pose a risk to the health and wellbeing for individuals with alcohol addiction and other mental illnesses.

TU391

Characteristics of Polybrominated Diphenyl Ethers Released from Primitive E-Waste Treatment

T. Li, Jinan University; J. Zhou, C. Wu, L. Bao, L. Shi, E.Y. Zeng, Jinan University / School of Environment

Primitive processing of e-waste potentially releases abundant organic contaminants to the environment, but the magnitudes and mechanisms remain to be adequately addressed. The present study conducted thermal treatment and open burning of typical e-wastes, i.e., plastics and printed circuit boards. Emission factors of the sum of 39 polybrominated diphenyl ethers ($\sum_{39}$PBDE) were 817–1.60 × 10^5 and RL–9.14 × 10^4 ng g^{-1} in thermal treatment and open burning, respectively. Airborne particles (87%) were the main carriers of PBDEs, followed by residual ashes (13%) and gaseous constituents (0.3%), in thermal treatment, while they were 30%, 43% and 27.2% in open burning. The output-input mass ratios of $\sum_{39}$PBDE were 0.21-10 in thermal treatment and 0.01–0.36 in open burning. All PBDEs were largely affiliated with fine particles, with geometric mean diameters at 0.61–0.83 µm in thermal degradation and 0.57–1.16 µm in open burning from plastic casings, compared to 0.44–0.56 and RL–0.55 µm from printed circuit boards. The main emission mechanisms for lightly and heavily brominated BDEs were suggested to be evaporation and mechanical formation, respectively. The difference between the size distributions of particulate PBDEs in emission sources and adjacent air implicated a noteworthy redisposition process during atmospheric dispersal.

TU392

How risky is the schoolyard? An approach from chemical composition of particulate matter

F. Sánchez Soberón, Universitat Rovira i Virgili / Chemical Engineering; J. Rovira, Universitat Rovira i Virgili; J. Sierra, Faculty of Pharmacy University of Barcelona / Faculty of Pharmacy, Soil Science Unit; M. Schuhmacher, Rovira i Virgili University / Departament d Enginyeria Química

According to last estimations, there are globally around 6.5 million deaths as a consequence of exposition to air pollutants. Among them, Particulate Matter (PM) is considered as the most harmful one. This material consists on solid particles and liquid droplets suspended in the atmosphere having a diameter smaller than 10 µm. Since they can come from a wide array of different sources (i.e. traffic, industries, indoor dust) their physicochemical characteristics are very heterogeneous, and knowing them becomes important in order to assess its damaging potential. To improve the knowledge regarding physicochemical characteristics of PM that children are exposed in schools we conducted a study focused on finding out chemical characterization of PM_{10}, $PM_{2.5}$, and PM_1 (i.e. particles smaller than 10, 2.5 and 1 µm respectively) in an industrial area in Tarragona (Spain). These three fractions of PM were collected in the schoolyard (high volume samplers TE-6070-DV, Tisch) and inside the classroom (low volume Sioutas cascade impactor, SKC) of 12 schools during two seasons (winter and summer). Subsequently, chemical characterization of the particles (through the analysis of metals, soluble ions and carbonaceous materials) was performed. Preliminary results show that both indoor and outdoor levels of PM are higher in winter than in summer. Indoor/outdoor ratios of particles are varying, showing some schools higher PM levels indoors, while the opposite phenomenon is observed in others. Consequently, concentrations of the different chemical constituents of particles are variable, although indoor particles trend to have higher shares of carbonaceous materials. Our results will be useful not only to schools managers and parents, but also to policy makers in order to assess risk coming from the inhalation of these materials.

TU393

Good news to lazybones kids: increasing sleeping time decreases exposure to airborne particulate matter

F. Sánchez Soberón, Universitat Rovira i Virgili / Chemical Engineering; F. Noardo, Universitat Rovira i Virgili / Department of Chemical Engineering; M. Mari, Universitat Rovira i Virgili / Chemical Engineering; V. Kumar, Universitat Rovira i Virgili / Departament d Enginyeria Química; M. Schuhmacher, Rovira i Virgili University / Departament d Enginyeria Química

Particulate matter (PM) is a complex mixture of extremely small particles (< 10µm) and liquid droplets suspended in the atmosphere. They are originated from a wide range of sources (such as traffic, industry, energy production or domestic combustion). Nowadays, the inhalation of this pollutant is a concern, due to its potential to cause irritation and inflammation of respiratory airways, asthma attacks, and lung cancer. These effects are especially pernicious in kids, since their inhalation rates are higher, and their immune system is still not fully developed. However, most studies dealing with human exposure to PM are focused on adults. Therefore, the objective of the present study was to evaluate the children's exposure to different sizes of PM. To do so, three fractions of PM (smaller than 10, 2.5, and 1 µm) were collected in the playground and inside a classroom of 12 schools within the Tarragona county (Spain), an area characterized by having one of the most prominent industrial clusters in southern Europe. To elucidate time-activity patterns of kids, and to know the characteristics of their dwellings, questionnaires were delivered to parents of kids attending these schools. Using an infiltration model (IAQX, US EPA) it was possible to calculate concentrations of PM inside houses. A subsequent run of a dosimetry model (MPPD2.11, ARA) made possible to know the deposition pattern of the different PM sizes within the distinct parts of the respiratory tract. Indoor/outdoor ratios of PM levels were variable among schools. Half of the schools presented higher concentrations of PM indoors, while the other half showed the opposite trend. Simulations indicated the great influence of PM_1 indoors, due to its easier capacity of infiltration from outdoors. Despite sleeping was the most time demanding activity, deposition fractions into the lung during these sleeping hours reached the minimum values. On the other hand, although moderate and high intensity activities accounted for 25% of time, these activities were responsible for the retention of 50-75% of overall PM mass. Most of this mass was addressed in the upper part of the respiratory tract, regardless of PM size, due to sedimentation processes. Tracheobronchial region registered the lowest values of deposited particles, while PM retained in the lung was mostly $PM_{2.5}$ and PM_1. Results from this study will help to take actions regarding indoor air quality and perform more accurate risk assessment studies.

TU394

Occupational Cement Dust Exposure: effect on blood level of some antioxidant enzymes and vitamins in Owerri, Nigeria.

C. Ikaraoha, J.A. Egeonu, Imo State University Owerri, Imo State, Nigeria / Chemical Pathology Unit Dept of Medical Laboratory Science; C. Unadike, Imo State University Owerri, Imo State, Nigeria / Medical Laboratory Science; N.C. Mbadiwe, University of Nigeria Teaching Hospital, Enugu, Nigeria / Medicine; J. Dike-Ndudim, Imo State University Owerri / Department of Medical Laboratory Science

ABSTRACT Despite that Cement dust with particulate matters are hazardous to humans, yet Occupational cement dust exposure continues among Cement workers/dealers in Nigeria. The Effect of cement dust exposure on the blood levels of some antioxidants enzymes and vitamins have not been adequately addressed especially in a black-African environment and particularly Nigeria. By random sampling method, 35 Cement workers, 35 Cement Dealers and 35 controls were selected for this study. Blood antioxidant enzyme such as glutathione peroxidase, superoxide dismutase (SOD) and catalase (CAT) were determined using ELISA while antioxidant vitamins such as vitamin E and vitamin C were determined by Spectrophometric techniques. There were progressive significant increases in blood level of vitamin C, vitamin E, catalase and glutathione peroxidase from Cement Workers to Dealers and Controls (non-cement workers), (P=0.0010, P=0.0011,

P=0.0001, P=0.001 and P=0.0011) respectively. There were significant decreases in blood levels of vitamin C, vitamin E , glutathione peroxidase and catalase (P=0.002, P=0.0004, P=0.0001, P= 0.0004) respectively in Cement Workers /Dealers compared to Controls. But no significant difference in SOD (P=0.627) on comparison of Cement Workers/Dealers with Non-Cement Workers (Controls). There was significant decrease in blood levels of vitamin C (P=0.0147), SOD (P=0.0001) and Catalase (P=0.0039) in Cement Workers, but no significant difference in both vitamin E (P=0.1676) and glutathione peroxidase (P=0.6987) when compared with Cement Dealers.There were non-significant positive correlation of Vitamin E with; Catalase, SOD, Glutathione peroxidase and Vitamin C, (r =0.2567, r = 0.3150, r =0.04598 and r = 0.2018 respectively). There were non-significant correlations of catalase with glutathione peroxidase, vitamin E, and vitamin C (r=0.058, r=0.256 and r=0.13) respectively, but there was a positive significant correlation of catalase with SOD (r=0.4173). This study suggest that Exposure to cement Dust may lead to reduction in blood levels of vitamin C, Vitamin E, Catalase , Glutathione Peroxidase, and SOD in Cement workers/Dealers. The observed reduction/decreases in the antioxidants were progressive from control to Cement Dealers and to cement Workers. **Key words; Cement dust, antioxidant, enzymes, vitamins**

TU395

Implementing NH3 mitigation strategies in a pig farm: different approaches to evaluate the environmental impact.

C. Baldini, Università degli Studi di Milano / Department of Environmental Science and Policy; J. Bacenetti, Università degli Studi di Milano / Department of Environmental Science and Policy; P. Fermo, Università degli Studi di Milano / Department of Chemistry; M. Guarino, Università degli Studi di Milano / Department of Environmental Science and Policy

Ammonia is an atmospheric pollutant causing acidification of soil, nutrient-N enrichment of ecosystems, and eutrophication of terrestrial and aquatic ecosystems. When in gaseous form, NH_3 has a short atmospheric lifetime and usually deposits near its source. In the atmosphere it reacts with other compounds to form ammonium sulfate and ammonium nitrate aerosols, leading to the formation of secondary inorganic aerosol ($PM_{2.5}$) that are a potential health hazard. Due to their smaller diameter and increased atmospheric lifetime, these particulates are able to travel long distances before being dry or wet deposited to the ground surface. This allows them to travel from rural areas to urban locations where they mix and build up in the atmosphere leading to smog or transportation to other areas. The particular unfavorable meteorological and orographic conditions of the Po Valley make this one of the most polluted region of Europe. Particulate matter pollution often exceeds the EU standards and WHO air quality guidelines for health protection. Being a main source of ammonia emissions, the agro-zootechnical compartment plays a key role in the secondary PM formation. Indeed, secondary inorganic aerosol from NH_3 accounts for 40% of PM_{10} mass at the urban sites, and its contribution is even bigger in the rural sites. This study aims at evaluating the environmental performance of different NH_3 mitigation strategies applied to Italian pig farms. Different mitigation scenarios are compared, considering the application of solutions suggested in the Best Available Technologies Reference document for the Intensive Rearing of Poultry and Pigs. Different strategies can be applied to determine the effectiveness of mitigation options: the SHERPA model or other approaches like Life Cycle Assessment can indicate the environmental benefits achievable with the different scenarios analyzed. Although techniques may be implemented and managed separately, they produce synergistic effect on the farm's environmental impact. However, all integrated measures to reduce emissions of NH_3 from pig farming will lead to a higher amount of nitrogen in the manure and to the amount that may potentially be emitted to air as NH_3during the downstream process of manure storage and spreading. The reduction of NH_3 emission from pig farming management steps can have a positive effect in NH_3-related impact categories, such as PM formation, terrestrial acidification and eutrophication.

TU396

Development of an In Vitro Method to Evaluate the Inhalation Bioaccessibility of Particle-Bound Hydrophobic Organic Chemicals and its Effects of Particle Size

s. xie, Jinan University; L. Bao, E.Y. Zeng, Jinan University / School of Environment

Bioaccessibility of particle-bound hydrophobic organic contaminants and its effects of particle size are significant for assessing the potential human health risk via inhalation exposure, but have not been clearly evaluated. To fill this knowledge gap, the present study develops an *in vitro* method to estimate the inhalation bioaccessibility of particulate organic contaminants using simulated human lung fluids, i.e., artificial lysosomal fluid (ALF) and Gamble's solution amended by dipalmitoyl-sn-glycero-3-phosphocholine, with Tenax as the absorption sink. Polycyclic aromatic hydrocarbons (PAHs) were selected as the target compounds and the assay parameters such as incubation time and the influence of particulate load membrane were examined. Results have shown that the bioaccessibilities of individual PAH compounds increased with the increasing incubation time and reached the steady state within 10 days. None of significant difference was found for the individual PAH bioaccessibilities between with and without adding glass microfiber membrane into the incubation system. Furthermore, the PAHs absolute recoveries, calculated by sum of PAHs masses in Tenax, artificial lung fluid, and residual particle dividing the initial masses, were from 92% to 112% in ALF and 75% to 99% in Gamble's solution, suggesting that this developed *in vitro* method could be well appropriate to evaluate the inhalation bioaccessibility to particulate hydrophobic organics matter. In addition, the PAHs bioaccessibility were found to increase with particle size, but decrease with the increasing hydrophobicity. It is noteworthy that the human health risk via inhalation exposure to particle-bound PAHs was reduced by more than 90% if the size-dependent PAHs bioaccessibility and deposition efficiency were involved into the assessment.

TU397

Toxicity does not vanish into thin air - molecular mechanisms of air pollutant mixtures

Z. Novakova, Masaryk University; J. Novak, Masaryk University / RECETOX; K. Hilscherova, Masaryk University, Faculty of Science, RECETOX / Research Centre for Toxic Compounds in the Environment   RECETOX; P. Kukucka, P. Pribylova, R. Prokes, Masaryk University / RECETOX; G. Lammel, Max Planck Institute for Chemistry / Multiphase Chemistry Department

Air pollution remains to be a hot environmental issue in many regions worldwide. It can directly affect human health and can contribute to higher incidence of pulmonary and cardiovascular diseases or cancer. Moreover, it has been shown that compounds occurring in the ambient air pose a hazard to disrupt our endocrine, immune or reproductive system. Since the air pollutants occur in complex mixtures, it is more appropriate to use an effect-based monitoring including a battery of *in vitro* bioassays which cover various interactions among mixture constituents. Bioassays represent an efficient approach for toxicological profiling and identification of pollutant modes of action. Together with chemicals analyses, they enable to identify main toxicity drivers. Two sites were selected, a heavily polluted urban site (industries, transportation, coal-based domestic heating) and a regional background. Samples were collected in summer and winter. To assess the more specific distribution of toxic potentials, gas phase, coarse particulate phase, and six PM_{10} size sub-fractions were sampled. Moreover, samples were also fractionated according to polarity. Human-based *in vitro* bioassays were employed to study endocrine-disruptive potentials, AhR-mediated induction of detoxification mechanisms, and cyto-/genotoxicity to the human respiratory tract. The results show that the studied effects were associated mainly with particulate phase. The most significant effects were attributed to the easily inhalable fine and ultrafine particles. This distribution pattern was found for example for AhR-mediated toxicity, estrogenicity, and androgenicity. The studied toxic potentials were elicited mainly by chemicals in the polar fraction containing relatively high levels of oxygenated-polycyclic aromatic hydrocarbons (oxy-PAHs). This study confirms that several molecular mechanisms of toxicity can be associated with air pollution and highlights the complexity of pollutant mixtures. For further understanding, the results will be discussed together with the results of the chemical analysis which focused on PAHs and their derivatives, nitro- and oxy-PAHs. This research was supported by project GACR P503 16-11537S

TU398

Human health assessment of air pollution exposure to tuberculosis risk in regions of Taiwan

H. Lin, National Taiwan University; Y. Lin, National Health Research Institutes / National Institute of Environmental Health Sciences; Y. Yang, C. CHEN, National Taiwan University / Bioenvironmental Systems Engineering; C. Liao, National Taiwan University / Department of Bioenvironmental Systems Engineering

BACKGROUND: Ambient air pollutants from anthropogenic sources play critical roles in the prevalence of tuberculosis (TB). Association between ambient air pollution and TB disease burdens should be clearly elucidated. Several studies found that air pollutants were highly likely to pose adverse effects on antimycobacterial immunity mechanisms, increasing risk of TB development. **OBJECTIVE:** The main objective of this study was to assess the contribution risks to TB incidences resulted from ambient air pollutant exposure. **METHODS:** A population-based probabilistic risk assessment framework was incorporated with air pollution concentration database and epidemiologic dose-response data to assess contribution risks of air pollutant-associated TB incidences in Taiwan regions. The contribution concept was quantified by using the population attributable fraction (PAF). The air pollution-PAF relationships were assessed by employing a three-parameter Hill model based on hazard ratio data of TB exposed to air pollutants. The contribution of air pollution exposure to TB was evaluated by applying multiplications of PAF likelihood and TB incidence rates. **RESULTS:** Both under severe and moderate scenarios, the results revealed that the most likely air pollutants significantly contributing to TB incidences were carbon monoxide (CO) and nitrogen dioxide (NO_2) in regions of Taiwan. Additionally, the particulate matter (PM_{10}) and nitrogen oxides (NO_X) also were likely to contribute to TB incidences in some regions. **CONCLUSIONS:** We suggested that the contributions of air pollutants mainly from diesel combustions (CO, NO_2 and NO_X) to TB incidences are of great concern. Furthermore, the human health risk assessment framework provides an alternative perspective to interpret the effects of air pollution on TB burdens. *Keywords:* Human health risk assessment;Air pollution; Tuberculosis; Population attributable fraction; Probabilistic risk assessment

TU399

Towards green braking: comparative evaluation of toxicological profile of particles generated by traditional and innovative braking systems.

S. Maiorana, D. Baderna, Istituto di Ricerche Farmacologiche Mario Negri / Department of Environmental Health Sciences; S. Gemma, L. Brunelli, F. Teoldi, IRCCS Istituto di Ricerche Farmacologiche Mario Negri / Department of Environmental Health Sciences; D. Dupin, Cidetec / Nanomedicine; M. Lodi, IRCCS Istituto di Ricerche Farmacologiche Mario Negri / Department of Environmental Health Sciences; A. Bonfanti, Brembo S.p.A.; E. Benfenati, IRCCS Istituto di Ricerche Farmacologiche Mario Negri / Department of Environmental Health Sciences

The protection and improvement of air quality are key critical points of environmental policies at national and international level. Inhalation of contaminated air and airborne particles is definitely the main route of exposure to the most important pollutants, secondary only to the diet. Several respiratory and cardiovascular diseases are associated with air pollution. Air pollution is responsible for 400.000 deaths per year in EU28. In 2013 outdoor air pollution was classified as carcinogenic to man (Group 1) by the International Agency for Research on Cancer, with a special attention to airborne dusts. In addition, powders can alter aquatic and terrestrial ecosystems when they reach the surface waters and soils as a result of transport and deposition events. Road transport and traffic contributes greatly to emissions of PM2.5 and PM10 and an important contribution to the non-exhaustive emission is due to the wear of brakes. The European Life+ COBRA (LIFE13 ENV/IT/000492) project aims to create a safer alternative to the pads currently on the market, replacing the phenolic binder with a new cementitious hydraulic binder. The study here presented evaluated the eco-and toxicological potential of particulate matters generated in laboratory conditions using test benches capable of simulating vehicle braking cycles. PM2.5, PM10 and deposited non-airborne particles were collected. Fine and coarse particles were detached from the filters and dispersed in an alcohol solution for *in vitro* assessment of their toxicological potential with non-tumor bronchial epithelium BEAS-2B cells as model of the respiratory tract. Effects on cell proliferation and cytotoxicity were assessed daily by the MTS assay and the evaluation of DNA-release from damaged cells. Sublethal responses were also measured including oxidative stress, DNA damage, mitochondrial membrane potential and metabolic alterations. In order to evaluate the ecotoxicological effects on seedlings growth and root damages, cress seeds were exposed to standard soil contaminated with non-airborne particles. The innovative cementitious pads were comparable to traditional pads in terms of braking performance but showed significantly reduced airborne particles production. Results showed different toxicity between particles generated from traditional and innovative braking systems: in our experimental models, particles derived from cementitious pads were less toxic than those from the resin based pads.

TU400

Toxic oxidation transformation products of phenanthrene measured in laboratory generated secondary organic aerosol particles

A. Kramer, Oregon State University / Environmental and Molecular Toxicology; S.L. Massey Simonich, C. Roper, Oregon State University / Department of Environmental and Molecular Toxicology; A. Zelenyuk, Pacific Northwest National Laboratory; K. Suski, Pacific Northwest Laboratories

Oxidized transformation products (OTP) of polycyclic aromatic hydrocarbons (PAHs) haven in some cases, been demonstrated to be more toxic than their parent PAH. PAHs are transported globally in sub-micron aerosols, the majority of which are secondary organic aerosol (SOA) particles. Recent laboratory studies indicate that SOA particles formed in the presence of gas-phase PAHs contain both parent PAHs and their OTP. In laboratory experiments of α-pinene SOA particles, OTP of phenanthrene, a model three-ringed PAH, were observed in varying ratios. Developmental toxicity testing with zebrafish (Danio rerio) will be conducted using embryos (n=32/treatment) that will be dechorionated and placed into 96-well plates containing OTP of phenanthrene in observed ratios at 6 hours post fertilization. Developmental toxicity will assessed by evaluating morphological changes, embryonic/larval photomotor behavior, and mortality at 24 and 120 hours post fertilization. Evaluation of the oxidative potential of the SOA samples as well as individual compounds and observed ratios of compounds is underway using the dithiolthreitol (DTT) consumption assay. The results from both assays will be discussed.

TU401

Chemical analysis and risk assessment for toxic compounds in PM2.5 in Gwangju, Korea

I. Kim, Gwangju Institute of Science and Technology; S. Kim, Gwangju Institute of Science and Technology / School of Earth Science and Environmental Engineering

Particulate matter (PM), a mixture of solid or liquid matter found in the air, has gained considerable attention as a major air pollutant in recent years. Recently, it is revealed particulate matter, especially PM2.5 (aerodynamic size < 2.5μm) cause numerous diseases such as respiratory, cardiovascular diseases, asthma and so on. The prime criteria for preventing the adverse effects of PM2.5 are based on the mass concentration, but recent research has shown that the chemical composition of PM2.5 can be a more important factor for determining PM2.5 toxicity than mass concentration. Many researchers reported the diverse chemicals in PM2.5 such as inorganic sulfates and nitrates, black carbon, metal, organic compounds, and others. In addition, a few studied detected organochlorine pesticides (OCPs) and polychlorinated biphenyls (PCBs) in PM2.5 of China. However, there is no researches on OCPs and PCBs within PM2.5 in Korea. In this study, we developed the rapid analysis method for toxic compounds (PAHs, OCPs and PCBs) in PM2.5 using accelerated solvent extractor system (ASE) and GC-MS/MS. PM2.5 of 4 different sites in Korea was collected from Oct. 2016 to Apr. 2017 (for 21 weeks). We determined the toxic compounds in the collected PM2.5 using the developed method and metal concentration in PM2.5 was also analyzed using microwave extraction. Cr, As and Cd showed high concentration in PM2.5 of all sites and several OCP and PCBs also detected. Lastly, we did the risk assessment on metals, PAHs, OCPs and PCBs in PM2.5 of Korea to determine the risk portion of OCPs and PCBs among the total risk of PM2.5. Our research is a valuable report on OCPs and PCBs in PM2.5 of Korea and suggests the practical method for screening trace toxicants in PM2.5.

TU402

Source apportionment study of PM10 and PM2.5 using selective wind direction sampling technique in the area of Civitavecchia (Italy)

r. gagliardi, Istituto Superiore di Sanità / Environment and Health; G. Settimo, m. Inglessis, Istituto Superiore di Sanità / Department of Environment and Health; g. marsili, osservatorio ambientale; m. soggiu, Istituto Superiore di Sanità / Deparment of Environment and Health

The concentrations of airborne particulate matter (PM) and their attribution to specific sources through the source apportionment methodologies is an important research topic in air quality study; in fact, the possibility to discriminate between different emission sources and between natural and anthropogenic contribution is a key issue for planning efficient air pollution reduction and mitigation strategies. Moreover, the knowledge of the chemical composition of PM for the different size fractions is recognized as increasingly important, in particular with respect to health effects of exposed population. The aim of the study is the characterization of PM10 and PM2.5 main sources located in the Civitavecchia harbour-industrial area (Central Italy), namely a large coal-fired power plant, a natural gas power plant, the harbour area, the vehicular traffic (due to both the local traffic and the highway crossing the area) and small industrial activities located in the town. To this purpose, the approach based on the use of PM samplers coupled with a wind-select sensor, allowing a selective PM10 and PM2.5 sampling downwind to specific emission sources, has been used. Furthermore, the chemical characterization of the PM collected has been carried out in order to explain specific emission patterns, and to assess the concentration levels of the micro-pollutants emitted by local sources and particularly toxic for health. Two sampling sites have been identified in the area, respectively urban and urban background site, and equipped with a PM10 and PM2.5 wind-select sampling device, designed to collect airflows from two directions, downwind respectively the coal-fired power plant and the port area. Samples of PM10 and PM2.5 were monthly collected for one year, and chemical analyses were performed to determine the concentrations of organic and inorganic species. A descriptive statistical analysis of data was performed, also verifying the occurrence of legislative threshold exceedances. Moreover, in order to highlight the contribution of specific sources, the differences in the measured micro pollutants concentrations between wind directions, PM size fractions and sampling sites have been investigated, as well as the seasonal trends of pollutants concentrations. These results allow to highlight that the applied methodology represents a valid support in source apportionment studies. **Keywords: source apportionment, wind select-sampling device, PM10, PM2.5**

TU403

Forecasting global atmospheric visibility based on air quality and meteorological data

H. Xiao, NUEORS, Chinese Academy of Sciences / NUEORS; J. Zhang, L. Tong, H. Yi, M. He, J. Zheng, IUE, Chinese Academy of Sciences

Simultaneous and continuous measurements of visibility, meteorological parameters, including relative humidity, and the concentrations of six atmospheric pollutants (PM_{10}, $PM_{2.5}$, SO_2, NO_2, CO, and O_3) were obtained for several cites around the global. The characteristics of visibility and relationships with air pollutants and meteorological factors were investigated using multiple statistical methods. Analysis demonstrated that within a certain relative humidity range, visibility is the exponential function of the $PM_{2.5}$ concentration. Thus, non-linear models combining multiple linear regressions with exponential regression were subsequently developed to describe the hygroscopic growth and the attenuation effect by the air pollutions. It was demonstrated that the derived models can quantitatively describe the reulationships between visibility, air quality and meteorological parameters around the whole globe.

TU404

Analyzing the Asian supply chain structure of health impacts with PM2.5 including secondary particle

F. Nagashima, Kyushu University; K. Nansai, National Institute for Environmental Studies; S. Chatani, National Institute of Environmental Studies; S. Kagawa, Kyushu University

Countries and regions in Asia have played an important role in producing intermediate products and final commodities today and supplied their products around the world. Productions and consumptions of goods and services in the Asian countries have clearly contributed to the economic growth in the world, whereas those economic activities brought about environmental loads. Particularly, China's emissions of air pollutants such as fine particulate matter ($PM_{2.5}$) have caused some serious environmental and induced health problems in Asian countries. With this research motivation,the health impacts associated with the $PM_{2.5}$ through the Asian supply chains have been estimated in the previous researches. While these analyses showed what production activities induced these health hazards caused by "primary" $PM_{2.5}$, almost of these results doesn't include the effects of "secondary" $PM_{2.5}$. This study developed the secondary $PM_{2.5}$ concentrations emitted on every industrial sector source in Asia using Emissions Database for Global Atmospheric Research (EDGAR) emission inventory data, Weather Research and Forecasting (WRF) model and CMAQ modeling system, and then estimates the induced mortality data in Asia. We further applied structural path analysis (SPA) to the Asian International Input-Output Table (AIIOT) to clarify the critical supply chains for the reduction of health impacts in Asia. The result shows that the Japan's consumption-based secondary $PM_{2.5}$ emissions in Asian were estimated 165 kt-C and we revealed top ranking supply-chain paths for $PM_{2.5}$ emissions induced by Japan's final demand. The most significant supply-chain path with the highest emissions was the path from transportation sector in China to Japan's final demand, and subsequently the path, other food products sector in Thailand --> food crops sector in Thailand --> Japan's final demand. We also argued the health impacts caused by the trans-boundary pollutions in Asian countries.

TU405

Source contributions to PM10 levels in a coastal area in northern France: a one year study

F. Ledoux, C. Roche, M. Borgie, L. Moufarrej, University of Littoral Côte dOpale / Unité de Chimie Environnementale et Interactions sur le Vivant UCEIV EA4492; G. Delmaire, University of Littoral Côte dOpale / Laboratoire Informatique Signal de la Côte dOpale LISIC EA4491; G. Roussel, M. Puigt, University of Littoral Côte dOpale / Laboratoire Informatique Signal Image de la Côte dOpale LISIC EA4491; C. Beaugard, ATMO Hauts-de-France; D. Dewaele, P. Genevray, University of Littoral Côte dOpale / Centre Commun de Mesures; O. Favez, INERIS; D. Courcot, Université du Littoral Côte dOpale / Unité de Chimie Environnementale et Interactions sur le Vivant UCEIV EA4492

The Hauts-de-France Region is one of the most concerned areas in France by exceedances of the PM_{10} daily mean limit value (50 µg.m^{-3}). For a better understanding of these phenomena, the identification as exhaustive as possible of sources contributing to PM_{10} concentration is an essential step. Numerous studies were performed on the identification of particles from terrestrial sources. The objective of this work is to fill the lack of knowledge about the impact of emissions resulting from the marine compartment. It includes natural emissions such as sea salts [1] and anthropogenic emissions linked to the marine traffic especially in the English Channel, that forms a narrow corridor with one of the greatest concentrations of shipping in the world (up to 800 vessels sailing per day). PM_{10} sampling and measurement campaign were performed continuously during one year in 2013 at Cape Gris-Nez, a coastal French site located in front of the Straits of Dover. PM_{10} levels were measured using MP101 analyzer (Environment SA®) and collected using the DA80 sampler (Digitel®, 30 m^3/h) on a daily basis. The characterization of PM_{10} was performed considering major and trace elements, water soluble ions, EC/OC as well as tracers of biomass burning (levoglusan), primary biogenic emissions (arabitol, mannitol) and marine biogenic emissions (methanesulfonate ions). These chemical parameters were used to explain PM_{10} levels on the coastal site, identify PM_{10} sources and estimate their contributions. Sources profiles were identified from the use of a Constrained Weighted non Negative Matrix Factorization (CW-NMF) model: fresh sea-salts, aged sea-salts, secondary nitrates, secondary sulphates, crustal, biomass combustion, primary biogenic emission, marine traffic, combustion, metal source. The monthly evolution of their contribution evidenced different behaviours between the sources: secondary nitrates were predominant during the cold season and appeared to be the most involved in the PM_{10} concentration peaks. The impact of marine traffic and a high proportion of aged sea-salts versus fresh sea-salts were mainly evidenced during the summer season. For the year 2013, the mean contribution of the different sources were 37% for sea salts and aged sea-salts, 43% for the secondary inorganic aerosols, 7% for biomass combustion, 5% for marine traffic. This distribution varies highly depending on the period and more particularly during exceedances of daily PM_{10} limits values.

TU406

Source-to-exposure assessment of industrial organic pollutants in Australia, using the Pangea multi-scale framework

C. Wannaz, The Unversity of Michigan, Ann Arbor / SPHEHS; P. Fantke, Technical University of Denmark / Quantitative Sustainability Assessment Division; J. Lane, University of Queensland, Brisbane; O. Jolliet, University of Michigan

Effective planning of airshed pollution mitigation is often constrained by a lack of integrative analysis able to relate the relevant emitters to the receptor populations at risk. Both emitter and receptor perspectives are therefore needed to consistently inform emission and exposure reduction measures. This presentation aims to extend the Pangea spatial multi-scale multimedia framework to evaluate source-to-receptor relationships of industrial sources of organic pollutants in Australia. Pangea solves a large compartmental system in parallel by block to determine arrays of masses at steady-state for 100,000+ compartments and 4,000+ emission scenarios, and further computes population exposure by inhalation and ingestion. From an emitter perspective, radial spatial distributions of population intakes show high spatial variations in intake fractions from 0.68 to 33 ppm for benzene, and from 0.006 to 9.5 ppm for formaldehyde, contrasting urban, rural, desert, and sea emission source locations. Extending analyses to the receptor perspective, population exposures from the combined emissions of 4,101 Australian point sources are more extended for benzene that travels over longer distances, versus formaldehyde that has a more local impact. Decomposing exposure per industrial sector shows petroleum and steel industry as the highest contributing industrial sectors for benzene, whereas the electricity sector and petroleum refining contribute most to formaldehyde exposures. The source apportionment identifies the main sources contributing to exposure at five locations of interest. Overall, this presentation demonstrates the relevance of addressing exposures both from an emitter perspective well-suited to inform product oriented approaches such as LCA, and from a receptor perspective for health risk mitigation. **<br clear="all" />**

TU407

Non-targeted screening of DNA adducts as biomarkers for human exposure to PAHs in the environment with liquid chromatography tandem mass spectrometry

Y. Feng, C. Yao, Health Canada; W. Foster, McMaster University

Humans are constantly exposed to thousands of contaminants in the environment. Polycyclic aromatic hydrocarbons (PAHs) are a group of organic compounds containing two or more aromatic rings. They are released into the environment from both natural and anthropogenic sources such as combustion of organic substances and incomplete burning of coal, oil, gasoline, tobacco products and wood. PAHs are known to be bio-transformed by phase I metabolic enzymes to chemically reactive intermediates that may bind covalently to DNA to form DNA adducts that interfere with DNA synthesis and transcription, leading to DNA mutations and/or toxicity. Furthermore, binding of electrophilic PAH metabolites to DNA is thought to be a key step in the initiation of cancer. Therefore, measurement of those DNA adducts could be an indicator or biomarker of human exposure to PAHs in the environment and of the dose of the ultimate reactive metabolite. Rapid non-targeted approaches are desired to explore a broader scope of new biomarkers associated with the contaminants in the environment. Previous non-targeted analysis with retrospective analysis of the full scan data to identify DNA adducts is time consuming. In this presentation, we will report a non-targeted screening method for identification of covalent DNA adducts using a combination of neutral loss scan and product ion scan in a Q-trap system. The method was applied to non-targeted screening of DNA adducts in follicular cells from isolated ovarian follicles that were exposed to cigarette smoke condensate (CSC). Four DNA adducts, benzo[a]pyrene-7,8-dihydrodiol-9,10-epoxide-dG(BPDE-dG), phenanthrene 1,2-quinone-dG (PheQ-dG), B[a]P-7,8-quinone-dG (BPQ-dG) and 4-aminobiphenyl-dG, were identified in the follicular cells. The results also revealed that two oxidative biomarkers, 8-hydroxy-2-deoxy guanosine (8-OH-dG) and 8-isoprostane (8-IsoP), had strong correlations with the three DNA adducts, BPDE-dG, BPQ-dG, and PheQ-dG, suggesting a strong link between the formation of covalent DNA adducts and DNA damaging oxidative stress. The method has also been successfully applied to investigate the selectivity of chemicals to modify the nitrogenous bases on DNA sequence. The results showed that each chemical had a different selectivity when it modified the DNA bases. The method has been demonstrated to be a potential tool to provide screening of unknown DNA adducts as biomarkers of human exposure to the parent contaminants in the environment.

TU408

Global inter-comparison of polyurethane foam passive air samplers evaluating variability due to sampler design and analysis

L. Melymuk, Masaryk University / RECETOX Research Centre for Toxic Compounds in the Environment; P. Bohlin Nizzetto, NILU - Norwegian Institute for Air Research / MILK; T. Harner, Environment Canada / Air Quality Research Division; J. Klanova, Masaryk University / RECETOX Research Centre for Toxic Compounds in the Environment; O. Arnador-Munoz, Universidad Nacional Autonoma de Mexico / Centro de Ciencias de la Atmosfera; B. Aristizábal Zuluaga, Universidad Nacional de Colombia Sede Manizales / Hydraulic Engineering and Environmental Research Group; M.Y. Tominaga, CETESB Companhia Ambiental do Estado de São Paulo; A.J. Sweetman, Lancaster University / Lancaster Environment Centre; B. Jimenez, IQOG-CSIC / Department of Instrumental Analysis and Environmental Chemistry; A. Dreyer, Eurofins GfA GmbH; M. Odabasi, Dokuz Eylul University; J. He, University of Nottingham Ningbo; W. Ma, Harbin Institute of Technology / China/International Joint Research Center for Persistent Toxic Substances (IJRC-PTS); J. Ma, Lanzhou University / College of Earth and Environmental Sciences; G. Zhang, Guangzhou Institute of Geochemistry, Chinese Academy of Sciences / State Key Laboratory of Organic

Geochemistry; J. Mueller, C. Paxman, X. Wang, The University of Queensland / Queensland Alliance for Environmental Health Sciences

Polyurethane foam passive air sampler (PUF-PAS) are the most commonly used passive air sampler for a range of semivolatile organic compounds (SVOCs) such as regulated persistent organic pollutants and polycyclic aromatic hydrocarbons, and emerging SVOCs (e.g. novel flame retardants, phthalates, current-use pesticides). PUF-PAS are used by numerous global/regional air monitoring programs as well as in case studies around the world. While the majority of PUF-PAS use similar double-bowl metal shielding, there is no standardized design applied in all studies in terms of bowl size, shape, deployment configuration. Many different PUF-PAS designs are used in regional or global programmes such as the Global Monitoring Programme under the Stockholm Convention and these data are compared for spatial or temporal variability and trend analysis. Yet, no information is available on the comparability of data from all the different designs. We brought together 12 types of PUF-PAS samplers from around the world and deployed them in a multi-part inter-comparison in order to evaluate the variability in reported concentrations introduced by different elements of PAS monitoring. Three sets of PUF-PAS were deployed in Kjeller, Norway in 2015-2016, as follows: (1) 3-month deployment of 15 PAS provided by international research groups, and returned to their respective research groups for analysis for SVOCs – this provides information on the overall variability in global monitoring data introduced by differences in sampler configurations and analytical methods; (2) 3-month deployment of 15 identical PAS, which were then distributed to international laboratories for SVOC analysis, to isolate the influence of analytical variability; and (3) 3-month deployment of 15 different PAS and analysis at a single laboratory (RECETOX Trace Analytical Laboratories, Masaryk University) to isolate the influence of PAS design on data comparability. Results indicate that while differences in sampler design (in particular the spacing between the upper and lower sampler bowls) account for 50-100% differences in masses collected by samplers, the variability introduced by analytical methods still significantly exceeds this amount, and this effect should be carefully considered when evaluating and comparing global monitoring data.

TU409

Microplastic Indoor Air Pollution Using a Simulated Breathing Mannequin - µFT-IR Imaging Quantification

N. van Alst, Aalborg University / Civil Engineering Department; A. Vianello, Aalborg University / Civil Engineering Department - Section of Water and Environment; R.L. Jensen, Aalborg University; J. Vollertsen, Aalborg University / Civil Engineering Department

Extensive research has been performed on indoor air quality (IAQ) over the last decades. This included determining mechanisms of deposition in lungs and on surfaces, as well as determining particle types and sizes. However, microplastic research in indoor air has been lagging behind. With the strides in microplastic research in the last years renewed interest has now arisen on microplastics as a form of indoor air pollution. This research focusses on microplastics in indoor air, with emphasis on the potential exposure to humans as a result of inhalation. This is simulated using a mannequin set-up built to imitate the human respiratory system. The mannequin takes in air through the mouth, which is led through a copper pipe to the filtering unit. The copper pipe meets a filter holder on which a 0,8 µm custom-cut 20 mm SterliTech silver membrane filter appropriate for µFT-IR imaging analysis is mounted. This is connected to a dual piston pump which simulates natural breathing. Samples have been taken in actively lived in apartments, as well as varied locations within in the universities' work environment. Samples have been divided up into continuous sampling and intermittent sampling under active living/working conditions. All samples have received active sampling for approximately 24h, either using continuous or intermittent collection. In each environment a catalogue and accompanying material for spectral identification is kept of the interior. The aim of the research is to ascertain the contribution of materials from the indoor environment as a function of activity, and determine possible exposures as well as contamination levels coming from indoor air. For identification and quantification of microplastics contained in the samples, an Agilent Technologies micro-Fourier Transform Infrared (µFT-IR) imaging system equipped with a 128x128 Mercury Cadmium Telluride (MCT) Focal Plane Array (FPA) is used. Samples are directly scanned on the silver filter at 3.3 or 5.5 µm pixel resolution, providing microplastic detection down to 6-10 µm in particle size. After collection, data is exported to in-house developed software for obtaining polymeric composition and quantifying size and mass of all µFT-IR imaged microplastics. Analysis of samples is ongoing and scheduled to be completed by March 2018.

TU410

Composite electrospun fibers based on sustainable and biodegradable polymers for monitoring air pollution

A. Macagnano, E. Zampetti, N. Pirrone, CNR-Institute of Atmospheric Pollution Research; G. Scarascia Mugnozza, University of Tuscia-DIBAF; C. Di Natale, R. Paolesse, University of Tor Vergata; F. De Cesare, University of Tuscia-DIBAF

Environmental monitoring is a growing concern in both developed and non-developed countries. Air quality monitoring is usually performed with specialized equipment and analytical methods by regulatory agencies and researchers. However EU projects guidelines report the need to involve also citizens in environmental monitoring, thus low-cost and easy-to-use technologies are required. To achieve this aim, novel sensors for environmental monitoring have been designed and developed to date to obtain reliable values comparable to those provided by standard methods and technologies. Currently, electrospinning is considered as one of the most versatile and inexpensive manufacturing technologies to design and develop nanostructured sensors to detect gases and volatile organic compounds (VOCs) in the air. Sensors based on polymeric fibers look extremely attractive for the low cost and great versatility of the raw materials that can be easily tunable, according to the transducer used and the application of interest, taking part to the resulting sensing features (selectivity and sensitivity). Therefore electrospun nanofibrous and environmentally friendly materials have been designed and fabricated for detecting specific atmospheric pollutants. The attention has been focused on the challenging goal of obtaining conductive sensors for the monitoring of air pollutants employing suitable scaffolds of eco-friendly (polyhydroxybutyrate) and sustainable (recycled) nanomaterials (polystyrene). Indeed biodegradability is a noteworthy feature to obtain sensing tools environmentally friendly and safe for health. However, sensors for gas monitoring must also be able to both persist intact for a useful shelf life and to preserve their sensing features over time, depending on the specific application and the working period. Finally, the selectivity of fibers can be tuned by introducing differently functionalized macromolecules (Me-tetraphenylporphyrins) that are sensitive to several classes of gas and VOCs. The conductivity of the planned sensors has been implemented by adding conductive nanoparticles (e.g. graphene's flakes). Rapid responses and good analyte selectivity were reported when the innovative sensors were tested to detect nitrogen oxides and ammonia in traces and VOCs, mainly due to both high porosity and high surface of interaction. Therefore, the use of polymers obtainable from recycled and biodegradable plastics sounds to be a promising and alternative strategy for the development of smart scaffolding for air pollution monitoring. Keywords: advanced sensors, sustainable and biodegradable polymers, nanofibers, air pollution

TU411

Determination of Cross Compartment Concentration Gradients of Polycylic Aromatic Hydrocarbons using PE Passive Samplers

J. Meierdierks, University of Tubingen / Center for Applied Geosciences; C. Zarfl, University of Tuebingen / Center for Applied Geoscience; P. Grathwohl, Eberhard Karls Universität Tübingen / Center for Applied Geoscience

Several polycyclic aromatic hydrocarbons (PAHs) are considered as human carcinogens or toxic to reproduction, and are thus a relevant class of "substances of very high concern" according to the European Chemicals Legislation REACH. Emission of PAHs is mainly caused anthropogenically by the incomplete combustion of fossil fuels. Due to the hydrophobic behaviour of these compounds a significant accumulation within soils has been observed. Recently decreasing atmospheric concentrations could trigger a change of equilibrium conditions between soil and atmosphere. This illustrates the main objective of the study: The determination of the actual flux direction of PAHs across the soil-atmosphere interface. Polyethylene (PE) passive samplers have been used to determine concentration gradients of the 16 EPA PAHs at this interface as well as the respective flux direction. Atmospheric monitorings have been conducted seasonally for two subsequent years using 80 µm thick PE sheets at three rural sites. During the second year an additional height has been implemented as well as active sampling. Soil samples were taken at each location at several intervals up to 50 cm depth and equilibrated ex situ with 30 µm thick PE sheets. A numerical approach, based on the double film diffusion model, was applied to the experimental data in order to deduce the atmospheric concentration over time. Seasonal deployments illustrate significant variations with 10 fold higher PAH concentrations in the atmosphere during winter compared to summer monitoring. Concentrations within the soil depicted homogenous profiles, considering Phe as representative PAH concentrations in the soil were in the range of 100 ng/g PE after equilibration. In contrast concentrations on the PE in the atmosphere vary between 70 ng/g during summer and 1200 ng/g during winter monitoring. This explicit difference between soil and atmosphere during colder months indicates a main flux direction into the soil.

TU412

Evaluating Computational and Structural Approaches to Predict Transformation Products of Atmospheric Polycyclic Aromatic Hydrocarbons

I. Titaley, MTM Research Centre, Orebro University / Department of Chemistry; D.M. Walden, M. Ogba, Oregon State University / Chemistry; P.H. Cheong, Oregon State University / Department of Chemistry; S. Simonich, Oregon State University / Depts of Chemistry and Environmental Molecular Toxicology

Polycyclic aromatic hydrocarbons (PAHs) undergo transformation reactions with atmospheric photochemical oxidants, such as hydroxyl radicals (OH•), nitrogen oxides (NOx), and ozone (O_3). The most common PAH-transformation products (PAH-TPs) are nitro-, oxygenated-, and hydroxylated-PAHs (NPAHs, OPAHs, and OHPAHs, respectively), some of which are known to pose potential human health concerns. We sampled four approaches for predicting the location of reactive sites on PAHs (*i.e.*, the carbon where atmospheric oxidants attack), and hence the chemoselectivity of the PAH-TPs. The four approaches are: 1) Clar's prediction of

Kekulé resonance structures, 2) thermodynamic stability of all possible OH-PAH adduct intermediate, 3) electron density at each carbon on the PAH, and 4) average local ionization energy (ALIE) at atom or bond sites. To evaluate the accuracy of these approaches, the predicted PAH-TPs were compared to published laboratory observations of major NPAH, OPAH, and OHPAH products in both gas- and particle-phases. We found that the Clar's resonance structures were able to best predict the least stable rings on the PAHs, but did not offer insights in terms of which carbon is most reactive. All other computational approaches provided specificity in their predictions, yet the ALIE approach was the most superior in accuracy, when compared to laboratory data. The high predictive capability of ALIE shows great potential for the prediction of the formation of previously unstudied PAH-TPs that are likely to form in the atmosphere. Furthermore, the results help: the environmental chemists to prioritize which PAH-TPs might be formed in the environment; the organic chemists to prioritize which PAH-TPs should be synthesized to verify their presence in the environment; and the toxicologists to prioritize which PAH-TPs should be analyzed for their toxicity and potential human health implications. Future direction of the study is to expand the prediction to screen for PAH-TPs from other parent-PAHs as well as alkylated-PAHs.

TU413

Spatial distribution of gas-phase Polycyclic Aromatic Hydrocarbons along South America and Antarctica

A. Azevedo, Instituto Federal do Rio Grande do Sul; K.S. Miglioranza, University of Mar Del Plata / Laboratorio de Ecotoxicología y Contaminación Ambiental, Instituto de Investigaciones Marinas y Costeras; P.G. Costa, FURG / Escola de quimica e alimentos; R. Barra, Universidade de Concepción; O.P. Amarante Jr, IFMA / DAQ; F. Wania, University of Toronto at Scarborough / Physical and Environmental Sciences; G. Fillmann, Universidade Federal do Rio Grande FURG / Instituto de Oceanografia

Polycyclic Aromatic Hydrocarbons (PAHs) are a large group of compounds with two or more condensed aromatic rings. These compounds are emitted from various sources to the atmosphere and some of them are known by their carcinogenic or mutagenic properties. However, quali-quantitative information is limited about PAHs in air, and normally rely on the availability of active sampling techniques, usually expensive and laborious, needing power source, inexistent in remote areas. Conversely, passive sampling allows cheap and easy handling atmospheric appraisal even in remote regions. Thus, the present study evaluated PAHs levels throughout the South American atmosphere employing XAD2-based passive atmospheric sampling (PAS). The Latin American Atmospheric Sampling Network (LAPAN) has begun in 2010 by deploying a pair of PAS containing one cartridge of XAD-2 resin on each site. Resins were deployed for 12 months during 3 consecutive years at42 sites (16 sites in Brazil and 26 distributed in Argentina, Chile, Peru, Uruguay, Venezuela and Antarctica) covering different backgrounds (rural, urban / industrial and remote). Passive samplers and XAD2 resins were prepared as described by Wania et al. (2003). XAD-2 resins were extracted by hexane:dichloromethane (1:1), purified and analyzed by gas chromatography/mass spectrometry (Perkin Elmer Clarus 680 GC-MS). The following PAHs were analyzed: naphtalene, 2-methyl-naphtalene, 1-methyl-naphtalene, acenaphthylene, acenaphthene, fluorene, phenanthrene, anthracene, fluoranthene, pyrene, benzo(a)anthracene, chrysene, benzo(b)fluoranthene, benzo(k)fluoranthene, benzo(a)pyrene, indeno(1,2,3-cd)pyrene, dibenzo(ah)anthracene and benzo(ghi)perylene. Results, reported as sequestered amount of each compound per PAS (ng PAH day^{-1}), indicated presence of PAHs at all sites. HPAs with higher molar weight (low vapor pressure) were below the detection limit or levels were very low, such as the potentially carcinogenic benzo(a)anthracene and benzo(b)fluoranthene. The urban and suburban areas showed higher levels compared to remote regions. This is a long term study looking forward to appraise temporal trends to PAHs along South America atmosphere.

TU414

Importance of Dermal Exposure to Polycyclic Aromatic Hydrocarbons Derived from Barbecue Fume

J. Lao, S. Xie, Jinan University; C. Wu, L. Bao, Jinan University / School of Environment; S. Tao, Peking University / Laboratory for Earth Surface Processes College of Urban and Environmental Sciences; E.Y. Zeng, Jinan University / School of Environment

Despite the ubiquity and carcinogenicity of polycyclic aromatic hydrocarbons (PAHs), its dermal absorption for the general population has not been adequately addressed. Aiming to verify the importance of dermal absorption for PAHs, barbecue (BBQ) in Guangzhou, China was chosen as a case study. Urine samples were collected approximately 17 h before exposure until 35 h after exposure from 20 participants and analyzed for nine hydroxyl (OH)-PAHs. Air, food, and cotton clothing samples were analyzed for 16 PAHs. Based on the occurrence of atmospheric PAHs, dermal absorption of low molecular-weight PAHs was greater than inhalation intake. In addition, the net excreted amounts of OH-naphthalene, OH-fluorene, OH-phenanthrene, and OH-pyrene via dermal contact were 367, 63, 98, and 28 ng respectively, comparable to those via combined dermal and inhalation exposure, which were 453, 98, 126, and 38 ng. The ratios of excretion to intake via dermal contact were 0.11, 0.036, and 0.043 for fluorene, phenanthrene, and pyrene, respectively, higher than those for inhalation (0.097, 0.016, and 0.025). These results indicate that dermal absorption is a significant exposure route of PAHs. In the case of BBQ fumes, dermal absorption is a more important pathway for intake of low molecular-weight PAHs than inhalation.

TU415

EDS Mapping of Particles As A Component of Lichen Biomonitoring in Seattle, Washington

G.T. Guddal, Western Washington University; J. Miller, A. Johnson, Western Washington University / Environmental Sciences Department; R. Sofield, Western Washington University

Lichen are an increasingly popular medium for conducting air quality monitoring due to their sensitivity to SOx and NOx, as well as their bioaccumulation of airborne material. This study incorporates characterization of particulate matter (PM) on the surface of lichen *Ramalina farinacia* to map exposure to air pollution in three industrial clusters in Seattle, Washington, USA. The PM was characterized using scanning electron microscopy with energy-dispersive X-ray spectroscopy mapping to determine PM size and composition. We also measured bioaccumulation of certain metals in the lichen and the biomarkers glutathione, chlorophyll degradation, malondialdehyde, and usnic acid. Principal components analysis has identified which geographic locations and particle types correlate the strongest with increased metal accumulation and physiological response in the lichen.

TU416

TBARS in horse hair as an indicator of oil industry pollution

M. Kovačević, Department of Biology, University of Osijek; T. Plavac, B. Kutuzović Hackenberger, University of Osijek / Department of Biology

Air pollution is a major problem today. Although there are many tests that measure the presence of certain substances in the air, it is important to measure the impact of various pollutant on living organisms as well. Horses that spend time outdoors are exposed to enviromental influences, and some of them are measurable in horse hair. The aim of this study is to assess how pollutants of the oil industry affect biological markers in permanent horse hair from mane and tail and whether the concentration of thiobarbituric acid reactive substances (TBARS) can be used as biomarker of oil industry air pollution. The horse hair samples from mane and tail were collected from two areas. One near Slavonski Brod, where an issue of air pollution is present due the outdated refinery plant in Bosanski Brod, and the other near Osijek where no apparent air industry pollution is present. The concentration of TBARS in samples was measured fluorometrically. The samples were cut into segments to detect differences in duration of exposure through the age of hair. The concentration of TBARS was significantly higher ($p < 0.005$) in horses exposed to polluted air (Slavonski Brod site). When segments were analysed according to the age of hair it was noticed a constant difference in concentration of TBARS between roots and top for mane and tail. A significant correlation between age of hair and concentration of TBARS was noticed. Although further research is needed, a concentration of TBARS in horse hair could serve as a simple and inexpensive method for monitoring air pollution by oil industry.

TU417

Morbidity for environment-related diseases in La Spezia, northwest Italy: an epidemiological analysis on hospital discharge rates.

F. Lucaroni, University Rome Tor Vergata / Department of Biomedicine and Prevention; A. Pietroiusti, University of Rome Tor Vergata / Department of Biomedicine and Prevention; N. Toschi, University of Rome Tor Vergata; A. Duggento, University of Rome Tor Vergata / Department of Biomedicine and Prevention; C. Alessandroni, C. Ambrosone, L. Palombi, University Rome Tor Vergata

Background. La Spezia Province, northwest Italy, is a potentially high risk area because of the massive presence of industrial and harbor activities. Only controversial evidence of their health impact in the area is available at the moment. Therefore, a comprehensive survey on potentially environment-related disease would be really useful. **Methods.** Liguria Region hospital discharge records from 2001 to 2013 have been collected. Only admissions for cardiovascular disease, respiratory disease or neoplasms as primary diagnosis were included. Hospital discharge rates, standardized by age and compared with Regional mean, were represented using geographic maps with a color scale identifying different disease distribution. For those disorders showing significant difference with regional mean, disease distribution was compared with emissions of air pollutants, estimated by Lichens biomonitoring. **Results.** La Spezia Province Hospital Discharge rates for potentially environment-realted disease were significantly different if compared with those of the Liguria Region. Malignant melanoma, chronic bronchitis and myocardial infarction rates were higher both in men and women (with a percentage increase of 84% and 87%, respectively, for malignant melanoma; +22% and +47% for chronic bronchitis and +40% and +41% for myocardial infarction). Conversely, hospital discharge rates for diabetes and hypertensive cardiopathy were lower than Liguria Region: -50% among males and -49% among women for the first and -56% and -54%, respectively, for the second. Comparing these results with Lichens maps, no evidence of a clear correlation between emissions of air pollutants and regional distribution of diseases was available. On the contrary, diseases differing the most from Regional mean were mainly distributed in areas with scarce anthropic activity. **Conclusions.** No clear morbidity trend is identifiable for La Spezia Province from 2001 to 2013. Also disorders belonging to the same pathological class –like myocardial infarction and hypertensive cardiopathy - showed a different behavior. Comparing results with Lichen maps helps putting excesses of morbidity into a Regional context, being cautious to clearly correlate such disease with industrial and Harbour activity. Indeed, most of the disorders showed the higher increases in areas with less signs of anthropic activity, according to Lichens biomonitoring.

TU418

Risk Assessment of Polyethylene Residues and Organoleptic Attributes of Bambara nut pudding (Okpa) Samples prepared using Alternative Cooking Materials

T. Otitoju, University of Nigeria Nsukka / human nutrition and dietetics; O. Otitoju, federal University Wukari / Department of Biochemistry; G. Arts, Wageningen Environmental Research (Alterra) / Environmental Risk Assessment; S. Baiyeri, Federal University OyeEkiti / Agronomy

Polythene residues are chemical components that are left over as monomers and end products after the thermal degradation of polythene. However, the use of plastic as cooking materials in bambara nut pudding (Okpa) a well-cherished food, especially among the inhabitants of the Eastern part of Nigeria may pose more danger than envisaged. The use of banana leaves in cooking food has been in existence in Nigeria before the introduction of the technological cooking materials such as polyethylene, cellophane plastic, tin and foil. In this study, we evaluated the risk assessment of polyethylene residues (volatile organic compounds - VOCs) and organoleptic attributes of Bambara nut pudding (Okpa) samples prepared using alternative cooking materials. Purge and trap method using Gas Chromatography and Mass Spectrometry instrument were used to estimate the concentrations of VOCs in the different pudding cooked using some alternative cooking material (cellophane, tin, foil and plastic) while banana leaves were used as control. Organoleptic evaluation was done using A-Point Hedonic Scale, standard methods and ANOVA was used to compare means of the results. The result showed the presence of some Volatile Organic Compounds such as Argon, Allene, Acetic acid, Propane-1-ol, difluoramine, Hexanoic acid, Amyl nitrite, Toluene, Butenenitrile,2-Butenal,Thiirane,Nonanoic acid, Ethylenediamine, Furfural, Hydrogen azide, 2-pentene, Formic acid, and octanoic acid; with Acetic acid occurring the most and Argon, Allene, and Difluoramine occurring the least. Pudding made with cellophane had the highest VOCs with 45% D-mannoheptulose, 45% hexanoic acid, 25% propane-1- ethenylthio and had other VOCs ranged from 4-9%. All the cooking materials had hexanoic acid at high concentrations of 25-42%. The result also showed that acetic acid and 2-butenenitrile ranged from 4-7% in all samples except Banana leaves pudding. Organoleptic evaluation of the Bambara pudding samples with different alternative cooking materials were generally acceptable ($p>0.05$) but pudding wrapped with banana leaf was significantly ($p< 0.05$) rated low for colour and taste while others were comparable ($p>0.05$). In conclusion, bambara nut pudding cooked with alternative cooking materials contained polythene residues

TU419

SETAC Human Health Risk Assessment Interest Group

B. Mulhearn, Ensafe Inc.

Fungicides - an overlooked compound group? Fate, effects, risk assessment and mitigation (P)

TU420

Ecological risk assessment of conazole fungicides in arable soils of the Czech Republic

J. Vasickova, M. Hvezdova, Masaryk University / Research Centre for Toxic Compounds in the Environment (RECETOX); P. Kosubova, Central Institute for Supervising and Testing in Agriculture; K. Brandstätter-Scherr, University of Natural Resources and Life Sciences / Institute for Environmental Biotechnology, Department for Agrobiotechnology (IFA-Tulln); Z. Simek, Masaryk University / Research Centre for Toxic Compounds in the Environment (RECETOX); J. Hofman, Masaryk University, RECETOX / Faculty of Science, RECETOX

Application of pesticides, including conazole fungicides (CFs), is an indispensable part of modern agricultural management, contributing to food security and safety. Conazoles are a class of azole-based fungicides, commonly used to prevent fungal growth on turf grass and agricultural crops. CFs are still widely used despite their reported ecotoxicity in water, chronical toxicity to mammals with hepatoxicity, carcinogenicity, reproductive toxicity and endocrine disruption. For example, in the EU classification, epoxiconazole and flusilazole are suspected carcinogens [1]. Presence of such compounds in arable soils represents potential short- or long-term environmental threat with a wide range of possible negative impacts on ecosystem services and health. Hence, risk assessment considering variable non-target taxonomic groups of vertebrates, invertebrate and plant species, should be performed. The objective of the present work was to assess the ecological risks of conazole fungicides based on the data from the comprehensive survey of pesticide residues in 75 agricultural topsoil floodplain locations in the Czech Republic acquired in early 2015 [2]. In this study, 51 currently used pesticides and 9 transformation products were analysed by multi-residue pesticide analysis on LC-MS/MS after soil QuEChERS extraction. The data indicated that over 70% of soils contained at least one CF and the total concentration of CFs exceeded 0.01 mg/kg in 53% of soils. Epoxiconazole and tebuconazole also frequently exceeded 0.01 mg/kg (in 25% and 11% of soils, respectively). The most frequent CFs were epoxiconazole (48% of soils) and tebuconazole (36%), followed by flusilazole (23%), prochloraz (21%), propiconazole (13%), cyproconazole (8%) and difenoconazole (7%). Overall, the CFs fungicides are of environmental concern because they exceeded risk based thresholds, tend to form long-term residues in soil and rank among suspected carcinogens and endocrine disruptors. [1] EU pesticide database. ec.europa.eu/food/plant/pesticides/eu-pesticidesdatabase. [2] M. Hvezdova, et al., Sci. Total. Environ., vol. 613–614, pp. 361–370, 2018.

TU421

Concentration- and time-dependent dissipation, partitioning and plant accumulation of selected fungicides, insecticide, herbicide and transformation product in sand and soil

N. Neuwirthová, Masaryk University; Z. Bílková, Masaryk University / RECETOX; J. Vasickova, Masaryk University / Research Centre for Toxic Compounds in the Environment (RECETOX); J. Hofman, Masaryk University, RECETOX / Faculty of Science, RECETOX; L. Bielska, Masaryk University / Faculty of Science RECETOX

In this study, the dissipation and partitioning dynamics and the extent of biouptake was measured and modeled for selected hazardous current-used fungicides (prochloraz, tebuconazole, flusilazole, epoxiconazole), insecticide (chlorpyrifos), herbicide (pendimethalin) and for a transformation product (2-hydroxyatrazine) in agricultural soil and quartz sand as representatives of a real and a worst-case scenario. Dissipation, uptake to *Lactuca sativa* and the freely dissolved concentration along with the organic carbon-normalized sorption coefficients (K_{oc}) were determined on day 12, 40, and 90 following the application of compounds at three fortification levels (0.1 - 1.0 - 10 mg/kg). Dissipation of tested compounds differed in soil and sand and was influenced by compound concentration. 2-hydroxyatrazine showed the longest persistence in soil among the tested compounds. The four fungicides showed very similar dissipation patterns and were more persistent in sand than in soil which implies that their main elimination mechanism in the environment is biodegradation. Plant roots were shown to accumulate higher amounts than shoots with root-to-shoot translocation factors (TFs) of 0.007-0.14 where the extent of root uptake was driven by compound partitioning. This was evidenced by the ability of C_{free} to reliably ($r^2 = 0.94$) predict root uptake. Concentration in leaves did not exceed the maximum residue levels (MRLs) for lettuce. K_{oc} values were in the range of literature values and were shown to increased (from day 0 to day 40) as well as decreased for some compounds (from day 40 to day 90) with time probably as a result of compound sequestration and competitive sorption, respectively. From the results, it follows that the tested compounds pose limited risks when presented in the soils for a given time as they were shown a) to be not persistent (except for 2-hydroxyatrazine), b) to accumulate in lettuce to extents below MRLs, c) to sorb effectively to soil, even in the presence of other co-solutes.

TU422

Evaluation of pesticides and fungicides transport using passive sampling devices in a vineyard catchment in South West France

J. Gaillard, Université de Bordeaux / EPOC UMR 5805; M. Dévier, University of Bordeaux / EPOC / LPTC UMR 5805 CNRS; K. Le Menach, P. Pardon, UMR CNRS EPOC Universite Bordeaux / EPOC UMR 5805; G. DUPORTE, Université de Bordeaux / EPOC UMR 5805; F. Macary, Irstea Bordeaux; H. Budzinski, University of Bordeaux

In vineyards of South West France, fungicides account for nearly 8 out of 10 pesticide applications which increases the likelihood of chronic exposure in adjacent ecosystems. The objective of the present study was to investigate the dynamics of pesticides and fungicides from agricultural drainage ditches to a third order stream in a vineyard catchment. Monitoring was a combination of passive and grab water sampling. Six sites with continuous-flowing were monitored during a year using polar organic chemical integrative sampler (POCIS) retrieved monthly together with grab water samples collected monthly or bimonthly. Four sites with intermittent-flowing were monitored using grab water samples only. Passive samplers such as POCIS enable the improvement of limits of quantification (LOQ) and estimation of time-weighted average concentrations over the exposure period for hydrophilic compounds. To provide a complete screening, more than 150 pesticides were targeted including 23 fungicides currently applied in the studied catchment. Extracted samples were analysed by liquid chromatography-tandem mass spectrometry (LC-MS/MS) and gas chromatography-tandem mass spectrometry (GC-MS/MS). Among the currently used pesticides in the catchment, preliminary results obtained from a sampling campaign conducted in spring allowed the quantification of 19 fungicides, 3 herbicides and 1 insecticide in passive samplers. In water samples, 9 fungicides and 1 insecticide were detected. Highest concentrations (> 1 µg/g) were measured for the fungicides benalaxyl and dimetomorph. Fungicides such as cyprodinil, kresoxim-methyl and iprovalicarb were detected in passive samplers but were not detected in water samples suggesting the importance of combined sampling techniques to provide a more complete assessment of fungicide exposure in vineyard catchments.

TU423

Assessment of secondary exposure to fungicide residues in fruit-growing workers: analysis, levels and sources

G. DUPORTE, J. Gaillard, Université de Bordeaux / EPOC UMR 5805; E. Barron, University of Bordeaux, CNRS / EPOC UMR 5805; K. Le Menach, P. Pardon, UMR CNRS EPOC Universite Bordeaux / EPOC UMR 5805; I. Baldi, University of Bordeaux / ISPED, EPICENE; F. Macary, Irstea Bordeaux; M. Dévier, University of Bordeaux / EPOC / LPTC UMR 5805 CNRS; H. Budzinski, University of Bordeaux

European Union countries have about 12 million agricultural holding, and approximately 172 million hectares (39% of the total European land surface) of agricultural areas. Pesticides are used extensively in agricultural production to prevent pests, diseases, weeds or other plant pathogens to reduce yield losses and to guarantee a good harvest. In the recent decades, numerous studies have suggested adverse health effects associated to long-term pesticide exposure.[1,2] Serious concerns have been raised about health risks resulting from occupational exposure. Nevertheless, the knowledge of occupational exposure levels and determinants to pesticides are still limited. The CANEPA project (Cancers and Exposures to Agricultural Pesticides) aims to characterise external contamination of agricultural workers and environmental contamination by pesticides in arboriculture. In this work, assesment of secondary exposure to pesticide residues was performed in several apple holding, situated in south-west of France. Dislodgeable foliar residues (DFR) and pesticide residues on equipements or apples (wipe sampling) were studied during the different activities of apple growing (treatments, re-entry tasks, harvests). Atmospheric levels of pesticides (outdoor and indoor) were also determined using passive samplers (Polyurethane Foams, PUF) and low-volume samplers in this work. This study was mainly focused on two fungicides (captan and dithianon), extensively used in apple growing, and their metabolites. High sensitive analytical methods were developed and validated, in this work, for the different collected samples based on gas or liquid chromatography coupled to hybrid high resolution mass spectrometry and to tandem mass spectrometry (NCI-GC-QTOF-MS and HPLC-ESI-MS/MS). Levels of pesticide residues and source characterisation will be presented. These findings provide a better understanding of current practices and may help for reducing pesticide occupational exposure and health risks for fruit-growing workers.

TU424

Intra-tracheal administration of the disinfectant, chloromethylisothiazolinone/methylisothiazolinone (CMIT/MIT), in a mouse model to evaluate a causal association with death

Y. Park, GLP Center, Catholic University of Daegu / APT; H. Kim, Graduate School of Medical Health Science, Catholic University of Daegu; B. Kang, Catholic University of Daegu, Graduate school of toxicity assessment

Objectives: The deaths of Korean victims who were exposed to the disinfectant, CMIT/MIT, have remain unsolved. This is mainly due to a lack of concordance between the few available toxicity tests and the abundant epidemiological data, thus making it difficult to establish a cause-and-effect relationship. Therefore, this study was carried out to investigate any potential associations between CMIT/MIT exposure and death. Methods: Groups of experimental and control C57BL/6 mice were instilled (in the trachea) with chloromethylisothiazolinone/methylisothiazolinone (CMITMIT), using a visual instillobot . CMIT/MIT was instilled over a period of 3 days and 8 weeks, respectively, to achieve acute and chronic exposures. A threshold dose-response model was applied for estimating the threshold level as one line of evidence for a causal association between CMIT/MIT and death. Results: An acute exposure of 1.2 mg ai/kg/day of CMIT/MIT was estimated to reflect the threshold for death. The dose-response curve with this threshold showed a very steep slop and a narrow range of CMIT/MIT exposures. A narrow range of CMIT/MIT exposures, in particular, indicated an evident boundary between survival and death, thus implicating a strong causal association. A similar threshold dose-response relationship observed following acute exposure was also seen following chronic exposure to CMIT/MIT. Airborne disinfectant exposure was visible as mimimal or mild lung damage with no fibrosis, as shown by histopathological tests. However, many observations are considered to be functional respiratory tract, as observed in necropsies of the mice that died due to CMIT/MIT exposures. Conclusions: There are two strong lines of evidence for a causal association between death and CMIT/MIT exposure; 1) The threshold dose-response curve, with a very steep slope and a narrow range of CMIT/MIT exposures showing a visible boundary between survival and death, and 2) functional respiratory tract failure except lung fibrosis. Thus it is concluded that CMIT/MIT exposure would cause the death without lung fibrosis.

TU425

Genotoxic response and alteration of intracellular redox balance in Hep-2 cell line by exposure to Iprodione

G. Chaufan, University of Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Química Biológica CONICET-UBA, Instituto de Química Biológica - Ciencias Exactas y Naturales (IQUIBICEN); C. Galvano, Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Química Biológica; M.D. Mudry, Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Ecología Genética y Evolución Facultad de Ciencias Exactas y Naturales; M.d. Rios de Molina, Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Química Biológica, CONICET-UBA, Instituto de Química Biológica - Ciencias Exactas y Naturales (IQUIBICEN); N. Andrioli, Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Ecología Genética y Evolución.

The use of fungicides represents one of the most important factors in the control of pests and diseases, which affects the production systems of fruits and vegetables. It is known that most fungicide residues remain stable in food for long periods of time, increasing exposure risk for the general population. The aim of the present study was to evaluate the oxidative damage, the antioxidant response and the genotoxic effect in a human cell line (HEp-2) against the exposure of sublethal concentrations of the fungicide Iprodione. For this proposal, we determine the content of protein carbonyls a marker of oxidative damage, the equivalent content of glutathione (GSH), the activity of antioxidant enzymes superoxide dismutase (SOD) and catalase (CAT) and from detoxifying enzyme GSH-S-transferase (GST), in 3 concentrations of Iprodione (1.5, 7 and 25 µg/ml). The cell division index, the replication index, the frequency of chromosomal aberrations and micronuclei were also determined in the presence of 7.5; 17.5 and 25 µg/ml of Iprodione. The cells were cultured in minimal essential medium supplemented with 10% fetal bovine serum (v/v), penicillin (100 U ml), streptomycin (100 mg/ml), amphotericin B (2.5 mg/ml) in a humid environment. with 5% CO_2 (v/v), at 37 ºC. For the cytotoxicity assays, the cells were seeded in 96-well plates, for enzymatic determinations and protein damage in Petri dishes (7.5×10^6 cells) and for genotoxicity parameters in 6-well plates. From the MTT assays, the LC50 was determined (29.88 (25.98 - 34.37) µg/ml Iprodione). The activity of SOD decreased significantly 40% ($p < 0.05$) to 25 µg/ml of Iprodione, while no effect on the activity levels of CAT and GST was observed. The content of protein carbonyls increased 30% ($p < 0.001$) at the highest concentration of Iprodione tested. In addition, it was observed that Iprodione induces tripolar and micronucleus divisions at 17.5 and 25 µg/ml and bridges with all concentrations tested. Both the index of division and the index of replication indicate that the cells maintain their proliferation capacity, which allows to study the biomarkers of genotoxicity in this system. These results confirm that Iprodione produces genotoxicity and an alteration in the redox equilibrium at the concentrations tested, which indicates the potential risk of exposure to this xenobiotic.

TU426

Toxicological effects of commercial fungicides on the earthworm Eisenia fetida (Savigny, 1826): laboratory and field investigations

T. Campani, I. Caliani, C. Pozzuoli, L. Poggioni, University of Siena / Department of Physical, Earth and Environmental Sciences; S. Casini, University of Siena / Scienze Fisiche della Terra e dellAmbiente

The use of plant protection products in agriculture can affect non-target soil organism and have a negative effect on the health of the ecosystem. In particular, information on toxicological effects of fungicides are very poor or absent, despite their widespread use. The aim of this study was to evaluate the potential toxic effects of 4 commercial fungicides (Prosaro®, Amistar®, Mirador® and Icarus®) on the earthworm *Eisenia fetida* (Savigny, 1826) species. We choose commercial products with the aim of testing the simoultaneous effect of active principles and additives present in the products. Laboratory experiments were conducted using the filter paper test (FPT): *E. fetida* was exposed to increasing concentration of Prosaro®or Amistar®, being the highest dose of treatment the recommended one for the usage in wheat farming. Field investigations were conducted transplanting *E. fetida* in cages in the soil of wheat and durum wheat fields before and during treatment with different combinations of the 4 fungicides. *E. fetida* specimens from laboratory and field work were analysed to evaluate vitality, potential neurotoxic effects (inhibition of acetylcholinesterase activity (AChE)), phase II enzymatic defense (glutathione S-transferase (GST)), oxidative stress (lipid peroxidation (LPO) and catalase (CAT) activity), genotoxic effects (Comet assay) and effect on the immune system (lysozyme activity). Laboratory studies with Prosaro® and Amistar® showed alterations in organism's vitality which increased with increasing treatment doses. Significant alteration of phase II metabolising enzymes (GST induction) and significant DNA fragmentation (Comet Assay) with respect to controls were detected at environmentally relevant doses of Prosaro®. Data from the field study underlined a statistically significant induction of GST in earthworms transplanted in the fields treated with Amistar® alone and Amistar®+ Prosaro®. This study represents a first step towards a better understanding of commercial fungicides toxicological potential to non-target organisms. Data obtained indicate that deeper investigations are needed which should include long term artificial soil tests (AST) and further field studies.

TU427

Potential Salinity Enhanced Impacts of the Phototoxicity of the Fungicides to Inland Silversides, Menidia beryllina

E.N. Vebrosky, Louisiana State University / Department of Environmental Sciences; W. Xu, Louisiana State University AgCenter / Renewable Natural Resources; K. Armbrust, Louisiana State University / Environmental Sciences School of the Coast and Environment
Dicloran and chlorothalonil are two active ingredients in fungicides commonly used in the United States that readily undergo photolysis in the presence of sunlight. Both pesticides have reported half-lives in seawater and freshwater. While the rate of degradation and half-life of dicloran is not impacted by the salinity of water (7.5 hours), the distribution of intermediate products is altered significantly; 2-chloro-1,4-benzoquinone forms at nearly double the concentration in seawater as opposed to freshwater. Chlorothalonil quickly degrades to 4-hydroxychlorothalonil via soil degradation and hydroxychlorothalonil can desorb back into the water column where it can be photochemically degraded. The degradation rate and half-life of hydroxychlorothalonil is very short, but differs significantly between freshwater (32.5 min.) and seawater (301 min.). Both dicloran and hydroxychlorothalonil have similar proposed photodegradation pathways, therefore the potential for enhanced phototoxicity due to salinity variation is possible. Dicloran has shown to be phototoxic to inland silversides at concentrations as low as 0.10 mg/L, with >90% mortality at 0.75 mg/L. Adverse sub-lethal impacts have also been observed, such as an upregulation in the CCL28 and PTGS2 genes. The effects of salinity on chemical toxicity may warrant changes to future chemical assessments.

TU428

From mother to offspring: multigenerational effects of carbendazim at individual and subcellular levels in Daphnia magna

A.R. Silva, University of Aveiro / Dept.of Biology & CESAM; C.S. Santos, Ghent University (UGent) / Terrestrial Ecology Unit (TEREC) - Department of Biology; N.G. Ferreira, Aveiro University   CESAM / Departamento de Biologia and CESAM; R. Morgado, University of Aveiro / Department of Biology and CESAM; D. Nunes Cardoso, CESAM, University of Aveiro / department of Biology & CESAM; A. Cruz, University of Aveiro; S. Mendo, A.M. Soares, University of Aveiro / department of Biology & CESAM; S. Loureiro, Universidade de Aveiro / Biology
Anthropogenic activities such as the use of pesticides may have indirect disastrous consequences to aquatic organisms. An example of a widely used fungicide is carbendazim, which has a high potential to end up in aquatic ecosystems mainly through runoff. The deleterious effects observed at the population level can often be depicted or explained by changes in homeostasis at cellular and individual levels. In the present study, an isoclonal population of *Daphnia magna* (clone k6) was exposed to an environmentally relevant concentration (5 µg/L) of carbendazim during thirteen generations. The effects of carbendazim on survival/longevity, reproduction, parental length, DNA damage (determined by comet assay), biochemical biomarkers (cholinesterase, catalase and glutathione *S*-transferase), lipid peroxidation and energy-related parameters (carbohydrates, lipids and proteins jointly with energy available and energy consumption) were assessed in some generations. The long-term exposure to carbendazim presented no effect on the intrinsic rate of natural increase (*r*) and length of adult *D. magna*. However, daphnids longevity decreased at F12 generation and an increase in DNA damage from generation F3 to F13 was found when compared to daphnids in clean medium. Cholinesterases and glutathione *S*-transferase activities and lipid peroxidation showed differences between non-exposed and exposed populations to carbendazim. However, for catalase and energy related-parameters (except lipids) no differences were observed between these two *Daphnia* populations. Overall, at the tested concentration, carbendazim induced low effects under a long-term exposure to a daphnid population.

Prioritisation and Intelligent Testing of Pharmaceuticals in the Environment (P)

WE001

Development of a modelling framework for estimating the sorption of pharmaceuticals in soils

L. Carter, University of York / Environment Department; J. Wilkinson, The University of York / Natural and Built Environments; A. Boxall, University of York / Environment Department
Ionisable pharmaceuticals comprise a significant and increasing proportion of chemicals used in Europe. At typical environmental pH, ionisable pharmaceuticals can become charged. Speciation can alter the fate and behaviour of a chemical in the environment including its sorption potential to soils and sludge. It is essential that this behaviour is recognised within chemical risk assessment and predictive approaches are able to account for how speciation alters chemical sorption. Several authors have proposed approaches to predict the sorption of ionisable chemicals in soils. However, these models are typically based on training sets containing a multitude of organic chemicals and their ability to predict ionisable pharmaceutical sorption specifically needs to be evaluated. We therefore evaluated a range of predictive approaches, that take into account sorbent properties (i.e. soil characteristics), for their suitability for estimating sorption of pharmaceuticals in soil. The evaluations were done using a database of high quality experimentally-determined pharmaceutical sorption coefficients provided by industry partners. Models developed for specific classes of ionisable chemicals (i.e. cations or anions) performed better in comparison to simple generic models, which assume that hydrophobicity is the key sorption mechanism and neglect to take into account of the effects of chemical speciation. Nevertheless, model predictions for anionic pharmaceuticals still performed poorly ($r^2 < 0.5$). Sorption coefficients for organic cations were typically within an order of magnitude of experimental values when sorption was considered as the sum of sorption to organic matter and to clay minerals. As sorption of neutral and anionic compounds were not well explained by the evaluated models, further model development was required for adequate prediction of soil sorption coefficients for these classes of molecule. A decision tree framework to guide the selection of appropriate sorption models by taking into account soil pH and ionisable functional groups has been created. This incorporates previously published models that performed well in our analysis and the development of new sorption models. Work is currently on-going to review sludge sorption models and will be presented. The authors acknowledge EU/EFPIA Innovative Medicines Initiative Joint Undertaking (iPiE grant n° 115735) for the financial support.

WE002

Photochemical transformation and intermediate formation processes in surface waters, in the context of climate change

D. Vione, M. Minella, C. Minero, University of Torino / Chemistry
Sunlight illumination of surface waters induces several photochemical reactions that play an important role in the transformation of naturally occurring compounds and of xenobiotics, in the inactivation of pathogens and in biogeochemical cycles. These processes involve both the direct photolysis of the target molecules (directly triggered by absorption of sunlight, if any), and their indirect or sensitised transformation. In the latter case, sunlight is absorbed by naturally-occurring photosensitisers (e.g. chromophoric dissolved organic matter or CDOM, nitrate and nitrite) to produce several reactive transient species that are involved in transformation reactions. The transients include, among others, the hydroxyl ($^{\cdot}OH$) and carbonate ($CO_3^{\cdot-}$) radicals, singlet oxygen ($^1O_2$) and CDOM triplet states (^{3}CDOM*). Their occurrence in surface-water environments is linked to irradiance and to key water parameters such as chemistry and depth [1,2]. The phototransformation of dissolved compounds involves an interplay between molecular photoreactivity and environmental features. Water chemistry and depth can affect both xenobiotics persistence and the possible formation of toxic or mutagenic intermediates. If an hazardous compound is preferentially produced by a certain photoreaction pathway, the environmental conditions can enhance or inhibit its formation in different surface-water environments [3]. The role of climate change on water chemistry and, as a consequence, on photochemical reactions is just starting to be investigated. The main difficulty is to disentangle climate effects from other disturbance factors (e.g. wastewater inputs) that may also operate and vary on the long term [4]. Climate change has the potential to deeply alter the photochemistry of freshwaters, but its effects could be very different in boreal vs. temperate environments. In the former case the main effects would involve water chemistry (browning), while in the latter case a range of phenomena (warming, treeline shifts, extended drought periods) would play key roles depending on the context. [1] Vione D, Minella M, Maurino V, Minero C. 2014. Chemistry Eur. J. 20:10590-10606. [2] Rosario-Ortiz FL, Canonica S. 2016. Environ. Sci. Technol. 50:12532-12547. [3] Avetta P, Fabbri D, Minella M, Brigante M, Maurino V, Minero C, Pazzi M, Vione D. 2016. Water Res. 105:383-394 [4] Minella M, Leoni B, Salmaso N, Savoye L, Sommaruga R, Vione D. 2016. Sci. Total Environ. 541:247-256.

WE003

How Pharmaceutical Industrial waste can make your medicines ineffective

N. Verma, Baddi University of Emerging Sciences & Technology / Pharmacy
\nSpread over 380 square kilometres in Himachal Pradesh's Solan district, the Baddi-Barotiwala-Nalagarh (BBN) industrial area is one of India's largest pharmaceutical manufacturing hubs. The region hosts around 500 small, medium and large pharma units and accounts for 35 per cent of Asia's total medicine production. But rapid industrialisation and a lax attitude towards safe disposal and management of pharma waste have raised concerns about the effects of pollution on the environment and health. Liquid waste from these units is also discharged through pipes and other outlets that open behind the plant or run underground and open into bushy areas. This released wastewater accumulates in or flows through nallahs, canals and rivulets into the Sirsa river. Effluents are also injected into the ground at night by digging bore wells or released during rains. Due to such practices, the BBN region remains prone to antibiotic pollution. Many companies manufacture formulations, or finished products such as tablets and syrups. Some companies also manufacture active pharmaceutical ingredients (APIs) or the main biologically active ingredient used in formulations, including antibiotics. These APIs can enter the environment due to insufficient treatment or improper disposal of waste and weak environmental regulations. They are among environmental persistent pharmaceutical pollutants which have not degraded completely during treatment. They may influence the genetic makeup of bacteria, leading to the survival of resistant bacteria and spread of antimicrobial resistance (AMR), a public health threat. The result of our study showed that all gaps leading to the release of

APIs in the environment are plugged. The discharge of pharmaceutical effluents should take place through proper waste management techniques and stringent environmental regulations. Currently, effluent standards are limited to chemical contaminants such as heavy metals. The government must adopt a new AMR-centric approach of waste management which considers APIs as a chemical contaminant. Laws must be made to ensure that there are no APIs in treated effluents. The government should support small-scale manufacturers to install and implement environmentally sound waste treatment and disposal techniques. Manufacturers with high-end WWTPs should also be strictly monitored. The SPCBs should conduct surveillance of APIs or antibiotic residues in the treated effluents and make data publicly available.

WE004

The environmental concentration and evaluation of active ingredients in pharmaceuticals in rivers flowing through urban area in Japan

T. Nishimura, Teikyo Heisei University / Faculty of Pharmaceutical Sciences; T. Suzuki, Y. Kosugi, K. Watanabe, Tokyo Metropolitan Institute of Public Health / Division of Environmental Health; A. Hirose, National Institute of Health Sciences / Division of Risk Assessment

The active ingredients in pharmaceuticalsare discharged into the aquatic environment after use mainly through sewage treatment facilities. There is concern about adverse effects on wildlife due to the possibility of the presence of highly susceptible species. However, we still know little about what kind of impact on wildlife in the environment. Based on these backgrounds, we set up environmental impact assessment methods for pharmaceuticals according to AMED's strategy and evaluated the risks. We have measured the environmental concentrations (MEC) of 31 kinds of active ingredients in marketing medicine, using liquid-chromatography with mass spectrometry, in representative seven urban rivers in Japan, once every four seasons in 2015-2016. The maximum detected concentrations of the active ingredients exceeded 100ng/L are shown below,olmesartan (571ng/L), valsartan (405ng/L), irbesartan (162ng/L), candesartan (113ng/L), losartan (117ng/L) for antihypertensive agent, and sulpiride (546ng/L) for antipsychotic agent, cralythromycin (445ng/L) for antibacterial agent, ketoprofen (150ng/L) for analgesic antipyretic agent, bezafibrate (200ng/L) for hyperlipidemia treatment drug, crotamiton (845ng/L) for antipruritic agent. Among target ingredients, the detect concentration of active ingredient contained in pharmaceuticals for the lifestyle-related disease, hypertension and lipid metabolism related disease, tended to be higher. The concentrations in the winter or spring was observed a higher tendency, but the detected concentrations of active ingredients greatly varied depend on river according to the type of lifestyle and the type of pharmaceuticals been spread. It was indicated that the detected active ingredients were derived mainly from sewage treatment water as it depends on the concentrations of sucralose measured at the same time. The pharmaceuticals whose maximum detected concentrations of active ingredients in each river water exceeded the predictive environmental concentrations (PEC) were five ingredients of candesartan, olmesartan, lorazepam, rosuvastatin and epinastine, even when the dilution ratio was doubled. This result suggests that in some circumstances it is necessary to consider lowering the dilution rate in the environment from 10 which sets it. Regarding the health effects on humans, the actual concentration for each daily minimum dose for each pharmaceutical ingredient was from 0.086% of lorazepam to 0.000001% of clofibric acid.

WE005

Evaluation of simple exposure models used for environmental prioritisation of active pharmaceutical ingredients

J. Wilkinson, The University of York / Natural and Built Environments; A. Boxall, L. Carter, University of York / Environment Department; E. Burns, University of York

Targeted quantification using analytical methods such as high performance liquid chromatography followed by tandem mass spectrometry (HPLC-MS/MS) are effectively used to monitor trace-levels (ng/L) of active pharmaceutical ingredients (API) in the aquatic environment. However, as more than 1500 chemicals are currently in-use as pharmaceuticals, the high cost of HPLC-MS/MS prohibits its widespread use in the monitoring and prioritisation of APIs. Predictive exposure models offer clear advantages to costly and analytically-intensive targeted API quantification. Predictive models are not without limitations and their assumptions and defults are, at times, not representative of actual environmental conditions. Here we evaluate the accuracy of simple exposure models used to generate predicted environmental API concentrations (PECs) and their suitability for prioritisation of APIs in the aquatic environment. Water samples (n=60) were collected in triplicate on a monthly basis for six-months both upstream and downstream from five wastewater treatment plants (WWTPs) discharging into four rivers in the UK. Measured environmental concentrations (MEC-values) of 33 APIs were determined by HPLC-MS/MS. PEC-values were determined using pharmaceutical use data from the National Health Service, the fractions of chemical excreted from the body and degraded during sewage treatment, the population equivalence of each WWTP, mean regional per-capita water use and the dilution ratio of treated sewage effluent in receiving rivers. API-specific PEC ranges were compared to complementary MEC ranges observed over the 6-month sampling campaign and PEC:MEC ratios were determined. PEC:MEC ratios were generally low (< 0.5), indicating that predicted API concentrations were lower than measured. Between rivers, PEC:MEC ratios were generally closest to measured values in the lowest flow (smallest) rivers and in stretches near the headwaters indicating that locations with minimal upstream contributions of sewage effluent produced the most accurate PECs. In terms of prioritisation, predicted concentrations successfully identified eight of the ten APIs measured at highest concentrations across all 5 study locations (metformin, gabapentin, atenolol, tramadol, desvenlafaxine, fexofenadine, sitagliptin and paracetamol). PECs may be best-used for prioritisation over use in more sensitive applications, such as risk assessment, as PECs were consistently shown to underestimate API concentrations.

WE006

The role of the water-sediment simulation test and its outcome in the environmental risk assessment (ERA) of pharmaceuticals

D. Gildemeister, Umweltbundesamt / German Environment Agency / IV2.2 Pharmceuticals; S. Schmitz, S. Zahorszki, German Environment Agency UBA / IV Pharmaceuticals; A. Hein, I. Rönnefahrt, German Environment Agency - UBA / Section IV 2.2 Pharmaceuticals

In view to the revision of the 'Guideline on the environmental risk assessment of medicinal products for human use' (EMEA/CHMP/SWP/4447/00, June 2006, rev. 2) current fate assessment is highly discussed concerning the water-sediment simulation test in tier A of the guideline. At the moment, only the partitioning into sediment triggers further assessment in the next tier of the guideline. Derived half lives were not used in refined predicted exposure concentration (PEC) calculations and transformation products (TP) were not specifically considered in the ERA. Now, UBA evaluates the data received in the last 10 years in order to determine the gaps and the benefit of the current fate assessment for the overall risk assessment e. g. classification of persistence. Especially the role of TP in the environment due to their frequent higher mobility compared to the parent compound is considered in the presented research. As a first step an overview is prepared on the overall persistence of pharmaceutical compounds in the aquatic environment. It is clearly demonstrated that total system half-lives already show a high persistence of pharmaceuticals in the aquatic environment. Furthermore it should be considered that especially for the sediment compartment often no kinetic model fits well enough to predict DT50 values. The risk of ground water contamination by bank filtration will be estimated by the physico-chemical characteristics e. g. lipophilicity / hydrophilicity by comparing parent compound and TP. The occurrence and identification of TP is often appeared to be something which is only "nice to have" but not really relevant for the risk assessment of human pharmaceuticals. TP are often more polar and stable in environmental compartments than the parent compounds. This is of high relevance for groundwater contamination. The identification of relevant TP is still often missing in provided studies. The water sediment simulation study is the only experimental study in the ERA which gives information about the possible behaviour and occurrence of parent compounds and TP in surface waters. Such results are relevant for monitoring and for the understanding which compartments are affected by pharmaceuticals. Instead of waiving OECD 308, the results should be better included in the ERA and communicated.

WE007

Expert System to Inform BCF Testing Strategies for Pharmaceuticals

A. Agatz, IBACON GmbH / Environment Department; L. Carter, University of York / Environment Department; P. Andrews, A. Nellis, SimOmics; S. Owen, AstraZeneca / Safety Health Environment; J. Snape, AstraZeneca UK Ltd. / AstraZeneca Global Environment; J. Timmis, SimOmics; A. Boxall, University of York / Environment Department

An important part of the environmental risk assessment (ERA) of pharmaceutical ingredients is the identification of the persistent, toxic and bioaccumulative properties (PBT assessment) of the molecule. Regulations and guidelines on how to conduct this assessment and what empirical data are required to do so are increasingly complex. Currently a large number of fish are used as part of the ERA process, particularly for experiments to determine the bioconcentration factor (BCF), even though research developments and guidelines already contain opportunities to significantly reduce the number of fish used via alternative methods and/or optimisation of the testing strategy. We developed a new software tool to support the PBT assessment of pharmaceutical ingredients by interpreting European regulatory needs and considering existing guidelines and the wider literature. The system generates transparent and evidence-based compound specific PBT assessment reports and BCF testing strategies if testing is required. In our strategy, the P and T assessments are conducted before the B assessment because the latter is currently only required to be conducted to categorise the compound as PBT or vPvB. Thus empirical BCF values are not always required as decisions are made according to specific trigger values which are either exceeded by a compound or not. This means that in many cases the use of appropriate BCF prediction models prevents the need for experimentation. If a fish BCF test is required, our tool suggests an experimental design with the ultimate aim of reducing the number of test organisms needed without sacrificing the test validity criteria. The novelty of our system is that it illustrates, in a transparent manner, how the system made its conclusions by incorporation of the argumentation tool ArtooPro. This tool visualises the system's decision incorporating what regulatory and guideline background was used to support that decision and what data, modelling approaches and assumptions were used in addition to the sources of data. Preliminary analysis of those compounds for which empirical fish BCF data are available in the literature against our new strategy revealed that if our strategy was followed in at least 19% of these cases an empirical study would have not been required.

WE008

Development of a quantitative Adverse Outcome Pathway-informed model to predict the risk posed by mixtures of non-steroidal anti-inflammatory drugs to fish

P. Marmon, J. Kayode, Brunel University London / College of Life Sciences; S. Owen, AstraZeneca / Safety Health Environment; L. Margiotta-Casaluci, Brunel University London / Institute of Environment, Health and Societies

The presence of low concentrations of non-steroidal anti-inflammatory drugs (NSAIDs) in the aquatic environment has raised the concern that chronic exposure to these compounds may cause adverse effects in wild fish populations, similar to those observed in human patients. This potential scenario has led to the inclusion of diclofenac in the European Union Watch List of emerging pollutants. Although the effects of diclofenac in fish have been investigated in over twenty published studies to date, the complexity of NSAIDs toxicology is such that many uncertainties still exist about the significance of those findings for environmental risk assessment. We hypothesise that the perturbation of cyclooxygenase (COX) activity in healthy fish tissues is the leading cause of adverse effects, as it is in humans. On the basis of this mechanistic starting point, we propose a quantitative Adverse Outcome Pathway-informed model that incorporates both pharmacokinetic and pharmacodynamic aspects of NSAIDs toxicology. After extracting all NSAIDs toxicity data available in the scientific literature, we applied drug uptake models to predict the plasma concentrations at which different effects would occur in laboratory studies. As all NSAIDs act by inhibiting the enzymes COX1 and/or COX2, we applied mixture pharmacology approaches to express the plasma effect concentrations of different NSAIDs as diclofenac-equivalents. A similar approach was used for measured and predicted river concentrations of NSAIDs, which were used to predict plasma concentrations of NSAIDs in wild fish. The overlay of the two approaches led to a visual model that enables a rapid assessment of the risk posed by environmental levels of NSAIDs to trigger multi-scale adverse effects. The major strength of the model is the ability to predict the toxicological potential of NSAIDs mixtures, expressed as diclofenac-equivalents. We propose that this mechanistic approach may provide a useful predictive tool to support the implementation of effective NSAIDs ecopharmacovigilance strategies and facilitate the regulatory interpretation of past and future toxicity data.

WE009

Evolution in the lab - How can we study the chronic exposure to pharmaceuticals over multiple generations?

K. Heye, Goethe University Frankfurt/ Main / Aquatic Toxicology; A. Schmidt, Goethe University Frankfurt / Aquatic Ecotoxicology; J. Oehlmann, Johann Wolfgang Goethe-Universitat Frankfurt / Aquatic Ecotoxicology

Long-term exposure of non-target organisms is rarely covered in the assessment of chemicals. Especially pharmaceuticals play an important role when it comes to long-term exposure. They enter the environment throughout the year and therefore pose a continuous risk to organisms. One of these ubiquitously detected pharmaceuticals is the antiepileptic drug carbamazepine (CBZ). Hardly degraded during conventional wastewater treatment, it contaminates a majority of waterbodies. How do organisms react to continuous exposure to pharmaceuticals like CBZ? To answer this question, the non-biting midge *Chironomus riparius* was chosen as a test organism for a multi-generation experiment. 2400 chironomid larvae (< 24 h old) were taken from a laboratory culture to set up two exposure cages – one where larvae were continuously exposed to the LC_{10} of CBZ (0.4 mg/L, nominal concentration) and one control. When we were sure that a new generation had started, egg clutches were taked out of the cages to set up two chronic toxicity tests. Lethal and effect concentrations of mortality and mean time to emergence were calculated using a non-linear regression model (logistic curve). Sensitivity was compared by looking at overlaps of the 95% confidence intervals (CI). Two months after the beginning of the experiment, mortality seemed to be lower in the pre-exposed group compared to the control. However, CI of the LC_{50} still overlapped (0.506 to 0.882 mg/L for the control and 0.729 to 1.1 mg/L for the pre-exposed group). Four months later, sensitivity was compared again. LC_{50} of the pre-exposed group was higher than in the control, with no overlap of the CI (0.668 to 1.02 mg/L for the control and 1.08 to 1.96 mg/L for the pre-exposed group). After two and six months, control mortality of both groups was low and emergence in the cages stayed constant. Multi-generation experiments are a helpful tool to investigate long-term effects of chemicals on aquatic organisms. Within the first six months of the ongoing study, midges showed to be less sensitive to carbamazepine after long-term low-level exposure. Chronic toxicity tests to study the sensitivity of exposure groups should be combined with genome and transcriptome analyses to get a full picture of adaptation processes in midges. *Acknowledgement* - The authors thank the Federal Ministry of Education and Research (BMBF) for funding (NiddaMan, project support code: 02WRM1367A).

WE010

Effects of duloxetine and econazole on freshwater species towards individual and combined conditions

G. AMARIEI, Universidad de Alcala; K. Boltes, University of Alcala / Chemical Enginnering; J. Valimaña-Traverso, M. García, P. Letón, M. Marina, R. Rosal, University of Alcala

Thousand of biologically active pharmaceuticals ingredients (APIs) are used in human and veterinary medicine over the world. Nowadays, the occurrence of pharmaceuticals in aquatic environments is a well-established issue, there are still gaps in our knowledge on the fate and effects of these compounds in the environment[1]. Evaluating API ecotoxicology is even more challenging due to uncertainties about appropriate dosages, durations of exposure, range of sensitive taxa, sensitivity of developmental stages, and toxicological endpoints[2]. More attention should be paid on the non-target organisms and the chiral nature of contaminants[3]. This work assess the toxicity of the antidepressant drug Duloxetine and the antifungal Econazole, individually and combined, on three freshwater species—algae, crustacean and duckweed, using APIs concentration from 0.039 to 100 mg L^{-1}. Level an type of drugs interactions were determined using the Combination Index-isobologram method. The enantiomers concentration of the target componds in the culture media were also measured to analize the relation between degradation profile and the observed toxicity on organisms. Results reveal toxic effects of Duloxetine and Econazole leading to growth reduction and significant changes in the morphology of duckweed fronds. The EC50 values obtained shown Duloxetine as very toxic for algae and toxic for crustacean and plants. Econazole appears as very toxic for all species evaluated. Mixed toxicity profile was different for the organisms studied, showing also dissimilar persistence of R and S enantiomers in culture media. [1] Minguez L, Pedelucq J, Farcy E, Ballandonne C, Budzinski H, Halm-Lemeille M.P. 2016. Toxicities of 48 pharmaceuticals and their freshwater and marine environmental assessment in northwestern France. Environ Sci Pollut Res 23: 4992. [2] Kostich M.S, Lazorchak J.M. 2008. Risks to aquatic organisms posed by human pharmaceutical use. Sci. Total Environ 25: 329-339 [3] Sanganyado E, Lu Z, Fu Q, Schlenk D, Gan J. 2017. Chiral pharmaceuticals: A review on their environmental occurrence and fate processes. Wat. Res. 124: 527-54 *Acknowledgement* - The research was co-funded by the Comunidad de Madrid, grants S2013/MAE_2716 REMTAVARES and grants CCG2016/EXP-037 by University of Alcalá.

WE011

Application of newly developed in vitro assay to detect physiological activities of antidepressants in wastewater

M. Ihara, M.O. Ihara, D. Kato, H. ZHANG, Kyoto University

Over recent years, growing numbers of human pharmaceuticals have been detected in effluents of wastewater treatment plants (WWTPs). Concern about their potential risks to aquatic species has been raised because they are designed to be biologically active. One of most concerned pharmaceuticals are antidepressants. For example, selective serotonin reuptake inhibitors (SSRIs) such as fluoxetine and sertraline could alter the behaviour of fish in vivo testing. Antidepressants such as SSRIs, serotonin-norepinephrine reuptake inhibitors (SNRIs), dopamine reuptake inhibitors (DRIs), and tricyclic antidepressants (TCAs) are now on the market worldwide. It is possible to measure the concentrations of selected antidepressants by chemical analysis, but such concentrations do not indicate the physiological activity of antidepressants in waters. For example, even if the concentration of each substance is low, through additivity compounds might produce a strong enough physiological activity to harm aquatic organisms. To determine whether antidepressants in aquatic environments alter the behaviour of aquatic organisms, we must know the extent to which such organisms may be exposed to antidepressants as determined by the inhibition of monoamine transporters. In this study, we measured the physiological activity of antidepressants in WWTP effluents for the first time by the in vitro assay (namely called antidepressant assay). We utilized fluorescence substrate, APP, for monoamine transporters (serotonin transporters (SERT), norepinephrine transporter (NET), and dopamine transporter (DAT)). By transfecting a plasmid expressing transporter into cultured cell line, and measuring the fluorescence intensity inside the transfected cells, inhibitory activity of antidepressant on the uptake of APP by transporter could be quantified. We applied the antidepressant assay to secondary effluents (SEs) of WWTPs in Japan, and succeeded to detect the inhibitory activity of antidepressants in SEs. Inhibition was detected in SERT or NET-expressing cells, but not in DAT-expressing cells, suggesting that detected inhibitory activity come from SSRIs, SNRIs, and/or TCAs, not DRIs in SEs. Activities detected in SEs could be quantified as antidepressant-equivalent quantities (EQs). By comoaring EQ values with the effective concentrations of antidepressants in vivo behavior testing, we can know whether antidepressants in environmental water is really risky to aquatic organisms.

WE012

Toxicology of pharmaceuticals to aquatic organisms: a meta-analysis of effects on development and reproduction

V.F. Fonseca, I.A. Duarte, MARE - Marine and Environmental Sciences Centre; H.N. Cabral, Faculdade de Ciências da Universidade de Lisboa / MARE-FCUL; B.M. Gillanders, School of Biological Sciences, The University of Adelaide / Southern Seas Ecology Laboratories,; P. Reis-Santos, MARE - Marine and Environmental Sciences Centre

The ubiquity of pharmaceutical compounds in the aquatic environment, their growing use and their potential to elicit biological effects even at low concentrations is a major concern to environmental health and safety management. In recent years an increasing number of studies have addressed the presence and toxicity of various pharmaceuticals, using various biological endpoints in different biological models, and reporting varying effects. Accordingly, a systematic quantitative assessment is key to improve current understanding of the ecological risks of pharmaceutical compounds to non-target organisms in the aquatic environment. To unravel patterns in biological responses across aquatic taxa a meta-analysis was performed on reported effects of exposure to pharmaceutical compounds (according to therapeutic class). Minimum response concentration and biological responses were collected from selected studies based on a set of objective criteria considering organisms' exposure to pharmaceuticals under controlled conditions. For a response sensitivity analysis various endpoints were considered, namely biochemical, developmental (e.g. growth), reproductive and behavioral responses, as well as lethality, in studies reporting effects on aquatic taxa. The comparative sensitivity analysis of biological endpoints highlighted the sensitivity of molecular responses, followed by individual level-responses (e.g. behavior and growth), yet variable sensitivity scores among taxa were observed when considering different pharmaceutical classes. Data availability and comparability limited quantitative analysis, yet the meta-analysis provided a key framework to compare effects influence on development and reproduction of crustacean and fish exposed to pharmaceuticals. Overall the implications of current findings for environmental monitoring and ecological risks of pharmaceuticals in aquatic ecosystems are discussed.

WE013

Leveraging Pharmacological Data for Prioritization of the Ecological Risks of Chiral Pharmaceuticals

E. Sanganyado, Shantou University / Marine Biology Institute; J. Gan, University of California, Riverside / Department of Environmental Sciences

Pharmaceuticals are frequently detected in wastewater and the environment at concentrations ranging from ng/L to µg/L. More than 50 % pharmaceuticals are chiral compounds. Enantiomers of chiral pharmaceuticals have been shown to exhibit differences in environmental fate, transport and toxicity. Since more than 2,500 pharmaceuticals are currently in use, it is implausible to carry out whole organism toxicity studies of all pharmaceuticals. However, there is a wealth of knowledge available from drug discovery and development research that can be leveraged for predicting potential environmental exposure and effects of chiral pharmaceuticals. Assuming evolutionary conservation of primary drug target, read-across method can be used to predict the potential effect of chiral pharmaceuticals in fish. In this study, we estimated the stereoselective effect of 11 chiral pharmaceuticals using the fish plasma model. We found metoprolol had high risk with an effect ratio, ER (ratio of human therapeutic plasma concentration to fish plasma concentration at steady state) that was less than 1.0, whereas propranolol, salbutamol, fluoxetine and venlafaxine were medium risk ($1.0 < ER < 30$). However, stereoselectivity was predicted in all compounds except atenolol and pindolol. In this study, we showed the fish plasma model has considerable potential as a tool for predicting stereoselective toxicity of chiral pharmaceuticals.

WE014

Effects of benzoylecgonine exposure at different levels of the biological hierarchy on Daphnia magna

M. Parolini, University of Milan / Department of Environmental Science and Policy; B. De Felice, Università degli Studi di Milano; C. Ferrario, University of Milano Bicocca; N. Salgueiro-Gonzalez, IRCCS Istituto di Ricerche Farmacologiche Mario Negri; S. Castiglioni, Mario Negri Institute / Environmental Health Sciences; P. Tremolada, University of Milano / Deprtment of Biomolecular Sciences and Biotecnology; A. Finizio, University Milano - Bicocca / Department of Earth and Environmental Sciences

A number of monitoring studies have shown that benzoylecgonine (BE), a metabolite of cocaine, is the main illicit drug residue measured in both wastewater and surface waters worldwide. Although the aquatic concentration of BE can be considered still low, the exposure to this molecule may cause diverse adverse effects. The few studies that have investigated the toxicity of this molecule towards invertebrate and vertebrate aquatic non-target organisms have shown different detrimental effects at low levels of the biological organization, mainly at biochemical, molecular and cellular levels. However, to date no one study has evaluated the consequences of BE exposure to the higher levels of ecological hierarchy. Thus, the present study was aimed at investigating the toxicity of a 48-h exposure to two concentrations of BE, similar to those found in aquatic ecosystems (0.5 µg/L and 1.0 µg/L), on the cladoceran *Daphnia magna* at different levels of the ecological hierarchy. We relied on a multi-level approach focusing on the effects at biochemical/biomolecular (biomarkers), individual (swimming activity) and population (reproduction) levels. As previous studies of BE have shown that this molecule can induce oxidative stress, we assessed the amount of reactive oxygen species and of the activity of antioxidant (SOD, CAT, and GPx) and detoxifying (GST) enzymes and the lipid peroxidation (TBARS) as oxidative stress endpoints. We also measured the acetylcholinesterase (AChE) activity because this enzyme is strictly related to behavioral changes in aquatic organisms. Alterations in the swimming behaviour of *D. magna* were investigated by a video tracking analysis, while the consequences on the reproduction were assessed by a chronic toxicity test. Our results showed that the exposure to two BE concentrations similar to those found in aquatic ecosystems induced oxidative stress and inhibited the activity of AChE, affecting the swimming behavior and the reproduction of *Daphnia magna* individuals.

WE015

Impact of the antidiabetic drug metformin and its transformation product guanylurea on brown trout (Salmo trutta f. fario)

S. Jacob, Universität Tübingen / Animal Physiological Ecology; L. Kundy, M. Biecker, University of Tuebingen; R. Triebskorn, University of Tuebingen / Animal Physiological Ecology

The last decades, the number of patients suffering from diabetes type 2 is increasing. Consequently, a rising consumption of antidiabetic drugs as metformin (MF) has become obvious. In wastewater treatment plants, metformin is only partially retained and also transformed to guanylurea (GU) leading to high concentrations of both compounds in surface waters. However, possible effects of MF and GU in aquatic organisms are far from being understood. The aim of this study is therefore to investigate influences of MF and GU on different metabolic pathways and behaviour in different life stages of brown trout (*Salmo trutta* f. *fario*). Juvenile trout (age: 8 month) were exposed for 4 weeks at 7°C to different concentrations of MF (0, 10, 1000 µg/L) and GU (0, 10, 100, 1000 µg/L). Additionally, eggs of brown trout in the eyed ova stage were exposed to different metformin concentrations (0, 1, 10, 100, 1000 µ/L) at 7°C & 11°C (regarding possible interactions of chemical toxicity & temperature). To show influences on the embryo development, mortality, hatching rate, and heart rate were recorded. Tissue samples were taken three and eight weeks after the end of the sac-fry stage. In all experiments, several endpoints characterizing fish health were investigated, including the histological condition of the liver, alterations in the stress protein level (Hsp70), changes in the intestinal microbiome and additionally the glycogen storage in the liver of MF-exposed fish. Besides, swimming and predator-prey behaviour were investigated. There was no influence of MF on the developmental parameters in brown trout larvae. Neither behaviour nor stress protein level were influenced by MF. The liver tissue of the MF-exposed trout was in a good condition. The glycogen storage was tendentially increasing in MF-exposed fish compared to the control, whereas the glycogen content of the trout exposed to 1000 µg/L MF was partially decreased. The intestinal microbiome of MF-exposed larvae showed a significantly different composition compared to the control. The results for the experiment with GU will be presented (analyses not yet finished). This work is part of the project Eff-Net (Effect Network in Water Research) funded by the Wassernetzwerk Baden-Württemberg. By a multi-disciplinary approach, antidiabetics, artificial sweeteners, antidepressants and their transformation products are studied from the mode of action at the molecular level up to the effects at the community level.

WE016

Effect of life-cycle exposure to environmentally relevant concentrations of metformin and its metabolite guanylurea on F1 progeny 28 days post hatch.

Z.P. Pandelides, University of Ontario Institute of Technology; E. Ussery, University of Ontario Institute of Technology / Biological Sciences; J. Guchardi, University of Ontario Institute of Technology / Faculty of Science Aquatic Toxicology; D.A. Holdway, University of Ontario Inst. of Tech / Science

Recent studies have demonstrated that the type 2 diabetic drug metformin and its only known metabolite, guanylurea, are common environmental contaminants found in the ng-µg/L concentration range in surface waters and wastewater effluent. This should be of concern as recent work in our lab shows that Japanese medaka (*Oryzias latipes*) exposed to environmentally relevant concentrations of metformin and guanylurea from embryo through 28 days post-hatch have a significant decrease in length and weight of both males and females when compared to control fish. Furthermore, our studies show that larvae exposed for 28 days to both compounds have a significant dysregulation in lipid and fatty acid metabolism, possibly leading to this stunted growth. A full life-cycle exposure to both compounds at environmentally relevant concentrations, alone and in combination, was conducted in order to examine the effects of chronic exposure to the F1 progeny. The effects of metformin and guanylurea on the length and wet weight were compared 28 days post hatch and will be discussed. Possible implications of exposure to metabolomics and gene expression will be explored

WE017

Life-cycle effects in Oryzias latipes exposed to environmentally relevant concentrations of metformin and its metabolite, guanylurea.

E. Ussery, University of Ontario Institute of Technology / Biological Sciences; Z.P. Pandelides, University of Ontario Institute of Technology; J. Guchardi, University of Ontario Institute of Technology / Faculty of Science Aquatic Toxicology; D.A. Holdway, University of Ontario Inst. of Tech / Science

One of the current most common contaminants in the aquatic environment is the type 2 diabetic drug metformin. Metformin has been measured in the ng-µg/L concentration range in both German and North American surface waters and wastewater effluent. As the majority (>90%) of metformin is metabolized into guanylurea during the waste water treatment process, it is found in the environment in higher concentrations than metformin, usually in the µg/L concentration range in surface waters. This is concerning, as our recent research shows that Japanese medaka (*Oryzias latipes*) exposed to environmentally relevant concentrations of metformin (1.0-100 µg/L) and guanylurea (1.0-100 ng/L) from embryo through 28 days post hatch have a significant decrease in length (mm) and weight (mg) of both males and females when compared to control fish, with guanylurea appearing to be roughly 1,000 times more potent than its parent compound, metformin. Furthermore, these studies show significant changes in the metabolome of 28 day old male medaka exposed to both metformin and guanylurea, indicating significant dysregulation in fatty acid and lipid metabolism. These results raised concern regarding the consequences of a full life cycle exposure, including the important reproductive phase. Thus, a full life-cycle continuous exposure experiment was undertaken utilizing both compounds at environmentally relevant concentrations, alone and in combination, was conducted in order to examine the effects of chronic exposure on growth, reproduction, steroid production, and gene expression. Results will be discussed.

WE018

Environmental Fate and Effects of the Antidiabetic Drug Metformin and Its Transformation Product Guanylurea

J. Straub, F.Hoffmann-La Roche Ltd / Roche Group Safety, Health & Environmental Protection; D.J. Caldwell, Johnson & Johnson / Environment Health Safety   Sustainability; V. D'Aco, Quantum Management Group, Inc.; T. Davidson, Bristol-Myers Squibb / EHS; K. Kappler, Johnson & Johnson; S. Owen, AstraZeneca / Safety Health Environment; B. Simon-Hettich, Merck KGaA; J.G. Tell, Merck & Company, Inc. / Global Safety   the Environment

Metformin (MET) is an active pharmaceutical ingredient (API) with very high patient use worldwide that is excreted in unchanged form. This has led to concern about the potential aquatic life impacts associated with the presence of MET in surface waters. MET is largely transformed to guanylurea (GUU) in WWTP, and both MET and GUU are further degraded in the environment. A comprehensive aquatic life risk assessment of MET and GUU in surface water is presented that is based on literature data, previously unpublished data from industry studies conducted to support new drug registration applications as well as new studies commissioned to fill data gaps. Predicted environmental concentrations (PECs) for MET were modelled based on documented usage for the USA with the PhATE model and for the European Union with the GREAT-ER model. These PECs were compared with measured environmental concentrations (MECs) for both the USA and EU. A predicted no effect concentration (PNEC) for MET was derived by deterministic procedures based on multiple chronic studies with algae (4), daphnids (5) and fish (5, two species). Both the PEC/PNEC and MEC/PNEC risk characterization ratios were well below 1, indicating no significant risk for MET with high Margins of Safety. However, since MET is known to be primarily degraded during wastewater treatment to GUU, relevant chronic studies for GUU were conducted to derive a PNEC. In addition, PECs were derived for GUU for the USA and EU as above for MET. Fate and removal/in-stream-loss parameters for both MET and GUU show alignment of PECs and MECs in the USA and Europe. The PEC/PNEC and MEC/PNEC risk characterization ratios for GUU were also below 1. We conclude there is no significant risk to aquatic life for both MET and its transformation product GUU.

WE019

Fluoxetine exposure modulated antioxidant and anxiety-related gene expression altering swimming activity in zebrafish embryos

B. De Felice, Università degli Studi di Milano; A. Ghilardi, L. Del Giacco, University of Milan; M. Parolini, University of Milan / Department of Environmental Science and Policy

The massive release of human pharmaceuticals into the aquatic ecosystems continues to be a serious environmental problem. Among pharmaceuticals, psychotropic drugs such as antidepressants are one of the main therapeutic classes detected in freshwater worldwide. Selective serotonin reuptake inhibitors (SSRIs) are commonly the first-line antidepressant drugs prescribed to alleviate anxiety disorders in humans, and fluoxetine (FLX), the active principle of the Prozac®, is one of the most used SSRI worldwide. After human consumption FLX enters the aquatic ecosystems, whereby it has been detected in the high ng/L to low µg/L concentration range. Although many studies have demonstrated that the exposure to FLX caused a plethora of adverse effects in aquatic species, the information regarding its molecular mechanisms of action and the relationship with organism behavior remains scant.Thus, the present study was aimed at investigating 1) the effects induced by two concentrations of FLX (50 ng/L and 500 ng/L) on the expression of genes related to oxidative stress response (*sod1, sod2, cat, gpx* and *gst*), stress and anxiety (*oxtl, prl2, npy* and *ucn3l*), as well as tranporters of main neurotransmitters (*slc6a3, slc6a4a, slc6a4b, slc6a11*) and 2) if changes in the expression of neurotoxicity-related genes was related to changes in the swimming behavior of zebrafish (*Danio rerio*) embryos at 96 hours post fertilization (hpf). Our results showed that FLX exposure overexpressed *sod1, cat* and *gpx*, suggesting that this drug can induce an overproduction of pro-oxidant molecules. In addition, changes in the expression of *slc6a4a, slc6a4b, slc6a11* genes indicated that FLX can affect neurotransmission and, consequently, alter swimming behavior of embryos, as demonstrated by the significant reduction of the distance moved by treated embryos in response to an external stimulus.

WE020

Bio-Optical probing of Bezafibrate toxicity in model marine diatom Phaeodactylum tricornutum

B. Duarte, MARE - Marine and Environmental Sciences Centre / Centro de Oceanografia; A. Matos, BioISIBiosystems and Integrative Sciences Institute / Plant Functional Genomics Group; T. Cabrita, IPMA IP; J.C. Marques, University of Coimbra / MARE, Dep. of Life Sciences, Coimbra University; I. Caçador, Faculdade de Ciências da Universidade de Lisboa / MARE - Marine and Environmental Sciences Centre; P. Reis-Santos, MARE - Marine and Environmental Sciences Centre; H.N. Cabral, Faculdade de Ciências da Universidade de Lisboa / MARE-FCUL; V.F. Fonseca, MARE   Marine and Environmental Sciences Centre

The occurrence and fate of pharmaceutical active compounds in aquatic environments has become a major cause for concern due to their effects on humans and aquatic ecosystems. The high consumption of blood lipid regulators is leading to increased occurrence of fibrates in natural streams and wastewater effluents. Fibrates are a class of drugs derived from fibric acid widely used to reduce plasma triglycerides and raise the level of high-density lipoprotein cholesterol. Specifically, Bezafibrate is extensively used as a lipid regulator with consumption greatly increasing over the years in developed countries. Due to its large use and its persistence, bezafibrate has been detected in surface and drinking waters as well as in wastewater effluents. This can have serious impacts on marine life, including on marine primary producers and thus impacting the whole system productivity and functioning. Exposing the model diatom *Phaeodactylum tricornutum* to a range of environmentally relevant concentrations of bezafibrate (0-60 mg/L) revealed no serious impacts on cell growth. Nevertheless, after 48h of exposure damages in the photosynthetic apparatus were detected using bio-physical probing Pulse Amplitude Modulated (PAM) Fluorometry. Bezafibrate exposure impaired both photosystems, which reduced the algae ability to harvest photonic light and convert it into an electron flow, and thus its chemical energy production (ATP). This may result from a direct effect of bezafibrate in membrane fatty acids from the chloroplast, since both photosystems are anchored in a lipidic membrane system. Moreover triglycerides (TAGs) are known to protect the photosystems against photoinhibition. The reduction of TAGs could lead to burnout of the photosystems due to excessive energy being absorbed, as observed by the high incoming photonic energy flux, this way reducing the number of active reaction centers in the algae and thus its photosynthetic ability. All these bio-optical parameters show a clear dose-effect relationship, indicating that *P. tricornutum* is a good candidate organism for fibrate toxicity testing in marine systems, screened by non-invasive high-throughput bio-physical probing tools.

WE021

Environmental Risk Assessment for the Active Pharmaceutical Ingredient Mycophenolic Acid in European Surface Waters

J. Straub, F.Hoffmann-La Roche Ltd / Roche Group Safety, Health & Environmental Protection; A. Haener, F. Hoffmann-La Roche Ltd / Group SHE (LSO)

An Environmental Risk Assessment (ERA) was performed for the active pharmaceutical ingredient mycophenolic acid (MPA) for Europe. MPA is an older immune inhibitor developed in the USA in the 1990s by Syntex, Inc., now a part of the Roche Group. So far, no sufficient dataset describing biodegradability, environmental fate or ecotoxicity for MPA nor an ERA for MPA have been available. The present ERA is based on old environmental data from Syntex and on new tests, all performed under GLP quality assurance, for physico-chemical characteristics, partitioning, environmental fate, biodegradability and (sub)chronic\necotoxicity and on sales amounts for the products containing MPA in Europe. Both a new biodegradation study and an older sediment/water fate test show that MPA is not recalcitrant but undergoes primary up to ultimate biodegradation in wastewater treatment and surface water models. A predicted environmental concentration (PEC) in Europe from all products containing MPA was calculated based on compound actual use data from IMS Health, Inc. per annum and country, incorporating population data from Eurostat, for the decade 2004–2014. A crude inital PEC was derived based on standard ERA assumptions of no removal in sewage treatment or surface waters. The crude PEC was refined by incorporating predicted sewage works removal, based on new biodegradability data, and by country-specific dilution factors. The lowest of the no observed effect concentrations from chronic and subchronic tests with algae, daphnia and fish was divided by an assessment factor of 10 to derive the chronic-based predicted no effect concentration (PNEC). Potential risk for surface waters was then quantified by dividing the PECs by the PNEC. Potential risk from MPA was also assessed for sewage works and bacterial populations. In addition, MPA is not expected to bioaccumulate nor to adsorb to sewage sludge or to sediment to a significant extent. Conclusions on potential risks of MPA are given in the poster.

WE022

Cytostatics in Dutch surface water - overview of use and potential risks to the aquatic environment

m. marinkovic, National Institute for Public Health and the Environment (RIVM); C. Moermond, RIVM / Centre for Safety of Substances and Products; B. Venhuis, RIVM; P.v. Vlaardingen, RIVM / Expert Centre for Substances; M. van Elk, RIVM / GZB; A. Oostlander, J. van Dijk, RIVM
After use, pharmaceuticals and their residues eventually end up in the sewage system. Sewage treatment plants reduce the nutrient load of waste water, and while organic micropollutant removal occurs concomitantly by bacterial activity and sorption, many contaminants, including pharmaceuticals, are only partly removed. Consequently, effluent containing pharmaceuticals and their residues is discharged into surface waters. A recent study showed that 29 of 80 monitored pharmaceuticals were regularly detected in Dutch surface water, and that five of these substances, i.e. the pain killer diclofenac, the antibiotics azithromycin, clarithromycin and sulfamethoxazole, and the anti-epileptic carbamazepine, pose a risk to the aquatic ecosystem (Moermond et al., 2016). This raises concern, perhaps even more so when considering that for many of the around 2000 pharmaceuticals that were authorized for the Dutch market in 2016, it remains unknown to what extent they are present in surface waters, how they behave in the environment, and to what extent they exert toxicity to aquatic species individually and jointly. A class of pharmaceuticals that has received increased attention in the Netherlands, but also in the EU, e.g. PHARMAS project and Cytothreat, are cytostatics. These potent substances are used to inhibit cell division in cancer patients, but the fraction released unchanged to surface water could affect aquatic species in a similar manner. This project aimed to provide an overview of the use of cytostatics in the Netherlands and to determine if cytostatics pose a potential risk to the aquatic environment. First, an inventory was made of cytostatics use in the Netherlands. A top 10 of cytostatics was compiled by taking into account the metabolic transformation of cytostatics in patients, the removal efficiency in sewage treatment plants, and the available monitoring data in Dutch surface waters. For these 10 cytostatics, available environmental fate and effect data were gathered and safe environmental concentrations were derived. Comparison to predicted and measured environmental concentrations will allow to conclude if the selected cytostatics pose a potential risk to the aquatic environment.

WE023
Environmental risk assessment of human pharmaceuticals - what can we learn from regulatory effect data so far?
S. Schwarz, German Environment Agency UBA / Section IV 2.2 Pharmaceuticals; J. Bachmann, German Environment Agency (UBA) / Section IV2.2 Environmental Risk Assessment of Pharmaceuticals; U. Brandt, German Environment Agency UBA / Section IV Environmental Risk Assessment of Pharmaceuticals
Since the coming into force of the guideline on the environmental risk assessment of medicinal products for human use (EMEA/CHMP/SWP/4447/00 corr 2), the German Environment Agency (UBA) is tasked with environmental risk assessment of human pharmaceuticals. Applicants seeking approval of medicinal products need to submit fate and effect data, in case predicted environmental concentrations exceed 10 ng/L in surfacewaters, or the substance is of specific concern through its mode of action or physico-chemical characteristics. Basic aquatic ecotoxicity data includes testing of three trophic levels, represented by algae, crustaceans and fish – usually following OECD-guidelines 201, 211 and 210. Both the applicant and the assessor evaluate the studies to assure adequate data quality. Over the last decade, this regulatory work resulted in a data base containing effect data on approximately 300 active pharmaceutical ingredients (APIs) – which was evaluated in our current project: More than 12 % of evaluated APIs show NOECs below 1 µg/L; with several NOECs in the low ng/L-range, particularly for substances with endocrine mode of action. The predominant part of substances with NOECs between 0.01 and 1 µg/L is non-endocrine, belonging to a diverse range of pharmaceutical classes. For approximately 2/3 of investigated APIs, valid effect studies on all three trophic levels were available – allowing a comparison of sensitivity. In over 60 % of cases, the effect value quotient of most and least sensitive test organism was greater than 10, in over 20 % of cases greater than 100. Fish were the most sensitive test organism in more than half of the cases, while algae and crustaceans were the most sensitive in one quarter, each. Detailed information concerning specific pharmaceutical groups/mode of actions will be given in the final poster. Our results will help to identify possibilities and limitations of the current regulatory approach, and provide information for future modifications of the regulatory framework.

WE024
Prioritisation of human pharmaceutical substances - a regulatory perspective
I. Rönnefahrt, German Environment Agency - UBA / Section IV 2.2 Pharmaceuticals; S. Konradi, Federal Environment Agency (Umweltbundesamt) / Section IV 2.2 Pharmaceuticals; A. Hein, S. Schwarz, German Environment Agency UBA / Section IV 2.2 Pharmaceuticals; K. Westphal-Settele, German Environment Agency (UBA) / Section IV.2.2 Pharmaceuticals; I. Ebert, German Environment Agency UBA / Section IV 2.2 Pharmaceuticals
Pharmaceuticals in the environment have been recognized by the European Commission as emerging issue. Possible actions to reduce their emission into the environment and the need for amendments of the legislation are currently discussed in the 'strategic approach to pharmaceuticals in the environment'. At the German market, there are currently about 2300 active pharmaceutical substances used in human medicinal products; at least 1200 of them are compounds of potential environmental concern. For the majority of these 1200 compounds data for an environmental risk assessment (ERA) are incomplete or lacking, with the result that their potential environmental impact cannot be assessed in an appropriate manner. The reason for this is simple: So called 'legacy products' have been authorised before the 'Guideline on the environmental risk assessment of medicinal products for human use' came into effect in 2006. According to the current legislation all new marketing authorisation applications have to be accompanied by an ERA. However, there are no statutory provisions in place how to deal with legacy products. Hence, there is a vital need to prioritise active substances used in legacy products for further investigations and evaluation of their environmental impact as well as risk management activities. This is of particular importance because many of them are frequently detected in surface water and other environmental compartments. Moreover, active substances which are persistent, mobile, bioaccumulative and/or toxic or have a specific mode of action as e.g. endocrine active substances, are in general of high environmental concern. We propose a step wise prioritisation concept that allows the identification of active substances with a high potential environmental impact and/or a high potential presence in the environment. The poster outlines parameters which should be considered in a prioritisation approach, as e.g. consumption data and their trends over the years, mode of action, monitoring data, available data on fate and effects in the environment and metabolism in patients. The proposed tiered prioritization approach considers also elements of the EMA Guideline for environmental risk assessment of human pharmaceuticals. It is important to recognise that any approach needs to be focused on the overall objective of the prioritisation and should be regularly adapted to the current state of knowledge.\n

WE025
SETAC Pharmaceuticals Interest Group
G. Maack, German Environment Agency / Ecotoxicological Assessment

WE026
What makes a chemical substance a 'natural substance'? A case study in the context of the EU veterinary medicines marketing authorisation procedure
T. Hahn, Fraunhofer Institute Toxicology and Experimental Medicine / Chemical Risk Assessment; C. Floeter, HAW Hamburg / Department of Environmental Engineering; S. Schwonbeck, G. Koennecker, Fraunhofer Institute Toxicology and Experimental Medicine / Chemical Risk Assessment
The marketing authorisation process for veterinary pharmaceuticals in the EU and other countries requires an environmental impact assessment (EIA) for each veterinary medicinal product (VMP). This EIA follows a phased approach with conservative estimates of environmental exposure in phase I, which may later require refinement by experimental data in phase II. Core of the phase I assessment is a catalogue of 19 questions on use and characteristics of the VMP under consultation. These questions aim at establishing an initial predicted environmental concentration, which, together with information on therapeutic use and targeted animal species, may trigger the necessity to perform an in-depth phase II assessment. According to question 2 in the phase I assessment a substance is exempted from further investigation when there is scientific proof that it is a natural substance "the use of which will not alter the concentration or distribution in the environment". At first sight this definition appears unambiguous. Nevertheless may it be a hurdle for applicants because no further guidance is given which criteria apply for acceptance or rejection of a given concentration as 'natural'. Here, we present a case study comprising two substances in order to highlight possible uncertainties for applicant companies, as well as for competent authorities.

Obesogens and lipid disruptors (P)

WE027
Unraveling distinct pathways of PFOS toxicity by combining morphological, metabolomic and transcriptomic analyses
R.M. López, Institute of Environmental Assessment and Water Research (IDAEA-CSIC) / Environmental Toxicology; L. Navarro-Martín, C. Luccarelli, IDAEA-CSIC; E. Ortiz, IDAEA-CSIC / Department of Environmental Chemistry; A.E. Codina, CNAG; D. Raldua, IDAEA-CSIC; C. Barata, CSIC / Environmental Chemistry; B. Pina, IDAEA-CSIC / Department of Environmental Chemistry; R. Tauler, IDAEA-CSIC / Environmental Chemistry
Exposure to PFOS (perfluorinated octyl sulfonate) has been related to toxic effects on lipid metabolism, immunological response, and different endocrine systems. We present here a combined metabololmic and transcriptomic analysis of zebrafish embryos exposed to different concentrations of PFOS (30-1000 ppb) from 48 to 120 hpf. While parallel morphological analysis showed no macroscopic changes below the 1000 ppb mark, some metabolomic and transcriptomic chages occured even at the lowest used concentration. Functional analyses of the observed changes revealed at least three major modes of action: alteration of PPAR signalling and lipid metabolism, effects on cell-cell interaction, perhaps linked to effects on the immuno response and neuronal system development, and a general alteration of the development, reflected by an alteration of different development- and metabolism-related signalling pathways, likely affecting to cell cycle functions, and

to the metabolism of proteins, nucleotides, and amino acids. The results suggest a complex, multiple endocrine disruption-like toxic effects, at a concentrations well bellow the 1 ppm considered the LOAEC/NOAEC for many of the macroscopic effects tradiitionally linked to PFOS toxicity in zebrafish embryos, including lipid disruption, effects to sensorial organs, and lethality. It is also remarkable the functional correlation between these macroscopic effects and the molecualr changes we observed at metabolic and/or transcriptomic levels at concentrations 10 to 100 below the macroscopic NOAEL.

WE028

Impacts of fatty acids and methylmercury on preadipocyte differentiation in rainbow trout (Oncorhynchus mykiss).

G. Tinant, Université Catholique de Louvain; I. Neefs, Université Catholique de Louvain / Institut des Sciences de la Vie; A. De Groote, Université Catholique de Louvain; J. Rees, Y. Larondelle, C. Debier, Université Catholique de Louvain / Institut des Sciences de la Vie

Fish can be exposed to nutritional and chemical stress. In aquaculture, fish oil is increasingly replaced by plant-derived oils, which results in a modification of the fatty acid (FA) composition of the diet. Also, pollutants such as methylmercury (MeHg) are still present in aquatic environments. Adipose tissue is an essential endocrine organ involved in energy homeostasis and can be affected by some stressors. However, there is a lack of knowledge about the effects of FA and MeHg on rainbow trout adipose tissue. In this context, an *in vivo* experiment was conducted and linoleic acid (LA) induced a lipid content increase in fish, while MeHg decreased it. To understand better these results, two *in vitro* experiments were carried out on trout primary cultured adipocytes to study the effects of FA and those of MeHg. *Effects of FAs* - During 2 days, differentiation of confluent cells was induced through a hormonal cocktail. Cells were then incubated during 13 days with 0, 75, 150, 300 and 600 µM of α-linolenic (ALA), eicosapentaenoic, docosahexaenoic (DHA), LA, arachidonic and docosapentaenoic (DPA) acids and 2 µL/mL lipid mixture. At day 13, for each FA, the higher the concentration, the more the lipid accumulation. At 600 mM, DHA and DPA were the most adipogenic FA, while LA and ALA (typical to plant-derived oils) induced less lipid accumulation. For all conditions, a clear enrichment of membranes and lipid droplets with the incubated FA was observed. *Effects of MeHg* - Confluent cells were incubated for 6 days with or without a hormonal cocktail, with 0, 0.5, 2.5 or 5 mM MeHg and with 4 µL/mL lipid mixture. No cytotoxicity was observed. At day 6, cells were collected to determine mercury concentration, FA content and composition, and gene expression. Analyses are in progress. Preliminary results showed that the hormonal cocktail combined to increasing MeHg concentrations tended to affect cell morphology, towards a more typical adipocyte phenotype. In conclusion, it seems that the effects of FA can be different at organism and cell levels. Regarding the effects of MeHg, we can highlight the presence of cells with typical adipocyte morphology in presence of hormonal cocktail and MeHg. For both experiments, analyses of expression of genes related to adipocyte differentiation, lipid metabolism and lipolysis are under progress and could provide helpful results to understand better the impacts of stressors in trout preadipocytes.

WE029

Obesogens in the aquatic environment

A. Capitão, CIIMAR   University of Porto; A. Lyssimachou, CIIMAR; F. Castro, CIIMAR - University of Porto; M.M. Santos, CIIMAR/FCUP / Biology/Endocrine Disruptors and Emerging Contaminants

The rise of obesity in humans is a major health concern of our times, affecting an increasing proportion of the population worldwide. It is now evident that this phenomenon is not only associated with the lack of exercise and a balanced diet, but also due to environmental factors, such as exposure to environmental chemicals that interfere with lipid homeostasis. These chemicals, also known as obesogens, are present in a wide range of products of our daily life, such as cosmetics, paints, plastics, food cans and pesticide-treated food, among others. A growing body of evidences indicates that their action is not limited to mammals. Obesogens also end up in the aquatic environment, potentially affecting its ecosystems. In fact, reports show that some environmental chemicals are able to alter lipid homeostasis, impacting weight, lipid profile, signaling pathways and/or protein activity, of several taxa of aquatic animals. Such perturbations may give rise to physiological disorders and disease. Although largely unexplored from a comparative perspective, the key molecular components implicated in lipid homeostasis have likely appeared early in animal evolution. Therefore, it is not surprising that the obesogen effects are found in other animal groups beyond mammals. Collectively, data indicates that suspected obesogens impact lipid metabolism across phyla that have diverged over 600 million years ago. Here we identify the knowledge gaps in this field and we set future research priorities.

WE030

The Environmental Causes of Obesity: Novel human in vitro models of adipocyte differentiation for studying the effects of chemical exposure

S. Ermler, Brunel University London / Institute of Environment, Health and Societies; B. Blumberg, University of California, Irvine / Department of Developmental and Cell Biology; J. Legler, Utrecht University / Institute for Environmental Studies; S. Jobling, Brunel University / Institute of Environment, Health and Societies

Obesity has become a worldwide challenge, with obesity rates not only increasing in adults, but also in children. Obesity is caused by an imbalance between caloric intake and energy expenditure. However, increased caloric intake due to changes in diet and lack of physical activity cannot solely explain the observed rise in obesity. Other factors, such as genetics or environmental stressors, also play a role. Exposure to endocrine disrupting chemicals (EDCs), which act as so-called obesogens during development, may impact on adipogenesis and susceptibility to obesity, and several such compounds have been found to stimulate adipocyte differentiation *in vitro* and *in vivo*. Recent systematic reviews in our group have shown that prenatal exposure to EDCs such as BPA and DEHP is related to increases in adiposity later in life in rodent models. To identify potential obesogens and study the effects of EDCs on adipogenesis, suitable model systems are required. Standard assay systems to screen for obesogens *in vitro* are either using reporter gene assays e.g. for activation of the peroxisome proliferator-activated receptor gamma (PPARgamma), a key regulator of adipogenesis; or differentiation assays using established cell lines such as 3T3-L1 preadipocytes. However, whereas these assay systems are good screening tools, they only assess effects of obesogens on specific receptor activation or differentiation of already committed, white adipocytes. Our research aims at unravelling the effects of obesogenic EDCs on human adipocyte development. The most critical effects of exposure to obesogens are elicited *in utero* and in early life. Therefore, *in vitro* models that mimic the earliest stage of adipogenesis are best suited to investigate how EDCs can disrupt normal cell differentiation during development. One promising cell culture model involves the commitment of mesenchymal stem cells (MSCs) to either adipose or bone cell lineages. Future research using MSCs cells as model for adipogenesis will not only look at the differentiation to white adipocytes. We also envisage to employ the model to gain an understanding of other processes such as the more recently described “browning” of white adipocytes and the differentiation of MSCs to brown adipocytes.

WE031

Comparing metabolomic responses in Oryzias latipes to environmentally relevant concentrations of metformin and its metabolite, guanylurea

E. Ussery, University of Ontario Institute of Technology / Biological Sciences; K. Bridges, B.J. Venables, University of North Texas / Advanced Environmental Research Institute; Z.P. Pandelides, University of Ontario Institute of Technology; J. Guchardi, University of Ontario Institute of Technology / Faculty of Science Aquatic Toxicology; A. Kirkwood, University of Ontario Institute of Technology; D.A. Holdway, University of Ontario Inst. of Tech / Science

In recent years, the occurrence and fate of pharmaceuticals in the aquatic environment has become a priority issue in aquatic toxicology. One of the most prevalent pharmaceutical contaminants is the type-2 diabetic drug metformin, which has been found in wide-ranging concentrations (ng/L - µg/L) in wastewater effluent and surface waters. Greater than 90% of metformin is metabolized into guanylurea during wastewater treatment, and it’s the metabolite guanylurea that is found in receiving waters in relatively high concentrations (µg/L). To improve our understanding of the toxicological effects of metformin and its metabolite guanylurea in developing fish, we found a significant decrease in length (~6%; mm) and wet weight (~22%; mg) of male Japanese medaka (*Oryzias latipes*) when exposed to 3.2 µg/L metformin from embryo through 28 days post hatch. When male medaka were exposed to an extremely low concentration of guanylurea (1.0 ng/L), there was a similar percent decrease in length and wet-weight. Using radio labelled metformin, we demonstrated that about 1% of the waterborne concentration of metformin could be taken up in both embryo and larval medaka after exposure windows ranging from 24 hours to 7 days. We also conducted a metabolomics assessment of metformin and guanylurea exposed fish to elucidate the sub-lethal biochemical mode of action for each contaminant exposure. Significant changes were detected in the metabolome of 28-day larval male medaka exposed to both metformin and guanylurea, indicating significant dysregulation in fatty acid and lipid metabolism. This biochemical effect is likely a contributing factor to the observed decreased growth in exposed fish. In combination, these results suggest that the current concentrations of metformin and guanylurea in receiving waters are of ecotoxicological concern for resident fish populations.

WE032

Levels of proteins, carbohydrates, lipids and cholesterol in the digestive gland of juvenile catarina clam Argopecten ventricosus (Sowerby, 1842), exposed to toxic metals

A. Sobrino-Figueroa, Universidad Autonoma Metropolitana Iztapalapa / Hidrobiology; C. Cáceres-Martínez, Universidad Autónoma de Baja California Sur

The analysis of the composition of the digestive gland, gives information on the energy level of the organism, this energy is mobilized in the different stages of its life cycle. When the organisms are subjected to severe stress conditions, it has been observed the mobilization of these reserves to maintain homeostasis, in short periods of time. In this work, an evaluation of the composition of the digestive gland of juvenile catarina clam exposed to the metals Cd, Cr, Pb and their mixtures was carried out to determine their energy content. Bioassays with water replacements were carried out. The organisms were exposed to 1 sublethal concentration of each metal (LC_{20}) (0.35, 5.0 and 3.0 mg L^{-1} of Cd, Cr and Pb respectively) and of the mixtures in proportion 1:1. The levels of proteins (Lowry, 1951), carbohydrates (Dubois, 1956), lipids (Bligh and Dyer, 1959) and cholesterol (Kit Biorad) were quantified at 24, 96, 144 and 168 hours after the start of the bioassay. The Kruscal-Wallis test showed that the difference between the concentrations of proteins, lipids, cholesterol and carbohydrates of the control group compared to the treatments was significant ($p < 0.034$). An increase in cholesterol levels was observed at 24 hours of exposure and a decrease in protein, lipid and carbohydrate levels of up to 75% in only 96 hours (4 days) of exposure to metals and their mixtures. This indicates that juveniles exposed to metals had high stress levels, (as was also observed in relation O:N). It should be mentioned that the surviving organisms of the tests, died 48 hours (2 days) after it was observed the mobilization of their energy reserves.

Environmental risk assessment and management of the spoil material produced in tunnelling excavation (P)

WE033

Environmental assessment of foaming agent persistence in conditioned soil for EPB-TBM tunnelling

P. Grenni, National Research Council of Italy (CNR) / Water Research Institute; L. Patrolecco, Water Research Institute-National Research Council / Water Research Institute; T. Pescatore, F. Spataro, N. Ademollo, M. Cardoni, M. Di Lenola, National Research Council of Italy / Water Research Institute; A. Barra Caracciolo, National Research Council / Water Research Institute

Earth Pressure Balanced Shields are currently the most used full face tunnelling machine thanks to the wide use of conditioning agents in different soil types that change the mechanical and hydraulic behaviour of the soil into a plastic paste, permitting soil pressure applications in the bulk chamber. The most frequently conditioning agents used for soil in the bulk chamber are various types of foams that are mixed to the soil from injection points located on the cutter head and inside the bulk chamber to give the conditioned soil properties necessary to guarantee that the EPB machine will work in the proper way. The excavation process produces a large amount of spoil material rich in foaming agents that can have an impact on ecosystems. The possible way-of-reuse of the excavation products strongly depend on the additive composition, on soil properties and environmental conditions. Currently, there are neither soil threshold limits in European legislation for these components nor comprehensive studies on their environmental risk and persistence for soil ecosystems in these exposure scenarios. In this context, the objective of this study was to evaluate the biodegradability of the sodium lauryl ether sulphate (SLES) contained as the main component of two commercial foaming agents in two different soils (S1: silty-clay; S2: gravel in a clay-silty-sand matrix soils) sampled from a construction site. Moreover, the degradability of one product was evaluated in the presence of an additive used to improve the compactness and subsequently transport of the spoil material. For this purpose, microcosms were set up using soil samples conditioned separately with the two foaming agents. Control microcosms, consisting of un-treated soil, were also set-up to compare the microbial community before and after the foaming agent addition. At selected times, soil samples were collected for assessing SLES concentration by ASE extraction followed by MBAS spectrophotometric method. Microbiological analyses were performed in order to assess microbial abundance, viability and dehydrogenase activity in the conditioned and control soils. Results showed that SLES degradation depended on the soil type, with DT_{50}, ranging from 11 to 19 days; the additive increased significantly the surfactant persistence especially in the S2 soil. In the latter case, the higher persistence of the product can be ascribable to the detrimental effect of the additive on the microbial abundance and activity.

WE034

Application of the Vibrio fischeri acute toxicity test to assess the environmental impact of spoil materials containing foaming agents

L. Mariani, CNR-IRSA / IRSA; E. Donati, National Research Council of Italy / IMC; L. Patrolecco, Water Research Institute-National Research Council / Water Research Institute; J. Rauseo, National Research Council / Institute of Water Research IRSA-CNR; L. Rolando, National Research Council of Italy / Water Research Institute; A. Barra Caracciolo, National Research Council / Water Research Institute; P. Grenni, National Research Council of Italy (CNR) / Water Research Institute

The rapid development of TBMs in the tunnelling industry has been mainly due to their advantages over conventional excavation methods, such as continuous operation, safer working conditions, reduced damage at surface level and higher tunnelling speed. The performance of TBMs relies on the use of appropriate soil conditioning foaming agents containing water solutions of anionic surfactants, mainly sodium lauryl ether sulphate (SLES) and in smaller concentrations other additives. In accordance with the Italian legislation, spoil material from excavation processes can be re-used as by-products if the chemical thresholds for organic and inorganic contaminants (e.g. heavy metals, hydrocarbons C>12; Italian Decree 120/2017) are not exceeded. However, there are currently neither SLES soil threshold limits in European and Italian legislation (Annex 4 of the Italian Decree 120/2017), nor comprehensive studies on its ecotoxicological effects on soil and water organisms. The use of ecotoxicological tests makes it possible to overcome the analytical limits to detect multicomponent commercial foaming products (of which the complete composition is often unknown), to save time by avoiding the designing of new analytical methods for the increasing number of chemicals used in new foaming formulations continuously being put on the market. Above all, they provide information about the different interactions between the mixture and the specific matrix and the possible ecotoxicological effects on biota. In several studies performed to evaluate the potential environmental impact of spoil materials the bacterium *Vibrio fischeri* showed to be very sensitive to the residual concentrations of the surfactant SLES in elutriates obtained from soil samples collected from excavation sites. The overall analysis of a set of chemical and ecotoxicological data showed that the bioluminescence inhibition was directly related to SLES concentration. Consequently, the ISO 11348-3:2007 test is a suitable tool to assess in a short time the occurrence of foaming agent residuals at effect concentrations in spoil material.

WE035

Biodegradability of the anionic surfactant sodium lauryl ether sulphate used as the main component in two foaming agents for tunnelling process

M. Cardoni, National Research Council of Italy / Water Research Institute; A. Barra Caracciolo, National Research Council / Water Research Institute; N. Ademollo, National Research Council of Italy / Water Research Institute; A. Di Giulio, Italian National Research Council / Institute of Environmental Geology and Geoengineering; P. Grenni, National Research Council of Italy (CNR) / Water Research Institute; T. Pescatore, National Research Council of Italy / Water Research Institute; J. Rauseo, National Research Council / Institute of Water Research IRSA-CNR; L. Patrolecco, Water Research Institute-National Research Council / Water Research Institute

The anionic surfactant sodium lauryl ether sulphate (SLES) is the main component of most commercial products used for soil conditioning in the excavation industry, in particular as lubricants for mechanized tunnelling. This excavation process produces a large amount of spoil materials that can have a potential impact on ecosystems. The lack of accurate information about SLES persistence in the environment has aroused increasing concern for their possible recycling as construction materials or as soil replacement for covering rocky areas. Currently, there are neither SLES soil threshold limits in European legislation, nor comprehensive studies on the environmental risk for soil ecosystems in these exposure scenarios. The objective of this study was to evaluate the biodegradability of the sodium lauryl ether sulphate (SLES) contained in two common commercial foaming agents (P1, P2). For this purpose, a set of microcosms was set up using two different soils (S1: silty-clay soil; S2: gravel in a clay-silty-sand matrix soil) from the construction site. Microcosm experiments were set-up with soil samples conditioned separately with the foaming agent P1 (85 mg/kg SLES concentration) or P2 (83 mg/kg SLES concentration). Some soil samples were previously sterilized in order to evaluate abiotic degradation in absence of the microbial community. Moreover, control microcosms, consisting of un-treated soil, were also present in order to compare the microbial community before and after the foaming agent addition. At selected times (0, 7, 14, 21, 28 d) soil samples were collected for assessing SLES concentration by MBAS spectrophotometric method. Moreover, microbiological analyses were performed in order to assess microbial abundance (DAPI counts), cell viability (Live/Dead method), dehydrogenase activity and the phylogenetic structure of the microbial community by the Fluorescent In Situ Hybridization (FISH) method. Although an initial negative effect on microbial abundance and viability was observed, at the end of the experiment SLES was no longer present in all soils and the dehydrogenase activity and cellular vitality were comparable between treated and control soil. SLES was completely biodegraded at day 28 and a shift in the microbial community was observed comparing the control vs treated soils. In particular, a significant increase in the Gamma-Proteobacteria group, which includes bacteria able to transform SLES, has been found.

WE036

Development of new foaming agents with better environmental impact for EPB soil conditioning - The Polyfoamer ECO line

a. boscaro, e. dal negro, Mapei SpA / Underground Technology Team; m. stefanoni, Mapei SpA / R&D

1. New foaming agents with better environmental impact: the POLYFOAMER ECO line Thanks to the development of new foaming agents carried out by the R&D Group, MAPEI have created the new product line Polyfoamer ECO, with the main goal to reduce the environmental impact on the tunnel muck, thus facilitating the re-use of the tunnel muck as by-products, in example for road constructions or old quarries refilling. All the new Polyfoamer ECO foaming agents have been classified by a third-party German laboratory as WGK = 1, representing the lowest class of risk againts waters and organisms associable to a chemical product, according to the German regulation. The new products Polyfoamer ECO/100 and Polyfoamer ECO/100 Plus are characterized by lower values of COD at the initial stage when compared to traditional products, meaning that their provision of organic material to the conditioned soil is lower. 2. Environmental results with soils conditioned with the Polyfoamer ECO products Various laboratory tests have been carried out with the new foaming agents of the Polyfoamer ECO line of products and samples of soil coming from different TBM projects. The results obtained with two samples of soil from an Italian project are described: the material called “M” (a

geological formation with alternance of limestone and marlstone) and the material called “S” (a cohesive clay). The amount of the surfactants inside the conditioned samples “S” and “M” have been measured with the instrument HPLC-MS (“High-Performance Liquid Chromatography Mass”) at different stages from the addition of the foam to the soil: at time 0, at 3 and at 7 days. Toxicity tests with the bio-luminecest bacteria Vibrio Fisheri (ISO 11348-2:2007) and the fish embrio Danio Rerio (OECD236) have been carried out. The results of the environmental tests with the new foaming agents Polyfoamer ECO confirm that these new formulations allow to reduce the impact on the soil and therefore to facilitate its re-use in short periods as a by-products. The main results obtained with the Polyfoamer ECO/100 and Polyfoamer ECO/100 Plus are: lower toxicity and lower content of organic material when compared to traditional foaming agents, fasta degradation of the surfactants inside the conditioned soil, low toxicity of the conditioned soils and tendency of toxicity decrease along the time. Values comparable to the natural soil toxicity are achieved in a short period.

WE037

Determination of anionic surfactants by Pressurized Liquid Extraction (PLE) followed by the modified Methylene Blue Active Substances (MBAS) method in spoil material from excavation processes

L. Patrolecco, Water Research Institute-National Research Council / Water Research Institute; N. Ademollo, T. Pescatore, National Research Council of Italy / Water Research Institute; J. Rauseo, National Research Council / Institute of Water Research IRSA-CNR; F. Spataro, National Research Council of Italy / Water Research Institute

Recent years have been characterized by a rapid worldwide growth in underground constructions in the form of new infrastructures such as pipelines and communication cables as well as road and railway tunnels. The utilization of underground space offers a new strategy to urban planning, including a huge development of the mechanized tunnelling industry by the use of Tunnel Boring Machine (TBMs). Performances of TBMs depend on the selection of appropriate soil conditioning products, principally foaming agents. Anionic surfactants are the main constituents of commercial foaming agents widely used as lubricating products in the mechanized excavation, improving the stability of the excavation face and reducing the friction between soil cuttings. Among the anionic surfactants, sodium lauryl ether sulphate (SLES) is the most utilised compound in the commercial products for the excavation industry. Significant amounts of rock broken into various sizes mixed with anionic surfactants are produced during the execution of engineering works. The potential re-use of this non-renewable natural resource, for example as land covering, depends on the assessment of its environmental compatibility; otherwise, the spoil materials must be treated as a waste. In this context, it is important to evaluate the residual concentrations of SLES in the excavated soils in order to evaluate their possible final destination. Given the absence of official analytical methods, it has become necessary to develop and validate a reliable and accurate methodology to quantify anionic surfactants in the spoil materials and, more in general, in the environmental solid matrices. For this purpose, the aim of the present work was the optimization of an analytical method for the determination of SLES in conditioned-soil samples. It consists of a first phase of extraction of anionic surfactants from the soil by the use of Pressurized Liquid Extraction (PLE) and the following analysis in the extract by the MBAS (Methylene Blue Active Substances) method using the water official method partially modified. The optimised method has been applied to real excavated soil samples because the determination of residual concentrations of the anionic surfactants in the spoil materials produced during excavation process, is currently one of the mandatory parameters for assessing their eco-compatibility.

WE038

Distribution and persistence of anionic surfactants in leachate and conditioned soil: mesocosm study for EPB-TMB applications

J. Rauseo, National Research Council / Institute of Water Research IRSA-CNR; N. Ademollo, National Research Council of Italy / Water Research Institute; A. Barra Caracciolo, National Research Council / Water Research Institute; A. Di Giulio, Italian National Research Council / Institute of Environmental Geology and Geoengineering; T. Pescatore, National Research Council of Italy / Water Research Institute; G. Mininni, National Research Council / Water Research Institute; L. Patrolecco, Water Research Institute-National Research Council / Water Research Institute

The development of the mechanized tunnelling industry by EPB-TBMs (*Earth Pressure Balance -Tunnel Boring Machines*), results in a wide use of foaming agents and polymers as lubricating products for soil conditioning. Anionic surfactants, and principally sodium lauryl ether sulphate (SLES), are the main components of foaming commercial products. Soil debris from excavation processes can contain residual concentration of SLES. The potential re-use of the spoil material for public green areas or industrial purpose (e.g. land covering) depends on the site-specific SLES persistence in the excavated soil and on the related environmental exposure scenario. In this context, we evaluated the SLES leaching in two different soils in order to simulate check whether underground water contamination may occur in a scenario where the spoil material is located close to a water body. For this purpose, we evaluated the persistence (DT_{50}) of SLES in two soils (S1: silty-clay soil; S2: gravel in a clay-silty-sand matrix soil) conditioned separately with two common commercial foaming agents, respectively F1 and F2, used at conditioning ratio giving final concentration in both the soils of about 150 mg/kg. The presence of strengthening foaming polymers (P1 or P2, 527 and 50 mg/kg respectively), needed in some cases to increase foam persistence, was also considered. After a preliminary phase at laboratory scale, a mesocosm experiment was conducted in order to entail the scale-effect, which is very significant when the soil behaviour is involved. Sixteen mesocosms were set-up mixing 100 kg of each soil with water, foam and polymer and then stored for 28 days in high-density polyethylene bins (HDPE diameter of 30 cm and height of 100 cm). The effect of soil type, grain size or aeration on SLES persistence was evaluated. For this purpose, eight bins containing S1 or S2 conditioned separately with the two foaming agents, were weekly turned to improve aeration, while the corresponding eight blends were not turned. At selected times (0, 4, 7, 12, 20 and 28 days), soil and leachate samples were collected from the bins for assessing SLES concentration by MBAS spectrophotometric method, preceded by ASE (*Accelerated Solvent Extraction*) in the case of the soil matrix. The results showed that residual SLES concentration in soil and in leachate is dependent both on the type of soil and on the nature of polymers.

WE039

Preliminary environmental risk assessment of sodium lauryl ether sulphate contained in foaming agents used in mechanized tunnelling

A. Finizio, University Milano - Bicocca / Department of Earth and Environmental Sciences; P. Grenni, National Research Council of Italy (CNR) / Water Research Institute; L. Patrolecco, Water Research Institute-National Research Council / Water Research Institute; E. Galli, National Research Council of Italy / IBAF; V. Muzzini, National Research Council of Italy / Institute of Agro-environmental and Forest Biology; E. Donati, National Research Council of Italy / IMC; I. Lacchetti, P. Gucci, Istituto Superiore di Sanità; A. Barra Caracciolo, National Research Council / Water Research Institute

Anionic surfactants (ANS) are a heterogeneous group of amphipathic compounds characterized by linear aliphatic chains (ranging from C8 to C18) with a polar group (sulphate or sulfonate) neutralized with a counter ion. Given the variability of their molecular composition ANS are considered mixtures. They are utilized in several applications (i.e. detergents, cleaning products, fracking or soil conditioning in the excavation industry). Among ANS, the sodium lauryl ether sulphate (SLES) is commonly utilized as a foaming agent to facilitate the excavation procedures in mechanized tunnelling. However, its use raises concern for the environment considering the presence of SLES residues in soil debris produced during the excavation. In addition, the absence of soil threshold limit for SLES in the EU legislation does not facilitate the re-use of soil debris as by products (e.g. land covering) and, consequently, a huge amount of such detritus can be discharged as a waste with high economic costs. In absence of a threshold limit, performing an environmental risk assessment (ERA) of foaming agents containing SLES can be a possible alternative. However, the ERA is hampered by both the rather scarce data on the effects of SLES and the site specific condition of use which lead to different levels of exposure. Indeed, the selection of the type and quantity of foaming agents depend on soil, geological conditions, and characteristics of the tunnel boring machines. Furthermore, several commercial formulations are available on the market with different percentages of SLES and several other components. This study is part of a wider project aiming to develop a methodology to be applied to identify environmental acceptable levels of SLES residues in soil debris produced during the tunnelling operations in Italy. Particularly, we report the results regarding the preliminary ERA which has been utilized to select, among all the available commercial formulations, the one leading the lowest level of risk for the environment in a specific condition of use. The risk has been characterized based on PEC/PNEC ratios. PECs were calculated by predictive models and considering the percentage of SLES in the commercial formulations as well as the required treatment ratios for tunnelling operations. PNECs (soil and surface water) for SLES were derived from ecotoxicological data (terrestrial and aquatic organisms) which were obtained from laboratory tests on several test organisms.

WE040

Ecotoxicological assessment of spoil material produced in mechanized excavation

I. Lacchetti, Istituto Superiore di Sanità / Environment and Health; P.M. Gucci, Italian Institute of Health ISS / Environmental and Health; P. Grenni, National Research Council of Italy (CNR) / Water Research Institute; L. Patrolecco, Water Research Institute-National Research Council / Water Research Institute; M. Cardoni, M. Di Lenola, National Research Council of Italy / Water Research Institute; J. Rauseo, National Research Council / Institute of Water Research IRSA-CNR; E. Galli, National Research Council of Italy / IBAF; V. Muzzini, National Research Council of Italy / Institute of Agro-environmental and Forest Biology; E. Donati, National Research Council of Italy / IMC; A. Finizio, P. Fumagalli, University Milano - Bicocca / Department of Earth and Environmental Sciences; A. Barra Caracciolo, National Research Council / Water Research Institute

Mechanized excavations using Tunnel Boring Machines (TBM) has consolidated in recent years. In order to facilitate the overall process, specific foaming agents and polymers are added to soil. The main component of many commercial foaming agent products are anionic surfactants such as the alkyl ether sulphates (AES). The possible re-use of huge amounts of spoil material produced during the excavation process as by-products (e.g. land covering) or its discharge as a waste depends on the residual concentration of AES in the soil. The first option has the undoubted advantage to lower the costs of disposal. However, there are concerns about the potential environmental risk related to the re-use of conditioned soil. In fact, even if anionic surfactants are generally considered biodegradable and not toxic, there is little information in literature on their environmental fate and the possible ecotoxicological effects of the commercial formulations of foaming products and of the conditioned soils. The aim of this study was to evaluate the environmental compatibility and the ecotoxicological effects of two different soils treated with two different foaming agents containing the anionic surfactant AES, applying a suitable battery of bioassays. For this purpose, a set of mesocosms was prepared, containing two soils with different geopedological characteristics, conditioned with two foaming agents at the same treatment ratios (TR, L/m^3) used for mechanized drills. Soil samples were collected at different maturation times (0, 7, 14, 28 days) in order to perform the ecotoxicological tests on the spoil material or in its aqueous extracts. The bioassays selected are representative of different trophic levels for the aquatic and terrestrial compartments: Microtox test with the bacterium *Vibrio fischeri*, Fish Embryo Acute Toxicity Test (FET) with the species *Danio rerio*, germination and growth test with the plant *Lepidium sativum* and test with the worm *Eisenia foetida*. In parallel, sub-samples of soil and elutriate from each mesocosm were analyzed in order to determine the residual concentrations of the anionic surfactant AES. The overall results obtained showed different ecotoxicological response depending both on the kind of conditioned soil and target organism tested. Therefore, this study highlights the importance of a site-specific ecotoxicological evaluation in the tunneling projects in order to have a real environmental compatibility of the spoil material.

WE041

Expeditious test for on-site monitoring activity in mechanized tunnelling applications

D. Sebastiani, G. Vilardi, Università La Sapienza; A. Di Giulio, Italian National Research Council / Institute of Environmental Geology and Geoengineering; S. Miliziano, L. Di Palma, Università La Sapienza; A. Barra Caracciolo, National Research Council / Water Research Institute; P. Grenni, National Research Council of Italy (CNR) / Water Research Institute; L. Patrolecco, Water Research Institute-National Research Council / Water Research Institute

In the vast majority of tunnel projects performed with TBM-EPB technology a key issue is currently represented by the disposal management of the spoil, huge amount of excavated material mainly composed by the natural soil, water and chemical injected during the excavation process. The opportunity to plan strategies for the spoil disposal management in a virtuous cycle of reuse of the resources leads to relevant economical and logistical advantages. The raising awareness about the chemical composition of the products injected during the excavation resulted in the development of experimental procedures aimed at studying the environmental impact of chemicals and their permanence in the soil during the excavation and after its completion. Commonly accepted standards have been developed to evaluate the effect of specific concentration values of these chemicals on terrestrial and aquatic environments, as well as to measure the reduction of the concentration of these compounds in each environment due to the action of the microorganisms inhabiting them. All these experimental procedures must be carried out in specialized laboratories equipped with sophisticated apparatuses, in which controlled environments are predisposed, so that at present it's not possible to measure the level of pollution through expeditious tests directly on site. A joint research activity between Sapienza University and National Research Council of Rome has developed a test procedure able to provide expeditious information on the presence in the spoil of the chemicals often used in mechanized tunnelling. The results of preliminary laboratory tests convinced that the expeditious assessment proposed can describe the amount of chemicals in the soil and their evolution in time, complementing the laboratory activities currently accepted. In fact, this fast procedure must be regarded as a first screening which can be run directly in site on a large number of samples without the use of expensive, delicate or complex instruments, to be used in combination with few more precise laboratory tests. Moreover, it seems to be particularly suitable for monitoring large volumes such as those involved in tunnel excavation. The intention for the future is to apply the procedure to real cases to verify, through a comparison with the most accurate laboratory tests, the actual effectiveness of this procedure.

WE042

Toxicity of several additives used in mechanized tunneling: effects on daphnids, algae and cress.

D. Baderna, S. Maiorana, A. Passoni, R. Bagnati, Istituto di Ricerche Farmacologiche Mario Negri / Department of Environmental Health Sciences; M. Lodi, E. Benfenati, IRCCS Istituto di Ricerche Farmacologiche Mario Negri / Department of Environmental Health Sciences

Surfactants and polymers are used in mechanized tunneling to facilitate the excavation and to preserve the tunnel boring machine (TBM) from wear, block and break. As a by-product of the process, several tons of rocky debris are produced. These excavation rocks are made of rock fragments contaminated by the additives such as anionic surfactants, glycols, sealants, polyacrylates and polyacrylamides. In the past, rocky debris were disposed as waste with a large economic impact on total excavation cost, while some disposal alternatives were recently applied, trying to recycle these complex materials as road filling material, covering rocky areas or river banks. These disposal solutions, however, have attracted the attention of regulators and environmental protection agencies, especially in Italy. In fact, the toxicology of these active mixtures is not yet fully known as well as the potential effects resulting from the simultaneous presence of additives with other regulated environmental contaminants such as, for example, metals and hydrocarbons. A preliminary study recently conducted by our group on three commercial TBM additives showed toxic effects on the aquatic ecosystem in concentrations comparable to those resulting from excavations carried out in Italy. This new study analyzes 8 surfactants and 4 commercial polymers, using a multidisciplinary approach to determine their reference thresholds for both water and soils, accounting of the effects on ecological targets. The chemical composition of the technical mixtures was determined by liquid chromatography coupled with high resolution mass spectrometry. The main chemical components were analyzed *in silico* to highlight the potential similarity with other pollutants, already listed in our environmental framework regulation. Finally, the toxicity of the various agents has been evaluated by tests with *Daphnia magna*, freshwater algae and cress. Chemical characterization identified 15 molecules present in all the surfactant mixtures, although in different proportion. No similarities with compounds already regulated by the Italian Environmental Act were found by the in silico analysis. All the tested surfactants were toxic for the aquatic organisms at concentrations comparable to those that can be found in leachates of conditioned rock debris. The additives resulted non toxic for the terrestrial plant at concentrations theoretically found in conditioned rock debris.

PBT/vPvB & PMT/vPvM substances and Non-extractable residues (NER): Scientific strategies, Analytical challenges and Regulatory Issues (P)

WE043

Bioaccumulation, tissue distribution, and trophic magnification of organic ultraviolet absorbents in freshwater ecosystem in the Pearl River catchment, China

X. Peng, Z. Zhu, S. Xiong, Guangzhou Institute of Geochemistry, Chinese Academy of Sciences

Bioaccumulation and trophic transfer of 13 organic ultraviolet absorbents (UVAs) were investigated in riverine wildlife organisms in the Pearl River catchment, South China. The UVAs were widely present in the fish with the highest level detected for UV 531. Generally, the UVAs concentrations were higher in the freshwater fish than in shrimp. Dietary habits of the fish showed effects on bioaccumulation of the UVAs with higher levels in the carnivorous species than in the herbivorous, planktivorous and detritus feeding fishes. Tissue distributions were generally in the order of liver > belly fat > muscle. Obviously higher concentrations of 2-ethyl-hexyl-4-trimethoxycinnamate were detected in eggs than in muscle, probably indicating maternal transfer of the compound. The calculated bioaccumulation factors(LogBAF)were usually > 3.3, suggesting potential of the UVAs in the freshwater fish. UV 531 showed tendency of bioaccumulation in the fish from the sediment indicated by the calculated BSFA >1. The estimated trophic magnification factors were > 1 for some benzotriazole stabilizers, suggesting potential of biomagnification of the UVAs in the freshwater fish.

WE044

Hyalella azteca as non-vertebrate alternative species for bioaccumulation studies

M. Habekost, BASF Corporation; N. Kreling, BASF SE / Crop Protection - Ecotoxicology; B. Kusebauch, M. Obermann, BASF SE Agrarzentrum Limburgerhof

Bioaccumulation is one of the PBT (persistence, bioaccumulation, toxicity) cut-off criteria for plant protection products (EC/1107/2009) in the EU; furthermore, high bioaccumulation is generally considered a critical parameter in other regions, too. The standard regulatory assessment of bioaccumulation is based on bioconcentration in aquatic species, i.e. for regulatory purposes in fish. However, standard fish bioconcentration studies are time consuming, expensive and they use a considerable number of vertebrate animals. Thus, there is a need for a relatively quick, cheap, and preferably alternative test method that enables the ranking of structurally clustered candidate molecules regarding bioaccumulation potential and the prediction whether a candidate molecule will exceed the BCF (bioconcentration factor) trigger value. Furthermore, *Hyalella azteca* might in the long-term perspective be able to replace fish for BCF testing. There is indication that experimental BCF values from flow-through bioaccumulation studies with *Hyalella* are similar to those obtained from fish (Schlechtriem, 2012). Further work is presented in order to (i) increase the data base of *Hyalella* – fish BCF data sets covering a wide range of BCF values (i.e. 100 to 20 000), (ii) to standardize and simplify the test system and (iii) to check the suitability of the test system for molecules with an insecticidal mode of action which poses inherent challenges since *Hyalella* as an aquatic invertebrate can be quite sensitive. The results from several BCF studies with *Hyalella* supports this species as suitable test species for bioaccumulation testing and supports planned activities on OECD level.

WE045

Bioaccumulation of ionizable organic chemicals in fish - The quest for reliable predictors

F. Polesel, Technical University of Denmark (DTU) / DTU Environment; Z. Zhang, Technical University of Denmark DTU / DTU Environment; K. Bittermann, Helmholtz Centre for Environmental Research GmbH - UFZ / Analytical Environmental Chemistry; l. linden, UFZ Helmholtz Centre for Environmental Research / Department of Analytical Environmental Chemistry; C. Schlechtriem, Fraunhofer IME / Department Bioaccumulation and Animal Metabolism; K. Goss, Helmholtz centre for environmental research - UFZ / Analytical Environmental Chemistry; S. Trapp, Technical University of Denmark DTU / DTU Environment

Dietary bioaccumulation in fish is quantified using biomagnification factors (BMFs), which are derived under controlled conditions according to OECD guideline 305-III. To reduce *in vivo* experimental efforts, pre-screening using statistical models for BMF prediction is becoming increasingly popular. While dietary bioaccumulation of neutral chemicals has been successfully associated to lipophilicity descriptors, no suitable predictor has yet been identified for ionizable chemicals. In this study, we investigated the capability of selected chemical properties (e.g., molar volume, adsorption to albumin, lipophilicity, solubility, topological polar surface area) to predict bioaccumulation of organic electrolytes in fish with specific focus on dietary exposure studies. Measured dietary BMFs were collected from existing literature, and empirical correlations with measured or estimated chemical descriptors were evaluated. The dataset includes dietary BMFs in whole fish obtained under laboratory-scale conditions closely resembling or directly referring to the OECD 305-III guideline. In total, BMF data were available for 29 ionizable chemicals (of which 10 are perfluorinated chemicals); including 24 acids and 19 permanently ionized chemicals at environmental pH (range 3 to 9). A parallel dataset was compiled with bioconcentration factors (BCFs) of the same chemicals derived in water exposure studies with fish (OECD 305-I guideline). Bivariate correlation analysis (Pearson and Spearman) revealed that a) $\log K_{OW}$ was not a sufficient predictor of BMF, although with significant positive correlation ($R>0.40$), and b) that significant correlation was shown only with $\log D$ at pH=3 ($R=0.35$). Furthermore, significant negative correlation was shown between BMF and solubility ($R< -0.60$). These preliminary results indicate that commonly used predictors for bioaccumulation (e.g., $\log K_{OW}$) are of limited relevance for ionizable chemicals, and other predictors should be identified. Ongoing research is focusing on the prediction of BCF from quantum-chemistry-based estimations of partitioning coefficients (to membrane lipids, structural proteins and albumin). Estimation of BCFs from BMF for the investigated chemicals will be also performed and verified with existing BCF measurements. Eventually, identified empirical regressions between BMF and chemical descriptors will be validated with *ad hoc* experimental data with radiolabelled test chemicals.

WE046

Evaluation of a tiered approach for the bioaccumulation assessment of fragrance substances: in silico, in vitro assays, invertebrate vs. in vivo fish bioconcentration test

S. Gimeno, Firmenich / Product Safety and Regulatory Affairs; V. Laubscher, F. Berthaud, Firmenich SA / DRAS; I. Bischof, Fraunhofer Institute for Molecular Biology and Applied Ecology IME; C. Schlechtriem, Fraunhofer IME / Department Bioaccumulation and Animal Metabolism; C. Kropf, University of Bern / Centre for Fish an Wildlife Health; H. Segner, University of Bern / Centre for Fish and Wildlife Health; H. Schug, Eawag Swiss Federal Institute of Aquatic Science and Technology; K. Schirmer, Eawag / Environmental Toxicology; F. Begnaud, Firmenich / DRAP

Bioaccumulation is a key end point in environmental hazard and risk assessment, especially for substances with a high octanol water partition coefficient (logKOW). To measure the BioConcentration Factor (BCF), a tiered approach is followed starting from the assessment of the octanol water partitioning coefficient as a measure for lipophilicity, which is often used as surrogate for lipid partitioning up to an experimental BCF value which is considered as the gold standard for fish bioaccumulation assessment. We have applied a series of non-animal tests to predict the BCF values and compared those outcomes to the results from a BCF test in order to validate this alternative approach. Several fragrances from the tetranorlabdane diterpenoids family, either composed of a single or a mixture of stereoisomers were tested. The logKOW predicted by QSAR ranges from 4.75-5.41 and is > 6.2 when determined by HPLC (OECD 117). The slow stir method (OECD 123) provides a logKOW of 5.09 which is retained as the reference value. Various structure-activity relationship models were used to predict the fish bioconcentration factor, which ranged from ~ 1000 to ~ 4500, not exceeding the EU criteria for (very) Bioaccumulative substances (vB), however, the structure was mostly outside the applicability domain of the models. Therefore in vitro assays were conducted on rainbow trout S9 fractions and hepatocytes confirming the potential of biotransformation; the refined BCF values calculated with IVIVE extrapolation models were < 1000. In addition the bioaccumulation potential of one isomer was investigated in a flow-through test on the invertebrate Hyalella azteca resulting in a BCFSS or kinetic < 500 L/kg. Finally an experimental fish BCF of ~500 (OECD 305) confirms that the fragrance composed of various isomers is not bioaccumulative, and supports the in vitro biotransformation findings. Histopathological results from toxicological studies showed liver hypertrophy consistent with the increased metabolism associated with detoxification processes. A tiered weight-of-evidence approach is clearly justified for the current bioaccumulation assessment, confirming that the tests described in the abstract may offer alternatives to animal testing when sufficient and supportive evidence is provided.

WE047

Proposal for a freshwater trophic magnification study based on a comprehensive literature evaluation

V. Kosfeld, Fraunhofer Institute for Molecular Biology and Applied Ecology IME / Bioaccumulation and Animal Metabolism; H. Ruedel, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Department of Environmental Monitoring; C. Schlechtriem, Fraunhofer IME / Department Bioaccumulation and Animal Metabolism; C. Rauert, Umweltbundesamt / International Chemicals Management; J. Koschorreck, Umweltbundesamt

The bioaccumulation potential is a critical property used for the risk assessment of chemicals and is usually expressed by parameters derived from laboratory or field experiments, in particular bioconcentration-, bioaccumulation- and biomagnification factors . A relatively new approach is the determination of so-called trophic magnification factors (TMF) which integrate enrichment processes in a food web. Reliable TMF can provide valuable information to answer different questions in regulatory and monitoring affairs. The TMF can be used in the evaluation of the biomagnification potential of chemicals under REACH. However, TMF may be also applied in the context of the Water Framework Directive to normalize chemical monitoring data of fish to a common trophic level as well as to derive environmental quality standards for the protection goal ‘secondary poisoning of predators’. To date, only a few detailed TMF studies have been performed and the investigated endpoints have shown considerable variation. The aim of this study is to define a sound concept for TMF investigations to enhance both, the reproducibility and accuracy of TMF estimates to allow the regulatory usage of this endpoint. The developed concept focuses on freshwater habitats, covers different invertebrate and fish species and will be tested in a pilot field study. A water body will be selected under consideration of several aspects such as the chemical burden of the water body, the type of contamination source, and species diversity in the water body. Invertebrate and fish species will be collected in the water body during spring/summer 2018. The trophic levels of the species will be determined applying different methods such as comparison of stable isotope patterns in the collected species against a baseline organism (including stable isotope analysis of different amino acids.). Sample handling will follow the protocols applied by the German Environmental Specimen Bank (ESB) including cryo-milling, homogenization, sub-sampling, and long-term storage. During all processing steps samples will be kept constantly at a temperature < -150°C. The sample material obtained will be analyzed to derive TMF estimates for different contaminants found in the selected water body. Different normalization-, benchmarking-, and statistical methods will be applied. The validated concept may provide the framework for a new TMF testing scheme integrated in the German Environmental Specimen Bank (ESB).

WE048

Obstacles in identifying PBT/vPvB-substances under REACH for high tonnage chemicals

A. Oertel, J. Menz, A.L. Kronsbein, K. Maul, A. Bruening, A. Schulte, German Federal Institute for Risk Assessment / Chemicals and Product Safety

Appropriate ecotoxicological and toxicological information provided by registrants is crucial for identifying substances with PBT/vPvB properties under REACH (Regulation (EC) No 1907/2006. Registration, Evaluation, Authorisation and Restriction of Chemicals). The availability of that information in REACH registration dossiers of substances manufactured or imported in quantities of 100 to 1 000 tons per year (tpa) is evaluated in the current project on REACH compliance. This is a follow-up project on substances registered in quantities of 1 000 tpa or more. For human health, the endpoints repeated dose toxicity, developmental and reproductive toxicity and mutagenicity are considered and for the environment information on biotic and abiotic degradation, bioaccumulation and ecotoxicity are taken into account. The stepwise approach applied in the preceding project is adapted to the lower tonnage band with the aim to receive a high conclusion rate regarding the compliance of the endpoints. The previously published findings for substances registered in quantities of 1 000 tpa or more suggest that the main obstacle in identifying substances with PBT/vPvB properties are shortcomings in data quality, data gaps or inappropriate data-waiving/adaptation approaches. A minimum of 12% (for abiotic degradation) and a maximum of 61% (for ecotoxicity) of the dossiers were found to be “non-compliant”. It was recommended that registrants should thoroughly review and update their dossiers in order to fulfil the information requirements. This can be achieved either by using appropriate standard tests, providing a sound justification to waive data or using appropriate surrogate data. The poster will show preliminary results on the dossiers of substances registered in the tonnage levels of 100 to 1000 tons per year and its

consequences on the PBT/vPvB-identification.

WE049

PBT/vPvBs: All equally bad or some worse than others? - How to inform risk management

K. Thiele, WUR; S. Gabbert, Wageningen University / Social Sciences

In the risk management of chemicals there is an increasing demand to assess the economic, social and environmental impacts of regulatory measures in order to facilitate political decision-making. Within REACH, socio-economic analysis (SEA) is the tool to assess and balance positive and negative impacts of different policy options. Currently, persistent, bioaccumulative and toxic (PBT) and very persistent, very bioaccumulative (vPvB) chemicals are regulated on the basis of their inherent properties. These imply a high and long-lasting damage potential for the natural environment. In reality, however, it is likely that PBT/vPvB are not of equal concern in terms of their damage potential to humans or ecosystems. The lack of knowledge on the actual effects of PBT/vPvB chemicals in the environment hampers the estimation of their risks and, in turn, a full quantification of all impacts. Consequently, a fundamental challenge for SEA is to adequately describe the regulatory concerns of PBT/vPvB chemicals, and to integrate specific information on a certain PBT/vPvB substance into a metric that informs policy-makers on their potential impacts. So far, there is no systematic approach on how available hazard and risk data as well as complementary information about the uncertainty due to data quality or lack of knowledge, can be used to assess the difference in damage potential of PBT/vPvB chemicals in SEA. Exploring the perspectives of experts from academia, industry and regulatory agencies may therefore be helpful to guide the development of approaches for comparative evaluations of PBT/vPvB substances. This poster proposes a research project that is going to explore how specific characteristics of PBT/vPvB chemicals can influence the concern. Based on surveys and structured interviews, it will examined what parameters are considered most relevant to characterise the concern of PBTs/vPvBs. Furthermore, it will be explored how experts in academia, industry, regulatory agencies rate the relative importance of different (sets of) characteristics of PBT/vPvB substances in terms of their environmental impact potential. The results can be used to inform cost-effectiveness analyses and ranking schemes for PBTs/vPvBs. This will, ultimately, facilitate comparative evaluations of PBT/vPvB substances for SEA and regulatory decision-making.

WE050

Modelling Persistent & Mobile Organic Compounds using an updated Multimedia Urban Model: A Toronto Case Study with Organophosphate Esters (OPEs)

T.F. Rodgers, University of Toronto / Chemical Engineering and Applied Chemistry; L. Jantunen, Environment and Climate Change Canada; P.A. Helm, Ontario Ministry of Environment and Climate Change / Environmental Monitoring and Reporting Branch; M.L. Diamond, University of Toronto / Department of Earth Sciences

Organophosphate Esters (OPEs) are a group of chemicals found at relatively high levels as environmental contaminants. The usage of OPEs has increased in recent years following the listing of penta- and octa- BDEs as POPs under the Stockholm Convention. In contrast with BDEs, OPEs highly polar molecules which can be considered persistent and mobile organic compounds (PMOCs). We modified the Multimedia Urban Model (MUM) of Diamond and co-workers by using polyparameter linear free energy relationships (ppLFERs) to represent partitioning, and we modified the wet deposition processes to account for intermittent rainfall. We looked at three chlorinated (Cl-OPEs) and three non-chlorinated OPEs (non-Cl-OPEs) in Toronto, Canada. Our goal was to estimate their emissions to Toronto air and to evaluate their environmental pathways. Air emissions were estimated by from measured outdoor air concentrations and model resuls were evaluated against measured water and rain concentrations in Toronto tributaries. Based on estimated emissions to air, modelled water and rain concentrations were within an order of magnitude of the measured concentrations, with an RMSE of 140% of the mean measured water and rain concentration. Since the water and rain concentrations were taken independently of the air concentrations, these results gives some credence to the model estimates and showed that the emissions estimates were accurate to approximately an order of magnitude. Estimated aggregate emissions to outdoor Toronto air of OPEs for 2010 ranged from 110 (TDCPP) to 1,200 (TCEP) kg/y and were significantly higher than emissions of Σ_5PCBs (90 kg/y) and Σ_5PBDEs (9 kg/y) for 2008, calculated using the same model. The results show that using modelling techniques developed for polar, hydrophilic compounds can provide estimates of emissions and fate to a similar level of accuracy as was possible previously for persistent, bioaccumulative and toxic compounds. These model estimates provide evidence of relatively high emission rates to air and, by showing OPE mobility in water, support the hypothesis of long-range transport of Cl-OPEs by rivers. The major route of transfer for Cl-OPEs to surface water systems is through stormwater runoff, either through the washoff of films on impervious surfaces or soil wash-off.

WE051

An approach for the evaluation of PBT and vPvB substances subject to authorisation and restriction procedures in the context of socio-economic analysis

S. Gabbert, Wageningen University / Social Sciences; S. Hahn, Fraunhofer ITEM / Chemical Risk Assessment; M. Klein, Fraunhofer Institute for Molecular Biology and Applied Ecology IME; F. Oosterhuis, Vrije Universiteit Amsterdam / Institute for Environmental Studies; M. Nendza, Analytisches Laboratorium

A key objective of the European chemicals legislation REACH is to ensure that the risks caused by substances of very high concern (SVHC) are adequately controlled. The two regulatory procedures adopted in REACH to control the risks arising from SVHCs are authorisation and restriction. Both regulatory instruments make use of socio-economic analysis (SEA), which is generally defined to be a tool for assessing all relevant positive and negative impacts from substances' use or non-use, and for comparing these impacts across different scenarios. Impacts arising from chemicals' use, including PBTs/vPvBs, are use-specific. Furthermore, due to stock pollution properties of PBTs/vPvBs, impacts may last for long periods and even long after emissions have ceased. In addition, information about (long-term) impacts needs to be balanced with costs of emission reduction and abatement. Acknowledging that monetary valuation of impacts using stated or revealed preference methods is not possible for a broader set of PBT/vPvB substances, the evaluation of PBT/vPvB substances in a SEA has to rely on cost-effectiveness analysis (CEA). This requires specifying benchmark values, i.e. target values (standards) by means of which it can be determined whether or not the costs of a control measure are excessive. This paper suggests an approach for the evaluation of PBT/vPvB substances by means of CEA that accounts for the complex environmental distribution patterns, and that allows balancing (long-term) impacts from PBT/vPvB use against costs for emission reduction and abatement. The approach starts with a grouping and ranking of PBT/vPvB substances (stage 1). Following to this, exposure dynamics are analysed with a multimedia stock pollution approach (stage 2). The evaluation of impacts arising from the stock can be evaluated via different routes (stage 3). To assess the cost-effectiveness and proportionality of possible (policy-) measures for PBT/vPvB control, the routes to impact evaluation are linked to assessments of costs for restricting or stopping a specific or multiple uses of a PBT/vPvB substance (stage 4), and to benchmarks, being standard values of a specific parameter to which the actual/estimated value of that parameter can be compared (stage 5). The approach offers a ready-for use framework for a concern-based evaluation of PBT/vPvB substances to be used as decision-support in REACH authorisation and restriction processes.

WE052

Polymers: The Next Frontier in Environmental Hazard Assessment

A. Carrao, Kao USA / R&D; T. Suzuki, Kao Corporation, S.A. / Product Safety & Regulations; I. Davies, Personal Care Products Council / Science; J. Coleman II, Kao USA

Polymers are a very large and diverse class of chemicals widely used in cosmetic and personal care products. Their use and function are essential in creating high performing products that meet the needs of consumers. As used in cosmetic formulations, polymers can act as thickeners, emulsifiers, conditioners, opacifiers, film formers, rheology modifiers, etc. In the simplest terms, according to the Oxford Dictionary, a polymer is a "substance that has a molecular structure consisting chiefly or entirely of a large number of similar units bonded together." They have a full range of physical-chemical properties including a wide breadth of solubility and molecular charge, for example. Currently, the majority of large molecular weight polymers are exempt from chemical regulations around the world (e.g. REACh) or are largely considered of low concern based on a minimum set of physical-chemical properties (US EPA's FIFRA, Australia's NICNAS). However, there is a speculation that these regulatory exemptions, specifically the REACh exemption, could be removed in the next 5-10 years. If this is the case, many previously untested chemicals would then need an environmental hazard assessment supported by an ecotoxicological dataset. This dataset may include aquatic toxicity testing, read-across to structurally similar chemicals that have been tested, weight of evidence toxicity estimates based on physical-chemical properties, or all of the above. However, the same variety of physical-chemical properties that allows polymers to have so many functions in cosmetic formulations also makes these substances difficult to test in aquatic systems – varying absorption properties, molecular charge, insolubility, etc. Therefore, safety assessors evaluating polymers must look to new and novel approaches for filling environmental hazard data gaps in order to create a robust environmental hazard assessment. This poster will examine the current polymer landscape for cosmetic uses, identify common data gaps, provide possible solutions to fill those data gaps, and offer a prioritization scheme for future testing of polymers. Ultimately, the objective is to suggest a more modern approach to substantiating the environmental safety of the large variety of polymers used in cosmetic and personal care products.

WE053

A consistent Approach for PBT/vPvB Assessment for Pharmaceutical Products

E. Nfon, Smithers Viscient / Department of Regulatory Affairs; K. Malekani, Smithers Viscient / Environmental Fate and Metabolism

Keywords: Persistence, bioaccumulation, Toxicity, Pharmaceuticals Track 6: Environmental policy, risk management, and science communication. Session 6.7: PBT/vPvB Assessment: Update on regulatory guidance,

requirements, strategies and challenges. Abstract A PBT substance is one that that is persistent (P), bioaccumulative (B) and toxic (T) or very persistent (vP) and very bioaccumulative (vB). The PBT assessment approach is well described under the REACH regulation (Regulation EC No 1907/2006) starting with a screening assessment based on available data and when a potential PBT is identified, then a definitive assessment is required. While this procedure is clearly understood for industrial chemicals regulated under REACH, the situation is quite different for pharmaceuticals. There is no definitive PBT/vPvB guidance for pharmaceuticals from the European Medicines Agency (EMA), although it is recommended that the assessment be made according to REACH criteria. Application of the REACH guidance to the PBT assessment of pharmaceuticals is not straight forward. A PBT evaluation of a substance is triggered within REACH if more than 10 tonnes of the substance is used per year. There is no established trigger value for performing a PBT assessment for pharmaceutical products, although it appears that a PBT assessment is applicable to pharmaceutical products that go into Phase II. However our experience is that there is room for interpretations during the review process – especially for products that end at Phase I. Some RMS have consistently rejected the use of all available data; especially data derived from QSARs and instead treat the product as a potential PBT. Furthermore, although there is currently no information on the regulatory consequences of the PBT-assessment for any given product, the situation may change in the future. It is our hope the EMA will issue clear guidance on how a PBT/vPvB assessment should be performed for pharmaceutical products and the consequences for products which fulfil the PBT/vPvB criteria. This presentation will describe our experiences and the challenges we have encountered in performing PBT/vPvB assessments for pharmaceutical products, including an overview of typical review comments from Co-(Rapporteur) are discussed. We also discuss review options for the improvement of PBT/vPvB assessment for pharmaceuticals.

WE054

Evaluation of new assessment methods and enhancement of PBT/vPvB criteria for ionisable substances

H. Holzmann, RWTH Aachen University; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics

The identification of persistent (P) bioaccumulative (B) and toxic (T) substances under the EU regulation REACH (Registration, Evaluation, Authorisation and Restriction of Chemicals) was developed to assess neutral organic compounds. However, nearly 50 % of the chemicals pre-registered at the European Chemicals Agency (ECHA) are partly or completely ionised under environmental conditions (Franco et al., 2010). Since the charge of chemicals strongly influences their properties and environmental behaviour, the currently valid concept under the REACH regulation does not allow a sufficient assessment of ionic or ionisable substances. The objective of the project is to refine the P assessment of ionic and ionisable substances under REACH. For this purpose, simulation tests following OECD guidelines are conducted using two different types of environmental compartments: Aerobic and Anaerobic Transformation in Aquatic Sediment Systems (OECD 308) and Aerobic Mineralisation in Surface Water (OECD 309). As model substances serve three ^{14}C-radiolabelled surfactants, which solely differ in their polar head group: 4-*n*-dodecyl phenol (DP), 4-*n*-sodium dodecylbenzenesulfonate (DS^-) and 4-*n*-dodecylbenzyltrimethylammonium chloride (DA^+). Sediment and surface water were collected from a rainwater detention basin in Aachen, Germany. Preliminary studies using DP and DS^- were performed according to OECD 308 and 309. In the water-sediment system, formation of non-extractable residues (NER) was 13 % for DP and 10 % for DS^- after 65 days. The amount of $^{14}CO_2$ was 48 % (DP) and 63 % (DS^-). In surface water, about 40 % (DS^-) and 30 % (DP) of the initially applied amount of radioactivity was mineralised to $^{14}CO_2$. Degradation studies with DA^+ are in progress and the results will be presented. The results will be used to refine the evaluation of the P criteria for ionic and ionisable chemicals in the PBT assessment. **\n References** Franco, A., Ferranti, A., Davidsen, C., & Trapp, S. (2010). An unexpected challenge: ionizable compounds in the REACH chemical space. The International Journal of Life Cycle Assessment, 15(4), 321-325.

WE055

Assessment of the persistence of ionic or ionisable organic chemicals under REACH

D. Classen, RWTH Aachen University / Institute for Environmental Research; J. Ackermann, Federal Environment Agency Umweltbundesamt / Chemicals; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics

For the protection of humans and the environment, the identification and regulation of chemicals with persistent (P), bioaccumulative (B) and toxic (T) properties are central elements within the environmental assessment. The criteria for the identification of PBT substances under REACH (Registration, Evaluation and Authorization of Chemicals) (EU Nr. 1907/2006) and the guidance for the PBT-assessment have been developed for neutral organic molecules, and do not properly address charged chemicals. Due to their charge, ionic and ionisable substances seem to behave differently in the environment compared to neutral substances. With the addition of cationic, anionic or amphoteric characteristics, the chemicals intrinsic properties (e.g water solubility, log Kow) change as a function of the environmental pH. This depency affects the distribution of these substances within environmental compartments. The ionic function may also lead to different interactions between organic or mineral solid particles and the substance, influencing their bioavailability for potentially decomposing microorganisms, which are governing biotical degradation. In order to improve the evaluation of the persistence of ionic and ionisable substances in the PBT-assessment, sorption and degradation patterns of three 14C model substances carrying either a positive, negative or non-charged functional group will be investigated. The fate and behavior of 14C-labelled 4-n-Dodecylphenol, 4-n-Dodecylbenzenesulfonicacid sodium salt and 4-n-Dodecylbenzyltrimethylammonium chloride in soil simulation tests (OECD 307) will be investigated according to their mineralized, extractable and non-extractable fraction. Non-extractable residues will be investigated further. In addition, the sorption behavior of the model substances will be determined in a sorption study (OECD 106). The results will enable the estimation of the effect of the positive and negative charged groups within the molecule structure regarding the biodegradability and will improve the evaluation of the persistence of ionic and ionisable substances in the PBT-assessment.

WE056

Interaction of sulfonamide with soil humic acid: ESR investigations with nitroxide spin label

A. Ricke, E. Bondarenko, H. Steinhoff, University of Osnabrueck / Physics; G. Úr, K. Hideg, T. Kálai, University of Pécs / Organic and Medicinal Chemistry; M. Matthies, University of Osnabrueck / Institute of Environmental Research

Sulfonamides (SAs) are veterinary antibacterial agents, which are widely used in animal husbandry for treatment of infections. After application of manure of treated livestock to soil, SAs interact with organic soil components, e.g. by reversible sorption or irreversible formation of non-extractable residues (NER). The latter is attributed to physical entrapment (sequestration) or/and covalent binding to soil organic matter. The paper presents a new approach of using stable paramagnetic spin probes to investigate the kinetics of covalent binding of SAs to soil humic acids and to distinguish between sorption, sequestration and covalent binding.Leonardite humic acid (LHA) was mixed with laccase to enhance the amount of reactive quinone groups of LHA and then incubated with nitroxide spin labelled analogues of sulfadiazine (SDZ; HO-4888) and N^4-acetyl derivative of HO-4888 (N-Ac-SDZ; HO-4917). The labeling at the pyrimidine moiety of SDZ leaves the aniline moiety susceptible to covalent binding to LHA, which is blocked by the N-acetylation. A broadened ESR signal was observed for HO-4888, which indicates strong restriction of the reorientational motion of the spin probe, i.e. immobilzation due covalent binding of the aniline moiety of SDZ to reactive quinone sites of LHA. This signal increased immediately after incubation and was used to determine the first-order reaction kinetics of the covalent bond formation. A fast reaction with a half-life of 0.108 h and a slower reaction with a half-life of 14.9 h of covalent binding as well as a reduction half-life of 642 h for the unpaired electron were determined. The treatment of LHA with laccase corroborates the covalent bond formation by oxidizing non-reactive hydroquinone to reactive quinone moieties, which could react via a nucleophilic addition with the amino group of HO-4888. A broadened ESR signal was also recorded for HO-4917 immediately after incubation with LHA. However, this signal declines in contrast to the increase of the signal of HO-4888. This immobilization is caused by unspecific sorption to LHA, not by covalent binding, which is blocked by the N-acetylation. The decrease is attributed to the reduction of the nitroxide spin label and has a half-life of 98.4 h. In a further experiment with the antioxidant Na-ascorbat the reaction rate constants of the bound and free SDZ were found to be almost identical demonstrating that SAs are probably not physically entrapped, at least with soil humic acids.

WE057

The role of non-extractable residues in the environmental risk assessment from regulatory perspective - requirements and challenges

A. Wiemann, UBA Umweltbundesamt; J. Hogeback, Federal Institute of Hydrology; G. Speichert, German Environment Agency UBA; D. Gildemeister, Umweltbundesamt / German Environment Agency / IV2.2 Pharmceuticals; D. Löffler, T. Ternes, German Federal Institute of Hydrology

Non-extractable residues (NER) play different roles in regulatory frameworks. In some cases NER are integrated in the calculation of predicted environmental concentrations (PEC) or are an issue in the authorisation decision. The significance of NER in the assessment of persistence (e.g. PBT, vBvP, POP classification) has been more or less neglected in the past. However, new developments as reflected in guideline revisions (e.g. ECHA R.11, 2017) highlight the importance of NER in tests on transformation in water/sediment and soil. NER can either be reversibly bound to the soil/sediment and pose a potential risk to the environment or irreversibly bound which can be interpreted as sink. Hence, the potential release of parent or transformation products from NER in soil or sediment should be considered. However, distinguishing between these types of NER presents a challenge up to now. Standardised or commonly accepted extraction schemes or analysis techniques are not available due to the broad range of substances and soil/ sediment characteristics. A general classification for NER was proposed by Eschenbach (2013) based on a literature survey dividing NER into four types: type 1 (heavily sorbed fraction) and type 2 (physically entrapped fraction) are considered to be possibly remobilised in the environment. NER Type 3 (covalently bound residues) and type 4 (biogenic bound NER) are considered to be irreversibly bound to soil/sediment or transformed into biomass and therefore a risk to the environment is not anticipated. Furthermore, a stepwise extraction scheme is proposed which would allow the determination of these different NER types. The comprehensive scientific assessment of this extraction scheme supported by experimental data was the aim of a research project funded by UBA. Transformation tests in soil with ^{14}C-labelled substances were carried out in accordance with the OECD 307 guideline. Different extraction methods and chemical breakdown procedures were performed and compared in order to characterise the formation of the different NER types. Under consideration of these results, a refined extraction scheme will be proposed with respect to the general applicability for an adequate risk assessment including NER, in which the reversibly bound fractions have to be considered in assessment of the persistence. An UBA evaluation of regulatory data demonstrate the effects of several influencing factors (e.g. extraction methods, soil type) on NER formation.

WE058

Sorption properties of Ionic organic chemicals: Correlations between ion exchange chromatography retention factors and environmental sorption coefficients

S. Endo, Osaka City University / Urban Research Plaza & Graduate School of Engineering; L. Henneberger, Helmholtz centre for environmental research - UFZ / Cell Toxicology; K. Goss, Helmholtz centre for environmental research - UFZ / Analytical Environmental Chemistry

A common metric for the extent of equilibrium sorption and partitioning, such as the octanol-water partition coefficient (Kow) for neutral organic chemicals, does not exist for ionic organic chemicals. Finding a suitable reference sorption system for ionic organic chemicals should enhance the evaluation, modeling, and prediction of environmentally-relevant sorption coefficients for such chemicals. In this study, we considered synthetic ion exchange materials as possible reference sorption phases and compared their sorption properties against those of various environmental and biological materials. Retention times on commercial ion exchange chromatography columns were measured in fully aqueous eluent and were converted to retention factors (k'), which are proportional to the ion exchanger-water partition coefficients. In the end, we established a data set for retention factors of 61 cations on a strong cation exchange column (SCX), 24 cations on a weak cation exchange column (WCX), and 66 anions on a weak anion exchange column (WAX) measured in consistent experimental conditions (i.e., pH, T, co-existing ions, injected amount). The obtained retention factors were compared to soil organic carbon-water (Koc), clay minerals-water (KCM/w), bovine serum albumin-water (KBSA/w), and muscle protein-water partition coefficients (KMP/w) from the literature. Relatively good correlations (R2 = 0.5-0.6) were found for some cases such as log Koc, log KMP/w, and log KBSA/w against log k' for WAX. For comparison, similar correlation analyses were performed using experimental and predicted log Kow instead of log k'. In most cases, the correlation with log Kow were lower than the correlation with log k'. Notably, log k' has a clearly larger applicability domain than log Kow, because log Kow is unavailable for ionic chemicals derived from strong acids/bases (e.g., sulfonates, quaternary ammoniums), whereas log k' can be measured for such ions too. This study offers a step forward to the development of accurate prediction models for sorption coefficients of ionic chemicals in the environment.

WE059

Simulation of the fate of co-labeled 13C3-15N-glyphosate in a water-sediment system and formation of biogenic non-extractable residues

A. Brock, DTU Environment / DTU Environment; A. Rein, Technische Universität München / Chair of Hydrogeology; F. Polesel, Technical University of Denmark (DTU) / DTU Environment; K. Nowak, TU Berlin / Institute for Environmental Research (Biology V); M. Kästner, Helmholtz centre for environmental research - UFZ / Department of Environmental Biotechnology; S. Trapp, Technical University of Denmark DTU / DTU Environment

The combination of dynamic simulation and stable isotope techniques allows tracking the assimilation of pesticides into biomass [1]. Here, we simulated the fate of co-labeled $^{13}C_3{}^{15}N$-glyphosate in an OECD 308 sediment-water degradation test [2]. The mathematical model used consisted of two compartments for sediment (slow and rapid ad-/desorption), one compartment for dissolved mass, and microbial growth and metabolism. The flow of both ^{15}N and ^{13}C were balanced. The model considers two biodegradation pathways for glyphosate, namely the sarcosine-pathway with complete mineralization, and the incomplete pathway with AMPA as a stable transformation product. Kinetic input parameters were partly estimated from the data, while others were calculated. The microbial growth yield was predicted from the MTB method, using thermodynamics and chemical structure [3]. The model can capture the dynamics of the system, including degradation of glyphosate, formation of AMPA and CO_2, formation of living and dead biomass (proteins) and chemical adsorption. At the end of the experiment (80 days), non-extractable residues accounted for 23% of the ^{13}C and 26% of the ^{15}N; 10% of the ^{13}C and 12% of the ^{15}N were recovered from the protein fraction (mostly non-living amino acids), which is equal to the biogenic non-extractable residues (NER). Biogenic NER consist of assimilated $^{13}C/^{15}N$ and are thus considered to be 'irreversibly bound' as proposed in the updated ECHA guideline for PBT/vPvB assessment [4]. This is the first study simulating the formation of biogenic NER using experiments with ^{15}N-labeled molecules. [1] Kästner, M., Nowak, K. M., Miltner, A., Trapp, S., & Schäffer, A. (2014). Classification and Modelling of Nonextractable Residue (NER) Formation of Xenobiotics in Soil – A Synthesis. Crit Rev Environ Sci Technol, 44(19), 2107–2171. [2] Wang, S., Seiwert, B., Kästner, M., Miltner, A., Schäffer, A., Reemtsma, T., Q. Yang, Nowak, K. M. (2016). (Bio)degradation of glyphosate in water-sediment microcosms - A stable isotope co-labeling approach. Water Res., 99, 91–100. [3] Brock, A. L., Kästner, M., Trapp, S. (2017). Microbial growth yield estimates from thermodynamics and its importance for degradation of pesticides and formation of biogenic non-extractable residues. SAR QSAR Environ Res, 28(8), 629–650. [4] European Chemical Agency (2017). Guidance on Information Requirements and Chemical Safety Assessment. Chapter R.11: PBT/vPvB assessment, Helsinki, Finland.

WE060

Determination of persistent organic pollutants (POPs) in soil from sites adjacent to landfills: different provinces of the Republic of Armenia

A. Aleksandryan, Hazardous Substances & Waste Policy Division / Head of Division; Y. Bunyatyan, Environmental Monitoring and Information Center / Division of waste inventory, classification and technology investigation; V. Khachatryan, National Institute of Oncology / Thoracic Surgery Department; F. Petrosyan, UNIDO BAT/BEP Project(Armenia)

Sources of environmental pollution by persistent organic pollutants (POPs), either used or previously applied pesticides, include landfills, many of which do not meet sanitary and hygienic standards and, at open burning of wastes, become a source of dioxins and furans. Investigation was carried out on soil samples taken in different marzes (provinces) of the Republic of Armenia at the boundaries of landfills and agricultural lands or water basins in the vicinity of towns Ararat (Ararat Marz), Hrazdan (Kotayk Marz), Sevan (Gegharkunik Marz), Gavar (Gegharkunik Marz), Armavir (Armavir Marz), as well as Sasunik village (Aragatsotn Marz). The obtained soil samples were analyzed for determination of the following POPs: - 4 Hexachlorocyclohexane (HCH) isomers: α-, β-, γ-, δ-HCH; - DDT isomers: 2,4'-DDT, 4,4'-DDT; - DDT metabolites: 2,4'-DDE, 4,4'-DDE , 2,4'-DDD, 4,4'-DDD; - Hexachlorobenzene, - Heptachlor, - Aldrin - Dieldrin - Heptachlor epoxide A and Heptachlor epoxide B, - Endosulfan 1 and Endosulfan 2, - Endrin, - Mirex - 14 Dioxin-like polychlorinated biphenyls: congeners No. 77, 81, 105, 114, 118, 123, 126, 156, 157, 167, 169, 170, 180, 189. Quantification of POPs was done using chromatograph with electron capture detectors (ECD) equipped with glass capillary column with stable phase DB–5MS UI and the following parameters: 60 m x 0.250 mm x 0.25 µm. Special attention was paid to the total concentrations of HCH isomers, DDT isomers and their metabolites, as well as the total amount of polychlorinated biphenyls, as maximum allowable concentrations (MACs) are set for the aggregate amount of these compounds. On the analogy, concentrations of other POPs pesticides and all studied POPs are also considered on the whole (summary concentrations) as obvious integral indicators of soil pollution by the studied POPs. In the investigated soil samples HCH isomers, DDT isomers and metabolites, certain Dioxin-like polychlorinated PCBs were detected at concentrations exceeding the established norms.

WE061

Improving the interpretation of Non-Extractable Residues (NER) in degradation assessment

A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics; M. Kaestner, Helmholtz Centre for Environmental Research UFZ / Dept Environmental Biotechnology; S. Trapp, Technical University of Denmark DTU / DTU Environment

Criteria for the assessment of chemical properties, potential (eco)toxicity, and environmental behaviour of industrial chemicals in general, and particularly for REACH substances, biocides, pesticides, and veterinary medicines are summarized in specific European legislations. The regulatory views on NER formation differ considerably, with two extremes of assuming them as either degraded residues of no environmental concern or as bioavailable and non-degraded residues. This may be changed if clear indications for ultimate degradation or irreversible immobilisation are available. Recent research indicated that three types of NER of chemicals in environmental matrices can be experimentally discriminated: sequestered (strongly sorbed and entrapped) residues (type I), containing the parent compound or transformation products or both and having the potential of release. Type II NER are residues that are covalently bound to organic matter in soils or sediments or to biological tissue in organisms. Such residues are considered strongly bound with very low remobilization rates like that of humic matter degradation. However, providing the proof for type II NER is a critical issue in NER assessment. Harsh extraction conditions may release both types of NER but for type II this will rarely happen under physiological conditions. Type III NER comprises biogenic NER (bioNER) after complete metabolization of the xenobiotic and anabolic formation of natural compounds like amino acids etc. The formation potential of bioNER can be predicted by using the theoretical microbial yield, which can be estimated using the Microbial Turnover to Biomass (MTB) method. In addition the amount of bioNER can be experimentally quantified by labelling with stable or radioactive isotopes. bioNER are of no environmental concern. Type

II NER and type I NER should be considered as potentially releasable residues in persistence assessment but the probability of type II release is much lower. For these types the potential of remobilization needs to be tested and evaluated. Our concept is to consider the total amount of NER minus bioNER as the amount of potentially remobilisable xenoNER (type I + II). If a clear differentiation of type I and type II is possible and the latter is irreversibly bound, only type I NER needs to be considered in the persistence assessment. If no characterization of NER is available, we recommend to assess the total amount as potentially remobilizable.

WE062
Photodegradation of Atrazine in the Presence of Indole-3-acetic Acid and Natural Montmorillonite Clay Minerals
C. Gu, Nanjing University / School of the Environment; L. Zhang, Nanjing University
In this study, a new natural degradation pathway of atrazine and the potential mechanism are proposed. Atrazine was oxidized under simulated solar irradiation by indole-3-acetic acid at the environmentally relevant concentration under aerobic condition. The reaction was initiated by the production of hydrated electrons generated from the photoionization of indole-3-acetic acid, and then this species transformed into hydroxyl radical after a series of radical reactions with proton and dissolved oxygen. During this process, the presence of montmorillonite greatly enhanced the yield of hydrated electron and promoted the further degradation of atrazine by hydroxyl radical. The novel reaction is to some extent affected by pH and the type of exchangeable cation present on montmorillonite. Based on our results, montmorillonite saturated with potassium ion (K^+-montmorillonite) exhibited a superiority in the promotion of the degradation rate compared to that saturated with calcium ion (Ca^{2+}-montmorillonite), and this reaction was conspicuously suppressed as pH increased. In this process, montmorillonite not only increases the yield and prolongs the lifetime of hydrated electrons by stabilizing radical cations through electrostatic attraction with the negative charges embedded in the interlayer, but also provides a confined space where the probability of contact between atrazine and active radicals is markedly increased. In this study, we also investigated the influence of smectite hydration status on the degradation efficiency of atrazine, which revealed that freeze-dried K^+-montmorillonite promoted the degradation process to a greater extent than freshly-prepared K^+-montmorillonite, while the freeze drying process had no significant influence on the hydration status of Ca^{2+}-montmorillonite.

WE063
Photodegradation Half-lives of a Fragrance Ingredient in Natural Waters at Depth Calculated from Laboratory Study Results
J. Lin, Firmenich Inc / Research & Development; K. McNeill, ETH Zurich / Institute of Biogeochemistry and Pollutant Dynamics; M. Emberger, A. Casilli, V. Hewins, Firmenich, Inc / Research & Development; S. Gimeno, Firmenich / Product Safety and Regulatory Affairs
Photodegradation, an important abiotic degradation process, is rarely considered in the persistence assessment of chemical substances. This is due to the difficulties and the lack of knowledge to extrapolate results from laboratory studies to the complex environment. As the first step of our undertaking to develop methodologies to extrapolate laboratory data to the aquatic environment, we chose a fragrance ingredient, MyrrhoneÒ as an example, and used laboratory study results to calculate its photodegradation half-lives at depths in natural waters. Direct photodegradation was revealed to be the dominant photodegradation process of MyrrhoneÒ and the quantum yield was determined to be 0.51 in the laboratory. Four main volatile photodegradation products, which continued to photodegrade, were observed, isolated by preparative multidimensional GC and identified using NMR. Direct photodegradation half-lives of MyrrhoneÒ in natural waters at different depths were calculated based on its molar absorption spectrum, quantum yield, and the irradiance at depth of natural waters. The irradiance values at depth were obtained by applying diffuse attenuation coefficients (K_d) to the irradiance at the surface of water, generated using a solar irradiance calculator as a function of time, date and location. K_d are empirical values determined by the interaction of a number of factors, including absorbance by dissolved organic matter and particulate matter, scattering, and the angular distribution of the light field. A correction factor for cloud cover was applied at the last step of calculation, which gave rise to half-lives that ranged from 1.3 to 9.1 daysfor small size lakes and, 6.3 to 45 daysfor large size lakes under realistic conditions in Australia. The calculation of half-lives at the surface of water was validated by two outdoor photolysis experiments. The calculated half-lives of 0.38 h and 1.14 h were in agreement with the measured half-lives of 0.40 h and 1.15 h, respectively. This agreement indicated that mathematical models can be developed to define complex environmental conditions for the extrapolation of laboratory data to the environment. The next step is to design experiments to measure half-lives at depth in natural water to refine and validate the calculation of half-lives. Thoroughly validated models can be valuable tools for the persistence assessment of chemicals based on photodegradation information.

WE064
The Photolytic Fate of Fungicides
J. Apell, MIT / Civil & Environmental Engineering; K. McNeill, ETH Zurich
The use of pesticides has allowed for increased crop productivity in agriculture. However, because of their frequent application onto agricultural lands, their environmental fate is of particular interest. Pesticides have been ubiquitously detected in natural waters throughout Europe as well as in water supplies, which suggests these pesticides may be more mobile and persistent than their initial evaluations indicated. A potential reason for the mischaracterization of these pesticides' persistence is that environmental fate models can oversimplify the degradation pathways in the environment. For example, only direct photolysis in pure water can be considered, but the presence of dissolved organic matter can significantly affect the photolytic fate of pesticides in natural water. The photolytic degradation can either be enhanced by indirect photolysis reactions with photochemically produced reactive intermediates (e.g., triplet state dissolved organic matter, single oxygen, hydroxyl radical), or it can be suppressed by the light screening effect of organic matter once the light enters the water column. This research focused on the photolytic fate of fungicides, which account for approximately half of pesticide usage in Europe, in natural waters. For the several commonly used fungicides investigated, the results show that both direct and indirect photodegradation are important to quantify in order to accurately characterize the environmental fate of fungicides.

WE065
Study Design Considerations for E-Fate Testing of UVCB Substances
C. Lowrie, Charles River / Environmental Fate and Metabolism
Substances of unknown or variable composition, complex reaction products or biological materials (UVCB Substances) are products which are often not fully identfiable and their composition may be difficult to predict. Under REACH Registration, it may be necessary to test these substances, which from many perspectives represents significant challenges. Reference is made to REACH Regulations and in particular Annex VII Section 9.2 where a series of degradation studies are proposed including simulation testing in surface water, soil and sediment with an endpoint in each case including identification of degradation products. It is relatively straight forward to generate e-fate data on a substance of known composition (chemical identity and purity) based on results from simulation studies in soil, sediment or water. OECD test guidelines 307, 308 and 309 used to describe experimental designs for simulation testing require the use of high purity material (>95%) and the use of a radiolabelled substance is highly recomended if the researcher aims to study the degradation products. It is therefore fundamentally not possible to perform simulation tests to determine the e-fate characteristics of UVCB substances due to the variable, unknown or unrepdictable nature of the starting material. If testing of the "substance" is flawed then there are 2 options - the first is to isolate, purify and identify individual substances from the UVCB and then determine the e-fate characteristics of each component independently of each other...this seams unrealistic in most cases. Incorporation of a radiolabel to this test is likely to be prohibitive in terms of scale. An alternative approach would be to consider the chemical structures in a UVCB in groups and to test exemplar molecules or structures from within the mixture. This approach is possible when the indivudal structures have similar physico-chemical properties and structures are related. In this case hazard data is produced for the exemplars which can be used to predict the overall persistance of the UVCB substance. Choosing the most appropriate exemplar molecule may be challenging and examples are given. If the exemplar molecule is persistent then reasoned logic would dictate that the UVCB substance was persistent and further testing implimented accordingly.

WE066
In silico investigation of the triplet-sensitised phototransformation of phenols induced by chromophoric dissolved organic matter
E. Papa, University of Insubria / Department of Theoretical and Applied Sciences (DiSTA); L. Motta, University of Insubria / Department of Theoretical and Applied Sciences DiSTA; A. Sangion, P. Gramatica, University of Insubria / Department of Theoretical and Applied Sciences (DiSTA); M. Minella, D. Vione, University of Torino / Chemistry
Chemical reactions driven by sunlight are important processes in surface freshwaters, where they are involved in the transformation of xenobiotic molecules and of naturally occurring compounds. The relevant reactions are generally divided into direct photolysis and indirect phototransformation. Direct photolysis involves molecules that absorb sunlight and are transformed as a consequence. Indirect phototransformation involves reactive transients such as $^{\cdot}OH$, $CO_3^{\cdot -}$, $^1O_2$ and the triplet states of chromophoric dissolved organic matter ($^3CDOM^*$). They are generated by irradiation of photosensitisers such as CDOM (producing $^3CDOM^*$, $^1O_2$ and $^{\cdot}OH$), nitrate and nitrite (producing $^{\cdot}OH$). Among these transient species, $^3CDOM^*$ is certainly the least understood one in terms of its nature (which is a consequence of the poorly known nature of CDOM) and reactivity. Still, $^3CDOM^*$ is involved into the transformation of several organic pollutants. In this work different triplet sensitisers that may be used as surrogates to estimate second-order rate constants with CDOM have been studied *in silico*. In particular, the experimental second-order reaction rate constants measured for the photodegradation of 49 phenols with four model photosensitisers used as CDOM proxies (1-nitronaphthalene (1-NN), riboflavin (Rbf), 4-carboxybenzophenone (4CBP), and anthraquinone-2-sulphonate (AQ2S)) have been used to derive Quantitative Structure-Activity Relationships on the basis of theoretical molecular

descriptors. The choice of phenols is motivated by the fact that they are the most likely compounds to undergo triplet-sensitised phototransformation in sunlight surface waters. Results show that the reaction rate constants with ^{3}AQ2S* and ^{3}4CBP* give the best QSAR models that can be used to simulate the photodegradation of phenols and similar compounds in the presence of CDOM. Quality differences in the QSARs generated for these reactions are probably due to differences in the chemical structure of the four sensitisers. These results provide additional insight into the mechanisms that regulate the fate of potential organic pollutants in surface waters. They will be used to design future experimental tests by focussing on one/two among the studied sensitisers, and to predict the photodegradation of new and existing substituted phenols and similar compounds on the basis of their chemical structure.

WE067

In silico Tools to Assess the Confidence of QSAR Model Predictions

R. Kühne, Helmholtz centre for environmental research - UFZ / Department of Ecological Chemistry; S.S. Kutsarova, O. Mekenyan, University of Zlatarov / Laboratory of Mathematical Chemistry; G. Schuurmann, Helmholtz centre for environmental research - UFZ / Department of Ecological Chemistry

For the regulatory acceptability of QSAR predictions solid information about the reliability of the applied models is crucial. This regards the model in general as well as the particular prediction for a certain chemical. The presented study provides computerized tools to support the assignment of prediction reliabilities. The first topic of concern is the applicability domain. Generally, the applicability domain comprises various aspects. The focus of this presentation is on the mechanistic domain and on the chemical domain in terms of structures and substructures. In particular, the application of atom-centered fragments (ACF) is demonstrated. While ACF characterization of the general structural domain of a training set is established already, the approaches shown here provide more specific information about the reliability of a prediction. On one hand, this is achieved by extensions of the ACF approach with data sets beyond the training set. Furthermore, data sets are separated into subsets with regard to performance or value ranges, and these subsets are employed to derive advanced reliability indicators. Secondly, automatically obtained model selectors can assist in selecting the presumably best-performing model from a model suite for a certain chemical depending on its structure and properties. Model selectors in this regard are computed scores derived from application of the model suite to chemicals with known experimental data. Thirdly, consensus modelling strategies are presented and examined to compensate for individual model errors. When combining predictions from different models, consensus outcomes can accordingly increase the levels of confidence, while conflicting outcomes are indicating lower reliabilities. In this respect, specific indicators can be achieved by in silico methods for particular model suites, and consensus approaches contribute to weight of evidence assessments. For all three aspects, working tools will be presented, and their performance will be demonstrated via examples from existing models and data sets. Acknowledgment: This study was financially supported by the European Union 7th Framework Program SOLUTIONS (FP7-ENV-2013) of the under grant agreement no. 603437.

WE068

Data Gap filling with ECOSAR in K-REACH compliance, a limitation and weakness

J. Ra, Korea Institute of Industrial Technology / Environmental Science and Engineering; H. Park, Korea Institute of Industrial Technology; S. ok, Kitech / Regulatory Chemical Analysis & Risk assessment Center

ECOSAR is a computer based QSAR program developed by US EPA within the regulatory constraints of the TSCA. However, it is also used in the other country or organization such as EU, Korea, OECD, etc. for their regulatory purpose. We introduced ECOSAR program to generate toxicity data and fill the data gap for developing species sensitivity distribution of 20 organic compounds. However, ECOSAR shows more pragmatic than theoretical characteristic. Thus, we investigated whether this model shows acceptable results on the deficient data of 20 organic compounds or not. Therefore we collected published data for fish and daphnia available and compared their geometric value to the output of ECOSAR. Some chemicals show similar output value to experimental data within double scale but others show very large difference of 1,400 times higher value in ECOSAR output. The least predictable substance is acrylic acid where 4 experimental data are used for the geometric mean value. On the contrary, methyl hydrazine shows almost equal value in fish toxicity. According to the results, ECOSAR may not applicable to all types of chemicals within the acceptable limit of regulatory system. In this study, we are going to further investigate ECOSAR program to categorize chemical class showing higher applicability, which may reduce the error of ECOSAR in regulatory area.

WE069

Innovative analytical method to enhance POPs and emerging pollutants extraction in water samples by micelles using GC-MS/MS

s. giannarelli, University of Pisa / Chemistry and Industrial Chemistry; I. De Angelis, A. Lazzarelli, University of Pisa / Department of Chemistry and Industrial Chemistry

In this paper solubilization of persistent organic pollutants, PAHs, PCBs, pesticides and emerging pollutants, as PBDEs or PCN, was investigated in water samples, using some anionic, cationic and non-ionic surfactants. These pollutants are well known for their considerable toxicity, persistence and bioaccumulation toward both human health and environment in addition to their low aqueous solubility. However, the use of surfactants to water solution enhances solubilization of hydrophobic organic compounds. Above the critical micelle concentration (CMC), surfactants exist as aggregates in solution and hydrophobic compounds move to hydrophilic micellar core region. This technic was used as alternative method to traditional liquid-liquid extraction with hexane. The physical and chemical properties as size, shape, ionic strength and hydrophobicity are important to identify the appropriate surfactant depending on the type of compound to be removed. The method consists in two steps: a) removal of analytes from aqueous solution to the micelles; b) movement of micelles from aqueous solution to the organic solvent followed by micellar deformation with addition of NaCl. Both steps are effective, easy and with high recovery of pollutants. Furthermore, the samples are already in the solvent for quantitative analysis. The removal of analytes from aqueous solution was verified by comparing UV-Vis spectra in the range 240-360 nm before and after extraction in isooctane and then, has been quantified with capillary gas- chromatography-triple quadrupole mass spectrometry. Results reveal better extraction by micelles than traditional method, mostly more interesting are binary system of surfactants, i.e. cationic-nonionic, anionic-nonionic. It was found that solubilization of analytes increases with increasing hydrophobicity of surfactant. Therefore, the aim of this study is to improve this method comparing solubilization capability of these surfactants based on different number of ethylene oxide groups in nonionic surfactants, tail length and presence of ethyl or benzyl groups in cationic and anionic surfactants in order to optimize recovery of pollutants and minimize the quantity to be used in environmental monitoring programs.

WE070

Water Treatment - A Regulatory Challenge under Regulation (EC) No 1107/2009

F. Schnitzler, S. Dorn, J. Wilbuer, Dr Knoell Consult GmbH

Regulation (EC) No 1107/2009 aims to protect humans and the environment and lays down rules for the authorisation of plant protection products in commercial form and for their placing on the market, use and control. Plant protection products consist of or contain active substances, i.e. the molecules or materials responsible for the action against the target pest, weed or fungal. Ozonation and chlorination are primary disinfection processes for central water treatment. Metabolites of certain active substances were found to react during ozonation of drinking water and to form by-products with toxic, carcinogenic and genotoxic characteristics. During the approval process of active substances, data gaps have recently been identified by EFSA regarding Article 4, 3(b) of Regulation (EC) No 1107/2009: *A plant protection product ... shall have no immediate or delayed harmful effect on human health ... directly or through drinking water (taking into account substances resulting from water treatment)…\n* In contradiction, water treatment processes are not implemented in the data requirements (Reg. 283/2013 or 284/2013) and no guidance document for experimental testing is available. Published EFSA conclusions of active substances submitted under the 3rd group of the Annex I Renewal Programme (AIR3) are evaluated for different applied ways of addressing ozonation and chlorination during the registration process. A statistical overview of the results shows whether water treatment processes have been addressed successfully. An overview of these regulatory challenges is provided with intermediate recommendations to address water treatment processes for active substance approval.

WE071

The identification of persistent, mobile, toxic (PMT) chemicals as SVHC based on their equivalent level of concern to persistent, bioaccumulative, toxic chemicals defined in Article 57(f) of REACH

S. Hale, Norwegian Geotechnical Institute; H. Arp, NGI / Environmental Technology; L. Vierke, German Environment Agency / Chemicals; M. Neumann, German Environment Agency (UBA) / Section IV 2.3 Chemicals

The identification of polychlorinated biphenyls in top predators nearly 50 years ago lead to the establishment of many environmental chemistry regulations and chemical regulatory frameworks directed towards persistent, bioaccumulative, toxic (PBT) or a very persistent, very bioaccumulative (vPvB) chemicals. In order to protect human and the environment, additional physico-chemical properties to PBT and vPvB may be relevant to consider. Mobility is one of these additional properties and persistent, mobile, toxic or very persistent very mobile compounds are highly problematic for human health and the environment. Once mobile compounds have entered the environment they have the potential to impact remote pristine water bodies and raw water used for drinking water owing to their ability to spread spatially. Mobility has been qualitatively defined in REACH as "the potential of the substance, if released to the environment, to transport to groundwater or far from the site of release" (Annex II of REACH). However, a commonly accepted quantitative definition of mobility is not given within REACH and registrants/manufacturers are not obligated to carry out an assessment of mobility. Here we present a case for the consideration of PMT and vPvM as substances of very high concern (SVHC) based on their identification through article 57(f) of REACH based on a demonstrated equivalent level of concern (ELoC) as PBT or vPvB substances. In order to identify PMT as a substance of ELoC it must be demonstrated that there is "scientific evidence of probable serious effects to human health or the environment which give rise to an equivalent level of concern" and that there is evidence from a risk based considerations that the substance may cause serious effects during use and after through consideration, it should have been established that the inclusion of the substance on the candidate list is the most effective management strategy. With the protection of drinking water and pristine water bodies in mind, both bioaccumulation and mobility of persistent chemical substances are non-desirable. The environmental effect felt by bioaccumulative and mobile chemical substances varies both temporally and spatially, where mobile compounds can potentially accumulate in semi-closed drinking water cycles, whilst bioaccumulative compounds are able to accumulate in a closed loop system. vPvB can accumulate in food chains over time while vPvM can accumulate in pristine environments over time.

WE072

How many vPvM/PMT substances have been registered under REACH? - vPvM/PMT screening by using the Danish QSAR database

R. Holmberg, Danish EPA / Chemicals; E.B. Wedebye, N.G. Nikolov, Technical University of Denmark (DTU) / Division of Diet, Disease Prevention and Toxicology, DTU Food; K. Tyle, DK EPA / Chemicals

UBA, Germany, has initiated work to develop criteria to identify substances which are very persistent and very mobile (vPvM), and persistent, mobile and toxic (PMT). These substances are of particular concern for surface water organisms and for ground water used as drinking water (human health concern). QSAR screenings using the free online Danish QSAR DB were performed on 2,372 mono-constituent organic substances. For persistency (P) algorithms as used for the persistency screening under PBT assessment was used. A new screening algorithm for very persistent substances (vP) was set up by adjusting the P screening algorithm. For mobile (M) and very mobile (vM) substances new screening algorithms were developed using the substance properties of water solubility (Sw) and the soil/sediment organic carbon-water partition coefficient (log Koc) by equating log Koc and Sw values based on >64.000 substances. The pH-dependent octanol-water partition coefficient (log Dow) was used as an alternative value for substances which ionize at environmentally relevant pHs. For ecotoxicity, similar QSAR algorithms as used for the ecotoxicity screening under PBT assessments were used. In addition, new algorithms for predicting CMR properties from ongoing work on the updated Advisory Self-classification List (expected to be published by end of 2017 by the Danish EPA) were included to also take potential chronic mammalian toxicity into account. The selected QSAR based T-related algorithms were employed on top the screening algorithms for P and M properties as a screening tool for substances with a potential for mammalian and non-mammalian toxicity to wildlife species and human health. The results of the screenings identify the potentially vPvM and PBT substances currently registered under REACH in tonnages >10 tpa per manufacturer or importer. The screenings identify substances according to criteria proposed by UBA and the developed QSAR algorithms were also used to explore potential alternative sets of criteria for vPvM/PMT properties. The results provide input for current and future work with the concept of vPvMs/PMTs. The Danish QSAR DB contains 650,543 substances, of which 80.085 currently are pre-registered and/or registered under REACH. Future vPvM/PMT screenings can be refined to address specific substance groups of interest; substances registered after the last REACH registration deadline; or address future modifications, if relevant, in the proposed vPvM/PMT criteria.

WE073

Identifying PMT substances amongst REACH registered substances

H. Arp, NGI / Environmental Technology; S. Hale, Norwegian Geotechnical Institute; A. Striffler, denkbares; D. Sättler, UBA / Section IV Chemicals; I. Schliebner, UBA; M. Neumann, German Environment Agency (UBA) / Section IV 2.3 Chemicals

The chemicals that have the greatest chances of appearing in drinking water are those that are mobile in the aquatic environment enough to enter drinking water sources and persistent enough to survive water treatment processes. Despite the growing number of organic chemicals released on the market, there has been very little consideration as to how to identify or categorize which of them are persistent, mobile and toxic, (PMT) and thereby pose a potential threat to drinking water. Herein the list of REACH registered substances as of May 2017 was independently evaluated for their likelihood of being a PMT. The evaluation of persistent (P), very persistent (vP) or potentially persistent (i.e. Pscreen) was performed according to REACH guidelines. For mobility, a criteria of a measured (or estimated) log Koc

WE074

Recent Advances in Toxicology, Safer-Alternatives Assessment, Value-in-Use and Best Practice Guidance of Short-Chain Fluorotelomer-based Products for AFFF, Textiles and Other End-Uses

S. Korzeniowski, BeachEdge Consulting; J. Bowman, FluoroCouncil

Per- and polyfluoralkyl substances (PFAS) is a term that describes a wide and diverse array of chemistry containing fluorine and carbon. The focus of this poster presentation will be on the fluorotelomer-based products of the PFAS group with six or less fluorinated carbons ("short chain"). Fluorotelomer-based products can be in either the polymeric or non-polymeric PFAS categories. Within the polymeric PFAS category, the fluorinated repellent products, including durable water repellents (DWRs), are found. These are normally side-chain fluorinated polymers typically applied in combination with other finishing auxiliaries. The side-chain polymeric fluorotelomer-based products perform exceptionally well and provide essential and critical properties on high-end performance garments, work-wear, first responder gear and in military uses. Within the non-polymeric PFAS category, fluorotelomer-based surface active agents (e.g. "fluorosurfactants") are used in complex multi-component formulations such as Cleaning Products, Paints, Coatings and Aqueous Film Forming Foams (AFFF). The non-polymeric fluorotelomer-based products provide superior surface wetting and leveling properties and unmatched AFFF fire-fighting performance on high-hazard Class B (i.e., hydrocarbon & polar solvent liquids) fires. The remarkable strength of the C-F bond provides products with superior resistance to extreme thermal, chemical and environmental conditions. While this unique stability makes these products ideal in many end-use applications, as well as in protecting people, equipment and property, it also makes them resistant to degradation and persistent in the environment. Each of these noted fluorotelomer-based chemistries and end-use applications have the potential to be released during use and could create an environmental footprint. Thus it is essential to follow published best practice guidance (BPG) in handing these products. This poster will highlight recent advances in toxicology, including multiple endocrine evaluations, safer-alternatives assessment methodology, analytical advances, challenges and success in the development of short-chain fluorotelomer-based products and an overview of their value-in-use including some critical uses. A perspective on when and how best to use these products, while at the same time minimizing the environmental footprint will be featured in this Poster Presentation.

WE075

LIFE project PHOENIX: a new project for the management of water pollution from short chain perfluoroalkyl acids in Veneto region (Italy)

F. Russo, M. Vazzoler, V. Groppi, Regione Veneto, Direzione Prevenzione, sicurezza alimentare, veterinaria; F. Zanon, F. Da Prà, R. Lava, M. Mazzola, G. Onofrio, L. Da Rugna, ARPA Veneto; M. Bonato, University of Padua, Department of Biology and Department of Industrial Engineering; L. Guidolin, P. Irato, G. Santovito, L. Tallandini, University of Padua, Department of Biology; M. Carrer, L. Palmeri, University of Padua / Department of Industrial Engineering; N. Tormen, University of Padua, Department of Biology; S. Valsecchi, F. Cappelli, Water Research Institute - Italian National Research Council IRSA-CNR; S. Polesello, Water Research Institute- CNR / Water Research Institute

In 2013 a significant episode of PFAS pollution of surface, ground- and drinking water has been discovered in a large area of the Veneto region, in Northern Italy. The most important source of pollution was identified in a fluorochemical plant, sited in an area of groundwater recharge. The Veneto Region immediately put in place mitigation actions that were more effective for long chain PFAS than for short chain ones. For that reason, within the framework of the Community Life Program, a project on the management of short-chain perfluorinated compounds (PFAS), coordinated by the Department of Health Protection, Food and Veterinary Safety of the Veneto Region, in association with CNR IRSA, ARPAV and University of Padua, has been proposed and then funded. The activities of the LIFE-Phoenix project, acronym for "Perfluorinated compounds Holistic Environmental Institutional eXperience" started on 2017 and whih will end in 2020. LIFE PHOENIX project aims at demonstrating how a new interinstitutional governance system, supported through innovative forecast tools based on ongoing monitoring, can manage risks related to the diffusion of persistent mobile organic contaminants (PMOC) such as short chain PFAS. This project will develop a set of institutional procedures and tools to assess and possibly prevent as well as respond to risks for environment and human health with the contribution of multidisciplinary specialists who will develop tools, protocols, guidelines and indications to assist policy-makers in taking decisions and implementing effective prevention measures for environment, human health and the socio-economic context. All project activities will focus on a real scale case constituted by the PFAS pollution in the provinces of Vicenza, Verona and Padova (an area of 930 km^2), and involve regional authorities managing risks and emergency. The project will validate and compare some innovative technological tools for the mitigation of PFAS concentration in the water through a pilot plant adopting different technics for the purification of irrigation water and drinking water, using full-scale plant (wetland system) and physico-chemical plans breakdown system (filters). The technologies applied to these experimental sites will be incorporated into an integrated management system that will serve as a model for managing analogous chemical pollution events from persistent and soluble polar substances.

WE076

Ecotoxicological characterization of aquifers at Junin Formation and Pampeano from Hydrogeological Sub-Region II, Buenos Aires Argentina

W.D. Di Marzio, CONICET-PRIET / PRIET; M. Saenz, PRIET CONICET, National University of Luján; J. Alberdi, priet conicet unlu; s. curieses, S. Martinez, CONICET PRIET UNLU; A. Silva, UBA Fac Cs Exactas; D. Galassi, Universita LAquila; T. Di Lorenzo, ISE CNR

The Groundwater of Hydrogeological Sub-Region II at Buenos Aires province, is well characterized from a physico-chemical perspective. Until today is well established that fluoride and arsenic are the main concerns related with the potential use as source of drinking water. However, an ecosystemic perspective that focus on their invertebrate communities and the ecotoxicity potential is missing at least in this subregion. In this work we showed the preliminary results obtained after the sampling and analysis of ten on 20 total wells projected to be evaluated. This area is known as Junín Formation, which consists of sandy loam sediments to silty loams of reddish brown to light brown color, very friable, and with scarce calcareous bodies of pedogenetic origin. The Junín Formation of wind morphology constitutes an alternation of low elevations and depressions. Aeolian sediments, which belong to the Junín Formation (Aeolian Platense), normally do not exceed 5 m in thickness and usually have calcareous (coarse) levels. The alluvial and colluvial deposits (sandy silts, sands, gravels and blocks) have a reduced vertical and areal expression. The samples were characterized according their main anionic and cationic constituents, presence of glyphosate and chlorpyrifos, TOC, arsenic and fluoride. Also, cyto and genotoxicity of concentrated waters were studied by comet assay using coelomocytes of *Eisenia fetida*. Water quality was analyzed in combination with the dominant taxon of invertebrates founded. They were mainly epigean Copepods, Acari, Collembola, Insecta, Oligochaeta, Nematoda. A preliminary biotic and ecotoxic index were created to characterize each sampling well.

WE077

Chemical analysis, monitoring and toxicological evaluation of very polar compounds in drinking water and drinking water sources

R. Sjerps, KWR Watercycle Research Institute / Chemical Water Quality and Health; P. Kooij, KWR Watercycle Research Institute; K. Baken, KWR Watercycle Research Institute / CWG; A. Kolkman, KWR Watercycle Research Institute; P. de Voogt, University of Amsterdam / IBED

Very polar organic compounds are of special interest for drinking water utilities, since these substances are likely to end up in drinking water. The distribution of persistent and mobile compounds in the urban water cycle is widespread since they can leach to groundwater and/or pass wastewater and drinking water treatment. Currently there is an analytical gap, a monitoring gap and a lack of toxicity data for persistent and mobile organic compounds (PMOC). We aimed to close these gaps by the implementation of a target HILIC-MS screening method for very polar compounds and quaternary ammonium compounds and a non-target HILIC screening. With these methods 45 samples from surface water, river bank filtrate, groundwater and drinking water in The Netherlands and Flanders have been analysed. Detected compounds include known contaminants melamine, urotropin, metformin and guanylurea and newly detected compounds cotinine, cyanuric acid and n-methyldiethanolamine. Despite of the high removal rates during drinking water treatment (>70%), these compounds were found in drinking water. One compound is introduced during treatment: dichloroacetic acid. Most compounds and highest concentrations were detected in surface water and drinking water produced from surface water. The monitoring data of the very polar compounds gives insight into the seasonal variation of surface water quality. For the detected polar compounds human toxicological risk assessment is performed and results will be presented.

WE078

Beyond DEHP: High-molecular-weight phthalates and non-phthalate plasticizers in German Rivers

R. Nagorka, Federal Environment Agency (UBA) / Water and Soil; J. Koschorreck, Umweltbundesamt

The phthalate plasticizer Di(2-ethylhexyl) phthalate (DEHP) is a well-documented ubiquitous contaminant and frequently regarded as the predominant plasticizer in the environment. The use of DEHP and other phthalates was banned in the EU because of their endocrine-disrupting activity. As a result, the production of these phthalates decreased significantly. In contrast, the global plasticizers demand is continuously growing. Market data show that high-molecular-weight phthalates are now extensively substituting DEHP together with various types of non-phthalate plasticizers. Previous investigations on plasticizers in the aquatic environment have largely focused on DEHP and selected low-molecular-weight phthalates. However, little attention has been given to the presence of other phthalates and non-phthalate plasticizers. In consequence, data are scarce on the occurrence, fate and effects of these alternative plasticizers in the environment. The objective of this study was to obtain spatio-temporal trends for DEHP and its substitutes in freshwater systems. We analyzed suspended particulate matter samples (SPM) for the presence of 23 plasticizers, i.e. 17 phthalates and 6 non-phthalates. Samples from recent years were retrieved from the German Environmental Specimen Bank (ESB), which covered 16 sampling sites from major rivers including Rhine, Elbe and Danube. Retrospective trend monitoring with archived ESB samples that were collected over the last decade enabled trend assessment for DEHP and its non-regulated substitutes. Today, the high-molecular-weight plasticizer Diisononyl phthalate (DINP) is the most abundant plasticizer detected in the SPM-samples. Our results indicate a fast appearance of new plasticizers like Diisononyl cyclohexane-1,2-dicarboxylate (DINCH) in freshwater environments and we identified several compounds, for instance Di(2-propylheptyl) phthalate (DPHP), as potential chemicals of emerging concern with increasing levels.

Wastewater effluents: How research can improve risk assessment and regulation (P)

WE079

Acute and chronic toxicity of Direct Blue 15 on microalgae and cladocerans: a comparative study

M. Hernández Zamora, Escuela Nacional de Ciencias Biologicas-I.P.N / Laboratory of Experimental Hydrobiology; F. Martínez-Jerónimo, Escuela Nacional de Ciencias Biológicas-I.P.N. / Laboratory of Experimental Hydrobiology

Aquatic pollution resulting from industrial activities, especially textile, leather, food and agrochemicals, is a major concern. Dyeing process of fabrics produces approximately 90% of the total textile wastewaters, containing significant concentrations of residual dyes. Colored wastewaters reduce light penetration in the water column, and affect photosynthesis of phytoplankters. In addition, azo dyes are synthetized from carcinogenic compounds, such as benzidine; this can threat the aquatic biota. The environmental impact caused by the discharge of textile dyes effluents has been scarcely studied; therefore, our study was aimed at evaluating the toxic effect of the azo dye Direct Blue 15 (DB15) on a primary producer (*Pseudokirchneriella subcapitata*) and on a primary consumer (*Ceriodaphnia dubia*). The microalga was exposed to 4, 8, 16, 32 and 64 mg L^{-1} DB15 (96 h, 25°C, and continuous illumination of 120 μmoles $m^{-2} s^{-1}$); the effects of DB15 on photosynthetic pigment and macromolecules content (proteins, carbohydrates and lipids) were assessed. The acute toxic effects of DB15 dye in cladocerans were determined at 48 h; tested concentrations were 100, 200, 300, 400 and 500 mg L^{-1}, at 25°C, 16:8 h photoperiod, with no food supply during the assays. In chronic toxicity tests *C. dubia* individuals were exposed to 5, 10, 15, 20 and 25 mg L^{-1} DB15 (7 days at 25 °C, 16:8 h photoperiod, $1x10^6$ cell mL of *P. subcapitata* as food). *P. subcapitata* was more sensitive to DB15 (IC_{50}: 13.30 mg L^{-1}) than *C. dubia* (LC_{50}: 450 mg L^{-1}). Chlorophyll-*a* and -*b* were significantly increased in the algae exposed to all the dye concentrations, comparing with the control, but carotenoids were significantly reduced in all the DB15 concentrations. Concentration of proteins, carbohydrates and lipids per cell in *P. subcapitata* exposed to all DB15 concentrations were significantly higher than that measured in the control. In the highest DB15 concentrations, total progeny, number of released clutches, and reproduction were significantly decreased in *C. rigaudi*; but age at first reproduction was significantly increased at 20 and 25 mg L^{-1} DB15. Results demonstrated that DB15 dye caused toxic effects of different magnitudes on aquatic biota (primary producer and primary consumer), for this reason, the azo dyes must be regulated to prevent environmental impacts caused by the discharge of textile dyes into waterbodies.

WE080

Integrated biomarker response calculation as a useful tool to assess the impact of effluents on the health status of fish

S. Wilhelm, University of Tuebingen / Animal Physiological Ecology; S. Jacob, Universität Tübingen / Animal Physiological Ecology; M. Ziegler, R. Triebskorn, University of Tuebingen / Animal Physiological Ecology

Wastewater treatment plants (WWTPs) are considered as one of the major sources of micropollutants in the aquatic environment. Many compounds have been linked to toxic and endocrine effects in aquatic organisms. The present study examines the impact of three WWTPs situated on different streams in Southern Germany on the health status of fish. Two of the investigated facilities are conventional treatment plants, combining mechanical, biological and chemical treatment. The third one was equipped with an additional powdered activated carbon filter unit, which has been in operation since October 2013. In order to examine the effects of the different effluents on fish, one-year old rainbow trout (*Oncorhynchus mykiss*) were exposed in cages upstream and downstream of each WWTP effluent. Furthermore, the impact of the WWTP upgrade with activated carbon was investigated by comparing results of caging exposures conducted prior and subsequent to the upgrade. Several biomarkers, including histopathological alterations, the formation of micronuclei and binuclei, changes in vitellogenin levels, induction of hepatic EROD activity, and changes in stress protein levels were examined, and the integrated biological responses (IBR) were calculated for the downstream exposure sites according to Sanchez *et al.* (2013), using the respective upstream site as a reference. IBR values for the conventional treatment plants (WWTP 1 and 2) differed slightly from each other, with WWTP 2 showing three to five times higher indices than WWTP 1. However, the highest IBR values were detected for male fish exposed downstream of the third WWTP prior to the upgrade with an activated carbon filter unit. After the installation of the additional treatment technology, a pronounced reduction of this IBR was observed. The integration of different biomarkers proved to be a suitable approach to assess the impact of WWTP effluents on the health status of fish. Furthermore, it was a helpful tool to reveal the advantages of WWTP upgrading with powdered activated carbon.

WE081

Application of eco-genotoxicological and microbiological parameters for the assessment of the quality of wastewater industrial reuse.

S. Cacioli, Italian Institute of Health ISS / Department of Environmental and

Health; I. Lacchetti, Istituto Superiore di Sanità / Environment and Health; M. Carere, Italian Institute of Health ISS; C. Puccinelli, A. D'Angelo, Italian Institute of Health ISS / Department of Environment and Health; S. Marchini, Italian Institute of Health ISS; R. Giuseppetti, E. Pierdominici, E. Volpi, Italian Institute of Health ISS / Department of Environment and Health; L. Mancini, Italian Institute of Health ISS

In industrial sectors, water is used in large amount in production cycles. In the field of Medical Devices (MDs) production, for example, water is used, both as an ingredient and as a necessary element for production. The re-use and the recycle of the wastewater represents a priority area in the strategical plan of the European Commission for a sustainable water management and also considering the scarcity of water resources caused by the climate changes. The aim of this study is the assessment of treated wastewater quality discharged from Medical Device and pharmaceutical industries, by identifying a suitable set of tools in order to support the potential re-use of treated wastewater, taking into account both microbiological and eco-/geno-toxicological parameters. The study was carried out on three MDs industries in Italy, which perform the recycle of the wastewater in their own system processes. Samples were collected inlet and outlet of wastewater treatment plants. The microbiological parameters investigated were Bacterial Total Count (BTC), E. coli, Enterococci, Staphylococcus spp., Pseudomonas spp. A set of ecotoxicological bioassays was selected for this study, namely the bacterium *Vibrio fischeri*, the algae *Selenastrum capricornutum*, the crustacean *Daphnia magna*, the fish *Danio rerio (Fish Embryo Toxicity Test)* and the plant *Sorghum saccharatum*, in order to represent different trophic levels and thus to assess any potential effects on the aquatic ecosystems. Seeds of *Vicia faba* were exposed for assessing the possible genotoxic effect of wastewater using the Micronucleus test (MN-test). The study results show a significant decrease in treated water samples of all microbiological parameters and the absence of E. coli. The ecotoxicological assays highlight a significant toxicity of the wastewater before the treatment while an evident decrease has been recorded after it. Sublethal effects for *Danio rerio* embryos and genotoxic effects for *Vicia faba's* micronucleus frequencies have been reported. Even though not yet conclusive, this approach can be considered a useful and promising tool in the reuse management of industrial wastewater and an initial support to the policy in this field.

WE082

Comparative effects of the azo dye Congo Red on the green microalgae Ankistrodesmus falcatus and Scenedesmus incrassatulus

A.A. Chávez-Vargas, Instituto Politécnico Nacional. Escuela Nacional de Ciencias Biológicas; M. Hernández Zamora, Escuela Nacional de Ciencias Biologicas-I.P.N / Laboratory of Experimental Hydrobiology; F. Martínez-Jerónimo, Escuela Nacional de Ciencias Biológicas-I.P.N. / Laboratory of Experimental Hydrobiology

Azo dyes account for up to 40% of commercially available colorants. Approximately 10–15% of the total production of azo dyes is released into the aquatic environment, affecting light diffusion, gases solubility -including oxygen concentration- and are toxic to aquatic biota. The azo dye Congo Red (CR) has several applications however it is most extensively employed by textile industry as a direct dye; hence it has become an important pollutant in water bodies. This study was aimed to evaluate the toxic effects of CR on the green microalgae *Ankistrodesmus falcatus* and *Scenedesmus incrassatulus*. The median inhibitory concentration (IC_{50}) was determined for each alga following the OECD guideline 201, using Bold's Basal Medium (BBM), at 25°C, under constant illumination, in static conditions; 4, 8, 16, 32 and 64 mg L^{-1} of CR were tested. Once IC_{50} values were determined, *A. falcatus* and *S. incrassatulus* were exposed to subinhibitory CR concentrations (IC_{20}, IC_{30}, IC_{40}, IC_{50} and IC_{60}) during 96 h, starting with cell densities of $5x10^4$ cell mL^{-1}, at 25°C, under continuous illumination and constant air bubbling. In this case, photosynthetic pigments, proteins, carbohydrates and lipids concentrations were determined. Results indicate that *S. incrassatulus* (IC_{50} = 9.27 mg L^{-1}) was more sensitive to CR than *A. falcatus* (IC_{50} = 19.25 mg L^{-1}). At subinhibitory concentrations, algae were more affected by higher concentrations of CR. In *A. falcatus* carbohydrates, carotenoids and chlorophyll-*a* contents significantly increased in treatments IC_{30} to IC_{60}, while lipids and chlorophyll-*b* contents significantly increased in IC_{40} to IC_{60} values. In *S. incrassatulus* only carotenoids content significantly increased when algae were exposed to the highest CR concentration (IC_{60}). Our results suggest that the sensitivity of primary producers to azo dyes could be species-specific; therefore the negative effects of CR discharge on water bodies may change depending on microalgal assortments in impacted environments.

WE083

Effluent ozonation treatment: effects on adult zebrafish fecundity, behavior and vitellogenesis in a 21 day exposure study

J. Pohl, Swedish University of Agricultural Sciences (SLU) / Department of Biomedical Sciences and Veterinary Public Health; S. Örn, G. Carlsson, Swedish University of Agricultural Sciences / Department of Biomedical Sciences and Veterinary Public Health; B. Björlenius, KTH Royal Institute of Technology / Industrial Biotechnology Division; J. Fick, Umea University / Department of Chemistry; J.D. Larsson, University of Gothenburg, Sweden / Department of Infectious Diseases; L. Norrgren, Swedish University of Agricultural Sciences / Department of Biomedical Sciences and Veterinary Public Health

Pharmaceutical residues in sewage effluents pollutes the aquatic environment and may adversely affect fish populations. Conventional sewage treatment plants (STP) are not equipped to efficiently remove pharmaceuticals. Ozonation is emerging as a method to improve sewage treatment. Ozonation can however potentially create toxic by-products (OBP) that may have deleterious effects on fish. In this study we screened the concentrations of 103 pharmaceuticals and biological effects at a Swedish STP with a full scale parallel ozonation line. The major aim was to investigate endocrine, reproductive and behavioral effects in fish and its progeny exposed to the conventional STP effluent. Furthermore, we sought to elucidate if any such effects were either abated or amplified by effluent ozonation (7 mg O_3 h^{-1}). We exposed zebrafish to dechlorinated tap water (n=3), STP effluent (n=3) and ozonated STP effluent (n=3). The fish were exposed during 21 days (12:12 day/night cycle) in continuously replenished tanks (0.1 L/min, 25°C). During the last seven days, the fecundity (number of eggs produced per female) was measured and fertilized eggs were gathered from each replicate. The eggs were kept in our laboratory facilities and checked for mortality, malformations and locomotor activity at 6 days post fertilization. At the termination of the experiment, we recorded adult fish swimming activity and liver tissues was sampled for subsequent mRNA extraction and expression analysis. Results from the chemical screening showed that on average 77% of the screened pharmaceuticals were removed by ozone treatment. However, on the contrary to our assumptions, the biological effect screening revealed male liver *VTG-2* gene expression, a marker of estrogenic endocrine disruption, was induced by the ozonated effluent. This indicates that ozonation possibly created estrogenic OBPs. Furthermore, the ozonated effluent treatment group exhibited a two-fold increase in fecundity. A suspected anxiety related behavioral phenotype was recorded in the adult fish exposed to the ozonated effluent. No adverse effects on the fish progeny was noted. Whether these biological effects would have an adverse impact on the population level remains speculative. Ozonation is a capable method for removing pharmaceutical residues from sewage effluents. Yet its implementation should be carefully monitored in order to minimize undesirable toxic side-effects.

WE084

Toxicity evaluation during secondary effluents treatment by UV/H2O2 using Eruca sativa and Artemia salina

J. Malvestiti, R.F. Dantas, University of Campinas / Technology

When advanced oxidation processes are applied there is the concern of not forming more toxic compounds as a result of the oxidation and transformation of organic compounds. Therefore, the presence of contaminants of industrial origin may affect disinfection and form more toxic by-products. Thus, a detailed study of by-products formation and toxicity assessment during the oxidation process contributes to a correct level of treatment. The composition of the effluents varies according to the characteristics of the region, time of year, etc. In the specific case of the city of Limeira SP, there is a high concentration of compounds of industrial origin and metals (Al, Fe, Zn, Cr, Ni, Cu and Pb) above that allowed by the Brazilian Legislation (CONAMA) in sewage due to the presence of many jewelry semi-jewelry industries. This work evaluated the toxicity of a secondary effluent, fortified with organic and inorganic compounds after an advanced oxidation treatment (UV/H_2O_2), through tests with arugula seeds (*Eruca sativa*) and *Artemia salina*. Samples of secondary effluent from a pilot plant located at the School of Technology campus (UNICAMP, São Paulo, Brazil) were collected immediately after the secondary treatment and then they were treated and analyzed in triplicate. The biological reactor was a hybrid: septic tank - anaerobic filter. After collection, 20-L bottles of the samples were transported to the laboratory and stored at 4ºC. Based on the preliminary tests it can be concluded that the secondary effluent treatment by UV/H_2O_2 in presence of high concentration of metals, nitrate, carbonate and industrial contaminants has no significant increase on toxicity.

WE085

Hospital effluent induced oxidative stress on Xenpus laevis larvae

I. Pérez-Alvarez, Universidad Autonoma del Estado de Mexico / Environmental Toxicology; H. Islas-Flores, Universidad Autonoma del Estado de Mexico / Toxicología Ambiental; L. Gómez-Oliván, Universidad Autonoma del Estado de Mexico / Farmacia; M. Galar-Martinez, Instituto Politécnico Nacional, Escuela Nacional de Ciencias Biológicas; N. SanJuan-Reyes, Autonomous University of the State of Mexico / Chemistry

Hospitals are one of the main sources of emerging pollutants to wastewater treatment plants (WWTP) that usually are fairly equipped to treat this kind of compounds. Activities performed in hospitals require the use of several compounds, which are potentially toxic, they can reach municipal wastewater, afterward they get into municipal waste water treatment plants, in some cases the procedure carried out in this WWTP is not able to remove all the contaminants, when they are not properly disposed, exposure to them can generate harmful effects on aquatic organisms. Physicochemical and pharmaceutical (11 pharmaceuticals) characterization of the hospital effluent were made, results shown a high concentration of mercury, and pharmaceuticals on concentrations of µg/L. Also oxidative stress was evaluated on *Xenopus laevis* larvae exposed to this hospital effluent; twenty oocytes were selected for each exposed group (control, 0.1, 0.3, 0.5, 0.7, 0.9 and 1%) in the middle blast stage, they were maintained at constant temperature 23 ± 2 ° C, for 96 hours until they reached the larval stage. They were weighted, homogenized and centrifugated for the determination of hidroperoxides, lipoperoxides, carbonilated protein content, and the antioxidant activity of superoxide dismutase and catalase, results shown statistically significative increments regarding control group in all the biomarkers evaluated, thus indicates that the hospital effluent tested in this work can generate oxidative stress on *Xenopus laevis* larvae, based on the results obtained, hospital effluents can generate oxidative stress in other species and due the lack of appropriate WWTP hospital effluents can represent a risk for aquatic organisms.

WE086

An assessment of (anti-)androgenic activity in sludge from a rain spillway basin of the WWTP Aachen Soers as well as in sediments from the catchment area of the recipient water, the river Wurm

K. Schröder, RWTH Aachen University; A. Shuliakevich, Institute for Environmental Research (RWTH Aachen University) / Institute for Environmental Research; Y. Müller, RWTH Aachen University / Institute of Environmental Research; S. Hotz, RWTH Aachen University / Institute for Environmental Research; S. Schiwy, RWTH Aachen University / Department of Ecosystem Analysis; H. Hollert, RWTH Aachen University / Institute for Environmental Research

Hormonally active micropollutants (MPs) are a well-known problem in aquatic environments. They can severely alter entire ecosystems by disrupting the endocrine system of its organisms. Adverse effects can extend to invertebrates and vertebrates including humans. As many MPs cannot be completely eliminated during conventional treatment in municipal wastewater treatment plants (WWTP), they enter the recipient waters. So far, mostly the effects of estrogenic active compounds have been investigated. However, a crucial part of the occurring hormone-equivalents is formed by androgen receptor inhibiting or activating compounds. Thus, the scientific attention has increased to account for their importance. Due to their strong lipophilicity, the main part of these compounds accumulates in sediments. A promising tool to reduce endocrine-disrupting MPs is the implementation of an additional treatment step like ozonation. Within the DemO$_3$AC Project, the WWTP in the catchment area of the river Wurm in Aachen Germany will be equipped with a large-scale ozonation. It is unknown how the concentrations of (anti-)androgens in sediments will be altered by this new treatment step. Up to now, controversial results were published regarding the elimination of (anti-)androgens by ozonation in effluents. Furthermore, sediments were rarely studied. To fill this scientific gap, this study will evaluate the status quo of (anti-)androgenic potentials in sediments and samples from the WWTP before the implementation of the ozonation at the WWTP in 2018. Investigation of samples from a rain spillway basin was conducted to measure the potential endocrine impact after a heavy rain event. Additionally, a sewage sludge sample was tested, to gain more information. Assessment of (anti-)androgenic activity was performed by testing sample extracts using the (anti-)AR-CALUX® assay. These studies will be conducted associated to the DemO$_3$AC Project as part of an exploratory study. First results revealed an anti-androgenic potential as well as cytotoxicity in the highest concentrations of the samples. The full depiction of the (anti-)androgenic activity in the catchment area of the Wurm will be available at the time of the conference.

WE087

Processes underlying the environmental fate of pharmaceuticals in the Nairobi River Basin "impact zone": implication for environmental risk assessment.

S. Bagnis, M. Fitzsimons, A. Tappin, Plymouth University; J. Snape, AstraZeneca UK Ltd. / AstraZeneca Global Environment; A. Gachanja, Jomo Kenyatta University of Agriculture and Technology / Chemistry; S. Comber, Plymouth University / Environmental Science

Recent investigations have highlighted the widely-spread occurrence of active pharmaceutical ingredients (APIs) in African water bodies. Wastewater (treated or otherwise) is the main source of APIs to the environment and identified heavily contaminated areas have been ascribed to the poor African wastewater treatment facilities (WWT). The formation of so called “impact zones” is severe in urban areas characterized by informal settlements with little in the way of wastewater treatment. The Nairobi River basin in Kenya is a an example of such contaminated areas. The wastewater generated from the city’s informal settlements and the insufficient WWT is directly discharged in the Nairobi River basin leading to a large-scale “impact zone” characterized by occurrence of high levels of ammonia, biochemical oxygen demand and low dissolved oxygen and commensurately elevated levels of APIs. Also, the presence in this area of industrial wastewater discharges from pharmaceutical factories also contributes to the occurrence of APIs. Hitherto, the published investigations provided evidence of APIs occurrence at concerning concentrations, but little is known about the processes underlying the E-fate of such contaminants. Processes such as distribution to suspended solids, colloids and dissolved organic matter, the quality of these sorbents, and the ability of the biodegrading bacteriological community to adapt to and degrade a contaminant are key in determining its E-fate. The impact zone generated by sewage and industrial wastewater together with the local natural sorbents abundance influences the APIs distribution and E-fate. Data will be presented on sorbent concentration and quality related to distribution processes of APIs in the impact zone and the individuation of eventual bacteriological community shifts as an effect of the direct discharge of untreated wastewater loaded with APIs. In addition, since preliminary studies on the biodegradation of the antiretroviral nevirapine, commonly used in Africa, has showed persistency (similarly to studies on the antiepileptic carbamazepine in Western countries) the occurrence of nevirapine will be studied to test its suitability as indicator of sewage pollution in African river bodies. The information obtained from this study contributes to the development of a methodology for the determination of the impact zone spatial boundaries and a dedicated environmental risk assessment approach for APIs in the impact zone.

WE088

Occurrence of pharmaceuticals, metabolites and transformation products from combined sewer overflows in London measured by high resolution targeted, suspect screening and untargeted chemical analysis

L. Barron, Kings College London / Analytical and Environmental Science; K. Munro, Kings College London; T.H. Miller, Kings College London / Analytical and Environmental Sciences; D.A. Cowan, Kings College London / Drug Control Centre; C. Martins, Thermo Fisher Scientific; J. Pereira, University of Aveiro / Department of Biology Centre for Environmental and Marine Studies CESAM

Combined sewer overflows (CSOs) are controlled releases of raw, untreated wastewater to a river during times of heavy rainfall to avoid back-flushing of buildings and streets. The impact of CSOs on a river catchment with respect to pharmaceutical residues is not well understood. In London, CSOs occur ~1-2 times per week as its Victorian sewage network struggles to cope. Herein, a temporal study of the River Thames is presented to identify CSO-related occurrence of pharmaceuticals, including metabolites and transformation products. Daily samples of river water, influent and effluent wastewater were analysed using a validated method involving solid phase extraction (SPE) and liquid chromatography and high resolution accurate mass spectrometry (LC-HRMS). The work was divided into four parts: (a) the identification of CSO markers based on quantitative differentiation of 30 pharmaceutical/metabolite occurrences in influent and effluent wastewater; (b) determination of CSO markers in receiving river water over a six-week period; (c) suspect screening to identify metabolites/transformation products; and (d) classification of samples using untargeted data analysis. By differentiating influent and effluent, four CSO markers were identified including caffeine, bezafibrate, benzoylecgonine and furosemide which were present only in influent at relatively high/consistent concentrations. Following this, targeted analysis of the River Thames samples revealed that CSO marker concentration increased mainly during wet periods where the tide was low. A further 14 compounds were also determined to observe any ‘dilution effects’ related to CSO influx and this was evident for tramadol and carbamazepine. The potential occurrence of known metabolites/transformation products is also presented for a selection of compounds via machine learning prediction of LC retention times and mining of HRMS data [1]. Finally, untargeted analysis revealed that river samples could be differentiated based on climate and/or tide height using principal component analysis and volcano plots. The use of several different modes of data analysis captured using the same instrumental method facilitated a deeper understanding of complex occurrence data potentially influenced by timed CSO events. [1] K Munro, TH Miller, CPB Martins, AM Edge, DA Cowan, LP Barron, *J. Chromatogr. A*, 1396 (2015) 34–44

WE089

Occurrence, fate and bioactivity of pesticides in wastewater

V.V. Yargeau, McGill University / Chemical Enineering; P. Westlund, D. Nasuhoglu, S. Isazadeh, McGill University

Unlike for contaminants of emerging concern (CECs), data available for the occurrence, fate and bioactivity of pesticides (herbicides, fungicides, and insecticides) in wastewater treatment plants (WWTPs) is limited. Our research showed that of the 18 compounds investigated only imidacloprid, was not detected at the three WWTPs included in the study, confirming that municipal wastewater discharges contribute to the presence of pesticides in the aquatic environment. Using a suite of bioassays (high-throughput bioluminescence assay using the target species *Vibrio fischeri*, yeast estrogenic screen (YES) and yeast androgenic screen (YAS) assays) the bioactivities of the pesticides as well as wastewater effluents treated using secondary treatment or ozonation was investigated. It was found that of the 12 pesticides showed either antiestrogenic or antiandrogenic activity and 7 compounds showed pleiotropic effects. This study was first to confirm endocrine activities based on yeast-based assays of recent neonicotinoids. The use of extended time points for the *Vibrio fischeri*, beyond the traditional 30 minutes, highlighted that toxicity for some compounds was underestimated using only the acute test. Using a structure-activity relationship approach similar to the one used in hazard assessments, the relationship between toxicity and key physicochemical properties of the pesticides was investigated and trends were identified. This work also provided new knowledge on the removal of some fungicides (climbazole, myclobutanil and tebuconazole) by ozonation and demonstrated the recalcitrant nature of pesticides during ozonation. This study is one of the first to investigate androgenic activity during the ozonation of a mixture of pesticides and an increase was reported. These findings further demonstrate the importance of combining bioanalytical tools to analytical chemistry in the evaluation of wastewater quality and treatment performance.

WE090

Fate of perfluoroalkyl substances within a small stream food web affected by sewage effluent

D. Cerveny, University of South Bohemia in Ceske Budejovice / Laboratory of Environmental Chemistry and Biochemistry; R. Grabic, University of South Bohemia in Ceske Budejovice / South Bohemian Research Center of Aquaculture and Biodiversity of Hydrocenoses; G. Fedorova, University of South Bohemia in Ceske Budejovice / Laboratory of Environmental Chemistry and Biochemistry; K. Grabicova, University of South Bohemia in Ceske Budejovice / Faculty of Fisheries and Protection of Waters; J. Turek, University of South Bohemia in Ceske Budejovice / Laboratory of Environmental Chemistry and Biochemistry; V. Zlabek, University of South Bohemia in Ceske Budejovice / Faculty of Fisheries and Protection of Waters, LECHB; T. Randak, University of South Bohemia in Ceske Budejovice / Laboratory of Environmental Chemistry and Biochemistry

Within our experiment, the fate of fourteen PFASs was studied in an ecosystem of a small stream affected by STP´s effluent. The unique field experiment design was carried out to allow long-term study focused on bioaccumulation of PFASs in indicator organisms and their ability of adaptation to the polluted environment. Two hundred brown trouts (*Salmo trutta*) originating from clean site within the same stream were tagged and stocked downstream the source of pollution. Those fish were recaptured after one, three, and six months they spent in the environment affected by the effluent of that local STP. Besides the fish stocked into the polluted locality from the clean site, also fish originally inhabiting the downstream locality and macroinvertebrates from both sites were sampled and analysed. Passive sampling approach using polar organic chemical integrative samplers (POCIS) was applied to determine occurrence of PFAS in water soluble fraction over the course of the experiment instead of conventional grab water samples. Twelve of the fourteen target PFASs were found in concentration above the LOQ in at least one of the studied matrices from downstream locality while only three were present in samples from clean site. The compound pattern varied significantly between fish, species of macroinvertebrates, and POCIS indicating several exposure pathways leading to bioaccumulation of PFASs in fish body. Concerning the accumulation of PFASs in fish, the highest concentrations were found in the liver of individuals sampled after three months of exposure. These concentrations rapidly decreased after six months although there was no significant change in occurrence of PFASs in water during the experiment. Such finding can be linked to both increase of water temperature leading to higher enzymatic activity and adaptation of studied fish to the polluted environment. Based on our results we also suggest that the process of adaptation might be related to the gender of fish as we found significantly higher accumulation of target compounds in males during the first month of exposure.
Acknowledgements: The study was financially supported by the Ministry of Education, Youth and Sports of the Czech Republic - projects “CENAKVA” (No. CZ.1.05/2.1.00/01.0024) and “CENAKVA II” (No. LO1205 under the NPU I program) and by the Ministry of Agriculture of the Czech Republic (NAZV “KUS” No. QJ1530120).

WE091

Patterns of natural and human-made interacting processes on source, transport and fate of trace metals in the Adriatic Sea basin

M. Rocha, University of Bologna; T. Combi, Instituto Oceanográfico da Universidade de São Paulo / Instituto Oceanográfico; L. Langone, National Research Council of Italy CNR / Institute of Marine Sciences National Research Council ISMARCNR; R. Guerra, University of Bologna / Environmental Sciences; S. Miserocchi, National Research Council of Italy CNR / Institute of Marine Sciences National Research Council ISMARCNR; P. Giordano, ISMAR CNR

The Adriatic Sea has been under intensive influences of human activities, which are pressuring the marine ecosystems as a whole. The morphology of this land-locked marginal sea and the intensity of human pressure on its coastline influence enhances trace metal concentrations on sediments giving rise to pollution effects especially in the northern Adriatic Sea. This work was developed under the PERSEUS EU Project (Policy-oriented marine Environmental research in the Southern European Seas), guided by the Marine Strategy Framework Directive (MSFD), which aims to achieve a Good Environmental Status until 2020 in European water bodies. Spatial and historical trends of sedimentary trace metals (Zn, Pb, Cu, Cr and Ni) and their characteristics (e.g., grain size and organic carbon) were assessed in recent layers and dated cores from western Adriatic Sea to reconstruct their sources, transport and accumulation. Our findings suggest that the Po River prodelta acts both as a bypassing and accumulation zone and exports ~30% of trace metals associated with fine particles southward, being mainly accumulated in the coastal mud wedge of the Central Adriatic. Based on the outcomes of this study, the area in the Po River vicinity could be considered an area of concern especially related to Zn and Pb accumulation. According to the requirements of the Marine Strategy Framework Directive (MSFD), we proposed a long-term monitoring plan, with a pluriannual temporal frequency (e.g., 5 years), suitable to point out possible changes in metal accumulation in the western Adriatic Sea. The anthropogenic signal of Pb and Zn can be recognized in sediment cores from the northern down to the Gargano Promontory, ~500 km away from the metal sources, with a delay of ~10 years. In line with many systems around the world, we observed a recent decrease in trace metal excess and concentration in the sediment cores from the Adige prodelta down to the Gargano, which has been mainly related to the environmental regulations enforced by governments. Finally, the main transfer process of trace metals from coastal waters to the open sea is attributed to the cascading of the North Adriatic Dense Water (NAdDW) in deep sea areas of the southern Adriatic, which would be able to quickly transfer suspended sediments (and, therefore, particle-binding contaminants) during episodic events and supports the inference that this region may act as the final repository for contaminants within the Adriatic Sea.

WE092

Photocatalysis as a potential pre-treatment process to reduce organic pesticide entries

F. Seitz, Inst. for Environmental Sciences / Institute for Environmental Sciences; R. Rosenfeldt, Uniersity of Koblenz-Landau, Institute for Environmental Sciences / Institute for Environmental Sciences; P. Garcia Munoz, N. Keller, D. Robert, V. Keller-Spitzer, Université de Strasbourg / Institut de Chimie et Procédés pour lEnergie, lEnvironnement , et la Santé (ICPEES), CNRS; B. Altmayer, M. Twertek, State Education and Research Center of Viticulture, Horticulture and Rural Development / Institute of Plant Protection; S. Lüderwald, Universität Koblenz-Landau / Institute for Environmental Sciences; R. Schulz, University of Koblenz-Landau / Institute for Environmental Sciences

For protecting their crops many wine growers apply plant protection products (PPPs), which may accidentally end up in the aquatic environment e.g. after being washed off from agricultural machinery (AM). Once there, PPPs can negatively impact aquatic life which provides important ecosystem services. This is contrary to major goals of the EU Water Framework Directive, aiming to increase the quality of surface waterbodies. Hence, tools to counteract this difficulty are needed. A targeted application of photocatalysis as pre-treatment is one promising approach to reduce PPP loads directly in wash waters from AM. Thereby, PPP concentrations and associated toxicity can be reduced prior entering aquatic ecosystems via wastewater treatment plant effluents. The present study evaluated the applicability and efficiency of different commercial TiO_2 photocatalysts (Aeroxide P25 and Hombikat UV100) by separately and simultaneously treating five different PPPs in aqueous solution under artificial UV irradiation (UVA: 40 W/m^2 for 60 min). The pesticides were chosen as representatives, being frequently used for viticulture in the Trinational Upper Rhine Area, a region where vine growing is one major form of agriculture. To evaluate product dependent efficiencies of TiO_2 based photocatalysis, treated and untreated PPPs were analyzed for remaining PPP concentrations and major metabolites before and after a combined $TiO_2 \times$ UV treatment. Therefore, UV-visible absorbance spectroscopy, Total Organic Carbon (TOC) measurement and liquid chromatography was used. Further, to assess for the photocatalytic efficiency of both TiO_2 products in terms of environmental toxicity, acute toxicity tests were conducted. In detail, *Daphnia magna* was exposed for 48 h to (un-)treated PPPs according to the OECD guideline 202. Gained immobility data was statistically analyzed to detect significant differences among photocatalytic treatments. Preliminary results of both, analytical and ecotoxicological investigations, show the suitability of TiO_2 to reduce PPP concentrations and associated toxicity in water when being irradiated by UV light. Further, a product related difference in the degradation potential of TiO_2 for the selected PPPs was observed. However, for a final statement whether TiO_2 can be used for an efficient photocatalytic pre-treatment of wash water from AM, further studies considering different environmental conditions – potentially hindering this process – are urgently needed.

WE093

Study of the efficiency of removal of organic load and generation of energy through a bioelectrochemical system coupled to a constructed wetland

J.H. Cardenas, Centro de Investigación y Desarrollo Tecnológico en Electroquímica / Microbiologia Ambiental; M.Y. Reyes Vidal, B. Lázaro Rodriguez, Centro de Investigación y Desarrollo Tecnológico en Electroquímica / Microbiología Ambiental y Energética; I. Robles Gutierréz, Centro de Investigación y Desarrollo Tecnológico en Electroquímica / Electrpquímica Ambiental; F. Espejel Ayala, Centro de Investigación y Desarrollo Tecnológico en Electroquímica / Electroquímica Ambiental

Bioelectrochemical systems are alternative options for energy generation through the degradation of industrial and domestic effluents using electroactive microorganisms, these systems coupled to build wetlands (BES/CW) acquire the capacity to treatment effluents of various kinds. Although several studies have been developed to evaluate the efficiency of these systems for chemical oxygen demand (COD) removal and energy generation. To this aim, it was implemented two plastic containers with a volume of 20 L and an operating volume of 0.96 L, packed with river gravel and planted with phragmites sp., carbon felt was used for the construction of anode and cathode, both with a surface of 0.72 cm^2. The distance between both electrodes was 6 cm without proton exchange membrane, both electrodes were connected by a resistance of 1000 Ohms. The feeding was performed by gravity applying four pulses of 1.5 L/h, using synthetic water whose composition was similar to date reported by Yadav et al., (2012). The effect of two types of contaminants was evaluated azo dye (AD) and alkylphenols (AP), the voltage measurements were made daily after 20 days of operation, the time necessary for the development of the biofilm (from 9:00 h in intervals of 2h). The

COD measurement was performed weekly on the influent and effluent of both systems. After 40 days in operation, the systems showed voltage average values of 673 and 580 mV, maximum current densities of 20.8 and 37.5 mAm^{-2} and COD removal of 38.5 and 36.71% for effluents AZ and AP, respectively. A significant increase in the current density was observed in the measurements taken after 13:00 h, which shows an effect of temperature on the generation of voltage and therefore current flow in the system. The results obtained represent a sustainable option for the generation of energy from domestic waters from secondary effluents, which would not only improve the quality of the water before being discharged to the receiving bodies but also take advantage of the high concentrations of nutrients contained in these wastes.

WE094

Adsorption of Crystal Violet from Quaternary Basic Dye Mixture onto A Sawdust-Based Adsorbent

A.A. Giwa, Cape Peninsula University of Technology / Department of Pure and Applied Chemistry; K.A. Abdulsalam, Adeleke University, Ede. Nigeria / 1 Department of Basic Sciences, Chemistry Unit; F. Wewers, Cape Peninsula University of Technology / Chemistry; I.A. Bello, Ladoke Akintola University of Technology / Department of Pure and Applied Chemistry

Wastewaters from textile and other dye-using industries usually contain more than one dye. In such an aqueous matrix therefore, the adsorptive characteristics of a dye species are likely to be affected by the presence of the other dye species in the medium. The removal of crystal violet from aqueous solution by sulphuric acid-treated sawdust of locust bean tree, and the influence of malachite green, methylene blue and rhodamine B on its adsorption from binary, ternary, and quaternary dye systems were studied. The combined effect of mixture components and process parameters on the adsorption was studied and optimized using response surface methodology. The adsorbent was characterized and the experimental data obtained were fitted to different kinetics and isotherm models. The experimental results were analyzed using the Analysis of variance (ANOVA) statistical concept. The optimum contact time, pH, adsorbent dose and temperature were found to be 275.10 min, 9.94, 0.99 g and 60 ºC respectively for the maximum decolorisation of 68.39 mg/L CV (97.2%). A linear model was obtained for the decolorization process through this design. The experimental values obtained were in good agreement with predicted values, and the model developed was highly significant, with correlation coefficient of 0.985. The adsorption in all the dye systems investigated followed Freundlich isotherm, and the maximum monolayer adsorption capacity was between 18.87 - 24.39 mg/g, depending on the composition of the adsorbate matrix. The adsorption kinetics was well described by the pseudo-second order model ($R^2 > 0.95$). All the eight adsorbate systems investigated were endothermic (?H positive; 35.30 to 43.60 $kJmol^{-1}$), thermodynamically feasible (?G: -2.30 to -6.13 $KJmol^{-1}$) and had increased entropy.

WE095

Diurnal patterns and removal of selected elements in two Norwegian wastewater treatment plants with primary treatment

J. Farkas, SINTEF Ocean / Environmental Technology; F. Polesel, Technical University of Denmark (DTU) / DTU Environment; M. Kjos, P.A. Carvalho, SINTEF Materials and Chemistry; A. Booth, SINTEF Ocean / Environmental Technology

Discharges from wastewater treatment plants (WWTPs) are a source for pollutants to the environment. Here we studied influent patterns and removal of selected elements in two full-scale WWTPs, Ladehammeren (LARA) and Høvringen (HØRA) in Trondheim, Norway. Both WWTPs have significant industrial loading contributions (up to 40% in LARA), employ preliminary and primary treatment steps, including chemically aided flocculation ($ClFeO_4S$/polyamine in LARA, polyacrylamide in HØRA), and discharge directly into Trondheimsfjord. In a 7 day sampling campaign, 24 h composite samples of influent and effluent wastewater, as well as sludge samples, were taken to determine influent concentrations and removal of Al, P, S, Cr, Fe, Ni, Cu, Zn, As, Cd and Pd. To study release patterns and gain information about potential sources, diurnal variations of elemental concentrations were determined and modelled in 8 h composite samples of raw influent wastewater from morning, evening and night discharges. Element concentrations in 24 h composite influent samples were highest for S>P and Al and lowest for Cd< As< Cr and Pd. Concentrations of Al, P, Cr, Cu and Cd were higher in HØRA than LARA, with Fe loadings being approximately double. Removal efficiencies varied between the analysed elements, and were highest for Al (86%), P (74%) and Cu (57%) in LARA, which utilises both inorganic and organic flocculants. In contrast, removal rates were below < 50% for P, Cu and S in HØRA. However, in LARA, concentrations of Fe, Ni and S were significantly higher in the treated effluent compared to the raw influent, deriving from the use of inorganic flocculant. This was also reflected in Fe and S concentrations in treated sludge. Elemental concentrations in 8 h composite samples mostly followed general diurnal discharge patterns, with higher concentrations in mornings and evenings and lower concentrations at night. In HØRA, concentrations of most elements further correlated well with total suspended solid concentrations (TSS), with the strongest correlations observed for P, S and Cu (R^2>0.9). Correlations with TSS were less pronounced in LARA, and were weakest for Pb, Fe and Cu (R^2< 0.6), which can be potentially attributed to the higher industrial loading contributions in LARA. Enrichment factors were high for P>Cu>Zn>Cd>As, and were still above 10 for Cr and Ni in biosolids, indicating anthropogenic sources for these elements. Several elements also occurred as nano- and micron-sized particles.

WE096

Rapid detection of E. coli in wastewater effluent and impact of effluent discharge on riparian invertebrate diversity

P.M. Mosolloane, University of the Free State / Zoology and Entomology

Effluent samples from wastewater treatment plants are expected, due to the treatment processes, to have lower concentrations of toxicants and pathogens than the influent. As such, they are expected to have a lesser impact on the environment than the influent. The aim of this study was to seasonally assess the efficiency of Phuthaditjhaba's wastewater treatment plant in removing pathogens (*E.coli*) from its effluent and to determine the impacts of that effluent on invertebrate diversity along the riparian zone of the receiving watershed. Loop mediated isothermal amplification technique was used to screen for the presence of *E.coli* in effluent samples. There was negative identification of *E.coli* in the samples collected. Impacts of effluent on invertebrates will be determined by analysing the changes in population dynamics of the invertebrates found in the riparian zone, above and below discharge point of the Wastewater Treatment Plant. At least, 13 morph species of eight insect families have been identified. Taxon diversity will be calculated using the Simpson-Yule Index, from where significant difference analysis would be calculated for environmental variable, species abundances and diversity data. More screening for the presence of *E. coli* will determine the efficiency of Phuthaditjhaba wastewater treatment plant in removing pathogens from its effluent. Further identification and presence or absence of invertebrates in the sampled riparian zone will determine if effluent has any impacts on invertebrate diversity.

WE097

The DemO3AC-project: Chemical and ecotoxicological investigations of the wastewater treatment plant Aachen

S. Schiwy, Institute of Environmental Research-RWTH Aachen / Department of Ecosystem Analysis; Y. Müller, RWTH Aachen University / Institute for Environmental Research; A. Shuliakevich, Institute for Environmental Research (RWTH Aachen University) / Institute for Environmental Research; S. Könemann, Institute for Environmental Research RWTH; S. Oster, RWTH Aachen University, Institute for Environmental Research; K. Klaer, R. Dolny, Institut of Environmental Engineering, RWTH Aachen; S. Classen, Research Institute gaiac; M. Hammers-Wirtz, gaiac Research Institute for Ecosystem Analysis and Assessment Aachen; I. Brückner, Eifel-Rur Waterboard; J. Pinnekamp, RWTH Aachen University / Environmental Engineering; H. Hollert, RWTH Aachen University / Institute for Environmental Research

Micropollutants (MPs) in municipal wastewater, like pharmaceuticals, are not sufficiently removed by conventional cleaning methods in wastewater treatment plants (WWTPs). As a consequence, complex mixtures of MPs discharged into the receiving streams and may cause various effects on the aquatic biota. To decrease the release of MPs into the environment, a full-scale ozonation is implemented into the WWTP Aachen Soers, Germany within the $DemO_3AC$-project. A crucial part of this project is the evaluation of the ecological and chemical state of the receiving stream, the River Wurm. This study focuses on the *status quo* of this river. To assess the impact of MPs, water samples were taken at 3 treatment steps within the WWTP and at 4 sampling sites (upstream and downstream the WWTP) along the river. These samples were tested as both native samples and extracts. To include various adverse effects a broad test battery was conducted (acute, chronic, mechanism-specific, *in vivo* and *in situ*) along with chemical analysis. The analysis of 60 MPs showed mainly the presence of pharmaceuticals and plasticisers/phthalates. In total 52 substances could be detected. A general elimination rate of about 55 % was determined. The toxic potential was high in the WWTP inlet for *D. magna, D. subspicatus* and *D. rerio*. *A. fisheri* showed no effect. However, the toxicity was already markedly reduced at the outlet of secondary clarifiers within the WWTP. No acute and chronic toxicity was detected in any of the Wurm samples. Similar results were obtained in *in situ* feeding experiments with *G. pulex*. No significant differences in feeding rate between the sampling sites were recorded. On the other hand, significant mutagenic and endocrine effects were observed at the inlet and outlet of the WWTP as well as at all sampling sites within the Wurm. The *P. antipodarum* reproduction assay showed also a significant increase in embryo production downstream of the WWTP. In parallel, experiments with river sediments and samples of an additional WWTP (upstream of the WWTP Soers) will be conducted to clarify to which amount the mutagenic and endocrine effects originates by different sources as the WWTP Soers. The second part of this project will contain comparative studies investigating the situation after the implementation of the full-scale ozonation. This project is funded by the Ministry for Climate Protection, Environment, Agriculture, Conservation and Consumer Protection of North Rhine-Westphalia.

WE098

To use or not to use: sewage overflow dredgings

M.H. Wagelmans, Bioclear earth

In a densely populated country like the Netherlands, with a dense sewage system, many sewage overflows into surface water are present. Sediment located 250 m before and after the overflow needs to be discarded and burnt after dredging while 'normal' sediment can be reused as soil. Discard and burning is expensive for water boards, besides the fact that it costs a lot of enrgy and causes air pollution due to transportation, which is not sustainable. Also it causes a loss of raw materials. The water board Noorderzijlvest has started a pilot for reusing sewage overflow dredgings as new soil, for instance for cnstruction puposes or agriculture. For this purpose two depots have been set up: one with reference sediment and one with overflow sediment. During one year the concentrations of pharmaceuticals and pathogens will be monitored as well as anitiotic resistance and endocrine disruption. The basis of the project is the hypothesis that pathogens will be killed during the process of drying and pharmaceuticals will be (biologically) degraded which makes resuse of the dredgings possible. In June 2017 the sediments were sampled and analysed. In October dredging was performed and the pilots have been set up. The first monitoring round was performed one week after set up. In each monitoring round chemical analyses are performed on pharmaceuticals. Pathogens are identified by both culturing methods and Next Generation Sequencing (NGS), combined with viable PCR analyses to quantify specific pathogens that have been identified by NGS. Antibiotic resistance will be monitored by menas of ESBL (extended Spectrum Beta-Lactamase) measurements. Endocrine disruption will be monitored by means of ER-Calux tests. In the presentation the background of the project will be sketched. then the results of the monitoring up till May 2018 will be presented. Also preliminary conclusions will be drawn and an outlook for resue possibilities will be given as well as the meaning of the project for other water boards.

WE099

Assessing wastewater processes at oil refinery industry in Kazakhstan

I. Radelyuk, Lund University / Department of Building and Environmental Technology; K. Tussupova, LTH, Lund University / Department of Building and Environmental Technology

This paper presents the first attempt to assess the wastewater treatment processes at the oil refinery sector in Kazakhstan and evaluate to what extent these processes follow international and national regulations regarding industrial waste water treatment. The assessment was performed considering wastewater discharge from refining processes at three factories in the country. While Kazakhstani environmental regulation promote the polluter pay principle and follow WHO guidelines, oil refinery factories in Kazakhstan still exceed the permissible concentration of pollutants in discharged wastewater. The national regulation allows discharge of wastewater to natural or artificial ponds by not exceeding the pollutant concentrations already existing in the pond. Therefore, the factories use ponds with already high concentration of pollutants, consequently allowing discharge of high concentration of pollutants (total petroleum hydrocarbons (TPH) exceeds concentration by 30-80 times, ammonia (NH_4^+) by 25 times, total dissolved solids (TDS) by 6 times, biochemical oxygen demand (BOD) by 6 times and surfactants by 5 times) to pond. The reason for the initial high pond concentration is a result of a time gap between the start of pollution discharge by the factories and start of the environmental regulations. This leads to no incentive to treat wastewater in an efficient way. Additionally, the national law lacks regulations regarding detailed methodology to assess the pollutant substances in the discharged wastewater. Thus, the assessment by environmental authorities for each oil refinery is negotiated separately between the factory and the governmental body, giving the factory a strong position to define the parameters assessing the wastewater. As such, none of the factories provides analyses of, e.g., heavy metal contamination in discharged wastewater. TPH concentration in wastewater is often exceeded at each factory and there is no analysis done for different hydrocarbon fraction. Consequently, it is strongly recommended to provide a unified and transparent methodology for the country´s oil refinery industry to assess all important pollutants in discharged wastewater and to include all types of hydrocarbon fractions.

Antibiotics and Antibiotic Resistance in the Environment: Fate and Ecological Effects, Resistance Development and Implications for Human Health (P)

WE100

Accumulation of Enrofloxacin in the sea lettuce Ulva lactuca

J. Rosa, University of Coimbra / Department of Life Sciences; S. Leston, CFE-Center For Functional Ecology / Department of Life Sciences University of Coimbra; A. Freitas, J. Barbosa, INIAV- Instituto Nacional de Investigação Agrária e Veterinária; M.F. Lemos, Instituto Politécnico de Leiria / MARE IPLeiria; M.. Pardal, CFE   Centre for Functional Ecology / Department of Life Sciences University of Coimbra; F. Ramos, Faculty of Pharmacy University of Coimbra

The demand for food products is pushing aquaculture to increase its production throughout the world. The increase in production can lead to negative effects since much more fish are growing in much smaller places. Aquaculture is still highly associated with the frequent use of chemical compounds in water, either to treat or prevent disease outbreaks in culture ponds. Integrated multitrophic aquaculture systems (IMTAs) can be a suitable approach to fish production, since one can have several species with different trophic levels growing together, where each species have its own economical value. Macroalgae can be used in such systems, usually at the exit point of the ponds, acting as biofilters and reducing the nutrient loading released to the environment. Contaminants such as antibiotics used in aquaculture can bioaccumulate in these organisms, passing to higher trophic levels. More recently, with the inclusion of macroalgae in human diets not only in Asia but also in other regions, we can ultimately ingest high levels of these contaminants, which are not legislated the same way as other fish products. Exposure tests were performed with the macroalgae *Ulva lactuca* in order to evaluate the effects of Enrofloxacin in growth. Antibiotic concentrations were measured in seawater and macroalgae discs at several sampling points, after immersion in an Enrofloxacin bath at two different concentrations. These results can help comprehend how IMTA systems should work in order to prevent contamination with antibiotics. As biofilters, these organisms are located at the exit point of fishponds or near cages, potentially accumulating pharmaceuticals.

WE101

Antibiotic resistance genes in manure, stored manure and soil after manure application

M. Virta, K. Pärnänen, University of Helsinki; R.D. Stedtfeld, J.M. Tiedje, Michigan State University / Center for Microbial Ecology Department of Civil and Environmental Engineering; J. Muurinen, University of Helsinki / Food and Environmental Sciences

Production animal farms are proposed to act as reservoirs where genetic material from environmental bacteria might transfer to human- or animal-associated bacteria including zoonotic pathogens. In the present study we followed two swine and two dairy farms and quantified ARGs amd MGEs with 363 primer pairs using a high-throughput qPCR array. Samples were taken from fresh manure, stored manure, unfertilized soil, soil fertilized with manure, and tile drainage water from ditch, which was sampled before and after land application. We aimed to answer the following questions: do ARGs disseminate to the environment (field soil and surface waters), are ARGs enriched in stored manure, and are ARG abundances elevated in soils at crop harvesting time. For evaluating the ARG mobility potential, genes related to MGEs were also quantified. DNA was isolated from the soil and manure samples with the PowerSoil DNA Isolation Kit (MO BIO Laboratories). Quantitative PCR reactions were conducted using WaferGen SmartChip Real-time PCR system. The ΔCt values, ΔΔCt values, relative gene abundances (R), and fold changes (FC) were calculated with R version 3.2.3 and RStudio Version 0.98. In total 182 out of 363 ARG and MGE qPCR assays were positive in one or more samples. Out of the positive assays, 161 targeted ARGs and 21 MGEs. Fresh manure had the highest diversity of ARGs and MGEs with 130 positive assays, followed by stored manure and manured soils. The number of positive assays decreased in fertilized soil between the 2 and the 6 week sampling points. Only 29 assays were positive in unfertilized soil samples. Manure had the highest relative abundances of ARGs, and these manure-associated ARGs were not detected in unfertilized soil or ditch water sampled before fertilization. Likewise, ARGs abundant in unfertilized soil or in ditch water were not abundant or even detected in manure. After fertilization, the manure-associated ARGs and MGEs were present in soil, hence a consequence of fertilization. However, the abundance and number of these ARGs and MGEs clearly decreased from fertilized soil to fertilized soil samples taken 2 and 6 weeks after manure application. During manure storage the relative abundance increased more than 4-fold for 41 genes and more than 2-fold for 62 genes. The highest increase (up to 65-fold) was observed in tetracycline-resistance genes, followed by sulphonamide and aminoglycoside resistance genes with up to 45-fold and 41-fold increases.

WE102

Bioaccumulation, biochemical responses and gene expression in the marine clam Scrobicularia plana exposed to a pharmaceutical mixture at sub-lethal concentrations

C. Trombini, CSIC   Spanish National Research Council ICMAN / ECOLOGY AND COASTAL MANAGEMENT; M. Hampel, INMAR   University of Cadiz; J. Kazakova, R. Fernandez-Torres, M. Bello-López, University of Seville; J. Blasco Moreno, CSIC   Spanish National Research Council ICMAN / ECOLOGY AND COASTAL MANAGEMENT

Pharmaceuticals are pollutants of potential concern in the aquatic environment where they are commonly introduced as complex mixtures as a result of incomplete waste water treatment plant removal processes and improper disposal. Despite of pharmaceuticals occurring in the aquatic environments at trace levels (ng L^{-1} to low µg L^{-1}), they have been specifically designed to be biologically active at low concentrations in human and animals. Therefore it is reasonable to expect that aquatic wildlife may also be susceptible to their effects particularly under condition of combined and chronic exposure. Hence the need to characterize biological effects in non-target organisms exposed to sub-lethal concentrations of pharmaceutical mixtures. Ibuprofen (IBU) is one of the most used non-steroidal anti-inflammatory drugs: its ability to induce toxic effects (i.e. oxidative stress, neurotoxicity, endocrine disruption, immunological alterations) in aquatic organisms at environmentally relevant concentrationsis has been widely proven. Ciprofloxacin (CIP) and flumequine (FL) are broad-spectrum antibiotics of the fluoroquinolones class. Fluoroquinolones toxicity was observed in rodents producing among others oxidative stress, cyto, neuro and hepatotoxicity were induced after treatment with CIP in mice and rats. However, ecotoxicological effects on aquatic organisms of CIP and FL are practically unknown. In our study specimens of the clam *Scrobicularia plana* were exposed to control water and a mixture of IBU, CIP and FL at close environmentally relevant concentrations (10 y 100 µg/L each) during 21 days with the aim of studying toxicological reponses along the time. Recovery of organisms also was assessed after 1 week of post-exposure depuration. Bioaccumulation of pharmaceuticals in clams was examined and changes in a suite of molecular biomarkers was used to evaluate the biochemical status of clams during both exposure and depuration: biochemical responses related to oxidative stress (CAT, SOD, GR, T-GPx activities and LPO levels), detoxification (GST activity) and neurotoxicity (AchE activity), and mRNA expression of genes associated with regulatory system of xenobiotic exposure. Results obtained indicate a general activation of oxidative stress and neurotoxicity related features in enzymatic responses as well as changes in genetic profiles suggesting that selected pharmaceutical mixture in aquatic environment represent a risk for the clam *S. plana*.

WE103

Changes in the environmental risk of veterinary antibiotics after the introduction of antibiotics-reducing policies

E. Han, D. Lee, Seoul National University / Environmental Planning Institute Graduate School of Environmental Studies

In recent decades, pharmaceuticals in the environment have been concerns for environmental and human health. Especially, the residual antibiotics in the environment could lead to adverse effects on non-target organisms, contamination of food and drinking water supplies, and increased antimicrobial resistance (AMR). Since 2000s, in Korea, the policies for reducing antibiotics usage, such as Separation of Dispensing and Prescribing of Drug, or Restriction of Adding Antibiotics in Animal Feed, have been enforced in view of public health management. Though total antibiotics usage has shown a decreasing trend since the enforcement of these policies, the assessment for these policies in view of environmental risk has not been performed yet. In our previous work (1), an emission prediction model for calculating the predicted environmental concentrations (PECs) of the active pharmaceutical ingredients (APIs) used not only for human but for veterinary purposes was presented. For veterinary usage, the model covers pharmaceutical life cycle posterior to consumption, including direct discharge, manure composting, and land application over the agricultural soil. The emission model was combined with SimpleBox and SimpleTreat into a single spreadsheet-type model for calculating the PECs. In the present study, antibiotics which are of top 20s in veterinary consumption in each year from 2001 to 2016 were selected as target substances. In this period, the environmental risk posed by the regulated antibiotics (Tetracycline, Norfloxacin, etc) has decreased, however, the uses of non-regulated antibiotics (Florfenicol, Tilmicosin, etc) have steeply increased (255 times for Florfenicol, 13 times for Tilmicosin), implying emerging risk. The method used in the present work may serve as a quantitative tool to efficiently assess the policy about pharmaceuticals concerning their environmental risk. Reference (1) Eun Jeong Han; Dong Soo Lee, Application of emission estimation model to the environmental risk assessment of the pharmaceuticals commonly used for human and veterinary purposes, Annual Meeting of Society of Environmental Toxicology and Chemistry, Brussels, Belgium, 2017

WE104

Development of microplate based assay and its application to establish differences in cyanobacteria sensitivity to antibiotics

G.C. Le Page, University of Exeter / College of Life and Environmental Sci; M. Trznadel, L. Gunnarsson, University of Exeter / Biosciences; J. Snape, AstraZeneca UK Ltd. / AstraZeneca Global Environment; C. Tyler, University of Exeter / Biosciences College of Life and Environmental Sciences

Antibiotics are continuously entering the environment and pose a risk to environmental health through their direct effects upon microorganism communities and to human health through antimicrobial resistance. They are designed/selected to target bacteria but in current environmental risk assessment (ERA) only one species of cyanobacteria and the activated sludge respiration inhibition test (ASRIT; proven not to be sensitive for antibiotics) are used to represent all bacterial diversity. There is therefore concern that the potential impacts of antibiotics on environmental health are not fully considered in ERA. We have developed a microtitre assay that broadly follows and meets the validity criteria of the OECD 201 test guideline as a cost effective way to determine the effect of antibiotics on cyanobacteria growth. We applied this assay to determine growth-rate effects on 8 species of cyanobacteria of 9 antibiotics to establish differences in species sensitivity for the improvement of the ERA of antibiotics. Our key findings are: 1) the performance of the microtitre assay is suitable for accurate and reliable assessment of effects on growth inhibition in a wide range of bacterial species; 2) differences in cyanobacteria sensitivity to antibiotics can span several orders of magnitude; and 3) the current framework for ERA of antibiotics inadequately addresses the risk to bacterial populations and testing several diverse cyanobacteria species will increase confidence in the protection goals established.

WE105

Direct and indirect effects of antibiotics in the leaf-shredding macroinvertebrate Gammarus fossarum

M. Konschak, University Koblenz-Landau / Institute for Environmental Sciences; J.P. Zubrod, University of Koblenz-Landau / Institute for Environmental Sciences; P.T. Baudy, University of Koblenz-Landau / Institute of Environmental Sciences; S. Lüderwald, Universität Koblenz-Landau / Institute for Environmental Sciences; C. Jusi, K. Englert, University of Koblenz-Landau / Institute for Environmental Science; R. Schulz, University of Koblenz-Landau / Institute for Environmental Sciences; R. Bundschuh, SETAC Europe Office / Department of Aquatic Sciences and Assessment

Recent studies indicated that both leaf-decomposing microorganisms (i.e., bacteria and fungi) and macroinvertebrate detritivores (i.e., shredders) can be affected by antibiotics via direct and indirect pathways (i.e., via altered microorganism-mediated food quality). However, relatively little is known about these effects on shredders. Therefore, we performed a comprehensive study involving three experiments, which aimed at unravelling the importance of waterborne and diet-related effects of the antibiotic ciprofloxacin (CIP) on the model shredder *Gammarus fossarum*. During a 7-day feeding activity assay, we assessed the effects of waterborne CIP exposure on gammarids' survival and feeding activity, while alterations in leaf palatability for *G. fossarum* due to microbial colonization of the leaves in the presence of the antibiotic were investigated using food choice assays (i.e., diet-related pathway). Furthermore, during a long term assay of 24 days, sublethal effects (the shredders' energy processing and physiological fitness) were assessed when either subjected to a control treatment, to waterborne exposure, a treatment where the animals received leaves that were microbially colonized in the presence of CIP, or a combination of the latter two effect pathways. During the feeding activity assay, *G. fossarum* was rather tolerant towards waterborne antibiotic exposure with LC_{50} and EC_{50} values of 13.6 and 6.4 mg CIP/L, respectively. Furthermore, the shredder did not show statistically significant preferences for control over CIP-exposed leaves during the food choice assays. However, the fungal biomass (an important food quality parameter) was significantly reduced in the highest CIP-treatments (0.5 and 2.5 mg/L), which indicates that antibiotics might affect the quality of the food for shredders. This assumption will be supported by the results of the long term bioassay at 0.5 mg CIP/L: likely due to an alteration in fugal biomass, the shredders' leaf consumption and growth were significantly affected when subjected to the diet-related pathway. Our data indicate that indirect effects of antibiotics on shredders via the diet-related effect pathway could be more relevant than waterborne exposure. Since shredders play a key role in the leaf litter breakdown of heterotrophic stream ecosystems, diet-related effects might result in implications for the energy dynamics of these systems.

WE106

Efficacy of removal antimicrobial resistance genes during avian manure composting process.

F. Esperón, M. Delgado, M. Carballo, INIA - National Institute for Agricultural and Food Research and Technology; M. Ugarte-Ruiz, M. Moreno, UCM; J. Tadeo, INIA   National Institute for Agricultural and Food Research and Technology; A. de la Torre, INIA   National Institute for Agricultural and Food Research and Technology / Environmental Health

Antimicrobial resistance (AR) is an emerging and global problem. Therefore, there is currently a remarkable effort to understand the mechanisms of resistance, to promote the responsible use of antimicrobials and to seek effective therapeutic alternatives. While most livestock studies are focused along the food chain, there are few available studies about the role of livestock manure in the spread of AR. The direct application of animal waste (or slurry) to crops may favor the transmission of AR from cattle to vegetables. The objective of this work is to evaluate the impact of the composting process on the persistence of AR genes. For this, a composting of 10 weeks of duration has been carried out from straw and avian manure, from a laying hen production. Composting samples were taken in triplicate at the end of each week, and total DNA was extracted from each. 22 genes coding for resistance to tetracyclines, sulfonamides, phenicols, aminoglycosides, quinolones, beta lactams, vancomycin and colistin were detected and quantified by real-time PCR. 16 of the 22 genes were detected in at least one sample. Analysis of the temporal evolution of the resistances shows that there is a marked reduction (> 97%) in the genes coding for tetracycline, b-lactam, quinolone and macrolide resistances, while an increase in aminoglycoside and sulfonamide resistance genes is observed. These genes usually form part of integrons, which have more persistence into the environment. Besides, we have found positive correlations among almost all ribosomal protection genes and with the deactivation genes; whereas efflux pump genes were positively correlated among them, suggesting that the persistence of antimicrobial resistance genes could be related to their mechanisms of action. In conclusion, although the composting process does not end up eliminating the AR genes, it can be considered a alternative to the environmental management of the avian manure. RTA2014-00012- C03-02 and S2013/ABI-2747.

WE107

Environmental Assessment Of Multi-Class Pharmaceutical Residues In the Tejo Estuary

S. Leston, CFE-Center For Functional Ecology / Department of Life Sciences

University of Coimbra; A. Freitas, A. Vila-Pouca, INIAV- Instituto Nacional de Investigação Agrária e Veterinária; J. Rosa, CFE Centre for Functional Ecology / Department of Life Sciences University of Coimbra; J. Barbosa, INIAV- Instituto Nacional de Investigação Agrária e Veterinária; F. Ramos, Faculty of Pharmacy University of Coimbra; P. Reis-Santos, I.A. Duarte, M.P. Pais, MARE - Marine and Environmental Sciences Centre; H.N. Cabral, Faculdade de Ciências da Universidade de Lisboa / MARE-FCUL; V.F. Fonseca, MARE Marine and Environmental Sciences Centre
Environmental pharmaceuticals contamination is now a recognized threat in coastal and estuarine ecosystems representing unknown and potentially harmful effects to non-target organisms. It is therefore paramount to monitor their presence in the environment and to evaluate the extent of their influence. The Tejo estuary, located on the Atlantic coast of Portugal was established as a case study for the environmental occurrence of pharmaceuticals due to the proximity to very urbanized areas in the Great Lisbon Region, with sewage disposal and discharges from industries, hospitals, agricultural and fish farms in upstream areas as well as urban effluents, anticipating the presence of contaminants. A monitoring campaign was conducted during summer where water, sediment, macroalgae, invertebrates and fish were sampled for pharmaceutical determination. Thirty sampling stations were selected according to their proximity to discharge points where effluents are expected to be rich in pharmaceuticals but also including reference sites where contamination was expected to be very low. Multi-residue multi-class analytical methods developed for each matrix are being applied for the detection and quantification of 67 compounds. The compounds monitored included antiepileptic and anticonvulsant drugs, benzodiazepines, anti-inflammatories and analgesics, angiotensin receptor blockers, b-blockers and antibiotics (42 compounds) in a total of 67 drugs. Multi-residue multi-class analytical UHPLC-ToF MS methods developed for each matrix are being applied for the detection and quantification. The knowledge gathered will then be applied to exposure assays and antibiotic resistance studies using the pharmaceuticals detected at the highest concentrations and in mixtures. The knowledge gathered will then be applied to antibiotic resistance studies using the pharmaceuticals detected at the highest concentrations.

WE108

Environmental risk of enrofloxacin used in aviculture

M. Carballo, M. Delgado, F. Esperón, INIA National Institute for Agricultural and Food Research and Technology; M. González, INIA National Institute for Agricultural and Food Research and Technology / CISA; J. Tadeo, INIA National Institute for Agricultural and Food Research and Technology; A. de la Torre, INIA National Institute for Agricultural and Food Research and Technology / Environmental Health
The presence of antimicrobial residues in animal excreta supposes a health and environmental hazard associated with its agricultural reuse. Many of them have toxic potential for terrestrial and/or aquatic organisms. The environment can act as a reservoir not only for residues, but also for antimicrobial resistance genes, and may spread them into the food chain. This is a particularly serious in the case of antibiotics that can accumulate in soil, such as fluoroquinolones, which have a high adsorption capacity for organic matter and a high persistence. The objective of this work focuses on the environmental risk assessment of enrofloxacin (ENR) and its main metabolite (ciprofloxacin, CIPR), associated with its use in poultry farming in Spain according to the technical prescriptions of the authorized products. The environmental risk ratios (RQ) have been calculated following the European Guidelines on Environmental Risk Assessment of Veterinary Drugs (EMEA/CVMP/ERA/418282/2005). In the case of the CIPR, information has been used on the metabolism and excretion of the ENR in chickens, to establish the levels of CIPR in soil and later, to assess their environmental risk. The results indicates that the estimated PECsoil for ENR (443 µg/kg), implies risk for terrestrial organisms, specifically in plants (RQ> 1). No risk is identified for CIPR. Finally, an ENR environmental risk map has been generated in Spain. Allowing us to identify the "hot spots" where the greatest environmental management and surveillance efforts should be applied. This spatial analysis (ArcGIS 10.2) was carried out using a simple addition method (MultiCriteria Decission) and two risk factors were included: the avian density and the capacity of the soil to accumulate this antibiotic (De la Torre et al., 2012). The environmental relevance of these results is discussed and the effectiveness of composting field-scale composting in battery cages is indicated to minimize the risk of these drugs. This work is funded by RTA2014-00012- C03-02 and S2013/ABI-2747.

WE109

Evaluating the use of veterinary antibiotics in dairy environments to inform on antimicrobial resistance spread and development

R. Baena-Nogueras, The University of Nottingham / Physical Chemistry; T. Dodsworth, The University of Nottingham / Chemical and Environmental Engineering; C. Ortori, D. Barrett, The University of Nottingham; R.L. Gomes, The University of Nottingham / Faculty of Engineering
The University of Nottingham owns a high throughput dairy farm with around 200 milking cows, from which the pressed liquid waste ends up in a 3000 m^3 slurry tank. Contained within the slurry we can find antimicrobial foot washings, waste milk and bedding particulates (among other components). Large volumes of liquid slurry are produced in farms and this waste material is commonly applied to fields as a fertilizer in the UK. The health of a dairy herd is supported by the administration of antibiotics so the dairy farm setting works as an antimicrobial resistance (AMR) reservoir and as a route of entry into the environment. Antibiotics' persistence in dairy settings occurs through mediums such as slurry, milk and subsequently, in soil via spreading. These routes can act as a channel for the transference of compounds and the insemination of AMR within the food chain through intake by plants and migration to other sources via water run-off, possibly affecting the therapeutic potential against human and animal pathogens and posing a high risk to public health. Therefore, the increasing soil retention of polar substances after soil amendment and the high persistence found for some antibiotics in batch experiments (e.g. sulfonamides), make further research on exposure assessment necessary along with the analysis of veterinary antibiotics in dairy environments in order to assist in shedding light on the long established concern of the environmental fate and behaviour of veterinary antibiotics in farming and to propose better handling practices as a basis for future regulations. The main objective of this study is to measure the distribution of select antibiotics currently in use within the farm environment, relating their presence to the length of use and the last date of prescription. For this purpose, specific optimized analytical methods including 19 antibiotics belonging to 10 different families were employed for clean-up and pre-concentration of different matrices (slurry,soil,milk) by means of liquid-liquid extraction, followed by solid-phase extraction (SPE) and identification and quantification by liquid chromatography-mass spectrometry (LCMS). For instance, several veterinary antibiotics under study were detected in wastewater slurry with concentrations up to 18.6 µg L^{-1} for oxytetracycline suggesting a high persistence. Summarizing, this study is the first step towards the evaluation of the impact of antibiotics presence and fate in the AMR development.

WE110

How do marine and freshwater cyanobacteria react to long term exposure of antibiotics? Is there a potential for increasing antibiotic resistance in the environment?

J.H. Heseding, C. Floeter, Hamburg University of Applied Sciences / Environmental Engineering
An increasing amount of pharmaceuticals are detected in waterbodies all over the world. Antibiotics are of special concern for the environment: Due to the high amount of medication, its specific activity against prokaryotes and a possible antibiotic resistance formation in the environment. Cyanobacteria have prokaryotic target structures of antibiotics and are of high importance for the primary production and nitrogen cycle in marine waters. To investigate the long term effects of antibiotic exposure on limnic and marine cyanobacteria, the limnic cyanobacterial test according to OECD 201 and the marine cyanobacteria test developed by Heseding and Floeter in 2016 were performed several times. Exposed cultures were recultured at the end of the test and then reexposed to the same antibiotic active substance as part of a repeated test. As test organisms *Synechococcus leopoliensis* (limnic cyanobacteria) and *Synechocystis spec.* (marine cyanobacteria) were selected. The tests were carried out on a 24-well mircowell plate. In the test, the percentage inhibition of the growth rate (cell number) is determined in comparison to the negative control over a period of 72 hours. The derived EC50-values after repeated exposition were compared for different antibiotics of environmental concern. The investigations are carried out as part of the PharmCycle project.

WE111

Impact of antibiotics on the feeding rate of the freshwater shrimp Gammarus pulex

G. Consolandi, University of Portsmouth; M. Bloor, University of Portsmouth / School of Earth and Environmental Science; A. Ford, University of Portsmouth / Biological Sciences
Antibiotics are one of the main categories of pharmaceuticals and their release into the freshwater environment can impact the natural bacterial and fungal communities, which can threaten the survival of organisms that rely on them as a food source. One such organism is the freshwater detritivore *Gammarus pulex* that commonly feeds on detritus such as, naturally conditioned *Alnus glutinosa* leaves. The study aim was to establish if the feeding rate of *Gammarus pulex* was altered when their food source (*Alnus glutinosa*) was exposed to environmentally realistic concentrations of antibiotics during the natural leaf conditioning process. The investigation included three antibiotic scenarios (1) exposure to the bacteriostatic agent Tetracycline, (2) exposure to a mixture of Sulfamethoxazole and Trimethoprim bacteriostatic agents that are commonly prescribed together and (3) exposure to the bactericidal agent and broad-spectrum antibiotic Ciprofloxacin. 24 h feeding assays were performed using *Alnus glutinosa* leaf discs of 1.3 cm Ø and standardised *Gammarus pulex* specimens (n=60). The organisms were kept at 15°C under a 12:12 h light:dark cycle. 15 replicates were undertaken with three environmental realistic concentrations and a charcoal filtered tap water control (200 mg/L, 20 mg/L and 2 mg/L). The leaf discs were photographed at the start and finish of the investigation and these images underwent analysis with Image J software in order to calculate the area consumed. After 24 h, the *Gammarus pulex* were sacrificed by exposure to -20°C temperature before being dried at 60°C for 24 h and weighed. This protocol was performed with antibiotic scenario 1, 2 and 3. The results showed that Tetracycline (Z=0.198, p=0.897) and Ciprofloxacin (Z=1.568,

p=0.667) were not a concern in relation to feeding at environmentally realistic concentrations (scenario 1 and 3), (p=< 0.05). When exposed to a mixture of Sulfamethoxazole and Trimethoprim (scenario 2) there was an impact on the *Gammarus pulex* feeding rate (Z=13.239, p=0.004). However, further investigation would be required to investigate these drugs individually to identify if the obtained results were driven by one or the combination, and also to establish if there is a genuine environmental concern associated to this mixture or if the data is blurred in some way.

WE112

Persistence of the sulfamethoxazole antibiotic in a digestate-amended agricultural soil

L. Patrolecco, Water Research Institute-National Research Council / Water Research Institute; J. Rauseo, National Research Council / Institute of Water Research IRSA-CNR; W. Gaze, University of Exeter / Medical School; I. Stanton, University of Exeter; N. Ademollo, M. Cardoni, National Research Council of Italy / Water Research Institute; M. Di Lenola, National Research Council of Italy; P. Grenni, National Research Council of Italy (CNR) / Water Research Institute; F. Spataro, National Research Council of Italy / Water Research Institute; A. Barra Caracciolo, National Research Council / Water Research Institute

Thousands of tons of antibiotics are annually used in human and veterinary medicine worldwide. They are excreted, from the treated organism, either unaltered or as metabolites, reaching soil and water ecosystems. In particular, the use of wastewater, sewage sludge, livestock manure and digestate as agricultural amendments and fertilizers, introduce residual concentrations of antibiotics to soils. Livestock raising practice involves the use of antibiotics in feed; consequently digestate obtained by anaerobic digestion of manure may be an additional source of antibiotics and resistance genes in soil. Sulfamethoxazole (SMX) is one of the most commonly prescribed and consumed sulfonamide antibiotics, due to its ability to inhibit Gram-positive and Gram-negative bacteria it is used in veterinary practices, aquaculture and livestock breeding both for treating diseases and promoting growth. However, current knowledge about its persistence and possible environmental effects is poorly understood. In the present study, we investigated the persistence and the possible effects on the soil natural microbial community of SMX in an agricultural soil amended with solid anaerobic digestate from bovine manure anaerobic fermentation. Microcosms, containing soil and digestate treated with 20 mg/Kg of SMX, were set up in the presence/absence (sterilized soil) of the natural microbial community. Moreover, non-antibiotic-treated microcosms were used as microbiological controls. At fixed times (0d, 7d, 13d, 20d, 61d), SMX residual concentrations (ASE extraction and HPLC-UV detection) and microbiological parameters (cell viability, abundance and activity) were analysed. Finally, a molecular marker for antibiotic resistance, the *intI* 1 gene (class 1 integron) was investigated at the start and the end of the experiment using qPCR. Results showed that although an acute negative effect (0d) was observed on the microbial abundance and viability, the antibiotic was degraded in just a few days. Interestingly, the *intI* 1 gene was found in the soil where the digestate was added, showing its introduction through this agricultural practice.

WE113

Pollution in the Mooi River: Fluconazole and fluconazole resistant pathogenic yeasts species

M.E. Monapathi, North West University (Potchefstroom Campus) / Microbiology

The presence of yeast species in water sources that are associated with faecal pollution have been demonstrated. Some of the isolated species are potentially pathogenic and could cause superficial and life-threatening infections. Fluconazole on the other hand is the most used antifungal agent in the treatment of yeast infections as well as prophylactic agent to prevent *Candida* and *Cryptococcus* infection in HIV patients. The aim of the study was to determine yeast pollution, susceptibility of these to, and the levels of, fluconazole in the Mooi River, North West (South Africa). Yeast isolates were enumerated using membrane filtration, selective media and incubation at 37°C and identified using biochemical methods (staining and 26S rRNA gene sequencing). Resistance to fluconazole was determined by disc diffusion. Environmental DNA was isolated directly from water using membrane filtration and a commercial DNA isolation kit. Yeast levels as inferred by qPCR of 26S rRNA gene levels were determined. For fluconazole analysis water samples were extracted using solid phase extraction. The extracts were analysed with liquid chromatography coupled to a quadrupole time-of-flight mass spectrometer. The purified isolates identified included *Candida albicans*, *C. krusei*, *C. tropicalis* and *Saccharomyces cerevisiae*. The yeasts identified have been associated with polluted waters. Some isolates in the present study are pathogenic and occasional direct contact with polluted water could cause infections to immune compromised people and were all resistant to fluconazole. Quantitative PCR of the 26S rRNA gene indicated that a high number of gene copies were present at all sites. Fluconazole levels ranged from

WE114

Reactivity, mobility and degradation of the antibiotic Sulfametoxazole and its impact on the microbial communities of an agricultural soil amended with organic waste products

J.M. MARTINS, CNRS IGE UMR 5001, Univ. Grenoble / OSUG-IGE; E. François, L. Spadini, J. Granat, C. Humbert, E. Vince, M. Morel, Institut Geoscience & Environemment

The fate of Sulfamethoxazole (SMX), a sulfonamide antibiotic widely spread in natural soils and waters, was evaluated in batches and columns of a silty-loam soil under culture from Feucherolles (INRA Versailles, France). SMX revealed to be a weak sorbent since in batches only 10% of SMX sorbed at a solid/solution ratio of 0.1. It's sorption increased strongly with soil organic matter content (addition of manure), indicating that the essential feature of OM addition is an increase in sorption sites density at almost constant sorption strength, and confirms previous results about the strong influence of sorbed complexants such as Cu(II). The mobility of SMX evaluated in water-saturated columns showed higher mobility of SMX than expected from the sorption study. Unexpectedly, this increased mobility was even higher in the OM-amended soil, suggesting that soil OM contributes to antibiotics transport in soils. Batch degradation experiments revealed that SMX removal is quite fast with half-life values ranging between 18 and 350 days in non-sterile and sterile soils. This degradation process was shown to occur principally in both the coarsest and finest soil-size fractions, while almost no biodegradation was observed in the mass-dominant silty fraction of the soil in agreement with its low microbial biomass content. The impact of SMX on the soil bacterial community, evaluated through total biomass (16SDNA), qPCR (Antibiotic Resistance Genes, ARG, *sul*1 and *sul*2), DGGE fingerprinting and high throughput sequencing revealed important impacts of SMX on soil microbial biodiversity and species richness and the emergence of specific taxons, resistant to the antibiotic. These results permitted to characterize the global fate and impact of SMX in an agricultural soil. SMX appeared quite mobile in soil with enhanced mobility in presence of added OM. SMX appeared also quite readily biodegradable, especially when in contact with coarse and fine soil size fractions, where it had the strongest impact on soil bacteria. Keywords: Antibiotics, SMX, organic matter, impact, DGGE, Miseq, ARG, biodegradation

WE115

Risk assessment of antibiotic resistance and related genes in human impacted environments

J. Muurinen, University of Helsinki / Food and Environmental Sciences; K. Pärnänen, J. Hultman, W. Muziasari, University of Helsinki; R.D. Stedtfeld, J.M. Tiedje, Michigan State University / Center for Microbial Ecology Department of Civil and Environmental Engineering; M. Virta, University of Helsinki

The origins of antibiotic resistance in the environment is relevant to human health because of the increasing importance of zoonotic diseases as well as the need for predicting emerging resistant pathogens. Antibiotics are used in diverse settings for food production. Domestic animals are treated with antibiotics for both curing disease and promoting growth. Moreover, aquaculture relies on antibiotics to manage infectious disease. Wastewater treatment plants receive sewage from various sources, including hospitals and households which are both important sources of antibiotics and their residues, and antibiotic resistant bacteria. Risk assessment of antibiotic resistance is complicated. It should include at least quantitative information of the gene, sequence of the gene, host cell of the gene and genetic environment of the gene (e.g. presence in mobile DNA element). We have used the combination of different methods for obtaining that information: Parallel quantitative PCR array for high throughput quantification (1), epicPCR(2) for host information and Inverse-PCR(3) for analysis of the genetic environment. Inverse-PCR and epicPCR combined with DNA sequencing resolve also the sequence of the resistance gene. Samples were collected from different locations in Finland: manure from cattle and pig farms, soil that received the manure as fertilizer, sediments from aquaculture farms and effluent, influent and activated sludge from waste water treatment plant. Our results demonstrate that human activities results to the increase to the abundance of antibiotic resistance genes. In many cases the genes are located in mobile genetic elements with increases the probability of transfer of the them between bacterial species. The host range information obtained by epicPCR revealed wide diversity on the host range of the antibiotic reistance genes in different environments. Our results can be used for the development ecotoxicological risk analysis for antibiotic resistance. (1) Karkman, A., Johnson, T.A., Lyra, C., Stedtfeld, R.D., Tamminen, M., Tiedje, J.M. and Virta, M. (2016) FEMS Microbial Ecology 92 (3): fiw014 (2) Spencer, SJ., Tamminen, MV., Preheim, SP., Guo, MT., Briggs, AW., Brito, IL., Weitz, DA., Pitkänen, LK., Vigneault, F., Virta, MP. and Alm, EJ. (2016) ISME Journal 10:427-436 (3) Pärnänen, K., Karkman, A., Tamminen, M., Lyra, C., Paulin, L., Hultman J. and Virta, M. (2016) Scientific Reports 6: 35790

WE116

Risk of antibiotics in the environment

D.d. Silva Tavares Duarte, Radboud University / Department of Environmental Science; R. Oldenkamp, Radboud University Nijmegen / Department of Environmental Science; A.M. Ragas, Radbound University / Department of Environmental Science

For decades, pharmaceuticals have been fundamental tools against disease and infection targeting humans and animals. Antibiotics in particular have played a decisive role due to their ability to inhibit growth or eliminate microorganisms. Unfortunately, its misuse combined with bacterial capability to acquire antibiotic resistant genes, have significantly contributed to the escalation of life-threatening infections leading to worldwide antimicrobial resistance (AMR). This issue is most evident in artificial high selective pressure settings (e.g. hospitals, animal farms) but the increasing occurrence of antibiotics and resistance genes in the environment is spawning serious concern. The measured concentrations of antibiotics are relatively low, most are readily biodegradable and there are considerable resistance-associated fitness costs. However, intricate bacterial compensatory mechanisms, population dynamics and long-term persistence can lead to resistance gene emergence and enrichment (e.g. via horizontal gene transfer). Therefore, there is a need for a better understanding of how concentrations of antibiotic relate to the abundance of resistance genes in different environmental compartments under different conditions. In this study, we compiled this sparse information by conducting an extensive literature meta-analysis to evaluate global trends. Our initial results show distinct environmental matrices exhibiting a wide range of gene abundance (e.g. surface water). Interestingly, there are cases were gene variation is weakly correlated with antibiotic concentration (e.g. sediment) which challenges the common proportionality assumption between these two parameters. This indicates that AMR genes can be highly maintained throughout bacterial communities under certain environmental conditions. Whether detected gene levels are antibiotic-induced or the consequence of carrying-microbes emissions (e.g. via urban effluents) is still under debate. These results are expected to support the development of integrative models capable of providing meaningful risk assessment to support decision-making.

WE117

Sulfamethoxazole degradation in river water microcosms and effect on the natural microbial community

J. Rauseo, National Research Council / Institute of Water Research IRSA-CNR; L. Patrolecco, Water Research Institute-National Research Council / Water Research Institute; N. Ademollo, National Research Council of Italy / Water Research Institute; P. Grenni, National Research Council of Italy (CNR) / Water Research Institute; M. Cardoni, L. Rolando, C. Levantesi, M. Luprano, National Research Council of Italy / Water Research Institute; A. Barra Caracciolo, National Research Council / Water Research Institute

The widespread use of antibiotics causes concern on their occurrence and fate in different environmental matrices. Following administration, antibiotics are only partially metabolized and a large amount is excreted unaltered or as active metabolites, reaching wastewater treatment plants (WWTPs). Most WWTPs are not able to remove them efficiently and through their effluents, they reach surface water. The synthetic compound sulfamethoxazole (SMX) is one of the most prescribed and consumed sulphonamide antibiotics to treat urinary tract infections used both in human and veterinary practices. The widespread use of SMX implies its occurrence in different environmental matrices. Furthermore, SMX is not readily biodegradable and resistant to hydrolysis. The high frequency of detection and relative persistence of SMX in environment cause a potential risk of antibacterial resistance spread in ecosystems. Multiple mechanisms confer sulfonamide resistance in bacteria, although data on biodegradation and spread of antibiotic resistance genes (ARGs) in natural water ecosystem are quite scarce. The aim of the present work was to investigate the SMX degradation in natural river water in presence/absence of the microbial community and to identify the occurrence of *sul* genes associated to the antibiotic resistance. Microcosm experiments were set up using river water treated with 500 µg/L of SMX. At fixed times, water sample were collected for chemical (SMX residual concentrations) and microbial analysis. The disappearance time of 50% of the initial SMX concentration (DT_{50}) and the effects of the antibiotic on the natural microbial community were evaluated in terms of cell vitality and abundance. Moreover, the spread of sulfonamides resistance genes was evaluated by quantifying the *sul 1* gene. The antibiotic SMX was biodegraded with a DT_{50} of about 20 days. The microbial abundance not only was not affected by the antibiotic addition (t=0 days), but at the end of was significantly higher in treated microcosms than in control conditions. The abundance of *sul 1* increased after addition of SMX, suggesting that ARG spread is a physiological adaptation of natural microbial community to its presence.

WE118

The effect of antibiotics on representatives of aquatic algal and plant species

B. Aubakirova, L.N. Gumilyov Eurasian National University / Management and Engineering in environmental protection; R. Beisenova, L.N. Gumilyov Eurasian National University / Department of Management and Engineering in environmental protection

Nowadays, pharmaceuticals are pollutants of increasing interest. The volume of the production of pharmaceuticals has been increasing rapidly in the last decade in Kazakhstan. Antibiotics make up a significant proportion of the pharmaceuticals that are in use in the country. However, antibiotics as environmental contaminants have received little attention in Kazakhstan and the topic is a new field for research in the country. The aim of the present study was to the impact of priority antibiotics in use in Kazakhstan on representative aquatic species. *Lemna minor* and *Chlorella sp.* were selected for the ecotoxicological investigations. Five major use antibiotics in Kazakhstan (amoxicillin, clarithromycin, azithromycin, sulfamethoxazole, oxytetracycline) and their mixture were used in the experimental assessments. The compounds were selected based on a previous prioritization study based on the risks of active pharmaceutical ingredients (APIs) to aquatic environments in Kazakhstan. The study on *Lemna minor* was conducted according to the OECD Guidelines for the testing of chemicals 221. *Lemna minor* species were cultured in Swedish Standard (SIS) growth medium and effects of the antibiotics on growth assessed over 7 days. The results of the study showed EC_{50} values of each test compounds ranged from 2.8 to 21.8 mg/L. *Lemna minor* was most sensitive to the sulfamethoxazole, with its EC_{50} being below 10 mg/L. The test on algae was performed according to the OECD Guidelines for the testing of chemicals 201. *Chlorella sp.* were cultured in Tamiya medium and algae numbers were counted in Goryaev chamber under a microscope. The macrolide substances azithromycin and clarithromycin were found to be the most toxic compounds to the algae with EC_{50} values being lower than 1 mg/L. In the future, it is recommended to perform assessments on the sensitivity of other less well studied aquatic species to priority API in use in Kazakhstan as well as monitoring studies to establish levels of exposure in the country. This will then provide a basis for the risk of these substances to be established.

WE119

The Presence of Human and Veterinary Antibiotics in Urban and Rural Streams of North Carolina

A.D. Gray, University of North Carolina at Greensboro / Biology; D. Todd, University of North Carolina at Greensboro / Chemistry; A.E. Hershey, University of North Carolina at Greensboro / Department of Biology

Antibiotic pollution is of concern to environmental health sciences due to the implications associated with their presence in the environment. There is a great deal of literature drawing attention to antibiotics in the environment contributing to antibacterial resistance amongst bacteria. Antibiotic pollution in stream ecosystems in the United States remains a research area that has received little attention, while more research has focused on antibiotics in larger rivers, lakes and estuaries. Streams ecosystems play a crucial role in the transport of water from inland areas to coastal waters as well as to rivers and lakes, and have greatest potential to mitigate pollutants compared to larger systems. The presence of antibiotics in streams demonstrates the potential for antibiotics to be transferred from inland areas into larger water bodies. Antibiotics entering streams can arise from various sources. In urban areas, antibiotics of human and veterinary origin can enter streams due to runoff or leaching from surrounding areas, but most notably from wastewater discharges that release effluent directly into streams. In rural areas, antibiotics can enter streams from application in the maintenance of livestock, which due to runoff and leaching, can contribute to veterinary antibiotics being present in rural streams and groundwater. Work from the present study found human and veterinarian antibiotics in both urban and rural streams. Antibiotics detected include sulfamethoxazole, sulfamerazine, trimethoprim, danofloxacin, sulfaquinoxaline, ciprofloxacin, enrofloxacin, and tylosin, with several of the detected antibiotics being present in both urban and rural streams. Results from this work demonstrate that streams are key sites regulating discharge of antibiotics to larger bodies of water and that surrounding land use and infrastructure influences the presence of antibiotics in streams in urban and rural areas.

WE120

The Role of Water Quality Analysis: Understanding our process environment to inform on AMR.

T.P. Dodsworth, The University of Nottingham / Biosciences; R. Helliwell, The University of Nottingham / Social Sciences; E. King, The University of Nottingham / Biosciences; R.L. Gomes, The University of Nottingham / Faculty of Engineering

When researching antibiotic resistance (AR) in an environmental framework there will be a number of factors, antibiotics, metals and other selective agents, that are constantly in flux, which may facilitate or inhibit the selection and transfer of ARGs between microorganisms within a matrix. This article will outline the ways in which water quality analysis (WQA) can be used as a tool for understanding key components of systems under study outside the scope of microbiology. Specifically, how WQA can contribute additional understanding with regards to environmental variation of organic and inorganic compounds and metals, alongside the complexity of a given matrix. Samples drawn from the 3000m3 capacity slurry tank of a high input/high output dairy farm in the East Midlands were tested for 16 variables. These included Zinc and Copper, Dissolved Oxygen, Chemical Oxygen Demand, pH, and common environmental factors such as Ammonium, Nitrate, Nitrite and Phosphate. In addition, WQA was used to understand matrix variation within the slurry storage tank over different time periods, as a result of different management practices such as mixing and variation between different aspects of the slurry management system on the farm. This is supplemented with data from additional external influences; rainfall, temperature and farm practices, to further understand how the system as a whole can be considered when researching AR.

WE121

Safety and efficiency assessment of antibiotic administration by magnetic nanoparticles in Zebrafish

G. Chemello, C. Piccinetti, B. Randazzo, O. Carnevali, F. Maradonna, Università Politecnica delle Marche; M. Magro, E. Bonaiuto, F. Vianello, G. Radaelli, Universita degli Studi di Padova; A. Fifi, Biotecnologie B.T. Srl / Ecotoxicological and chemical; F. Gigliotti, CRO BioTecnologie BT; I. Olivotto, Università Politecnica delle Marche

The indiscriminate use of antibiotics in the aquaculture sector has raised public concern because of possible toxic effects, development of bacterial resistance, and accumulation of residues in individual tissues. Even if several countries have developed regulations about their use, it is clear that long-term growth of the aquaculture industry requires both ecologically sound practices and sustainable resource management. Alternative strategies for better management of antibiotic administration are of primary interest to improve absorption rates and, as a consequence, to reduce their release into the aquatic environment. The present study investigates, for the first time to our knowledge, a new methodology for oxytetracycline (OTC) administration through the use of iron oxide nanoparticles (NPs) (made of maghemite γ-Fe_2O_3) in zebrafish (*Danio rerio*). Fish were divided into 4 experimental groups: control; group A exposed to 4mg/L OTC (through water); group B exposed to the 100mg/L SAMNs@OTC complex (equivalent to 4mg/L OTC), and group C exposed to bare NPs. HPLC analysis, histological analysis and other methods were applied to perform different evaluations. No detoxification processes or anatomical alterations were observed in fish exposed to bare NPs. Exposure of fish to the SAMNs@OTC complex resulted in a 10 times higher OTC accumulation with respect to using water exposure. This new method for OTC administration seems more efficient with respect to the traditional way of exposure and shows the potentiality to reduce antibiotic utilization and possible environmental impacts.

Analysis and Fate of Emerging Contaminants in soils, water and plants under water scarcity (P)

WE122

Identifying and Controlling Sources of Ultra-Trace Metals in Control Blanks and Ensuring High-Quality Data for Sensitive Environmental Risk-Based Decisions

R. Vitale, Environmental Standards; L. Dupes, A. Reed, Environmental Standards Inc; M. Mc Anulty, BP Exploration

The control of blank contamination is a necessary requirement when evaluating the qualitative and quantitative accuracy of analytical results for decision-making purposes of site delineation, toxicological evaluations, and site remediation. This control is essential to differentiate actual site concentrations from potential sources of introduced field or analytical contamination, especially during development of background site metals conditions and cleanup criteria. Trace-level concentrations of total and dissolved metals were observed in several equipment rinsate blank samples collected at multiple sites during the field sample collection season of\n 2014 through 2016. The detection of total and dissolved metals in these field quality control (QC) samples resulted in qualification of both total and dissolved sample results resulting in questions regarding the presence or absence of low-level site contamination. Although equipment rinsate blank samples do not have specific contamination acceptance criteria due to the field collection process, the goal of equipment rinsate blanks is to verify that contamination was not introduced during the sample collection process or by sampling equipment. The purpose of the investigation and identification was to determine potential sources of metals contamination in equipment rinsate blanks that could be identified, reduced, or eliminated. Multiple avenues of potential contamination were investigated including a study of sample tubing, peristaltic pumps and in-line filters; field observation of equipment blank collection processes; initial and post water-quality monitoring; sample bottle cleanliness and storage of deionized water for use in collection of the equipment rinsate blanks. This presentation will provide details of the investigation process and results after implementation of several important corrective actions.

WE123

Comprehensive Analysis of Elemental Contamination in Environmental Samples utilizing Inductively Coupled Plasma Mass Spectrometry (ICP-MS)

S.M. Nelms, D. Kutscher, Thermo Fisher Scientific / marketing; S. McSheehy Ducos, Thermo Fisher Scientific GmbH / Product Management

The analysis of elemental contamination is a significant task for laboratories working in environmental analysis. Besides direct regulation of contaminant levels, for example in waters, also a number of other sample types must be screened, such as soils or sludges. Targeted elements comprise the “big four”, arsenic, cadmium, mercury and lead, but also many other elements. Particular challenges in this area include the need to often measure large numbers of samples and potential complications caused by a variety of spectral interferences. For example, if rare earth elements are present in a given sample, they can lead to severe interferences on arsenic and selenium. To avoid false positive results, triple quadrupole ICP-MS is ideally suited for effectively removing all potential interferences. However, the inherently higher complexity of a triple quadrupole based system in comparison to traditionally applied single quad systems is a barrier for most laboratories dealing with routine analysis. This presentation will highlight the use of ICP-MS, especially triple quadrupole ICP-MS, for the analysis of environmental samples. Dedicated software solutions, such as tools to simplify method development, increase productivity or tackle advanced applications, such as chromatography or nanoparticle analysis, will be presented to show the broadness of accessible applications using modern ICP-MS instrumentation.

WE125

ANALYTICAL METHOD FOR DETERMINATION OF FULLERENE (C60) NANOPARTICLES IN SEAWATER SAMPLES.

L.R. Diniz, Universidade Estadual do Maranhão / Agroecologia; L. Capellini, Universidade Federal de São Paulo UNIFESP / Departamento de Química

Fullerenes are allotropic forms of carbono produced in highly energetic processes of natural origin or anthropogenic sources. In the last years, the increasing application of nanomaterials in several areas of human endeavor besides their physical and chemical properties, contribute for the growth of the global economy. However, the growing production and application of nanomaterials is also promoting discussions about the possible risks of these compounds to the environment and human health. Data have already been reported on the occurrence of fullerenes in different matrices, including the atmosphere, soils and sediments, and fresh water. Despite this, little information has been related to marine environments while coastal areas and estuaries are suspected to be one of their major sinks. The purpose of this study is developed and optimize an analytical method to evaluate the presence of nanomaterial fullerene (C60) in seawater samples. It will be tested two methods of extraction: (1) dispersive liquid–liquid micro extraction (DLLME), and (2) QuEChERS, after, all the samples will be analyzed by liquid chromatography tandem mass spectrometry (LC-MS/MS). Keywords: Marine pollution. Fullerenes. Nanomaterials.

WE126

Screening of per- and polyfluoroalkyl substances (PFASs) and total organic fluorine in wastewater effluent from Nordic countries

F. Chen, MTM Research Centre, Orebro University / SCHOOL OF SCIENCE AND TECHNOLOGY; U. Eriksson, R. Aro, MTM Research centre Örebro University; L.W. Yeung, University of Örebro / Department of Chemistry; T. Wang, MTM Research Center; R. Kallenborn, Norwegian University of Life Sciences / Chemistry, Biotechnology and Food Sciences; A. Karrman, Orebro University / MTM Research centre

The discharge of per- and polyfluoroalkyl substances (PFASs) into the environment via wastewater is a pressing public health issue. Apart from the frequently detected PFASs, such as PFOS and PFOA, more and more novel PFASs have been reported recently. We used the target screening method to identify novel and legacy PFASs in the Nordic environment. The aim of this project is to determine as many PFASs and total organic fluorine as possible in wastewater effluents from the Nordic countries and self-governing areas, including Finland, Sweden, Norway, Denmark, Faroe Islands, Iceland and Greenland. Field collected effluent (250 mL) were extracted by solid phase extraction (SPE). Identification and quantification of target compounds was performed using liquid chromatography-electrospray ionization tandem mass spectrometry (LC-ESI-MS/MS), ultra-performance convergence chromatography (UPC^2) and combustion ion chromatography (CIC). The significance of the occurrence, levels and patterns of various PFASs in Nordic wastewater effluents are discussed.

WE127

Quantitative evaluation of lag effect in polar organic chemical integrative sampler (POCIS) and modified POCIS with polytetrafluoroethylene (PTFE) membranes

Y. Jeong, H. Kwon, KIST Europe / Environmental Safety Group; H. Jeon, KIST Europe; A. Meyer, E. Fünfrocken, H. Beck, Saarland University; K. Smith, RWTH Aachen University / Institute for Environmental Research; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics

Increasing occurrence of organic contaminants in the aquatic environment has heightened the need for reliable and efficient monitoring technique. Traditional grab sampling followed by laboratory extraction and instrumental analysis is well established method, but only provides a snapshot of the water quality status. Achieving representative sample with grab sampling takes considerable labour, time and cost. Here, time-integrative passive sampling technique is recognized as a promising monitoring tool. Passive sampling technique allows the simple sampler construction and application, provision of time weighted average concentration and in situ sampling. Various configuration of passive sampling devices are currently available, one of which is polar organic chemical integrative sampler (POCIS). POCIS consists of Oasis HLB® sorbent sandwiched between two polyethersulfone (PES) membranes and has been widely used for the detection of hydrophilic contaminants in the past decade. However, uncertainties in quantitation of POCIS measurements have been pointed out as a main limitation of POCIS. Compound specific sampling rate depends on sampler configuration and environmental parameters such as flow rate and temperature. Lag effect from membrane sorption within POCIS further complicates the transfer kinetics of analyte. In this study, modified POCIS (POCIS-PTFE) with polytetrafluoroethylene (PTFE) membrane instead of PES membrane was tested in an attempt to avoid or lower the PES membrane sorption. The primary aim of this study is to (1) introduce modified POCIS and (2) identify the membrane sorption within POCIS in order to better understand partitioning kinetics of POCIS. In the laboratory experiment, the analyte mass fraction in membrane relative to total POCIS (i.e., Oasis HLB plus membranes) and membrane-water partition coefficient were determined for a range of compounds (log K_{OW} from -0.03 to 6.26). Less membrane sorption was found in

PTFE membrane than PES membrane for all target compounds. Two types of POCIS were then deployed in a small river and the outflow of wastewater treatment plant for two weeks. Both POCISs showed similar chemical profile and 22 contaminants were detected including 6 priority substances enlisted in EU Water Framework Directive. Although PTFE membrane showed better permeation performance than PES membrane in laboratory experiment, the lag effect was still found from the field application of POCIS-PTFE.

WE128

Occurrence and Ecological Risk Assessment of Several Endocrine Disrupting Chemicals in Urban River Water and Sediment of South China

C. Huang, Jinan University; L. Wu, Y. Guo, Jinan University / School of Environment

This study mainly focused on the occurrence, distribution, and ecological risk assessment of eight selected endocrine disrupting chemicals (bisphenol analogues, parabens, and triclosan) in urban river water and sediment of south China. The eight target chemicals were detected in both water and sediment samples with concentrations ranged from not detected to 65600 ng/L and from not detected to 492 ng/g dw, respectively. Among this eight chemicals, the top three major chemicals were bisphenol A (BPA) (account for 35%), methyl paraben (MeP) (23%), and triclosan (TCS) (14%) in water, while BPA (43%), TCS (37%), and MeP (14%) in sediment. Significant correlations were found between most of the selected EDCs, especially MeP and TCS both in water and sediment ($p < 0.01$), indicating that these chemicals were in common sources and widely usage. After calculation, our results indicated that about 43.1 metric tons target substances flowed into Liuxi river annually based on the 89 primary stream. The ecological risk assessment showed that TCS was the most dangerous compound to aquatic organisms with average HQ = 1.57 (up to 11.5) in river water and average HQ = 0.74 (up to 3.36) in sediment. And the possible joint toxic effect of selected chemicals showed that aquatic organisms were severely exposed to diverse EDCs. This study suggested that compared to the main rivers, the endocrine disrupting chemicals in streams deserves more attention.

WE129

Occurrence, distribution and fate of pharmaceuticals as chemical markers of contamination from urban sources in the vulnerable area of the Ebro Delta (Spain)

M. Celic, M. Gros, Catalan Institute for Water Research ICRA; M. Farre, IDAEA CSIC Barcelona; D. Barceló, M. Petrovic, Catalan Institute for Water Research ICRA

The Ebro Delta and the upper part of the Ebro basin have widely been exposed to contamination from both agriculture and urban sources, being wastewater treatment plant effluent discharges the dominant contamination pathway. In order to determine the distribution and impact of contamination from urban sources in the vulnerable area of the Ebro Delta, water and sediment samples were collected at different sites, covering reaches of the Ebro river located upstream and downstream WWTPs, lagoons, irrigation channels and coastal areas. Water samples were analyzed using solid-phase extraction (SPE) while sediment analysis was performed by pressurized liquid extraction followed by SPE purification. The occurrence of 81 pharmaceutical compounds in the extracts was determined by ultra-high performance liquid chromatography coupled to tandem mass spectrometry, using a hybrid triple quadrupole–linear ion trap instrument (UPLC–QqLIT–MS/MS). In order to assess seasonal variations, distribution and fate of pharmaceuticals in the river-estuary ecosystem, three sampling campaigns in 2016 were performed, covering autumn, winter and spring-summer. Fifty seven and thirty five out of 81 pharmaceuticals were found in water and sediment samples, respectively. Analgesics/ant-inflammatory, lipid regulators, cholesterol lowering statin drugs and antibiotics were the most frequently detected pharmaceuticals, with the highest concentration found in river water, while the lowest concetration were found in sea water. The occurrences of pharmaceuticals detected in sediment samples showed lower frequency of detection than in water. Nevertheless, some compounds were only found in sediments, and not in water, such as the synthetic glucocorticoid (dexamethasone), the antidiabetic (glibenclamide) and the diuretic (furosemide). Salicylic acid was the most ubiquitous quantified compound in sediment, with a maximum concentration of 18.2 ng g^{-1}. These results pointed out that pharmaceuticals are widespread pollutants in coastal environments and that WWTP effluent discharges are the main source of contamination by these substances in the Ebro Delta. Results also revealed that sesonal distribution of target compounds was affected by the river flow. Thus, concentrations of selected pharmaceuticals in samples collected during dry seasons were generally higher than those detected during the wet season, due to lower dilution factors.

WE130

Occurrence of pharmaceuticals and personal care products, and their associated environmental risks in a large shallow lake in north China

h. Zhou, P. Zhang, China Institute of Water Resources and Hydropower Research IWHR; k. li, China Institute of Water Resources and Hydropower Research

Eighteen selected pharmaceuticals and personal care products (PPCPs), consisting of five non-antibiotic pharmaceuticals (N-APs), four sulfonamides (SAs), four tetracyclines (TCs), four macrolides (MCs), and one quinolone (QN) were detected in water, pore water, and sediment samples from Baiyangdian Lake, China. A total of 31 water samples and 29 sediment samples were collected in March, 2017. Caffeine was detected with 100% frequency in surface water, pore water, and sediment samples. Carbamazepine was detected with 100% frequency in surface water and sediment samples. Five N-APs were prominent, with mean concentrations of 4.90–266.24 ng/l in surface water and 5.07–14.73 μg/kg in sediment samples. Four MCs were prominent, with mean concentrations of 0.97–29.92 ng/l in pore water samples. The total concentrations of the different classes of PPCPs followed the order: N-Aps (53.26%) > MCs (25.39) > SAs (10.06%) > TCs (7.64%) > QNs (3.64%) in surface water; N-APs (42.70%) > MCs (25.43%) > TCs (14.69%) > SAs (13.90%) > QNs (3.24%) in sediment samples, and MCs (42.12%) > N-APs (34.80%) > SAs (11.71%) > TCs (7.48%) > QNs (3.88%) in pore water samples. The geographical differences of PPCP concentrations were largely due to anthropogenic activities. Sewage discharged from Baoding City and human activities around Baiyangdian Lake were the main sources of PPCPs in the lake. An environmental risk assessment for the upper quartile concentration was undertaken using calculated risk quotients, and indicated a low or medium high risk from 18 PPCPs in Baiyangdian Lake and its five upstream rivers.

WE131

Occurrence of perfluorinated compounds in air, water, soil, sediment, and fishes from the Asan Lake region, South Korea

J. Lee, Y. Lee, J. Lee, Seoul National University; S. Kim, Eulji University; M. Kim, Seoul National University / Department of Health Science; Y. Kho, Eulji University; K. Zoh, Seoul National University / Department of Environmental Health

Perfluorinated compounds (PFCs) are known to be endocrine disrupting chemicals and can cause adverse effects on human health and environment. In July and October 2017, ambient air (n=2), fresh water (n=24), sediment (n=24), soil (n=24) and freshwater fish (n=27) samples were collected in Asan lake region, and the levels of PFCs in samples were determined. The Asan lake is man-made lake with freshwater between Pyeongtaek and Asan cities in Gyeonggi Province, and provides water for nearby industrial complex and agricultural areas. Two large streams join the lake and there are many industrial complexes near the streams. To analyze 16 PFCs, 2 samples were taken at each stream and 8 samples were taken at main lake. Analyses were carried out using LC-MS/MS after solid-phase extraction. The results showed that concentrations of ∑PFCs were ND~19.6 pg m^{-3} (for $\sum_{16}$ PFCs) in air, ND~447.8ng L^{-1} (for $\sum_{16}$ PFCs) in water, ND~9.7 ng g^{-1} dry weight (dw) (for $\sum_{16}$ PFCs) in sediments, ND~7.7 ng g^{-1} dw (for $\sum_{16}$ PFCs) in soil, and ND~35.0 ng g^{-1} dw (for $\sum_{19}$ PFCs) in the fish, respectively. The predominant species among the PFCs were perfluorooctanoic acid (PFOA) in air, perfluoropentanoic acid (PFPeA) in water, perfluorooctanesulfonic acid (PFOS) in sediment, soil, and fish, respectively. PFOS and PFDA were detected in all water, sediment, and fish samples. In air, 85% of total PFCs mainly existed as the gas phase compared to particulate phase. In water and sediment, higher levels of some PFCs were observed at the confluence of two streams, implying that anthropogenic activities from industrial complex in the upstream are the main source of contamination. Our study first reported the data of 16 PFCs levels in multimedia environment including air, water, soil, sediment, and fishes in Korea.

WE132

Seasonal changes in water and sediments' microplastics in a Mexican estuary (Tecolutla).

L. Sánchez Hernández, P. Ramirez Romero, U.A.M. Iztapalapa / Hidrobiologia

Microplastics (MP) are persistent contaminants that measure less than 5 mm, they have additives that are vectors of other POPs and metals, which can cause deleterious effects to the organisms that ingest them. MP can increase the temperature and decrease the sediments permeability. On the other hand, plastic particles are efficiently transported through water. There are just a few MP studies in Mexican aquatic ecosystems, so the objective of the present work was to evaluate the seasonal changes in numbers, size, color and form of the MP present in water and sediment of Tecolutla's estuary. Water and sediment samples were collected in five different sites in three different climate seasons (northern storms, dry and rainy). In the laboratory water volume was measured and filtered through a cellulose (Whatman #40) filter, which was later dried at 50 °C for 24 h. Sediments were dried at 50 °C, a 40 g subsample was taken and hydrogen peroxide (30 %) was added to disintegrate all organic matter, followed by a zinc chloride solution (ρ= 1.5 g/l) to float the MP particles. Later the solution was decanted through a cellulose filter and dried at 50 °C for 24 h. MP particles on the filters were observed through microscope (Dissecting followed by digital Celestron) and photographs of the particles were obtain to measure them with ImageJ software. Validation of the polymer identity was done through Scanning electron microscopy. MP size in water range from 10 to 1,730 µm, and their presence was higher in the northern storms season, followed by dry and rainy seasons with the highest numbers in a small tributary. Sediments had more MP and higher numbers in the dry season, with size ranging from 30 to 2,500 µm with highest numbers in the boat dock. Black was the most abundant color in both matrixes followed by blue and red. Most MP were fibers. In conclusion MP were present in water and sediments year round, bigger particles were found in sediments and smaller in water. This is the first evaluation of MP in a Mexican estuary so continuing this type of research is important to understand the biological significance of their presence.

WE133

Simultaneous biodegradation of water treatment additives: Transformation and byproduct formation, impact of biocide shock dosing and salinity

T. Wagner, University of Amsterdam / IBED; J. Parsons, University of Amsterdam / IBED-ELD; A. Langenhoff, H. Rijnaarts, Wageningen University / Environmental Technology; P. de Voogt, University of Amsterdam / IBED

Securing the supply of fresh water to fulfil the demand of the rising world population is identified as one of the largest environmental challenges in the near future. The Water Nexus research program aims at developing integral solutions for problems with water scarcity in delta areas worldwide. A significant fraction of industrial fresh water uptake is used in cooling towers. Several treatment technologies such as reverse osmosis, electrodialysis and membrane distillation may facilitate the reuse of discharged brackish cooling tower water. However, cooling towers water contains different water treatment chemicals such as corrosion inhibitors, biocides and antiscalants that hamper the optimal functioning of the treatment technologies by, for instance, membrane fouling. An interesting water pre-treatment option is the use of natural treatment systems such as constructed wetlands (CWs). Biodegradation is one of the main contaminant removal mechanisms in CWs. However, the biodegradation potential of CWs for many of the water treatment chemicals is not well understood. In this study, the simultaneous biodegradation of different representative water treatment chemicals by bacteria from CWs is explored. The representative water treatment chemicals consist of 1H-benzotriazole (corrosion inhibitor), DBNPA (biocide), glutaraldehyde (biocide), PEG (surfactant) and HEDP (antiscalant). The following questions are addressed: Does shock dosing with biocides affect the CW biodegradation potential for the target chemicals? What is the influence of different salinities on the biodegradation of the target chemicals? Which signature microbial transformation products are being produced by single target chemicals that can be used to monitor biodegradation in CW systems? Do possible transformation products show ecotoxicological effects? Does the simultaneous biodegradation of multiple water treatment chemicals result in the production of new possibly harmful crosslinked products?

WE134

Fate of organic micropollutants in a small river: hydrological and chemical processes

C. Glaser, Center for Applied Geosciences / Center of Applied Geoscience; M.E. Müller, Eberhard Karls Universität Tübingen; F. Faltermeier, Eberhard Karls Universität Tübingen / Center of Applied Geoscience; C. Zwiener, Environmental Analytical Chemistry, Center for Applied Geoscience, University of Tuebingen / Geosciences; M. Schwientek, Eberhard Karls Universität Tübingen; C. Zarfl, University of Tuebingen / Center for Applied Geoscience

Knowledge on pollutant transformation from laboratory experiments often fails to describe observations in the field. Thus, the CRC-CAMPOS aims to describe the fate and metabolism of anthropogenic pollutants on the landscape scale in different compartments in the Ammer catchment. This study is part of the subproject ‘Rivers’ and will identify and quantify the dominant processes from hydrology and chemistry which influence the fate of organic micropollutants in river systems. Field investigations take place in the Schönbrunnen River close to Tübingen (Germany) in the southwest of Germany, which is mainly influenced by agriculture. Salt tracer tests are combined with measurements of conservative ions and chemical target screening. Water mass balances are calculated based on the results of the salt tracer tests and provide information about hydrological loss and gain for the Schönbrunnen River. Dilution, mixing and dispersion processes can be identified with tracer tests and determine the residence time available for pollutant transformation. The quantification of the mass transport of pollutants in the river is possible by analysing conservative ions. This helps to derive and characterize chemical processes like photodegradation, sorption to particles or biochemical processes in biofilms from target screening data, mainly on pesticides. With the collected information from different disciplines, we get a larger picture about the pollutant mass transport in the Schönbrunnen River and adjoining compartments.

WE135

Occurrence of pharmaceuticals at extremely high concentrations in surface waters in Nigeria

O.M. Ogunbanwo, University of Leeds / Geography (Physical); P. Kay, University of Leeds / School of Geography; L. Brown, University of Leeds / School of Geography(Physical); J. Wilkinson, The University of York / Natural and Built Environments; A. Boxall, University of York / Environment Department; C. Sinclair, The Food and Environment Research Agency / Centre for Chemical Safety and Stewardship; R. Shabi, Lagos State Environmental Protection Agency

Pharmaceutical pollution of surface waters is increasingly recognized as a global problem, but to date, there have been no detailed studies from most African countries. In this study, the occurrence of 37 pharmaceuticals belonging to 19 therapeutic classes was studied in surface water and effluent in Lagos State, Southwest Nigeria. Samples were collected year-round from 22 surface water sites, and 27 compounds were detected at least once, many at extremely high concentrations. Maximum concentrations for a range of compounds, including trimethoprim, sulfamethoxazole, cimetidine, atenolol, and paracetamol were in the order of ³150 microg L^{-1}. The mean concentrations for sulfamethoxazole, trimethoprim, cimetidine, paracetamol, lidocaine, metformin, carbamazepine and atenolol were 55.90 microg L^{-1}, 38.69 microg L^{-1}, 31.612 microg L^{-1}, 24.99 microg L^{-1}, 22.55 microg L^{-1}, 20.98 microg L^{-1}, 15.35 microg L^{-1}, and 15.10 microg L^{-1} respectively. Venlafaxine has the lowest mean of 4.231 ng L^{-1}, other than the 10 compounds that were not detected. When compared with published data from around the world, these values are several orders of magnitude higher than most studies of pharmaceutical occurrence but similar to some other peak concentrations measured in developing countries such as China and India. Seasonal variations were observed for certain pharmaceuticals, i.e., antibiotics, paracetamol, tramadol, metformin, lidocaine, and carbamazepine which may be related to the environmental conditions that influence their degradation and seasonal usage or discharge. Pharmaceuticals are indispensable in human lives, although, their usage and discharge into the aquatic environment could lead to ecological problems and antibiotics resistance. Africa governments need to enact policies to clean up sewage discharges to rivers urgently.

WE136

Assessment of emerging contaminants in the L'Albufera Natural Park (Valencia, Spain)

D. Sadutto, University of Valencia / Environmental and Food Research Group, CIDE (GV, UV, CSIC); M. Andrés Costa, Universitat de Valencia / Environmental and Food Safety Research Group; R. Alvarez-Ruiz, University of Valencia; Y. Pico, University of Valencia / Medicine Preventive

Wetlands play a critical role in maintaining natural cycles and supporting a wide range of biodiversity. They regulate water quantity, groundwater recharge, and can contribute to regulating floods and the impacts of storms, and provide the fish and rice. The Albufera Natural Park is one of the most representative and valuable coastal wetlands in the Valencia Region and the Mediterranean basin. Covering an area of 21.120 hectares, it is located just 10 km from Valencia city. The surrounding population (> 1600000 inhabitants) has introduced a number of emerging contaminants that threats this wetland. In this study, 42 drugs of abuse and 45 pharmaceuticals have been studied in influents and effluents of 10 Wastewater Treatment Plants (WWTP), 7 irrigation channels and the Lake of L’Albufera de Valencia (Valencia, Spain). Three matrices were analyzed water, sediment and biota. The isolation and concentration were carried out by solid-phase extraction (SPE) and the contaminants were determined by ultra-high pressure liquid chromatography-tandem mass spectrometer (UHPLC-MS/MS). Cocaine as its major metabolite (benzoylecgonine), followed by cannabis as its Δ9-Tetrahydrocannabinolic acid, were the main drugs detected in water samples. Regard to pharmaceuticals, caffeine and ibuprofen were the main compounds obtained in these samples. Nevertheless, other pharmaceuticals were detected at high concentrations in all samples. In spite of this, and its non-complete removal in WWTPs, nowadays there are not enough knowledge about how the presence of these pollutants can affect to aquatic ecosystems, and specially living beings.

WE137

EFFECTS OF URBANIZATION PROCESS ON WATER QUALITY OF RIVERS ON THE SANTA CATARINA ISLAND, BRAZIL.

M. Barbosa Xavier, Universidade Federal de Santa Catarina / Biochemestry; C.H. Soares, Universidade Federal de Santa Catarina / Biochemistry Department

The metropolitan region of Florianopolis has undergone an intense urbanization process in recent years, which has modified the landscape and the quality of life in this region. The objective of the present study was to evaluate the water quality of the Itacorubi river in its estuarine region, in order to evaluate the anthropic changes occurring in the surroundings. Three sites were chosen, in which sediments and water samples were collected. The sediments were analyzed for the presence of sterols and pharmaceuticals by GC / TOF-MS after extraction with methyl tert-butyl ether. The water samples were analyzed with respect to the parameters: ammonia concentration, total phosphate, total phenols, fecal coliforms and sulfide, according to the methodologies described in Standard Methods (APHA). TOF-MS chromatographic analyzes of sediments and water samples were also performed on extracts obtained using SPE (Strata-X)/dichloromethane). The results obtained showed high concentrations of ammonia and total phosphate, besides high fecal coliforms. Between the analyzed sterols, cholesterol and derivatives such as coprostanol were identified at varying concentrations in the sediments of the several sites. Estradiol derivatives and drugs such as anxiolytics and remedies for sleep control were prominent in GC / TOF/MS chromatographic analyzes. The results confirmed the high contamination of the waters of the Itacorubi River by the discharge of domestic sewage. Ecotoxicological tests using fish are being conducted, including assay to assess genotoxicity.

WE138

Presence of emerging contaminants in sewage sludge and assessment of their environmental risk for the Albufera National Park, Valencia, Spain

M. Andrés Costa, Universitat de Valencia / Environmental and Food Safety Research Group; A. Cuñat, University of Valencia / Environmental and Food Safety Research Group, CIDE (UV, GV, CSIC); R. Alvarez-Ruiz, University of Valencia; Y. Pico, University of Valencia / Medicine Preventive

The wastewater generated by the human activity contains several anthropogenic contaminants, including emerging pollutants. Sewage sludge is generated during the depuration process in the wastewater treatment plants (WWTPs). The study of emerging pollutants present in this sludge are far limited comparing with the water, mainly because of the challenge that involve their high content in organic matter making difficult their handling, storage and analysis. In Spain, the 80% of this sludge is used in the agriculture sector as fertilizer, and the presence of pollutants could affect to the surrounding ecosystems. The sludge samples were from different treatment plants next to the Natural Park of the Albufera in Valencia, an area surrounded by 14000 hectares devoted to rice crops. Samples were extracted by liquid phase extraction (LPE) with mixture of EDTA-McIlvaine buffer (pH 4.1) and methanol, assisted by ultrasound. Then, supernatant was cleaned up by the solid phase extraction (SPE) with Strata^{TM}X cartridges and analytes were eluted with methanol at gravity flow. Once extracted, the analytes were identified by liquid chromatography-quadrupole time-of-flight mass spectrometry (LC-QqTOF-MS). As a result 50 compounds were identified, being the pharmaceuticals the main group of pollutants with 31 compounds. Human metabolites were the most relevant, present in all samples, including nucleotides (adenosine triphosphate), amino acids (phenylalanine) or peptides (leucine-phenylalanine). On the other hand, several compounds were tentative identified and are pending of confirmation. The results of this study demonstrate the interest of high resolution mass spectrometry to draw the profile of contaminants in solid complex matrices. Furthermore, the data obtained provides information about the potential risk of use the sewage sludge for agriculture. Continue researching is needed to assess the real environmental risk related to this sludge and their possible effects in the surrounding ecosystems.
Acknowledgements This work has been supported by the Spanish Ministry of Economy and Competitiveness and the ERDF (European Regional Development Fund) through the project CGL2015-64454-C2-1 "Análisis de riesgos y viabilidad económico-ambiental de la eliminación de contaminantes. Diseño una herramienta de apoyo a la toma de decisiones (Eco2TOOLS-DSS, http://http://www.eco2tools.es/)" R. Álvarez also knowledge the same institutions for his FPI grant BES-2016-078612.

WE139

CHLORINATED BENZENES IN FISHES FROM DONGTING LAKE

k. li, China Institute of Water Resources and Hydropower Research; P. Zhang, China Institute of Water Resources and Hydropower Research IWHR

Chlorobenzenes (CBs) are of worldwide concern due to their persistence, toxicity, bioaccumulation,and long-range transport. Hexachlorobenzene (HCB) and pentachlorobenzene (PeCB) are listed as persistent organic pollutants (POPs) by the United Nations Environment Program (UNEP) . CBs production in China accounts for more than 50% of the worldwide CBs production. The production\nof 1,2-dichlorobenzene (1,2-DCB), 1,4-dichlorobenzene (1,4-DCB) and 1,2,4-trichlorobenzene (1,2,4-TCB) in China was 12000, 30000 and 1000 tons in 2003, respectively. HCB has never been used as pesticide in China, but it was still produced as an intermediate of pentachlorophenol in Tianjin Dagu Chemical Company until 2003 with a production quantity of about 2000 tons/yr. CBs have been detected in water, sediment, soil and sewage sludge. However, reports on CBs in aquatic organisms-especially the aquatic organisms in typical epidemic areas of schistosomiasis prevalence in China-are lacking. The release of CBs from the production and use of Sodium pentachlorophenate (Na-PCP) has been identified as one of the most important sources. Dongting Lake is the second largest fresh water lake of China, which is also an area with most widely distributed oncomelaniahupensis and has the most severe schistosomiasis epidemic situation in China. Na-PCP has been sprayed as molluscicide in Dongting Lake from 1960s to 1990s, it was estimated that over 9.8×106 kg of Na-PCP had been devoted into the lake; CBs were also carried into the lake with using of Na-PCP. The aims of this study were to investigate\ncurrent contamination status, distribution of CBs in fish from Dongting Lake.

WE140

Occurrence of bisphenol A in Mediterranean mussels (Mytilus galloprovincialis) sampled from the north Adriatic coastal waters (Slovenia)

V. Cerkvenik Flajs, University of Ljubljana, Veterinary Faculty / Veterinary Faculty; I. Fonda, Fonda d.o.o.; M. Gombač, University of Ljubljana / Veterinary Faculty

From January to October 2015 in total 27 samples of Mediterranean mussels (*Mytilus galloprovincialis*) and 10 samples of sea water were collected along the Slovenian coast in the north Adriatic sea to be tested for the presence of bisphenol A. Samples were collected at three shellfish farms, at the open sea and also from the harbour of Koper. In addition, one sediment sample from the harbour of Koper was also collected. Homogenised mussel tissue, shells, and sediment were extracted with acetonitrile and purified with the two solid phase extraction (SPE) steps, using at first hydrophobic polystyrene-divinylbenzene (PS/DVB) copolymer Chromabond HR-X and secondly molecularly imprinted polymer (MIP) AFFINIMIP® SPE Bisphenol A. After adjustment of pH of water samples to the value of 5, these were also applied onto the MIP SPE sorbent. Sample extracts were analysed by isocratic (sea water) or gradient (tissue, shells, sediment) reveresed-phase HPLC using water and acetonitrile components of mobile phase, Hypersil Gold C18 (3 µm particle size) analytical column and fluorescence detection at excitation and emission wavelengths of 230 and 315 nm, respectively. Mean recovery rate values for mussel tissue, shells and sea water were 47%, 73% and 84%, respectively. Concentrations of bisphenol A in tissues of mussels from the farms (n = 20), open sea (n = 6) and a harbour (n = 1) were < 0.03 – 0.28 µg/kg w.w., < 0.03 – 0.46 µg/kg w.w. and 0.21 µg/kg w.w., respectively, while shells of mussels, from farms (n = 20), open sea (n = 6) and a harbour (n = 1) contained 0.01 – 0.3 µg/kg w.w., 0.04 – 0.27 µg/kg w.w. and 0.18 µg/kg w.w. of bisphenol A, respectively, Sea water at shellfish farms (n = 5), open sea (n = 4) and a harbour (n = 1) was contaminated with < 0.003 - 0.013 µg/l, 0.004 – 0.009 µg/l and 0.016 µg/l of bisphenol A, respectively. The observed concentrations indicate a relatively low contamination of the Slovenian coastal waters as a part of the north Adriatic sea, with bisphenol A, compared to available publications about Mediterranean mussels.

WE141

Toxicity of non-steroidal anti-inflammatory drug and the behavioural response in Juvenile Catfish

N.O. Erhunmwunse, University of Benin, Benin City, Nigeria / Animal and Environmental Biology; I. Tongo, University of Benin / Laboratory of Ecotoxicology and Environmental Forensics, Faculty of Life Sciences, Department of Animal and Environmental Biology,University of Benin, Nigeria; A. Muhib, University of Benin Benin City / Department of Environmental Management and Toxicology

The increasing levels of Pharmaceutical products in surface and underground water in third world countries is on the increase. We examined the toxicity of one pharmaceutical and personal care products (PPCPs) Acetaminophen on juvenile life stage of Africa Cat fish *Clarias gariepinus* using OECD 210 guideline. A 96hrs acute toxicity test protocol for African catfish was established and adopted using a static renewal assay. Fish were exposed for 96 hours assay to varying concentrations of 50, 100, 300, 500, 700 and 800mg/L. Mortality and behavioural changes were used as endpoint for acute test. Behavioural changes were characterized by restlessness, loss of body balance, gulping of air, rapid up and down movements. Estimated LC_{50} value was 358.80mg/L and the derived safe concentration value was 35.80mg/L. With survival from the range Finding Test, NOEC was 100mg/L and LOEC was 150mg/L. No acute toxicity effects were observed for concentrations below < 100mg/L. The 24, 48, 72 and 96hr median lethal concentration LC_{50} values of Acetaminophen was 800, 700, 594.5 and 358.80mg/L respectively.

WE142

Reproductive and maternal effects of Tamiflu metabolites in medaka (Oryzias latipes)

L. Bing Heng, Y. Wu, I. Meng Ian, W. Chen, Department of Biomedical Science and Environmental Biology, Kaohsiung Medical University, Kaohsiung

Tamiflu is the most commonly used anti-influenza drug. Human intake Taimiflu and excrete the Tamiflu metabolites into the aquatic environment. The Tamiflu metabolites might pose a potential risk to aquatic organisms. The purpose of this study was to assess the reproductive effect of medaka (*Oryzias latipes*) under long-term Tamiflu metabolite exposures. This study carried out the 56-day long-term toxicity, 14-d reproduction, and 21-day hatchability trial bioassays to observe the survival, growth, and egg production of the adult medaka, and hatchability of embryo, and larvae body length of F1 medaka under the Tamiflu metabolite exposure concentration (0, 0.3 and 90 µg/L). Results showed that the survival and growth rates of adult medaka were no significant difference between the control and exposure groups. However, the egg production and F1 hatching rate of 90 µg/L exposure group had a downward trend compared with control group, but there were no significant decrease. This study found that larvae body length of exposure groups were significantly shorter than that of control group. This study concluded that Tamiflu metabolite could have a significant impact on larvae growth development.

WE143

Earthworms (Eisenia fetida) response to chronic exposure to triclosan

J. Zaltauskaite, Vytautas Magnus University / Department of Enviromental Sciences; D. Miškelytė, Vytautas Magnus University

Triclosan (TCS) is a broad-spectrum antimicrobial and antifungal agent extensively used in industrial, household and personal care products. TCS widespread use has resulted in its introduction into environment and it has already been detected in surface waters, sediments, soil, living organisms and humans as well.The aim of the present study was to determine the response of *Eisenia fetida* earthworms to chronic triclosan exposure. Earthworms *E. fetida* were exposed to 10-750 mg kg^{-1} of triclosan in soil for 56 days. The impact on survival, growth rate, reproduction and antioxidative system was evaluated. TCS severely reduced the growth rate of *E.fetida* and reproduction. Chronic exposure to TCS in the soil induced a significant increase in the activity of antioxidative enzymes and malondialdehyde concentration.

WE144

Predicting the fate of pharmaceuticals during wastewater treatment and crop irrigation with reclaimed wastewater

M. González García, C. Fernández-López, UCAM; F. Polesel, Technical

University of Denmark (DTU) / DTU Environment; S. Trapp, Technical University of Denmark DTU / DTU Environment
Wastewater represents an alternative option for use in urban areas, industry and, especially, agriculture. Pharmaceuticals may undergo incomplete elimination in wastewater treatment plants (WWTPs) and are found in reclaimed wastewater, possibly being uptaken in crops following wastewater irrigation. Among commonly consumed crops, vegetable production uses considerable amounts of reclaimed wastewater for irrigation. In this study, we investigated the fate of pharmaceuticals compounds in a wastewater treatment plant (WWTP) equipped for the elimination of carbon and nutrients. The primary treatment consists of a screen, an aerated bean extraction tank and a primary clarifier. The biologically treated wastewater from the conventional activated sludge is filtered through a layer of a continuously operating sand filter prior to being disinfected by ultraviolet radiation. Effluents are used for irrigation in agriculture. We used the simulation tool “Activity SimpleTreat - fate model for ionics in wastewater treatment plants” [1] to predict the fate of pharmaceuticals compounds in the municipal WWTP. Model parameters were adapted to the situation at site. Chemical data were estimated using ACD/i-Lab. Model predictions were verified with measurements from a monitoring campaign in the WWTP. Results showed a high measured removal efficiency of Diclofenac, Ibuprofen and Ketoprofen concentrations in the WWTP and the simulation tool confirmed the same conclusion. As to uptake in lettuce, empirical results were compared to simulation outcome. For plant uptake prediction, a new steady-state model with translocation and phloem flow was applied [2]. Addition of phloem transport was necessary because the investigated compounds include weak acids (pKa 4 to 5), such as ibuprofen and naproxen, which undergo ion trapping in the alkaline phloem fluid (pH 8). The preliminary results with the new steady-state model, showed the uptake capacity of pharmaceuticals in different tissues of lettuce. The assimilation and distribution of pharmaceuticals compounds in the edible part of the lettuce leaves and the subsequent passage to the harvested plant parts is investigated. [1] Franco A. 2011. Activity SimpleTreat - fate model for ionics in wastewater treatment plants.
homepage.env.dtu.dk/stt/Homepage%20anf/Website.htm [2] Trapp S. 2017. New release dynamic (numeric) coupled soil-plant uptake model for monovalent ionics, homepage.env.dtu.dk/stt/2017Release_Plant_Model/index.htm

WE145

Exposure Assessment of Residual Organochlorine Pesticides (OCPs) in Orchard Soils and Fruits in Korea

B. Park, RDA / Chemical Safety; S. Lim, National Institute of Agricultural Sciences,RDA; G. Choi, National Institute for Agricultural Science, RDA; S. Ryu, J. Park, International Institute Science, RDA
Residual organochlorine pesticides (OCPs) are chemical substances that are resistant to environmental degradation chemical, biological and photolytic process, and are bioaccumulated with potential significant impacts on human health and the environment. OCPs were designated as persistent organic pollutants (POPs) by the international community at the Stockholm Convention on Persistent Organic Pollutant. This study was conducted to investigate the OCPs residue in orchard (grape, peach, apple, and pear) soils and fruits. Extraction and clean-up method for the quantitative analysis of OCPs was developed and validated by gas chromatography (GC). The method was established using the modified QuEChERS method for OCPs in orchard soil and grape, peach, apple, and pear. Recovery and limit of detection (LOD) of OCPs in orchard soils and fruits were 74.4-115.6 and 74.7-92.43%, 0.04-0.08 and 0.2-0.4 ug/kg, respectively. The precision was reliable since RSD percentage (0.5-3.5 and 1.6-4.8%) was below 20, which was the normal percent value. The residue of OCPs in orchard soils was analyzed by the developed method, and endosulfan sulfate, 2,4-DDT, 4,4-DDT, 4,4-DDD, and 4,4-DDE were detected at 11.3-444.9, 2.2-31.9, 4.5-863.1, 1.9-48.0, and 2.3-119.3 ug/kg, respectively. But OCPs in grape, peach, apple, and pear were not detected in all samples. These results showed that the residue in orchard soil were lower level than bioaccumulation occurring.

WE146

PhytoCOTE project: Assessment of organic and inorganic contamination in vineyard soils

M. Pierdet, LPTC EPOC UMR5805; J. Gaillard, Université de Bordeaux / EPOC UMR 5805; M. Dévier, University of Bordeaux / EPOC / LPTC UMR 5805 CNRS; L. Denaix, INRA BORDEAUX; H. Budzinski, University of Bordeaux
Viticulture is one of the agricultural crops that uses the most important quantities of pesticides in France, in particular fungicides. These regular inputs may lead to a long-term contamination of ecosystems and thereby affect fauna and flora. Different processes in soils play a role in pesticide retention and transfer. In order to improve the knowledge about the evolution in time and scale of different chemical contaminants within different soil types, a state of contamination level in soil surfaces and a characterisation of trace element availability were assessed. 53 plots with important pedological diversity were sampled over the 0-15 cm horizon. The soils were characterised (organic matter, Fe and Al oxyhydroxides, CEC, granulometry, pH) and total copper, cadmium, lead, zinc and 205 organic molecules were measured. The characterisation of trace element availability was performed using passive samplers (Diffusive Gradients in Thin films). A copper contamination due to past and current uses of Bordeaux mixture (copper sulphate) has been put in evidence on the experimental site (until 197 mg/kg of dry soil). Concerning organic pesticides, a high diversity of molecules at different levels of concentration were found depending on crops. The results of the analyses will allow to show if: (1) the copper contamination level plays a role on the molecule degradation and contamination level; (2) the soil physical and chemical parameters play a role on the molecule degradation and on the copper and molecule retention; (3) the past and current soil uses impact the contamination levels.

WE147

Analysis and Assessment of Organic Contaminants in Materials Spread on Land in Scotland

E. Stutt, WCA Environment Limited; I. Wilson, G. Merrington, wca
Due to improvements in analytical capability increasing numbers of synthetic chemicals are being found in organic materials that may be recycled to agricultural land such as sewage sludge, animal manures, compost and digestate. Commonly occurring contaminants include pharmaceuticals, veterinary medicines, personal care products and persistent organic pollutants. Application of suitable organic materials to land is an attractive and apparently sustainable option that offers a range of agronomic and environmental benefits. However, there is a balance to be struck between the benefits of application to land and potential risks, such as the possibility of human and environmental health effects from trace constituents. It is critical though that consideration of this exposure pathway and any resulting regulatory decisions are risk-based and made using robust evidence and science. Previous assessments of risks posed by contaminants in materials applied to land generally have several limitations in that the analysis of materials such as sewage sludge, have not been made for the material directly prior to application to land and the assessments often have to make use of data from a different geographical locations and regulatory jurisdictions with mismatches in chemicals management policy. This project is seeking to address these issues on behalf of the Scottish Environmental Protection Agency by undertaking representative sampling and analysis for priority organic contaminants in a range of different organic materials that are frequently spread on land in Scotland. As a large number of organic compounds (in excess of 200) have now been identified in materials that are applied to agricultural land the first phase of this project has been to undertake a risk screening exercise. The purpose of this has been to identify organic chemicals likely to persist and/or bioaccumulate and to prioritise substances that are considered to pose a risk to human health or the terrestrial environment under reasonable worst-case assumptions for spreading. Results will be presented from the analysis of priority chemicals in organic materials prior to spreading and the data will be used to undertake a refined risk assessment and to calculate a maximum safe spread rate for each material considered for application to land.
Acknowledgement - The authors thank the Scottish Environmental Protection Authority (SEPA) for funding this work

WE148

Microplastics in Agriculture Soil.

K.B. Olesen, Aalborg University / Department of Civil Engineering; N. van Alst, Aalborg University / Civil Engineering Department; A. Vianello, Aalborg University / Civil Engineering Department - Section of Water and Environment; M. Simon, Aalborg University; F. Liu, J. Vollertsen, Aalborg University / Civil Engineering Department
Microplastic is an environmental pollutant of worldwide concern. However, neither microplastic concentrations nor their sources or sinks are completely known. Infrared spectroscopy such as micro-fourier transform infrared (µFT-IR) spectroscopy and attenuated total reflectance (ATR) enables a reliable identification and quantification of microplastics. Studies show the tendency of microplastic accumulation in wastewater sludge. This sludge is used as fertilizer in agriculture farming. This study focuses on the occurrence of microplastics in the size range 5000-10 µm in soils that received wastewater sludge as fertilizer. It presents the methods of sample preparation and presents field data. In Sweden 3 fields were sampled. Monitored amounts of sludge fertilizer have been spread over a period of 35 years. The fields have either received 3 tons/year, 1 tons/year or no sludge fertilizer. 40 kg of soil were sampled from each field. The microplastic concentration is in general low; therefore the plastic needs to be extracted from other materials present. Due to the large size range of interest, two different IR techniques are applied for microplastic identification. This requires two different plastic extraction methods. Therefore, two sample protocols were developed – < 500µm and >500µm. < 500µm More than 500g of soil was dried and sieved through a 500µm metal sieve. To remove the inorganic fraction a gravimetrical separation was used. For a sample of this size a custom made aerator-device was built. The sample was agitated with air for 1 hour in $ZnCl_2$ (density of 1.7 g/cm^3). After 2 days the valve in the top chamber was closed and $ZnCL_2$ was drained so the top chamber could be removed. The fluid from the top chamber was filtrated over a 10 µm steel mesh. The device was refilled with $ZnCl_2$ and the agitation sequence was repeated. To remove the organic fraction the filtered material was treated with enzymes for several days and oxidised with H_2O_2. The remaining particles were suspended in ethanol and a subsample was deposited on a window and scanned by a state-of-the-art µFT-IR Imaging system (128x128 pixel Focal Plane Array (FPA) microscope detector). >500µm 10 kg of soil was wet-sieved through an 8mm, 6mm, 4mm, 2mm, 1mm and 500µm sieve. After the soil was dried it was floated in a

$ZnCL_2$ solution. All floating particles were collected and individually analysed under a light microscope. Selected particles looking like plastic was analysed on the ATR.

WE149

Novel Analytical Strategies for Anthropogenic Compounds in Plants: Vegetable Biomonitors for Contaminants in the Environment

R. Wahman, J. Grassmann, Technical University of Munich; P. Schroeder, Helmholtz Zentrum Munchen / Microbe Plant Interactions; S. Bieber, Technical University of Munich / Chair of Urban Water Systems Engineering; T. Letzel, Technical University of Munich

Plants play an important role in the maintenance of life. Besides providing us with food, plants are capable of cleaning the environment, i.e. water, from compounds like diclofenac, which occur in waterbodies in concentrations up to µg/L levels. The assilimated compounds are not excreted by the plants but stored in vacuoles. This project will focus on whether plants can eliminate pollutants from the environment and whether plants are capable of metabolizing the pollutants and to detoxify them. These two points already have been partially clarified in phytoremediation research. However, a major problem related to this kind of research is not concerning the plant metabolite pathways, although knowledge about those pathways, the involved enzymes and the resulting transformation products is essential. Thus, our major goal was to figure out whether the biological degradation pathways can be reflected by the analytical data obtained from polarity extended RPLC-HILIC-MS analysis. There are several important research fields which give an original contribution to investigate the secondary metabolites in leaf and root extracts of various plants. This study aims to contribute to this growing area of research by exploring new or modified secondary metabolites that appear after addition of pollutants. To the best of the authors knowledge, no research is available up to now that surveyed the changes in the plant metabolite pathways of constructed wetland plants (CWP). Moreover, along with growth in CWP, due to possibly accumulated contaminants there is increasing concern about how those plants must be treated further, i.e. which points have to be considered regarding their disposal. We will provide a conceptual theoretical framework based on analysis of different plant extracts before and after exposure to different pollutants using novel RPLC-HILIC/ToF-MS technique. The plants were exposed in the laboratory to different pollutants. Initially, the prevalent diclofenac was chosen. After the plants reached their maturity, they were exposed to pollutants for a few days. To establish the concept at the beginning comparably high concentrations of pollutants were applied. Finally, we will be able to provide an open access database of plant metabolites (PHRAGMITES-IDENT) and implement it into an analytical platform constructed earlier (FOR-IDENT; see https://water.for-ident.org/). This work is supported by the Bavarian State Ministry of the Environment and Consumer Protection.

WE150

Pharmaceuticals uptake by spinach from seven soils mixed with sewage sludge

R. Kodesova, A. Klement, Czech University of Life Sciences Prague / Dept of Soil Science and Soil Protection; O. Golovko, University of South Bohemia in Ceske Budejovice / South Bohemian Research Center of Aquaculture and Biodiversity of Hydrocenoses; A. Nikodem, Czech University of Life Sciences Prague / Dept of Soil Science and Soil Protection; O. Koba, University of South Bohemia in Ceske Budejovice / South Bohemian Research Center of Aquaculture and Biodiversity of Hydrocenoses; M. Fer, Czech University of Life Sciences Prague / Dept of Soil Science and Soil Protection; R. Grabic, University of South Bohemia in Ceske Budejovice / South Bohemian Research Center of Aquaculture and Biodiversity of Hydrocenoses

This study was focused on a mobility of pharmaceuticals from sewage sludge in soils and their uptake by plants. Soil samples were taken from top horizons of seven different soil types (Stagnic Chernozem Siltic, Haplic Chernozem, Greyic Phaeozem, Haplic Luvisol, Arenosol Epieutric, Haplic Cambisol, Dystric Cambisol). Sewage sludge samples were taken from two wastewater treatment plants. Two experiments were performed. a) Soils mixed with sludge were packed in plastic columns, humidified to a value close to a field water capacity and 14 days incubated under laboratory conditions. Next, a ponded infiltration was applied and cumulative water outflow and solutes discharge from the bottom were measured. b) Spinach (*Spinacea oleracea* L.) was planted in soils mixed with sludge packed in plastic columns under greenhouse conditions. The amount of pharmaceuticals in plant parts (i.e., roots and leaves) was evaluated after harvesting. Compounds' discharges as well as their root uptakes were soil and sludge dependent. In general, mostly larger discharges were observed form the Arenosol Epieutric and Cambisols. Mobility of compounds depended on their sorption affinity to particular soil, but also on the contents of a dissolve organic matter in the seeping solutions. Measured concentrations in spinach showed selective uptake of mobile pharmaceuticals in soil water. For instance sertraline and carbamazepine were found in the discharged solutions and also in spinach. On the other hand, relatively large amount of fexofenadine and venlafaxine was found in the discharged solutions but very low or negligible concentrations were measured in the spinach parts.

WE151

Will spent mushroom substrate application affect the dissipation and plant uptake of phthalate esters?

J. Gao, Institute of Soil Science, CAS / Key Laboratory of Soil Environment and Pollution Remediation, CAS; F. Zhu, Institute of Soil Science CAS

To investigate whether spent mushroom substrate (SMS) amendment was an appropriate way to reduce di(2-ehylhexyl) phthalate (DEHP) and di-*n*-butyl phthalate (DnBP) contents in soil and whether SMS could reduce DnBP accumulation in bok choy (*Brassica rapa* subsp. *chinensis*).Microcosm and pot experiments were carried out to study the influence of spent *Agaricus bisporus* substrate application on DnBP and DEHP dissipation in soils and plant uptake of DnBP. Variations in soil pH and enzyme activities were determined. The concentrations of phthalate esters (PAEs) in soils, bok choy and atmosphere were examined with gas chromatography or gas chromatography–mass spectrometry. Adding sterilized or non-sterilized SMS can increase soil pH and urease activity, and non-sterilized SMS can promote soil laccase activity. The results show that the dissipation of DEHP is accelerated after incubation with SMS for 25 d, however little effect can be found with continuing incubation due to low DEHP bioavailability. In this research, SMS amendment exhibits no effect on DnBP dissipation in soils and DnBP accumulation in bok choy. It was proposed that atmospheric deposition of DnBP might be the main source of DnBP in bok choy in the study, since equivalent amounts of DnBP were detected in the vegetables grown in soils with or without DnBP spiking. This study indicates that the application of SMS as an organic fertilizer is less likely to affect the fate of PAEs in soils, and proper strategies should be conducted to reduce PAE levels in atmosphere to control PAE contamination in vegetables.

Plants: predicting and assessing direct, indirect effects and recovery of plants from chemical stress (P)

WE152

Experiences of demonstrating aquatic plant recovery following herbicide exposure using sloped mesocosms

F. Pickering, Cambridge Environmental Assessments

Recovery is important when considering short term exposure of aquatic non-target plants to herbicides in edge of field water bodies. Mesocosm studies allow the effects on both individual species and communities to be assessed simultaneously. Unlike indoor laboratory studies, where test item concentrations are artificially maintained, mesocosm studies allow for a more realistic application and dissipation of test item. Therefore, mesocosm studies can assess direct and indirect effects whilst a test item is present, and also monitor the period after dissipation has occurred to assess possible recovery. Using our sloped mesocosms at Cambridge Environmental Assessments (CEA), we are successfully able to test up to ten macrophyte species, each with different physical structures and characteristics (e.g. rooted; emergent). Some traditional measures of plant health can be seen as subjective (e.g. necrosis scoring and macrophyte mapping). At CEA, additional metric parameters such as number of nodes, number of leaves and stem lengths are routinely measured throughout the in-life phase. As a result, a combination of parameters are measured for each species tested. This ensures that the endpoints of a study are suitably robust and can be used to assess recovery. Here we will use results from our past studies to share experiences of assessing plant health. We will discuss which endpoints are most sensitive, reliable and therefore most suitable for determining effects on each plant species. Finally we will discuss which of these measurements are most relevant to assess recovery.

WE153

Impact of plant density on the end points (ER50) determined for crop protection products in Non Target Terrestrial Plants Studies conducted to OECD 227, Vegetative Vigour

E. Paterson, A. Thompson, Dow Agrosciences; G. Meregalli, Dow AgroSciences Italia s.r.l. / Ecotoxicology; K. Ralston-Hooper, Dow Agrosciences; G. Karaiskou, Dow AgorSciences

Non Target Terrestrial Plant (NTTP) studies conducted to OECD 227 test guidelines are submitted as part of the registration process for plant protection products in Europe and the US. Current planting densities described in OECD test guideline 227 for Vegetative Vigour Studies, recommends 1-2 large plants per 15 cm pot, three medium size species per 15 cm pot and 5-10 small species per 15 cm pot. The minimum recommended number of plants per treatment in OECD 227 is 20, as a result vegetative vigour studies conducted to this guideline, regularly exceed 750 pots. These studies require a large amount of glasshouse space and are very labour extensive. Establishing, spraying, assessing and handling data from such large studies has its own issues and can increase the probability of errors occurring, limiting the numbers of studies which can be conducted in a given time frame; and increasing plant variability within the study making data interpretation more difficult. OECD 227 is a guideline and different planting densities, which are considered adequate to generate robust data, may be used. However it needs to be assessed whether these different planting densities in the pots can impact the final endpoints determined in the Vegetative Vigour studies and ultimately the risk assessment. Data will be presented for key test species planted at three densities to evaluate any impact on the Vegetative Vigour Study endpoints (expressed as ER_{50} values) used in the risk assessment.

WE154

Interspecific competition impact on organism responses to chemical stress : an SSD-based approach.

V. BAILLARD, LIEC (CNRS UMR 7360, Université de Lorraine); C. SULMON, ECOBIO, CNRS UMR 6553, Université de Rennes 1 / UMR CNRS ECOBIO; A. BITTEBIERE, LEHNA, CNRS UMR 5023, Université Lyon 1; C. MONY, ECOBIO, CNRS UMR 6553, Université de Rennes 1; S. Devin, LIEC, CNRS UMR 7360, Université de Lorraine / LIEC, CNRS; E. Billoir, Université de Lorraine, CNRS UMR 7360

Organisms are not alone in the environment. They interact with other individuals of same or other species in different ways. Interspecific competition is an important interaction for herbaceous plants in grass strips. Such vegetated areas generally act as buffer strips against pollutant flows and are thus submitted to various chemical exposures. However, competition is rarely considered in environmental risk assessment. To address this point, we tested whether competition modifies the way plants respond to herbicide (isoproturon) toxicity in an attempt to link individual tolerance of organisms and community dynamic. Then we investigated the impact of competition on species sensitivity distribution (SSD), a widely used community-level risk assessment tool that usually considers monospecific bioassays only. To do so, we exposed during 25 days 6 herbaceous species (representing varied isoproturon tolerance and competition ability) to 6 isoproturon concentrations (0 to 1.75µM) in presence and absence of a selected competitor, *Bromus erectus* (choice based on its high resistance to isoproturon and its high competitiveness). For each condition, 8 replicates were realized. After exposures, 10 different traits corresponding to morphological, biomass and physiological responses, as well as the response profile of 50 metabolites were quantified for aerial and underground plant parts, then representing respectively soft (easy to acquire) and hard traits. The consequent dataset generated was used to model plant responses depending on isoproturon concentration and competitor presence/absence. For soft traits, dose-responses curves were built for each species, in presence and absence of competitor for each endpoint to define (1) their sensitivity, (2) their relevance to assess toxicity, (3) how competition modify points (1) and (2). In parallel, metabolomic data were treated the same way, using a workflow created to handle high-throughput dose-response datasets from omics experiments. We then calculated toxicity values and built SSDs with and without competitor presence in an attempt to quantify competition effects compared to competitor-free ecotoxicological data. A final experiment, involving a complex assembly of the same 6 species under various isoproturon exposures was carried out to assess our model validity using community scale data.

WE155

How to consider recovery of aquatic plants in risk assessments?

U. Hommen, Fraunhofer IME; G. Arts, Wageningen Environmental Research (Alterra) / Environmental Risk Assessment; E. Bruns, Bayer AG, Division Bayer CropScience / Ecotoxicology; H. Krueger, EAG Laboratories

Exposure of non-target plants to plant protection products or other stressors can be restricted in time if the substances show fast dissipation, e.g. by degradation or transport. In such cases, the plants might recover if the effects are reversible. Neglecting the recovery potential in the risk assessment is definitely protective but might be over-restrictive resulting in for example unnecessary losses of crop yields. The recovery subgroup of the SETAC Plants Interest Group aims to review the different approaches to analyse recovery of plants and to make suggestions how recovery of these could be included in a risk assessment framework. In this presentation, we will focus on aquatic algae and macrophytes and the regulation of plant protection products in the EU. Experimentally, recovery of algae and macrophytes can be assessed in single species laboratory tests or micro- and mesocosm studies. In refined exposure laboratory toxicity tests, usually the recovery of the growth rate is assessed since the populations are kept in the experimental growth phase. In micro- and mesocosm studies, it is possible to analyse also recovery of abundance or biomass and potential indirect effects. The differences of these two options and their potential consequences for risk assessment will be discussed. Effect modelling can be used to extrapolate from empirical data to other exposure scenarios or species. However, while the simulation of refined exposure laboratory toxicity tests seems to be straightforward, the prediction of effects under field conditions is still challenging. In addition, the use of such models in the risk assessment requires clearer criteria on which magnitude and duration of effects can be considered acceptable.

WE156

Rimsulfuron toxicity and recovery in duckweed (Lemna minor)

M. Opincarne, University of Florida / School of Natural Resources and Environment; P.C. Wilson, Z. Li, University of Florida IFAS / Soil and Water Science

Rimsulfuron is an herbicide for which very little is known about its toxicity to aquatic macrophytes. This study was designed to evaluate the effects of rimsulfuron on the model aquatic macrophyte *Lemna minor* at low concentrations. This study also evaluated recovery by *L. minor* following a 5 day exposure period. Growth rates were measured at 1, 3, and 5 days following exposure to rimsulfuron-fortified 10% Hoaglands media at concentrations of 0, 0.0003, 0.0006, 0.00125, 0.0025, 0.005, 0.01, and 0.02 mg/L. After 5 days exposure, growth rates were significantly lower for rimsulfuron concentrations ≥0.0006 mg/L. Following the 5-day exposure period, plants were rinsed with deionized water and placed in fresh Hoagland's nutrient media. Impact on growth rate was measured as percent impact compared with growth of the control. Growth rates for 00006 mg/L were reduced 25.4% relative to the controls. Interestingly, a hormetic response was observed at the 0.0003 mg/L treatment concentration. In this case, the growth rate *increased* 16.7% relative to the control. Following exposure, significant reductions in growth rate were observed on days 3, 5, and 10. However, 15 days after removal from the rimsulfuron treatment solutions, growth rates recovered to control levels. While rimsulfuron exposure significantly reduced growth rates of *L. minor* at all concentrations ≥0.0006 mg/L, effects were found to be reversible. Rimsulfuron was fast acting, with toxicity observed 1 day following exposure. In contrast, a longer period of time was required for growth rates to recover to control levels.

WE157

Toxicokinetic/toxicodynamic (TK/TD) modelling - Increasing the realism in risk assessments for aquatic plants

S. Heine, Bayer Ag / Effect modelling; E. Bruns, Bayer AG, Division Bayer CropScience / Ecotoxicology; A. Solga, Bayer Ag; T. Preuss, Bayer Ag / Environmental Safety

For assessing the risk of plant protection products (PPP) to aquatic ecosystems, environmental concentrations of the active substance need to be estimated. Throughout Europe different approaches are used to predict these environmental concentrations. To characterize the risk to non-target organisms, such as aquatic plants, ecotoxicological thresholds derived from experiments with constant exposure to an active substance over several days are compared to the predicted maximum environmental concentration. Although, it can be deemed conservative to only consider the maximum concentration, there are plenty of cases where the risk assessment becomes overly conservative due to this practice. This applies particularly to assessments for lotic water bodies (streams) in which critical peak concentrations usually last for a few hours or days, only. Since some models for the prediction of environmental concentrations do not only deliver maximum concentrations but also temporally explicit exposure (exposure patterns), a more detailed and realistic assessment of exposure is possible. To also increase the realism on the effect side, either an ecotoxicological threshold from a refined exposure experiment is needed, or the effect of the predicted exposure pattern on the organism is investigated by ecological modelling. We propose TK/TD modelling as a powerful tool to evaluate effects of time variable exposure on aquatic plants. TK/TD modelling refers to linking effects to the internal concentrations in an organism instead of the external one and by this being able to consider time-variable exposure patterns. For characterizing risks of active substances by TK/TD modelling, it is necessary to adjust the approach to a specific substance. Adjusting in this context means defining TK/TD parameters to describe the uptake/elimination and the internal dose-response relationship. Besides defining the parameters, it is also necessary to validate them by using the parameterized TK/TD model and by comparing predictions of the model to measured data. In this work we present the validation/calibration results of a *Lemna* TK/TD model that was parameterized to describe the effects of different sulfonyl-urea herbicides. The results demonstrate that the TK/TD *Lemna* model with its specific parameterization is able to reliably predict effects. Using the TK/TD *Lemna* model allows to perform a more realistic environmental risk assessment and to link time variable-exposure to effects.

WE158

Assessing soil toxicity of methylparaben using plants and collembola

D. Kim, J. Kwak, Konkuk University; Y. An, Konkuk University / Department of Environmental Heath Science

Methylparaben is an endocrine disrupting chemicals (EDCs) and is contained in personal care products such as cosmetics and quasi-drugs. Methylparaben is known to have low toxicity to mammals, but there is no data on hazard assessment for soil ecosystem. Methylparaben was mostly removed in the sewage treatment process, but was detected in soils of various countries. In addition, there is a possibility that personal care products may leak into aquatic or soil environments if they are directly disposed in the environment without any treatment. Therefore, it is necessary to evaluate the hazard assessment of methylparaben in soil ecosystem. This study assessed the toxicity of methylparaben to plants (mung bean and rice) and collembola. Plants were exposed methylparaben from 0 to 400 mg/kg for 14 and 21days. In plant toxicity tests, shoot and root growth, root development, stomatal opening size, chlorophyll contents and photosynthetic factors were measured. In the collembola test, methylparaben was exposed at 0 to 500 mg/kg for 5 days and mortality was observed. The most sensitive endpoint in mung bean was identified as stomatal opening size, and no-observed effect concentration (NOEC) was 10 mg/kg. The most sensitive factor in rice was chlorophyll contents, and NOEC was under 10 mg/kg. The half-lethal concentration (LC50) value for collembola was 440.53 mg/kg. Methylparaben appears to have significant physiological effects on plants even at low concentrations. The results of this study can be fundamental for soil risk assessments of methylparaben. This study was funded by the Korea Ministry of Environment (MOE) as the Environmental Health Action Program (1485014458), and the Graduate School of Specialization for

managing information related to chemical risk.

WE159

Evaluation of phytotoxicity for Bisphenol A with new endpoint, phytoestrogen

D. Kim, J. Kwak, Konkuk University; Y. An, Konkuk University / Department of Environmental Heath Science

While endocrine disrupting chemicals (EDCs) are known as chemicals that show hormone-like action or inhibit hormones, the phytotoxicity assessment of EDCs does not have any specific toxic endpoints for these substances. The factors (growth, photosynthetic activity, chlorophyll, etc.) used to evaluate common toxic substances such as heavy metals are also applied to EDCs. These factors are not suitable for EDC materials, which have relatively low toxicity to organisms, and productivity, an indicator of the reproductive system associated with hormones, takes a long time to assess toxicity. Therefore, we tried to evaluate phytoestrogen, a new toxic endpoint for EDC materials, using bisphenol A. Meanwhile, bisphenol A is known as a representative EDCs used in the production of consumer products and in various industrial fields. While it is used widely for various purposes, the soil ecotoxicity of bisphenol A is limited. Therefore, we evaluated the toxicity of bisphenol A to plant (mung bean) using traditional endpoints and evaluated the applicability of phytoestrogen, a new endpoint for EDCs materials. This study was funded by the Korea Ministry of Environment (MOE) as the Environmental Health Action Program (1485014458), and funded by the Graduate School of Specialization for managing information of chemical risk.

WE160

Soil toxicity of DEHP and Nonylphenol on mungbean and rice

D. Kim, J. Kwak, Konkuk University; Y. An, Konkuk University / Department of Environmental Heath Science

DEHP is used as a plasticizer and insecticide. Especially, it can be used as plastic vinyl applied in farmland. Nonylphenol is used as surfactant, and 4-nonylphenol is most widely used substance with various isomers which are used as insecticide and detergent. Although DEHP and nonylphenol are likely to release into the soil environments, soil ecotoxicity data are currently limited. Ecotoxicological researches in soil were reported in only three cases of DEHP and four cases of nonylphenol. This study was conducted to evaluate the effects of DEHP and nonylphenol on the growth and physiological changes of mung bean and rice. The toxicity tests were conducted on 14 days (acute) and 21 days (chronic). Shoot growth was measured in 14 days-acute experiment and physiological factors including stomata opening size, chlorophyll contents, and photosynthetic activity were evaluated in the 21 day-chronic experiment. This study is meaningful because the soil toxicity of the two substances to the plants was conducted using various factors, and the results of this study can be fundamental for soil risk assessments of DEHP and nonylphenol. This study was funded by the Korea Ministry of Environment (MOE) as the Environmental Health Action Program (1485014458), and the Graduate School of Specialization for managing information related to chemical risk.

WE161

Toxicity of a glyphosate based formulation on phytoplanktonic green microalgae.

J.G. Perez, Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Biodiversidad y Biología Experimental, Buenos Aires; A. Magdaleno, Universidad de Buenos Aires / Facultad de Farmacia y Bioquímica, Cátedra de Salud Pública e Higiene Ambiental; M.d. Rios de Molina, Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Química Biológica, CONICET-UBA, Instituto de Química Biológica - Ciencias Exactas y Naturales (IQUIBICEN); A.B. Juarez, Universidad de Buenos Aires / Facultad de Ciencias Exactas y Naturales, Departamento de Biodiversidad y Biologia Experimental

The emergence of transgenic crops led to an increase in the use of glyphosate and its presence in different ecosystems is a worldwide problem. Although it was designed to inhibit the aromatic amino acids synthesis in plants, glyphosate exerts toxic effects on non-target organisms, probably through other mechanisms. Its entry into waterbodies means a risk for biota, particularly for the phytoplankton microalgae community that sustains aquatic food webs. In this work the effects of a glyphosate formulation (≈44% monopotassium salt of N-phosphonomethyl glycine) on the growth, chlorophyll content and oxidative stress parameters of 4 phytoplanktonic green microalgae were evaluated. Cultures of *Senedesmus acutus*, *Ankistrodesmus fusiformis*, *Monoraphidium contortum* and *Parachlorella kessleri* were exposed to increasing glyphosate concentrations (0 – 75 mg glyphosate/L) and kept at 24 ± 1 °C, under continuous agitation and illumination. After 96 h, growth, IC50, chlorophyll *a* content and oxidative stress parameters were evaluated. The glyphosate caused a significant decrease of chlorophyll *a* in *M. contortum* and *P. kessleri*, but not in the other two species tested. The growth of the 4 strains was negatively affected and regarding the IC50 values *M. contortum* was the most sensitive strain (3.37 mg/L), followed by *A. fusiformis* (6.50 mg/L), *S. acutus* (14.74 mg/L) and *P. kessleri* (41.75 mg/L). In order to evaluate the relationship between antioxidant defenses and sensitivity, we analyzed parameters of oxidative stress in the least and the most sensitive strains. The exposition to 2-4 mg glyphosate/L in *M. contortum* and 30-75 mg glyphosate/L in *P. kessleri*, caused increases of reactive oxygen species, lipid peroxidation (TBARS), reduced glutathione, superoxide dismutase, and glutathione transferase. The level reached of TBARS in *P kessleri* was 10 times lower than in *M. contortum*, while the levels of antioxidant defenses were 3.5 - 7 times higher. These results suggest that oxidative stress would be involved in the toxicity of the glyphosate formulation and that the differences in sensitivity between strains could be due to differences in their defense antioxidant levels. According to the EU Directive 93/67/EEC, the IC50 values estimated indicate that the glyphosate formulation assayed should be toxic for aquatic biota. Besides, results also warn about its possible effects on the composition of phytoplankton, which would put at risk the balance of the aquatic ecosystem.

WE162

Indicator, indigenous and invasive species: the need of risk-benefit considerations in PPP risk assessment?

G. Meregalli, Dow AgroSciences Italia s.r.l. / Ecotoxicology; C. Vaj, V. Zaffagnini, A. Carone, Dow AgroSciences Italia srl

Indicator species are the basis of the ecological risk assessment framework. Endpoints derived for these species are used in the risk assessment to evaluate the safety of, e.g. plant protection products (PPP) to non-target species. In certain cases indicator species are also indigenous species to a particular region (e.g. *Myriophyllum spicatum* in Europe). Invasive species are non-autochthonous species, accidentally introduced in a given region, which, in absence of their natural predators, often grow uncontrolled and overcome indigenous species, completely devastating the biodiversity of the habitats they colonise. The uncontrolled growth of these species can be also a threat to ecosystem functioning, e.g. altering oxygen balance in the case of the aquatic environment or shifting the prey/predator equilibrium. In cases where other control means are not possible, PPP could be employed to control the spread of invasive species. As an example, in the USA some herbicides have been authorised to control *M. spicatum*, which is an invasive alien species in North America. On the other hand, recently in Europe there have been reports of *Myriophyllum aquaticum*, a new alien invasive species genetically related to the indicator *M. spicatum*. In Piedmont (Italy), *M. aquaticum* has been observed in the Po River, threatening aquatic biodiversity. In addition also rice cultivation, a very important crop for the region, is at risk, due to the uncontrolled growth of *M. aquaticum* in canals and ditches feeding water to rice paddies. This species has been added as an invasive species to the black lists of Piedmont and of the European Union and it is being monitored and controlled with local initiatives. To prevent the irreversible degradation or destruction of natural habitats by non-autochthonous species, a timely intervention may be necessary, even if this may result in a temporal impact on indigenous desirable species. Programs to control invasive species could be designed to restore habitats degraded by the presence of alien species. The use of a risk-benefit analysis, which can help in the decision of whether PPP are suitable for use in such programs, will be discussed in light of the two abovementioned examples. \n

WE163

Auxinic herbicides: the impact of water plants' root measurements on the risk assessment

G. Gonsior, Eurofins Agroscience Services Ecotox GmbH

Data requirements for the registration of plant protection products in the EU Regulation 1107/2009 indicate that a test on a *Myriophyllum* species is necessary for auxinic herbicides. The OECD 239 water sediment test with *Myriophyllum spicatum* was developed to assess this species under realistic exposure scenarios comparable to natural conditions. In this testing methodology, shoot length, as well as, fresh and dry weights need to be recorded. The OECD 239 guideline requires that only a qualitative assessment of the roots is undertaken. Auxinic substances are known to exert their herbicidal activity by affecting growing tissues. As such, roots of *Myriophyllum* plants may be affected after exposure to auxins. The methodology described in the OECD 239 guideline can be adapted to include measurements of fresh and dry weights for whole plants, rather than just the shoots, thereby assessing, indirectly, also possible effects on the roots. However, it needs to be evaluated if an indirect quantitative assessment of the roots in the *Myriophyllum* studies with auxinic substances would result in significantly different endpoints that may impact the overall outcome of the risk assessment for these substances. Results will be presented to clarify if effects on the roots in a water sediment system are providing additional information relevant for the risk assessment.

WE164

Testing the emergent macrophyte, Glyceria maxima in a water-sediment system : Results of a ring-test with Isoproturon

J. Davies, Syngenta / Environmental Safety; G. Arts, Wageningen Environmental Research (Alterra) / Environmental Risk Assessment; K. Kuhl, Bayer AG - Crop Science Division; J. Kubitza, BASF; M. Ratte, ToxRat Solutions GmbH & Co. KG

Under EU pesticide regulation, regulatory tests are required for the aquatic macrophyte, Lemna, and two algal species for herbicides and plant growth regulators. Data requirements introduced under EU Directive 1107/2009 stipulate that further tests may be required for compounds which show selectively higher toxicity to either dicotyledonous or monocotyledonous plant species in terrestrial plant tests. In these cases, the recommended dicot and monocot species are Myriophyllum and Glyceria, respectively. OECD Test Guideline 239 for testing Myriophyllum spicatum in a water-sediment system was adopted in September 2014 and this test method has since been adapted to facilitate growth ofthe emergent, reed grass, Glyceria maxima. During 2016 and 2017, 15 laboratories participated in a ring-test with the herbicide, isoproturon against Glyceria. The objectives of this test were to establish suitable plant propagation techniques, to determine the required test duration, to characterize control variability and inform test design and to identify appropriate validity criteria. Results of this ring-test will be presented alongside progress on a second ring-test with the herbicide imazapyr, scheduled for Spring / Summer 2018.

WE165

Study of the toxicity effects of Cd, Ni and Zn on macrophytes, antioxidant responses and time for steady-state bioaccumulation under constant metal concentrations exposures

s. martinez, CONICET PRIET UNLU; M. Saenz, PRIET CONICET, National University of Luján; W.D. Di Marzio, CONICET-PRIET / PRIET

Heavy metals represent an actual environmental problem because the industrial and commercial uses of them are continuously increasing, bringing on a widespread contamination. Examples of human activities that contribute in heavy metal contamination are mining, smelting, fossil fuel combustion and industrial applications. Human activities may also indirectly cause changes in the environment that mobilize metals that were otherwise bound in stable forms, making them bioavailable. Aquatic bodies directly or indirectly receive pollutant discharges and metals dissolve in water and are easily absorbed by organisms. This study was aimed to evaluate the individual toxicity effect of Cd, Ni and Zn on a macrophyte and it bioaccumulation capability. Cadmium is a hazardous environmental pollutant and is toxic to most organisms. Nickel and Zinc are essential trace elements needed in the nutrition of plants. Nevertheless, over certain threshold they can present phytotoxic activity. Macrophytes are key elements in aquatic ecosystems. Here, we worked with the submerged free-floating plant *Ceratophyllum demersum*. Following the OECD 2014 guideline for sediment-free toxicity test, plants were exposed to a range of concentrations (1-16 Ni, 4-64 Zn or 0,5-8 Cd mg/L) and fresh weight, main shoot length and total shoot length were chosen as endpoints. For the bioaccumulation assays, plants were exposed to a constant metal concentration during a period of time until internal metal concentration reached a steady state. To make sure of a constant external metal concentration, a daily renewal of the media was carried out. Besides, the influence of these metals on antioxidative enzymes activity was evaluated for the three lower concentrations of each one. These enzymes are involved in the plant defense mechanisms activated by heavy metal exposure. Determination of catalase (CAT), guaiacol peroxidase (GPOX) and ascorbate peroxidase (APOX) were performed from the total plant mass. In the main, fresh weight resulted in the most sensible endpoint reaching an inhibition of almost 50% for 64 mg Zn/L and presenting significant inhibition for concentrations higher than 2 mg Ni/L and 1 mg Cd/L. Cd exposed plants over 1 mg/L presented sings of chlorosis and disaggregated easily at the higher concentrations. Metal uptake reaches the steady-state between days 11-14 in all cases. CAT activity at test concentrations remained near control values, while APOX and GPOX enzymes showed an increase indicating possible sublethal effects.

WE166

Physiological responses of Thlaspi praecox (Brassicaceae) to Ni hyperaccumulation

T.D. Mišljenović, K. Jakovljević, S. Jovanović, University of Belgrade / Faculty of Biology Institute of Botany and Botanical Garden Jevremovac; N. Mihailović, University of Belgrade / Institute for the Application of Nuclear Energy; V. Maksimović, University of Belgrade / Institute for Multidisciplinary Research; D. Mišić, University of Belgrade / Institute for Biological Research Siniša Stanković

Thlaspi praecox is a well known heavy metal hyperaccumulating plant species. The ability of *T. praecox* to hyperaccumulate Zn and Cd have been extensively studied, while data on Ni hyperaccumulation are scarce. Our aim was to bring more understanding to the physiology of *T. praecox* exposed to increasing concentrations of Ni. Seeds of *T. praecox* were collected from an ultramafic site on Mt. Maljen (Serbia). Two - week old seedlings were transplanted to a peat based substrate amended with increasing concentrations of Ni (250, 500, 1000 ppm). Plants were grown for 3 months under controlled conditions. Content of Ni in plants was analyzed by AAS, while phenolics, sugars and organic acids have been analysed using UHPLC/DAD/MS^2 or HPLC-PAD. No visible toxicity symptoms were observed during plant growth, and Ni did not affect biomass production at applied concentrations. A dose-dependent response of *T. praecox* shoots to applied Ni concentrations was recorded. Ni hyperaccumulation threshold of 1000 ppm was exceeded in the shoots at all treatments, and the highest Ni content was 6786 ppm. Calculated values of translocation factor (shoot/root ratio of Ni concentration) above 10 in all Ni treated groups indicated active translocation of Ni from roots to the shoots. At the highest applied Ni concentration, statistically signifficant reduction of total chlorophyl content and carotenoids were observed. Contents of phenolic acids and flavonoids were generally low, and were not signifficantly affected by increasing Ni concentrations. Effects of increasing Ni concentrations on the content of sugars and organic acids in shoots have also been analysed. Understanding the physiology of *T.praecox* exposed to Ni and its` Ni tollerance limits might be relevant for the potential application of this species in phytostabilization or phytoextraction technologies at contaminated soils.

WE167

Phytoextraction of heavy metals in Cienega of Tamasopo wetland, México, by Typha latifolia

C. Wong, C. Carranza, Universidad Autonoma de San Luis Potosi / Laboratorio de ciencias ambientales; A.J. Alonso, Universidad de Guanajuato / Departamento de Farmacia

Heavy metals are persistent inorganic toxic pollutants that come from diverse anthropogenic activities [1]. They can be easily absorbed by the roots of plants due to their relatively high mobility in the soil-plant system. Aquatic plant species are known to have great importance, forming a substantial component of the primary production in many aquatic ecosystems, especially in wetlands. Plants can remove and accumulate metals from the solution by phytoextraction; however, the metals can also be precipitated or eliminated from the solution by ion exchange or by adsorption on organic and inorganic compounds. Concentrations of heavy metals in aquatic plants depend both on metal speciation and on the species of plant absorbing the metal [2]. High concentrations of some trace metals in aquatic plants have led many authors to believe that they accumulate from water and/or from sediments; the uptake is influenced by several factors, such as temperature, pH, light and the presence of other metals in the water, all of which alter the uptake of heavy metals into the tissue [3]. Metal uptake by plants has three patterns:(1) true exclusion, in which metals are restricted from entering the plants; (2) shoot exclusion, in which metals are accumulated in the root but translocation to the shoot is restricted; and (3) accumulation, where metals are concentrated in the plant parts [4]. The present research examines the phytoextraction *in situ* of heavy metals by *Typha latifolia* to determine the concentration of these metals in the plant, water and sediments. The experimental procedure consisted of: 1) sampling of five sites of the Ciénega de Tamasopo wetland: 10 plants were collected per site of *T. latifolia*, 3 samples of 5% acidified water with HNO_3 and one sample without acidification for physicochemical parameters, one sample of the first 10 cm of sediment; 2) plants: washing, separation in roots and leaves and drying at 70 ° C for 18 hours in Lindberg / Blue stove; 3) grinding and spraying of root and leaves in analytical mill (KIKA Werke M20); 4) acid digestion with HNO_3 in plate at room temperature of root and leaves [5] and sediments; 5) quantification of heavy metals by ICP-MS in digestion and water column samples. The results show that *Typha latifolia* accumulate Mn>Zn>Cr>Pb>Cu>As>Hg>Cd in roots. This study aimed to gain a better understanding of the importance of aquatic plants such as *Typha latifolia* in heavy metal accumulation and detoxification mechanisms.

WE168

Heavy metal removal by aquatic plants

M. Saenz, PRIET CONICET, National University of Luján; J. Alberdi, priet conicet unlu; s. martinez, CONICET PRIET UNLU; s. curieses, priet conicet unlu; W.D. Di Marzio, CONICET-PRIET / PRIET

Release of heavy metals into the environment due to industrialization and urbanization is a great problem worldwide, due to their toxicity to many life forms. Aqueous waste from metal plating, mining operations, tanneries, smelting, alloy industries, and storage batteries are sources of metal contamination. Biological methods have been recommended as and effective alternative for removal and recovery of heavy metals from aqueous solutions. Aquatic plant biomass represents an abundant biological resource that possesses an immense capacity to accumulate heavy metals and therefore have been exploited worldwide in the field of wastewater treatment technologies. Aquatic plant species including free floating and submerged, as *Lemna*, *Spirodella*, *Ceratophyllum* and *Myriophylum*, have shown potential for metal removal from wastewater. The aim of this study is to evaluate the efficiency and capacity of different species of aquatic macrophyte in removing heavy metals from an artificial wastewater in a multi-metal solution. The initial whole ecotoxicity of a multi-metal system composed by Cd, Ni and Zn was assessed by growth inhibition test with the green algae *P. subcapitata*, acute toxicity test with *D. magna* and *ex vivo* cytotoxicity test with *E. foetida* coelomocytes. An experiment was set up for 10 days, by the addition of 10 grs of fresh weight of plants from different species mentioned above, in the metals solution. Previous works have shown than metal uptake rates were faster within the first 48 hours, and decrease with time and with metal concentration solution, so, at this time and at 3 and 5 days, respectively, plants were removed and new plants were placed in the same multi-metal system. At these time intervals, samples of solution and plant were taken for metal determination. The harvested plants were dried in vacuum oven and a microwave acid digestion were carried out. Metal determinations in aqueous and plants sample were made by flame atomic absorption spectrometry. There was a gradual decrease in metal contents in the artificial wastewater at time intervals. The metal removal capacity was different for each specie and for each metal. At the end of the experiment, the multi-metal system treated with aquatic plants was assessed with the same battery of tests used in the beginning. The whole ecotoxicity of the artificial wastewater decrease after treatment with aquatic plants, demonstrating an efficiency method for metal removal and recovery.

WE169

Toxicity of the binary mixture Cd-Zn on Lemna gibba evaluated using morphological and oxidative stress enzyme endpoints

s. martinez, CONICET PRIET UNLU; W.D. Di Marzio, CONICET-PRIET / PRIET; M. Saenz, PRIET CONICET, National University of Luján

The presence of metals in the environment represents one of the mayor concerns as they are persistent in nature, non-biodegradable and can bioaccumulate in living animals and plants. Metals in aquatic ecosystems may have effects on primary trophic level composed partly by aquatic vascular plants, also called macrophytes. These organisms play a critical role in this environment. As a representative species of macrophytes, we worked with a rooted free-floating *Lemna gibba*. The metals evaluated here were Cd and Zn, individually and in mixtures. Exposures of plants were carried out in presence or absence of Cd and Zn for 7 days. Different endpoints were determined at the end of the assays. Number of fronds, fresh weight, fronds/colonies ratio, frond area and exes' length are the determined morphological endpoints. Physiological changes were evaluated as enzymatic activity of catalase, ascorbate peroxidase and guaiacol peroxidase, determined at the lowest concentrations. Both metal concentrations, bringing about a 50 % inhibition of frond number (EC50) was determined. In order to compare the sensitivities of the different endpoints, NOEC and LOEC toxicity indexes were calculated. For Cd, fresh weight and fronds/colonies ratio resulted in the most sensitive, while for Zn total area was the most sensitive. Even though there was no significant difference for guaiacol peroxidase activity for Cd, it presented an increase compared to control. While the other enzymes had activity levels similar to the control. In the case of Zn, catalase and ascorbate peroxidase activities where higher than control, however neither of both presented significative differences with it. For the mixture analysis, multiple regression was used to fit the observed %frond number inhibition (%FNI) to dissolved metal concentration ($[M_{dis}]$). The negative value of the parameter of the interaction between Cd and Zn indicates alleviation of %FNI and toxicity. The concentration addition approach was evaluated by calculating the sum of toxic units ($\sum$TU) for each mixture test case based on single EC50s. The average $\sum$TU of all test cases resulted 1,13 suggesting that this mixture presents an additive toxicity to *Lemna gibba.* Enzyme activity was also calculated at the lower concentrations of the mixtures. In general an increase in the enzymatic activity was observed. Ascorbate peroxidase and guaiacol peroxidase presented the maximum increase, while catalase had a moderated activity rise.

WE170

Increase of tolerance of green algae as a tool in metal bioremediation

M. Saenz, PRIET CONICET, National University of Luján; F. Cassani, S. Martinez, s. curieses, J. Alberdi, CONICET PRIET UNLU; W.D. Di Marzio, CONICET-PRIET / PRIET

Presence of various metals in aqueous streams arising from the discharge of untreated metal containing effluents into water bodies, is one of the most important environmental issue, as human health risks and harmful effect to living organisms occur. In the last decades the amount of Chromium in aquatic and terrestrial ecosystems has increased as a result of different human activities such mining, chrome plating, leather tanning and wood preservation. The aim of this study was to evaluate the use of preadapted strains to subletal concentrations of Chromium, into bioremediation of Chromium containing wastewater. Preliminary results will be shown related to the assessment of the potential of this strategy to increase tolerance of selected species in order to become an interesting tool in the field of bioremediation processes mediated by green algae. Two green algae species were used, *Scenedesmus quadricuada* and *Nannochloris oculata*. These two species differ in its morphological structure and organization level as the former has a cenobial feature while the second a free unicellular one. Both strains were maintained by a year under subletal concentrations of chromium ranging from 0,42 to1,73 mg/l. These concentrations were chosen base on previous experiments through range finding tests. Subletal solutions were renewed monthly and algal cells were subcultured in new medium. After the preadapted period, each subletal exposed algal population from both strain and one which was never exposed to the metal, considered as the control, were centrifuged. An inoculum of know cell density was prepared with each pellet, and the algae were exposed to a wide range of Chromium concentration solutions. Samples of solution and algal cells were taken for metal determination in order to dilucidated the mechanism of resistance origin.The harvested cells were centrifuged and a microwave acid digestion were carried out. Metal determinations in subletal solutions and in algal sample were made by flame atomic absorption spectrometry. Chromium accumulation and compartmentalization in algal cells would explained the increase resistance observed. Further studies relative to detoxification mechanisms and chelating internal molecules as phytochelatin will be conducted to unravel the tolerance mechanisms involved.

WE171

Ecotoxicological assessment of the iron mining waste from Mariana (Brazil) on terrestrial flora using different plant species

O.R. Alves, University of São Paulo USP / Department of Hydraulic and Sanitation; O.A. Bandeira, T.J. Pinto, L.P. Figueiredo, University of São Paulo USP; E. Espindola, University of São Paulo USP / Hydraulics and Sanitation

In Brazil it is very common to have mining waste placed in dams, especially in the Minas Gerais state. The deposit of mining wastes implies in risk of dam rupture, between other problems. In November 2015, the rupture of the "Fundão" Dam in the city of Mariana in Minas Gerais state was one of the worst environmental disasters in Brazil. The rupture caused severe impacts to the terrestrial and aquatic environments, where tons of the waste has been placed. It is now matter of concern to study the effects of the mining waste deposition in soil to terrestrial flora in order to understand the real consequences to the environment and so be able to propose actions for restoration and management of the affected area. The main goal of this study was to evaluate the ecotoxicity of the mining waste that outpoured the Fundão dam to ten different plant species (*Avena strigosa*, *Pennisetum glaucum*, *Crotalaria juncea*, *Canavalia ensiformis*, *Cajanus cajan*, *Cajanus cajan*, *Dolichos lablab*, *Mucuna pruriens* gray, *Mucuna pruriens* black and *Lupinus albus*). The ecotoxicological assays were run using five treatments, which consisted in the mixture of a natural soil (NS) from Mariana (uncontaminated) and the waste (W). The combinations of mixture were: P1: 75% NS and 25% W; P2: 50% NS and 50% W; P3: 25% of NT and 75% of the W and P4: 100% W. The parameters avaluated were: Fresh and dry biomass (shoot and root), height, length of the longest root and seed emergence. All species, except *Lupinus albus* and *Avena strigosa*, had EC_{50} and/or EC_{20} in at least one of the seven parameters evaluated. The species that presented 50% inhibition of root growth were *C. juncea* (73.07%), *P. glaucum* (82.68%) and *C. cajan* (97.54%). The height of the first two species was affected by 20% in the proportions 78.61 and 85.91%, respectively. The proportions 87.32 and 40.61% of waste affected 50% of the length of the longest root of *C. juncea* and *P. glaucum*. The results showed that: the species tested presented different indices of tolerance to the mining waste; the waste that outpoured of the Fundão dam caused phytotoxic effects in all tested species; the most sensitive and least sensitive parameters, respectively, were root growth (root length and dry biomass) and seed emergence.

WE172

Mitigation of CuO nanoparticles microbial ecotoxicity by plant in an agricultural soil: plant variety matters

J.M. Martins, IGE UMR 5001 / Université Grenoble-Alpes; A. Cantarel, Université Claude Bernard Lyon1 / UMR Ecologie Microbienne 5557; J. Gervaix, Université Claude Berrnard Lyon 1 / UMR Ecologie Microbienne 5557; A. Richaume, Université Claude Bernard Lyon 1 / UMR Ecologie Microbienne 5557

New types of pesticides based on nanoparticles (NPs) are now being used to optimize phytosanitary treatments. However, they can generate soil contamination by metal-oxide NPs such as CuO-NPs [3] which fate and impact on agro-ecosystems is still largely unknown. Several studies showed the deleterious effects of metal nanoparticles (NPs) on soil microbial communities (1) and reported the importance of organic matter (OM) content in the NPs ecotoxicity due to its role as dispersing and stabilizing agent (2). A high OM content is likely to increase NPs toxicity by favoring their dispersion. Based on this assumption, our goal was to assess 1) whether the plant modifies the microbial ecotoxicity of NPs because of organic matter enrichment in the rhizosphere through the root exudation and 2) whether the plant variety mitigates the ecotoxicity according to plant traits. Endpoints that relate to soil fertility (ie abundance and activity of microbial communities involved in carbon and nitrogen cycle) were used to assess NPs effects. The experimental design consisted in planted and unplanted soil microcosms contaminated or not with two doses of CuO-NPs (1mg and 100 mg/Kg) before the seedling of wheat. We compared the effect of two conventional varieties (Arrezzo® and Skerzzo®) exhibiting contrasted traits reflecting different root exudation. Ecotoxicological effects were assessed after 30 and 50 days by measuring plant traits on each variety, microbial activities (respiration, nitrification and denitrification) and microbial abundance by qPCR targeting 16S RNA gene and function genes. The main physico-chemical properties of NPs were characterized by Dynamic Light Scattering in rhizosphere and unplanted soil solutions in which ionic strength, pH and dissolved organic carbon were also measured. The results showed that the NPs hydrodynamic diameter was higher in planted soil solutions compared to unplanted one. Comparison between planted and unplanted soil showed that the plant hampered ecotoxic effects on the microbial activity of functional microbial groups without significant changes in their abundance. Arrezzo® limited the reduction of nitrification and denitrification suggesting that NPs ecotoxicity depends on the wheat variety likely because of the effect of roots on NPs and /or the microbial populations recruited in the rhizosphere that can be more or less sensitive to NPs.

[1] Simonin and Richaume. 2015. Environ Sci Pollut Res, [2] Zhang *et al.* 2009. Water Res

WE173

Use of Posidonia oceanica as a potential bioindicator species of metal pollutants: cellular and molecular responses to mercury exposure

G. Moltedo, ISPRA-Institute for Environmental Protection and Research / National Center for Laboratory Networking Ecotoxicology Area; B. Catalano, ISPRA Institute for Environmental Protection and Research; G. Martuccio, C. Sebbio, ISPRA Institute for Environmental Protection and Research / National Center for Laboratory Networking Ecotoxicology Area; F. Onorati, ISPRA Institute for Environmental Protection and Research; A. Cicero, ISPRA Institute for Environmental Protection and Research / National Center for Laboratory Network

The marine phanerogam *Posidonia oceanica*, thanks to its worldwide distribution and to its capability to accumulate certain environmental metal pollutants, would be a potentially valuable bioindicator species of metal pollutants. Mercury represents one of the most abundant marine pollutants in the Mediterranean Sea. In this study sublethal effects of this metal were investigated in *P. oceanica*. Several foliar shoots of this aquatic plant were treated for 4 days (96 h) with different mercury concentrations (0, 0.1 and 1 µgL-1 Hg Cl_2) under constant laboratory conditions. Biochemical markers of oxidative stress and of genotoxicity, such as the glutathione S-transferase activities, the ascorbate peroxidase activity, the total antioxidant capacity, the phenols content, the level of lipidic peroxidation and the micronuclei frequency were measured in different parts of adult leaves: the blades and the sheaths for antioxidant responses, the meristem and the sheaths for genotoxic effects. Although a limited effect of Hg was measured in analyzed tissues, a significant difference in localization of responses was found in leaves of all treatments.

WE174

Influence of toluene vapor exposure on plant metabolic changes

W. Kim, J. Park, Gwangju Institute of Science and Technology / School of Earth Sciences and Environmental Engineering; S. Kim, Gwangju Institute of Science and Technology / School of Earth Science and Environmental Engineering

The conventional damage diagnosis methodology for plants after chemical accident only relied on the change in their phenotype such as leaf-bronzing, so there had been lots of controversy because of uncertain causality and inaccuracy. The ministry of environment tried to characterize of plant damage by introducing metabolomics, which has emerged as a powerful tool for sensitive and diagnosis. However, target metabolite selection process was unclear and the exposure method did not reflect the chemical accident scenario, so the research results have not been put to practical use. Therefore, untargeted metabolomics and vapor exposure chamber were introduced in this study to overcome the limitations of existing research. The development potential of metabolomics-based damage diagnosis tool was studied using *Oryza sativa, Trticum aestivum,* and *Hordeum vulgare.* Toluene was selected as target compound based on the scoring system, which takes into account both accident frequency and hazards. To reflect the realistic chemical accident scenario, plants were exposed in vapor exposure chamber. In this study, the metabolomics responses of plants at early development stages (4th leaf stage) to toluene were evaluated by liquid chromatography quadrupole time-of-flight mass spectrometry (LC-QToF-MS) based untargeted metabolic profiling. The exposed concentration-based and recovery time-based metabolic response patterns were analyzed by Principal component analysis and Partial least squares Discriminant Analysis. Overall, the results of multivariate statistical analysis demonstrated a number of potential biomarkers that were characterized by metabolomic approach and provided an insight into quantitative chemical accident damage assessment.

WE175

Influence of soil organic amendments on the phenolic contents in rosemary (Rosmarinus officinalis L.) plants

I. Nogues, National Research Council of Italy / Institute of Agro-Environmental and Forest Biology (IBAF); A. Barra Caracciolo, National Research Council / Water Research Institute; P. Grenni, National Research Council of Italy (CNR) / Water Research Institute; M. DE LOS ANGELES BUSTAMANTE MUNOZ, University Miguel Hernández de Elche (Spain) / Department of Agrochemistry and Environment

Rosemary (*Rosmarinus officinalis* L., Lamiaceae) is an aromatic shrub native from Mediterranean regions, grown as a common herb around the world. This species constitutes an exceptional source of different bioactive compounds, mainly phenolic compounds, with proved antimicrobial and antioxidant properties. Furthermore, different studies have shown the potential and important role that this shrub can have in the rehabilitation of degraded soils such as agricultural ones with low levels of organic carbon, contributing to the reduction of erosion and improving soil quality. Within this context, different studies have shown that the metabolism of the phenolic compounds in plants has been associated to environmental factors, such as temperature, rainfall and ultraviolet radiation incidence, as well as soil composition. In this sense, plant nutrient balance in the soil could influence the production of secondary compounds, the concentrations of secondary metabolites in plant tissues depending on the concentration and availability of determined nutrients in the soil. Therefore, this study evaluates the effectiveness of the rosemary plant to improve soil quality and the effect of the incorporation of two composts derived from anaerobic digestates on the phenolic contents of rosemary plants grown a semiarid soil. In the study, two composts (CM, mainly composed by cattle manure anaerobic digestate and CS, mainly composed by pig slurry anaerobic digestate), at two different rates (30 t/ha and 60 t/ha respectively) were incorporated into a semiarid soil from central Italy. These organic amendments were compared with the soil without amendment (control treatment, B) and an inorganic treatment (I). Subsequently, plants of rosemary (*Rosmarinus officinalis*) were planted on these soils. The efficiency of the treatments was evaluated by analysing chemical characteristics in the soil and the total contents of phenolic compounds and flavonoids in the rosemary plants grown in the different treatments. The results obtained have shown that the incorporation of the organic amendments into the semiarid soil improved soil characteristics, by increasing organic matter and nutrient contents, but also implied a decrease in the concentrations of phenolic compounds in the rosemary plants, probably due to the nitrogen fertilisation increases growth, but also leads to decreased concentrations of carbon-based secondary metabolites, such as phenolic compounds.

WE176

Leaf litter originating from trees treated with systemic fungicides - a new exposure pathway for aquatic decomposer detritivore systems

K. Newton, Univesity of Montreal; J.P. Zubrod, D. Englert, University of Koblenz-Landau / Institute for Environmental Sciences; S. Lüderwald, Universität Koblenz-Landau / Institute for Environmental Sciences; T.C. Schell, IMDEA Water Institute / Ecotoxicology; P.T. Baudy, University of Koblenz-Landau / Institute of Environmental Sciences; M. Konschak, University Koblenz-Landau / Institute for Environmental Sciences; R. Schulz, University of Koblenz-Landau / Institute for Environmental Sciences; M. Bundschuh, Swedish University of Agricultural Sciences / Department of Aquatic Sciences and Assessment

Leaf litter decomposition, an important process in freshwater ecosystems, is mediated by microbial decomposers and leaf-shredding macroinvertebrates. This process can, however, be modified by chemical stressors such as fungicides. Although fungicide stress has increasingly been assessed in recent years, the systemic nature of some fungicides, which enables the uptake and distribution within treated plants, is not yet considered. Therefore, as a first step, we treated *Alnus glutinosa* with a mixture of systemic fungicides (SFs; azoxystrobin, cyprodinil, quinoxyfen and tebuconazole) via soil drenching at three levels (control, field application rate (FR), and 10 times the field application rate (FRx10)). During leaf fall, we collected the leaves and assessed the potential impact of the fungicides on microbial decomposers and leaf-shredding macroinvertebrates. We quantified microbial leaf litter decomposition, their community composition and the palatability of leaves after microbial conditioning for the model shredder *Gammarus fossarum* Koch. By assessing growth and physiological fitness of this species over multiple weeks, we additionally estimated the nutritious quality of leaf litter. Gammarids preferred conditioned FRx10 over control leaves, which may reflect changes in microbial community structure. This increase in palatability as a consequence of SF may be related to the fungicides' ability to reduce fungal pest pressure, allowing trees to divert energy and carbon from defense to growth or storage. The same treatment resulted in a 300% increase in gammarid growth, while FR elevated this variable non-significantly by 100%. These data suggest that SF may indeed have implications for microbial decomposers and leaf-shredding macroinvertebrates, while the underlying mechanisms are still not fully understood.

WE177

SETAC Plants Interest Group

S. Loutseti, DuPont De Nemour Hellas S.A.

Environmental Risk Assessment in Sediments (P)

WE178

Benthic invertebrate bioturbation activity determines species specific sensitivity to sediment contamination

M. de Baat, University of Amsterdam / IBED-FAME; T.V. van der Meer, University of Amsterdam IBED Institute / Department of Freshwater and Marine Ecology; P. Verdonschot, University of Amsterdam / Department of Freshwater and Marine Ecology; M. Kraak, University of Amsterdam / IBED-FAME

Bioturbation activity of sediment-dwelling organisms promotes the release of contaminants across the benthic-pelagic ecosystem boundary, thereby affecting the exposure to and uptake of sediment associated contaminants at the sediment-water interface by themselves and the entire community around them. This way, bioturbation activity may contribute to species specific sensitivities to sediment associated compounds. Therefore we assessed if invertebrate bioturbation activity determines species specific sensitivities to sediment contamination. For two metals, Ni and Cu, sufficient data were available to construct Species Sensitivity Distributions (SSD). The position of the species in the SSDs could indeed be linked to their bioturbation rate: the most active bioturbators being the most sensitive benthic invertebrates. Active bioturbators thus enhance their exposure and therewith their sensitivity to sediment associated toxicants. Moreover, active bioturbators can hence promote the release of sediment-associated contaminants across the benthic-pelagic ecosystem boundary, thereby stimulating delivery of contaminants from what is often the most polluted environmental compartment in freshwater ecosystems. It is concluded that trait based ecotoxicology offers a possibly potent tool for predicting sensitivity of benthic invertebrates and the benthic community to sediment-associated contaminants.

WE179

Effect based sediment quality assessment incorporating chemical fingerprinting

N. Wieringa, University of Amsterdam/IBED Institute / FAME; M. de Baat, University of Amsterdam / IBED-FAME; B. van Hall, F. Selhorst, University of Amsterdam / Department of Freshwater and Marine Ecology; S. Droge, University of Amsterdam/IBED Institute / IBED; M. Kraak, University of Amsterdam / IBED-FAME; P. Verdonschot, University of Amsterdam / Department of

Freshwater and Marine Ecology
The European Union Water Framework Directive does not require member states to monitor sediment quality. When performed at all, water authorities most often monitor sediment quality by means of chemical target analysis focusing only on target compounds, potentially overlooking ecotoxicological risks caused by (un)known mixtures of sediment associated compounds. Hence, there is an urgent need to incorporate effect-based monitoring and chemical fingerprinting into sediment quality assessment. Therefore, the aim of the present study was to innovate ecotoxicological sediment quality assessment by incorporating whole sediment bioassays and chemical fingerprinting of bioavailable compounds. To this purpose intact whole sediment cores were collected using a sediment core sampler at a reference and 11 contaminated sites grouped by land use: urban, agricultural, and wastewater treatment plant (WWTP) effluent sites. A *Chironomus riparius* 28-day life cycle whole sediment bioassay was performed with survival and emergence as endpoints. Simultaneously, SPME fibers were applied as sediment passive samplers to determine pore water concentrations of phenanthrene and pyrene, selected as model compounds for sediment PAH concentrations. Survival in the bioassay was unaffected at the urban sites, while significantly lower at all WWTP sites and two of the agricultural sites. Emergence was significantly delayed at the urban sites, agricultural sites exhibited an irregular emergence time, while WWTP sites induced accelerated emergence. Pyrene and phenanthrene concentrations were negligible at the reference site, very low at the agricultural and WWTP sites, and highest at the urban sites. Urban sites thus have a high chemical load, but survival was higher than on the agricultural sediments. Contrastingly, agricultural and WWTP sites have a lower chemical load, but significantly lower midge survival. This is likely attributable to the mode of action of the pesticides present at agricultural sites, that affect survival more than the non-specific toxicity of compounds at the urban sites. Employing bioassays allowed ranking of sediments based on biological responses rather than on the presence of target compounds. All contaminated sediments caused effects on the relatively resilient *C. riparius*, underlining that sediment contamination is presently understudied. It is therefore concluded that ecotoxicological sediment quality assessment needs to be included in the EU WFD.

WE180

Quantifying the Bioavailability of HOCs associated with Suspended Sediment to Daphnia magna

X. Xia, X. Zhang, School of Environment, Beijing Normal University

In natural rivers, hydrophobic organic compounds (HOCs) are mainly associated with particulates, especially for the rivers with high suspended sediment (SPS) concentrations. Suspended sediment will affect the bioavailability of HOCs in rivers. However, no research has been carried out to quantify the bioavailable fraction of HOCs sorbed on different compositions of SPS with various particle sizes. In this study, we chose pyrene as a typical HOC to study the bioavailability of HOCs associated with SPS of various compositions and grain sizes to D. magna. The passive dosing devices were made to control the freely dissolved concentration of pyrene in the exposure systems. The effect of pyrene associated with SPS of different compositions (including amorphous organic carbon, AOC; black carbon, BC, and minerals) and grain sizes (including 0~50 μm, 50~100 μm, and 100~150 μm) on the immobilization and enzymatic activity of D. magna was investigated to quantify the bioavailability of SPS-associated pyrene. The results showed that with C_{free} of pyrene ranging from 20.0-60.0 μg L^{-1}, the immobilization of Daphnia magna in the presence of 1 g L^{-1} SPS were 1.11-2.89 times that in the absence of SPS. The contribution of AOC-, BC-, and mineral-associated pyrene to the total bioavailability of SPS-associated pyrene was approximately 50%-60%, 10%-29%, and 20%-30%, respectively. The bioavailable fraction of pyrene sorbed on the three components of SPS was ordered as AOC (22.4%-67.3%) > minerals (20.1%-46.0%) > BC (9.11%-16.8%). This is because the SPS composition will affect the sorption of pyrene in water as well as the desorption of pyrene from SPS in Daphnia magna. The immobilization caused by pyrene associated with different grain size SPS was ordered as 50-100 μm> 0-50 μm> 100-150 μm. When pyrene C_{free} was 20.0 μg L-1, the immobilization caused by pyrene associated with 50-100 μm SPS was 1.42 and 2.43 times that with 0-50 μm and 100-150 μm SPS, respectively. The protein and enzymatic activities of Daphnia magna also varied with the SPS grain size. The effect of SPS grain size on the bioavailability of SPS-associated pyrene was mainly due to the difference in SPS ingestion by Daphnia magna and SPS composition, especially the organic carbon type, among the three sizes. According to the results obtained in this study, a model has been developed to calculate the bioavailability of HOCs to aquatic organisms in natural waters considering both SPS grain size and composition.

WE181

Sediment quality assessment in the Netherlands: Link between chemical, toxicological and ecological parameters

L. Lautz, Radboud University Nijmegen / Department of Environmental Science; J. Chai, Radboud University Nijmegen; R. Hoondert, Radboud University Nijmegen / Department of Environmental Science; A.M. Ragas, Radbound University / Department of Environmental Science; R. Van Zelm, Radboud University / Department of Environmental Science; J.A. Hendriks, Radboud University Nijmegen / Department of Environmental Science

Sediment quality assessment is often based on chemical analysis only, occasionally including toxicological assays. Full sediment quality assessment, including chemical and macrofauna analysis and toxicological assays, is not the standard procedure due to high costs. Based on chemical analysis only, it is not always clear whether sediment management in form of dredging and landfill or remediation is necessary. To reduce costs of sediment management on the one hand and to increase environmental benefits on the other, the right priorities need to be set. To do so, in-depth information is needed from chemical analysis, toxicological assays and macrofauna analyses. Chemical, toxicological and ecological data is available from freshwater/sediment monitoring campaigns in the Netherlands. A multivariate analysis was performed to identify contaminants with high impacts on the bioassays. From the 49 chemicals included in the dataset, 28 were significantly related to the outcome of the bioassay results of *Daphnia magna* and *Chironomus riparius*. The Species Sensitivity Distribution (SSDs) method was used to quantify the ecological risk associated with concentrations of contaminants. Based on the SSDs the potentially affected fraction (PAF) of species was calculated. These PAFs were used to calculate the multiple substances PAF, combining effects posed by multiple compounds. Such correlation analysis have not been conducted previously for a large dataset of field-collected sediments in the Netherlands. With our work we contribute to the quantification of relationships between chemical concentrations and toxicological assay in sediments. These relationships can be used in future analyses of predictive abilities of sediment quality and can be applied in an assessment tool for sediment management to determine management strategies.

WE182

Integrative approach to assess ecological risks of sediment metallic contamination in Lake Ohrid (Albania)

L. Minguez, LIEC (CNRS UMR 7360, Université de Lorraine) / Laboratoire Interdisciplinaire des Environnements Continentaux, CNRS UMR 7360; E.M. Gross, University of Lorraine / Laboratoire Interdisciplinaire des Ecosystemes Continentaux; D.A. Vignati, CNRS / LIEC UMR7360; S. Pain-Devin, Université de Lorraine - UL / LIEC - CNRS - UMR 7360; S. Devin, LIEC, CNRS UMR 7360, Université de Lorraine / LIEC, CNRS; F. Guérold, Université de Lorraine UL / Laboratoire interdisciplinaire des environnements continentaux LIEC CNRS UMR 7360; L. Giamberini, Université de Lorraine, CNRS UMR 7360 / LIEC, CNRS

It is now widely admitted that chemical monitoring of pollutants in waters and sediments is not sufficient to assess the risks caused by such pollution in aquatic ecosystems, since chemical data alone provide no indication of biological effects. Biological responses of exposed organisms need to be taken into account, allowing to define the ecotoxicological status of the studied system. The ancient lake Ohrid, the most biodiverse lake in Europe shared between Albania and Macedonia, was chosen as case study. The Albanian side, due to the presence of ultramafic rocks, was a large mining area exploited to produce nickel, chromium, and iron until the early nineties. Several ore dumps from this past activity still remain near the shoreline, representing one potential input source of these metals. Several creeks flowing across soils naturally rich in metals also contribute to metal inputs into the lake. We studied three sites along the Albanian shoreline of Lake Ohrid, and defined by different metal pressures: “Pog” in an urbanized area but considered as dimly contaminated by metals, “Mem” and “Poj” located in the ultramafic area of the lake, at the vicinity of a Fe-Ni dump site for “Mem”, or nearby the outlet of a creek for “Poj”. In the two sites under metallic pressure, sediments contained high levels of metals with concentrations reaching 93.8 mg/kg for Co, 345.1 mg/kg for Cr, 553.8 mg/kg for Ni, 49.9 g/kg for Fe and 872.9 mg/kg for Mn. Despite these high concentrations, metals are not necessarily bioavailable. This is why it is also important to combine chemical characterization (total and available pools) with the study of lethal and sublethal effects after acute and chronic exposure. In our study, we assessed ecological risks in Lake Ohrid using an integrative approach consisting in: (1) chemical and physical characterizations of sediments, (2) assessment of metal bioavailability, (3) ecotoxicological bioassays, and (4) the study of sub-lethal effects on organisms. During this presentation, the main results from this integrative work at Lake Ohrid will be presented.

WE183

Active Biomonitoring and DGT Passive Sampling: Holistic Assessment of metal bioavailability in sediments and associated risks

K. De Schamphelaere, Universiteit Antwerpen / Department of Biology (SPHERE and ECOBE Research Groups); H. Hetjens, University of Antwerp / Department of Biolgy (SPHERE Research Group); J. Teuchies, E. Amato, L. Bervoets, University of Antwerp / Department of Biology (SPHERE Research Group); P. Meire, University of Antwerp / Departement of Biology (ECOBE Research Group); R. Blust, University of Antwerp / Department of Biology (SPHERE Research Group)

Impacts of sediment metal contamination on aquatic ecosystems and their functioning remain a widespread problem. The ecotoxicological risk associated with metal contamination is dependent on metal speciation, sediment characteristics and the behavior and physiology of the affected organisms. Hence, bioavailable concentrations, rather than total metal concentrations, are often a critical factor in sediment risk assessment. Determination of bioaccumulation in organisms is a frequently used indicator for bioavailability. However, active and passive biomonitoring techniques are often time consuming and highly dependent on the exposed organisms, limiting comparability and standardization. Diffusive gradient in thin films (DGT) passive sampling is an innovative technique, allowing for the time-integrated measurement of potentially bioavailable metals in sediments or surface water. Divalent metals are selectively accumulated onto a Chelex-embedded hydrogel layer, providing a measurement of labile and weakly-bound metals. To evaluate DGT passive sampling measurements as a possible indicator of bioaccumulation in organisms, a field experiment will be carried out in April 2018 on 6 locations in Flanders (Belgium), of which 3 freshwater and 3 brackish aquatic systems. Bioavailability of metals will be assessed by active biomonitoring through a 4 weeks exposure of caged macroinvertebrates, after which bioaccumulation will be determined. The organisms will be exposed both at the sediment water interface and in the water column. During a pilot study, carried out in November 2017, 3 bivalves and a polychaete worm are exposed in the Zenne river (north of Brussels) to test for their active biomonitoring applicability. During the 4 weeks exposure period, DGT passive samplers will be deployed 3 times for a period of 24h at the sediment water interface and in the water column. Relationships will be determined between the bioaccumulation in the exposed organisms and the determined DGT fluxes. Bioaccumulation and passive sampling measurements at the sediment water interface and in the water column will be evaluated. The experiment aims at establishing relationships between bioaccumulation in different macroinvertebrate species and passive sampling measurements, and further validating the DGT passive sampling technique as a monitoring tool for sediment quality assessments in both freshwater and brackish aquatic systems.

WE184

Bioturbation in contaminated sediments: effects on exposure, toxicity and biogeochemistry.

T.M. Remaili, W. Bennett, Griffith University / Environmental Futures Research Institute; S.L. Simpson, CSIRO Land and Water / Centre for Environmental Contaminants Research; E.D. Amato, University of Antwerp / Department of Biology; D.T. Welsh, Griffith University / Environmental Futures Research Institute; E. Lombi, University of South Australia / Future Industries Institute; D. Howard, Australian Synchrotron; D.F. Jolley, University of Wollongong / School of Chemistry

Sediments are a major sink for a range of contaminants. Organism-sediment interactions such as bioturbation can alter sediment physicochemistry, and facilitate the diffusion of reactive chemical species (e.g. O_2) into deeper sediments, potentially changing the oxidation state of various redox-sensitive materials and the fate and toxicity of contaminants. We applied multidisciplinary to: (i) characterise influences of bioturbation on contaminant fate, exposure and toxicity to aid current sediment quality assessment frameworks; and, (ii) assess the potential use of bioturbation in the management and natural recovery of heavily degraded sediment ecosystems. Incresed bioturbation in predominantly metal-contaminated sediments increased bivalve (*Tellina deltoidalis)* and amphipod (*Victoriopisa australiensis)* survival from 53 to 100% and 42 to 93%, respectively; and reproduction in a second amphiod (*Melita plumulosa*) from 3 to 65%. This was attributed to the decreased concentrations of dissolved copper in the overlying water associated with bioturbation. Conversely, increased bioturbation in sediments contaminated by metals and hydrocarbons decreased reproduction (44 to 23%), which was attributed to an increased release of polycyclic aromatic hydrocarbons (PAHs). High-resolution chemical imaging dissolved oxygen and metals in a legacy contaminated sediment mesocosm with a bioturbator present showed the indtroduction of oxic overlying waters waters into sediments, and significant organism-induced fluxes of nickel and zinc into burrow and overlying waters. The presence of nickel and zinc in burrow and overlying waters demonstrated that organism exposure is likely to be greater from the burrow waters than from the pore waters. This is consistent with increased accumulation of zinc observed with co-habitation of bivalves and amphipods. Low copper and lead concentrations in burrow waters during bioturbation events was consistent with the results of previous tests, where copper concentrations were lower in the presence of high bioturbation intensities, possibly due to binding with iron-(oxy)hydroxide phases or to resuspensed particulate phases. These results highlight the importance of considering organism-interactions during sediment quality assessments, and the contributions they have to biogeochemistry and contaminant exposure to surrounding ecosystems.

WE185

The diffusive gradients in thin films (DGT) technique predicts toxicity of nickel contaminated sediments to a marine amphipod

M.L. Gillmore, CSIRO Land and Water / School of Chemistry; G.A. Price, University of Wollongong / School of Chemistry; L.A. Golding, CSIRO Land and Water; J. Stauber, CSIRO / CSIRO Land and Water; M.S. Adams, CSIRO; S.L. Simpson, CSIRO Land and Water / Centre for Environmental Contaminants Research; D.F. Jolley, University of Wollongong / School of Chemistry

Mining of lateritic nickel ore deposits within the Southeast Asia and Melanesia region is expected to intensify as sulphide nickel ore deposits become depleted. The close proximity of these mining operations to coastal ecosystems places marine benthic organisms at a potential risk of adverse effects related to nickel exposure. Currently, limited data exists for the effects of sediment nickel exposure on coastal marine organisms. The diffusive gradients in thin films (DGT) technique has emerged as a tool that allows for the rapid *in situ* measurement of the lability and dynamics of metals in sediment. The objective of this research was to determine effects thresholds for sediment-nickel by measuring reproduction of the estuarine-marine amphipod, *Melita plumulosa* in 10-d whole-sediment bioassays with three nickel-spiked sediments and two field-collected nickel-contaminated sediments with varying chemical and physical properties. We compared the concentration-response relationships obtained using traditional extraction methods of metals from sediments with DGT-labile nickel to determine whether DGT can be used to predict nickel bioavailability and toxicity. Effect concentrations of total recoverable nickel (TR-Ni) to cause a 50% impairment in reproduction (EC50) were 2000 (1200-2900), 1100 (580-1700) and 1100 (740-1500) mg/kg for the silty, sandy-silt and sandy sediments, respectively (95% confidence limits). TR-Ni was found to be a poor predictor of toxicity for the two field nickel-contaminated sediments. Site 1 (2000 mg/kg TR-Ni) and Site 2 (1300 mg/kg TR-Ni) had reproductive responses of 88% (±10) and 71% (±11) of the control, respectively. The EC50s based on DGT-labile Ni were 2.3 (1.7-3.4), 3.3 (1.7-4.9) and 2.0 (1.0-3.0) $mg/m^2/h$ for silty, sandy-silt and sandy sediments, respectively. Concentration-response relationships based on DGT-labile Ni fluxes had less variation and better-predicted toxicity in the field collected nickel-contaminated sediments. For Site 1 (0.4 $mg/m^2/h$ DGT-labile Ni) and Site 2 (1.0 $mg/m^2/h$ DGT-labile Ni) sediments, respective reproductive responses were 88% (±10) and 71% (±11) of the control. This demonstrates that amphipods were responding to the labile nickel as measured by DGT and further supports its use in nickel risk assessments.

WE186

Identifying key toxicants in sediment samples from urban waterways in Guangzhou, China using a integrated method of TIE and EDA

J. You, H. Li, F. Cheng, Jinan University / School of Environment

Determining causality of sediment toxicity is of great importance in aquatic risk assessment. Two approaches, namely toxicity identification evaluation (TIE) and effect-directed analysis (EDA) have been developed. Conventional sediment TIEs take the advantage of environmental relevance by using whole organism bioassays while suffer from lack of effective methods for specifically identifying major contributors from a universe of chemicals if organic contaminants are identified as main class of toxicants in phase I TIE. Alternatively, EDA is a powerful tool in identifying causes of sediment toxicity with sophisticated fractionation and chemical analysis of targeted and non-targeted toxicants, but it is short of environmental relevance due to the use of in-vitro bioassays and exhaustive solvent extraction. To better understand the cause of sediment toxicity in urban waterways in Guangzhou, China, an integrated method of TIE and EDA was applied. Whole-sediment TIE in combination with bioavailability-based extraction found that sediment mortality to the benthic invertebrate, *Chironomus dilutus* was caused by organics and metals jointly and organic pollutants contributed to the mortality for all samples. To better elucidate the roles of non-target organic contaminants in sediment toxicity in these sediments, EDA tests were performed. Bioaccessible contaminants in sediment samples were extracted by XAD resin. Cell viability of the extracts was assayed using the cell counting kit-8 assays. To take tissue specificity into consideration, four cell lines (HepG2, MCF-7, A549 and SH-SY5Y) were used to distinguish toxicants related to metabolism dysfunction, endocrine disruption, respiratory toxicity and neurotoxicity, respectively. All test sediment samples showed significant cell proliferation of SH-SY5Y cell line, but little effect on HepG2 and A549 cell lines. The results were further confirmed by toxicity tests using *C. dilutus.* One sediment sample impacted MCF-7 cell line. The proliferation of SH-SY5Y proliferation was partially explained by oxidative stress. The SH-SY5Y cell line was used for further EDA experiments after separating the extracts into 35 fractions using GPC and NPLC. In conclusion, n integrated method of TIE and EDA would provide an environmentally relevant and toxicant specific approach to effectively determine causality of sediment toxicity by combining the merits of the two methods.

WE187

Water discharges from the city of Lausanne during rainfall in Lake Geneva: Use of a triad approach to assess their influence on sediment quality

M. Casado-Martinez, T. Benejam, R. Vivien, Centre Ecotox; S. Pesce, Irstea Lyon-Villeurbanne / Microbial ecology of anthropised river systems; L. De Alencastro, École Polytechnique Fédérale de Lausanne / Central Environmental Laboratory; N. Dubois, Eawag; L. Rossi, Hydrique Ingénieurs; S. Höss, Ecossa / Animal Ecology; B.J. Ferrari, Centre Ecotox EAWAGEPFL

Sediment represents an important compartment in surface waters. It constitutes a habitat or spawning site for many organisms and is an essential trophic resource for higher level organisms. It can be impacted by anthropogenic activities, particularly through urban wet weather discharges like stormwater and combined sewer overflows. In Switzerland, the Vidy Bay located in the middle of the northern shore of Lake Leman, in front of the city of Lausanne, is of particular interest as it receives a large portion of stormwater from the city of Lausanne via the Flon River. In this context, this study aimed to evaluate the impact of the Flon river stormwater overflows on the sediment quality of the Vidy Bay using a triad approach combining chemistry, ecotoxicology and the study of in situ benthic communities.

To do this, a sampling grid composed of 15 sites was developed in the discharge area of the effluent from the Flon river into the lake. At each point, sediment samples were collected to measure metal concentrations and assess the ecotoxicological quality of sediments in the laboratory using a whole sediment toxicity test with ostracods. At six selected sites in the central transect of this sampling grid, corresponding to the extension of the outlet of the Flon river, a more detailed monitoring program was applied, with measurements of PCBs and PAHs concentrations, additional sediment toxicity tests with chironomids, macrophytes and nematodes, as well as the study of the structure of oligochaete communities and tolerance of microbial communities induced by pollution (PICT) were carried out on. The results obtained showed that contamination induced by urban stormwater discharges, identified mainly as copper, zinc, PCBs and PAHs contamination, was elevated near the outlet and moderate at other sites. Although ecotoxicological tests did not indicate significant toxicity in this area, the study of in situ communities revealed the presence of pollution-resistant species among oligochaetes and benthic microorganisms. For the most remote sites, the effects observed in the sediments do not appear to be directly related to the stormwater discharges. The origin and dynamics of the contamination still require further investigations, based in particular on a hydrodynamic model. Overall, this study paves the way for the development of practical tools for assessing the impacts of urban stormwater discharges in lakes in Switzerland.

WE188

Ecotoxicological profiling of sediments along the River Wurm by Aachen (North-Rhine-Westphalia, Germany)

A. Shuliakevich, Institute for Environmental Research (RWTH Aachen University) / Institute for Environmental Research; Y. Müller, RWTH Aachen University / Institute for Environmental Research; S. Hotz, RWTH Aachen University / Department of Ecosystem Analysis ESA; S. Schiwy, RWTH Aachen University / Department of Ecosystem Analysis; S. Karnatz, RWTH Aachen University / Institute for Environmental Research Department of Environmental Analysis; S. Oster, H. Hollert, RWTH Aachen University / Institute for Environmental Research

River sediments serve as a sink and source for micropollutants. Characterized by their semi-dynamic behaviour, sediments can assimilate contaminants. Naturally occurring events such as storms, currents and flood events, as well as human activities like dredging can cause resuspension of sediments and, thus, pose a threat to aquatic organisms. So far, many investigations have been conducted to assess the biological responses in the water phase of streams being impacted by effluent from waste water treatment plants (WWTPs). However, the impact of WWTPs to sediments is still unknown. The present study was taking place within the DemO$_3$ AC-project aimed to assess of the ecotoxicological status of the River Wurm near Aachen. This stream is heavily impacted by anthropogenic activities, such as urbanisation, agriculture, industry, etc. Moreover, the River Wurm receives effluents from various WWTPs within the catchment area. Two of them, the WWTP Aachen Soers and the WWTP Eilendorf, served as investigative objects. Special attention is paid to the WWTP Aachen Soers, which will be upgraded by a full-scale ozonation at the end of 2017. To evaluate a possible impact of the WWTPs on the current ecotoxicological status of the stream a comprehensive ecotoxicological profiling of the sediments from 7 sites up- and downstream of the aforementioned WWTPs was performed. The samples were tested both as native and freeze-dried samples in the sediment contact assay with *Danio rerio*. Sediment extracts (25 g SEQ/ml) were applied for the fish embryo toxicity test with *Danio rerio* as well as for the assessment of the endocrine-disruptive and the mutagenic potential. The results showed estrogenic and mutagenic potential in sediment extracts upstream the tested WWTPs. The embryotoxic potential (enlarged heart, insufficient blood circulation, oedema, etc.) was accounted for the sediment extracts upstream of the WWTP Aachen Soers only. Exposure to native sediments did not lead to any adverse effects in embryos of the zebrafish. However, exposure to freeze dried sediments revealed reduced reactivity of fish embryos. Observed neurological conspicuousness will be verified by further investigations. The described toxicological profiling of sediments will also be completed by chemical analysis. Phase 2 of the DemO$_3$ AC-project will contain comparative studies in order to evaluate the possible influence to sediment toxicology after implementation of full-scale ozonation.

WE189

Comparing conventional and integrative concepts for sediment classification systems

S. Faetsch, Hamburg University of Applied Sciences (HAW); S. Höss, Ecossa / Animal Ecology; S. Heise, Hamburg University of Applied Sciences / Life Sciences

Environmental regulations and guidelines in Europe for assessing the quality of aquatic sediments and dredged material predominantly demand chemical data, and decision making mostly does not integrate the information from different lines of evidence (1, 2). Ecotoxicological data requirements are often limited, with the final classification of the sample not preserving the information of all applied biotests (3). Improved, holistic characterization of sediments and dredged material is needed, to enable a better risk assessment that conserves the ecological quality, is practical and economically feasible at the same time. This poster will present the concept of a study in the scope of the Interreg project "Sullied Sediments" (http://northsearegion.eu/sullied-sediments/) and will discuss first results. The study aims at comparing and evaluating conventional and alternative, integrative and science-based sediment classification concepts for holistic assessments of sediment quality, such as fuzzy-logic based classification (4, 5). Selected concepts will be applied on the classification of sediments from inland waterways in the North Sea region. A sediment quality triad approach will assess the ecotoxicity, the ecology and the chemistry of sediment samples from three catchment areas in the North Sea region with management problems related to polluted sediments. The locations of the sites are in the Port of Hamburg and along the River Elbe in Germany, on the Rivers Hull in the UK and Scheldt in Belgium and in the Netherlands. The applied biotest battery for assessing the ecotoxicological potential of the sediments will cover different trophic levels, different sensitivities and different exposure pathways, considering the toxicity of sediment contact tests, elutriates and extracts. The analysis of the benthic meiofaunal community will assess the ecology of the sediments. The chemical analyses will comprise a broad range of historic contaminants and emerging pollutants, originating from industrial activities, agriculture and pharmaceuticals. This work will be the basis for developing an improved, integrated sediment classification system. **References**

Ahlf et al., 2002. JSS 2: 37–42 Deckere et al., 2011. JSS 11: 504–517 Duft et al., 2003. Federal Ministry for the Environment, Nature Conservation and Nuclear Safety (BMU) Hollert et al., 2002. Ecotoxicology 11: 311-321 Keiter et al., 2009. JSS 9: 168

WE190

Submarine sewage outfall adversely affects the sediment quality of Santos, Brazil estuary - An acute toxicity study

A. dos Santos, Faculdade de Ciências Farmacêuticas - USP / Departamento de Análises Clínicas e Toxicológicas; M. Artal, University of Sao Paulo - USP / Toxicology and Toxicology analysis; J.A. Vendemiati, F.I. Vacchi, University of Campinas / LEAL Laboratory of Ecotoxicology and Environmental Microbiology School of Technology; G. Umbuzeiro, School of Technology, UNICAMP / LAEG

One solution for sewage disposal in several countries is primary treatment followed by chlorination and its discharge in the sea. In Santos city, SP, Brazil, 1 million of cubic meters of urban effluent are discharged into the Santos bay every day, 4.5 km from the beach. To assess the toxicity of environmental samples it is important to use species that are representative of the ecosystem we want to protect. So, the aim of this work was to evaluate the acute toxicity of water and sediment samples collected in the area under the influence of this discharge using the native marine amphipod *Parhyale hawaiensis*. Three campaigns were conducted. Acute toxicity tests were performed in water and in fresh and dried sediment as well water and organic extracts (2.5DCM:1MeOH). Liquid samples were tested using 96-wells microplates, and the sediment using 12 wells-microplates containing sediment and salt water in 1:4 (w/v). Exposure conditions were 96h, 24±2ºC, 12h/12h light and dark. All water and water extracts samples did not present toxicity. Fresh and dried sediment were toxic ranging from 17 to 100% mortality as well the respective organic extracts. The observed toxicity is probably mainly related to organic contaminants adsorbed to the sediment particles. The sediment of the area seems to be adversely affected by the influence the outfall discharge. Acknowledgements: FAPESP 2015/24758-5 and CNPq 400362/2014-7.

WE191

Swimming in turbid water: impacts of suspended fine sediments on fish physiology

M. Lefranc, S. Amaudrut, L. Merle, J. ORourke, Hepia, University of Applied Sciences Western Switzerland / Ecology and Engineering of Aquatic systems research group; L. Espinat, INRA; S. Bony, INRA-CNRS / IPE; A. Devaux, INRA-CNRS / UMR LEHNA USC INRA IGH ENTPE; J. Guillard, INRA, Université Savoie Mont Blanc / CARRTEL Centre alpins sur les réseaux trophiques des écosystemes limniques; F. Cattaneo, R. Santos, Hepia, University of Applied Sciences Western Switzerland / Ecology and Engineering of Aquatic systems research group

Anthropogenic activities lead to increasing sediment deposition in many rivers worldwide which must be managed to preserve industrial activities and population safety. In this context sediment dredging and dam sediment flushing are common actions that release downstream accumulated sediments thus increasing sediment loads which in turn impairs freshwater biodiversity. Reported impacts on fish species vary from mortality, behavioral changes, to physiological and histological impairment depending on SSC and exposure duration. Juveniles of rainbow trout (*Onchorhynchus mykiss*) and roach (*Rutilus rutilus*) were exposed in semi-static conditions to 40, 200 and 1000 mg/L of non-contaminated fine sediments (mica) for 28 days, mimicking dredging operations in terms of duration and environmental concentrations. They were sampled weekly, and mortality, condition index and histological gill lesions were evaluated. Several physiological parameters were also investigated to assess the level of oxidative stress and genotoxicity. Oxidative damages in gills were investigated measuring the level of lipid peroxidation (TBARS) as well as superoxide dismutase activity (SOD) involved in oxygen radical metabolism. The level of primary DNA damage in erythrocytes was measured with the alkaline comet assay. This preliminary work highlight that 28 days of exposure to fine sediments at high concentrations do not induce drastic mortalities as predicted by models and the scientific literature. Physiological parameters investigated in rainbow trout acted as early signals of biological defects pointing out a high level of genotoxicity measured in erythrocytes in exposed individuals as well as in the control batch; these decreased during the experiment until a basal level pointing out the resilience of fish whereas they were exposed for 28 days to high fine sediment concentrations. Roach exposure to suspended fine sediments did not induce genotoxicity or an oxidative stress. These results meant that fine sediment exposure did not lead to a physiological stress through the alteration of respiration and osmoregulation homeostasis but suggested that trout experienced undesired past stressful conditions (aquaculture) independent from the sediment exposure. However, we cannot conclude that exposures of juvenile fish to such sediment concentrations would not lead to biological detrimental effects without further considering environmental sediment quality.

WE192

Assessing the bioavailability of metals in natural sediments by DGT passive sampling and bioaccumulation

H. Hetjens, SPHERE / SPHERE; K. de Schamphelaere, University of Antwerp / Department of Biology SPHERE and ECOBE Research Groups; J. Teuchies, E. Amato, L. Bervoets, University of Antwerp / Department of Biology (SPHERE Research Group)

Worldwide, high metal concentrations from recent and historic sediment contamination form a widespread problem and are of major concern for water system management due to their impact on the surrounding water quality and resident biota. Sediment related metals can be present in a range of different physicochemical forms, some of which may be unavailable, non-toxic and therefore not-harmful to organisms so that the interplay between chemical speciation and biological effects can be very site specific and hard to predict. Total sediment concentrations are therefore often found to be poor predictors of the actual risk and a measure of bioavailability should be considered in risk assessment procedures. In this regard however, the established technique of measuring the level of bioaccumulation in exposed organisms has been experienced to be highly disruptive, time consuming and limited in comparability as its results are strongly dependent on the analysed organisms themselves. Therefore, an increasing need for less disruptive, more reliable and standardized methods exists. Recently, passive samplers have been tested to estimate bioavailable contaminant fractions as well as the contaminant flux over sediment-water interface. Diffusive Gradient in Thin film samplers (DGTs) have been indicated to provide reliable predictions of metal bioavailability and toxic potential for single (benthic) invertebrate species under (semi-) controlled conditions. The main objective of this study is to further evaluate the use of DGT passive samplers as indicators for the bioavailability of metals for (benthic) macroinvertebrates and to test the robustness of the results from laboratory studies under field conditions. In an extensive field and laboratory study, which will be performed in April 2018, the impact of a range of contaminated natural freshwater sediments with known physicochemical characteristics and metal gradients on species performance will be tested. Bioavailable metal fractions will be determined by the use of DGTs and by measuring the metal body burden and mortality of different (benthic) macroinvertebrates. The results of this study are expected to increase the insights in the applicability of passive samplers for future sediment risk assessment and to be useful for the development of more standardized and integrated approaches.

WE193

Ecotoxicological effects of sediments influenced by a municipal wastewater treatment plant - state of a receiving river before implementing an ozonation treatment

N. Wilbrand, RWTH Aachen University; A. Shuliakevich, Institute for Environmental Research (RWTH Aachen University) / Institute for Environmental Research; Y. Müller, RWTH Aachen University / Institute for Environmental Research; S. Schiwy, RWTH Aachen University / Department of Ecosystem Analysis; H. Hollert, RWTH Aachen University / Institute for Environmental Research

Since our economic progress continues, the environmental pollution increases. The contamination of the aquatic environment with chemicals is one of the major concerns of our contemporary society. More than 300 million tons of compounds are applied in industrial processes and consumables every year. A considerable part of it enters waterbodies from diffuse and point sources. [1] Micropollutants originating from e.g. pharmaceuticals and personal care products may cause adverse effects on different biological and ecological levels. A major concern is the fact that these substances are not fully removed during common wastewater treatment and, therefore, released into surface waters. To minimize the discharge of micropollutants from wastewater treatment plants (WWTP) additional treatment steps are required. Ozonation has been shown to be an effective method with reasonable costs. Hence, ozone treatment of the entire effluent is implemented in the Aachen-Soers WWTP, Germany, within the DemO_3AC-project. In this context, the actual ecotoxicological state of the recipient water, the River Wurm, and the upstream tributary, the River Haarbach, was evaluated before the implementation of the facility. The River Haarbach receives effluents from the Aachen-Eilendorf WWTP. Therefore, water and sediment samples from various sites upstream and downstream the WWTPs were investigated. This study focuses on the assessment of the sediment samples of the both rivers, since they play an important role in e.g. binding and remobilisation of substances. After the extraction of the sediments via a pressurised liquid extraction, cell-based bioassays with reporter cell lines will be conducted to estimate the anti-estrogenic and oxidative stress potential. Following, both native samples and extracts will be tested in the behavioural light/dark transition test with *Danio rerio*. This test utilizes the fish's scototaxis (aversion to bright areas and natural preference for the dark) to evaluated effects of neurotoxic compounds within these matrices. [1] Schwarzenbach et al. (2006). Science.

WE194

Dredging sediment quality evaluation: a comparison of an ecotoxicological classification using an weight-of-evidence approach and a "pass to fail" criteria

V. Piazza, E. Costa, F. Garaventa, CNR ISMAR; D. Sartori, V. Vitiello, D. Pellegrini, ISPRA Institute for Environmental Protection and Research; I. Lanzoni, Department of Life and Environmental Sciences Polytechnic University of Marche Ancona Italy; F. Regoli, Università Politecnica delle Marche; M. Faimali, CNR ISMAR

Recently a new regulation for the management of dredging sediment has been introduced in Italian legislation (Decree of Italian Ministry of Environment n. 173/2016), establishing criteria and methodological procedures for dredging sediment characterization, their classification and identification of appropriate management options and monitoring. One of main novelties is represented by the priority role assumed by ecotoxicology in sediment characterization. A battery of bioassays that considers the use of three species belonging to different trophic levels has to be applied both to solid phase and liquid phase (pore water or elutriate) of sediment. The results of ecotoxicological analyses are then assessed as a whole at the level of "battery" (not of single bioassay), weighting the biological relevance of the measured effects, the sensitivity of organism, the statistical significance of measured results and the assay conditions in terms of tested matrix and duration of exposure. Chemical and ecotoxicological data are finally integrated for sediment quality assessment, following the weight of evidence (WOE) criteria, this representing an innovative approach respect to previous regulation, where chemical classification was determined by at least one parameter exceeding the threshold level and ecotoxicological classification was determined by the worst bioassay result of the whole battery. In this work, a comparison between "old" and "new" sediment quality assessment was performed, applying the two classification methods to ecotoxicological data obtained on dredging sediments from different study sites. Results obtained underline the importance of using an integrated and weighted approach (WOE) respect to a "pass to fail" criteria.

WE195

Toxicity of sediment-bound lufenuron to aquatic arthropods in laboratory bioassays

T. Brock, Alterra, Wageningen University and Research Centre / Environmental Risk Assessment Team; D. Belgers, Wageningen Environmental Research Alterra / Environmental Risk Assessment Team; M. Boerwinkel, Wageningen Environmental Research / Environmental Risk Assessment Team; M. Kraak, University of Amsterdam / IBED-FAME; J. Vonk, University of Amsterdam IBED Institute / Aquatic Environmetal Ecology; I. Roessink, Alterra / Environmental Risk Assessment

Lipophylic pesticides are frequently detected in sediments, potentially leading to toxic effects on benthic organisms. Currently, prospective sediment risk assessments for pesticides are mainly based on results of laboratory bioassays with a few standard test species (*Chironomus* sp. and *Hyalella azteca*). It is, however, uncertain whether these standard benthic test species are representative for a wider array of freshwater benthic organisms. We selected the benzoylurea insecticide lufenuron as one of the benchmark substances to evaluate the prospective environmental effect assessment procedure for sediment-associated pesticides. 10-day and 28-day toxicity estimates from sediment-spiked laboratory bioassays with benthic arthropods belonging to different taxonomic groups are presented. In the 10-d sediment-spiked toxicity tests the LC50 values showed the following order from low to high LC50: *Caenis horaria* > *Chironomus riparius* > *Gammarus pulex* > *Chironomus* gr. *thummi* > *Sericostoma personatum* > *Chironomus dilutus* > *Hyallella azteca* > *Asellus aquaticus* and *Sialis lutaria*. The Hazardous Concentration to 5% of the tested species (HC5 and 95% confidence limit) derived from these 10d-LC50 values was 2.2 (1.2-5.7) µg/g organic carbon (OC). This HC5 value is approximately a factor of 2 lower that the 10-d LC50 estimate (4.37 µg/g OC) for the most sensitive standard test species *Chironomus riparius*. Valid 28-d LC10 values could be derived for 7 benthic arthropods. These 28d-LC10 values showed the following order from low to high LC10: *Asellus aquaticus* > *Chironomus riparius* > *Caenis horaria* > *Ephemera danica* > *Hyalella azteca* > *Gammarus pulex* > *Sialis lutaria*. The HC5 and 95 confidence interval derived from these 28d-LC10 values was 0.13 (0.02-1.50) µg/g OC. This HC5 value is approximately a factor of 3 lower that the 28-d LC10 estimate for the most sensitive standard test species *Chironomus riparius* (0.49 µg/g OC). These data show that *Chironomus riparius* is a representative standard test species to assess the potential risks of sediment exposure to the insecticide lufenuron. The HC5 obtained from 28-LC10 values was a factor of 6 lower than the the NOEC for the most sensitive population (0.79 µg/g OC) in a sediment-spiked microcosm experiment, while the HC5 from 10d-LC50's was approximately a factor of 3 higher than this microcosm threshold concentration.

WE196

Application of an undisturbed sampling technique for depth related analysis of sediment particles and pore-water in OECD TG 219 sediment test systems

A. Dorn, Hochschule Niederrhein / Department of Chemistry; P. Dalkmann, Bayer AG Crop Science Division; D. Faber, Bayer AG, Crop Science Division / BCS D ETX Ecotoxicology; K. Hammel, Bayer AG, Crop Science Division / Environmental Safety; E. Hellpointner, Bayer AG, Research & Development, Crop Science / Environmental Safety; E. Bruns, Bayer AG, Division Bayer CropScience / Ecotoxicology; M. Jäger, Hochschule Niederrhein / Department of Chemistry

Sediment toxicity testing of plant protection products (PPP) is gaining an increasing awareness within the scientific and regulatory community. Currently, PPP concentrations in sediment and pore-water of Chironomid toxicity tests acc. to OECD test guideline (TG) 218/219 are determined as mean over the entire sediment layer of the test system. Hence, a depth-related measurement would contribute to a more accurate assessment of the effective exposure for the predominantly surface sediment dwelling test organism. Therefore, we developed an undisturbed sampling technique and processing enabling the depth-related analysis of active substances in pore-water and adsorbed to sediment particles. After removing the water phase, plastic tubes were stung into the ca. 15 mm-thick sediment layer, which was subsequently frozen by liquid nitrogen. By the use of a special developed cutting device, the sediment cores were cut into three slices providing a thickness of ca. 5 mm. Each sediment slice was centrifuged to isolate the pore-water. A sequential extraction was performed to extract the sediment adsorbed residues. After combining the sediment extracts, pore-waters and sediment extracts were analysed by LC-MS/MS. To validate this newly developed sampling technique, a Chironomid toxicity study acc. to OECD TG 219 was conducted. Two model compounds A ($\log P_{ow} < 1$) and B ($\log P_{ow} > 3$) were applied as mixture at nominal concentrations of 2 µg/L. To investigate the spatiotemporal behaviour of the compounds, test systems were incubated and processed 3, 7, 14, 21, and 28 days after treatment (DAT). The concentration of the applied compounds decreased in the overlying water during the experimental duration. Both compounds were primarily found adsorbed on the sediment phase (ca. 40-50 % of applied compounds). Approx. 0.2 - 1 % of the applied compounds were recovered in the pore-water at the same time. The analytical results of pore-water and sediment extracts show that the highest amounts of both compounds were in the upper layer of the sediment during the experimental duration. Nevertheless, the results indicate the concentrations differences between the top layer and the layers below will be equalized in the course of time. These first results indicate that the newly developed sampling technique can provide a substantial contribution to a more realistic determination of exposure concentrations in chronic water-sediment toxicity tests, leading to an improved sediment risk assessment.

WE197

SETAC Sediment Interest Group

P.K. Sibley, University of Guelph / School of Environmental Sciences

Improving the environmental risk assessment of the aquaculture 'Blue Revolution' (P)

WE198

Global overview of aquaculture production with a focus on the development and current status of the activity in Portugal

C.V. Rocha, MARE-FCUL; H.N. Cabral, Faculdade de Ciências da Universidade de Lisboa / MARE-FCUL; J.C. Marques, University of Coimbra / MARE, Dep. of Life Sciences, Coimbra University; A.M. Gonçalves, MARE, Dep. of Life Sciences, Coimbra University/Biologia Department & CESAM, Aveiro University

Aquaculture activity experienced true global development firstly at the beginning of the 1900's and then around 1970's, as a result of the awareness of the negative impacts that years of intensive fisheries brought to wild stocks' status, which contributed to the development of aquaculture as an alternative means of producing animal protein, releasing, at the same time, pressure over wild populations. Production from capture fisheries has relatively stabilized for the past decades, whereas aquaculture production of aquatic animals has followed a rising trend, amounting, by 2014, 73.8 million tonnes produced. Asia contributed for about 89 percent of that production, followed by America, with a production of around 5 percent and Europe, contributing for about 4 percent of the world's aquatic animals' production in 2014. Portugal is a traditional fishing country, with yet little expression in terms of aquaculture production, focusing mainly on the rearing of fish and molluscs, whose relative production has somewhat fluctuated over the years, but reached a similar production by 2014. Most of Portuguese aquaculture facilities operate in estuaries and coastal lagoons using mainly extensive and semi-intensive rearing systems. The main fish species reared in Portugal in 2014 in transitional environments were the Gilthead seabream and the European seabass; clam production accounted for nearly 50 percent of mollusc production, followed by mussel culture. Aquaculture activity in Portugal has presented a rising trend over the years, however, extensive and semi-intensive rearing systems are poorly controlled, raising questions not only about the influence of environmental factors on production, but also on the threats that the activity may pose to the surrounding environment. Among these are, for example, the destruction of natural habitats due to facilities extension and aquaculture effluent discharges with high nutrient input to the surrounding water bodies. Careful site selection and efficient waste management plans are imperative to minimize these potential threats of aquaculture practices. Although fish supply for human consumption from aquaculture has already surpassed that of fisheries, concerns about farmed fish quality have been raised. Fortunately, it has also resulted on the honing of aquaculture methods and practices, especially concerning the control of water quality and animal feeding, in order to achieve the highest quality product.

WE199

Characterization of the ontogenetic variation and nutritional composition of Gilthead seabream and European seabass reared in two Portuguese estuaries

C.V. Rocha, MARE-FCUL; H.N. Cabral, Faculdade de Ciências da Universidade de Lisboa / MARE-FCUL; C. Nunes, ICECO & QOPNA, Aveiro University; M.A. Coimbra, QOPNA, Universidade de Aveiro; F. Gonçalves, University of Aveiro / Department of Biology and CESAM; J.C. Marques, University of Coimbra / MARE, Dep. of Life Sciences, Coimbra University; A.M. Gonçalves, MARE, Dep. of Life Sciences, Coimbra University/Biologia Department & CESAM, Aveiro University

The nutritional value and potential ontogenetic variations of cultured fish was assessed for the European seabass and the gilthead seabream specimens reared in semi-intensive systems in two southern European estuarine systems, the Sado and Mondego estuaries in Portugal. Quantification of total protein, carbohydrate and fatty acid profiles were determined to assess differences between the organoleptic composition of organisms of the same species reared in four different aquacultures (two aquacultures in each estuary). No significant differences were found among groups of both species regarding protein content. A significant influence of the rearing site was found for the European seabass regarding saturated (SFA), monounsaturated (MUFA) and highly unsaturated fatty acid (HUFA) contents, either between estuaries and within each estuary. In the Gilthead seabream, SFA, MUFA, polyunsaturated fatty acid and HUFA contents were also influenced by the rearing site. Eicosapentaenoic acid, docosahexaenoic acid, arachidonic acid and linoleic acid content in adults specimens were dependent on the fish rearing site. In general, seabass and seabream fatty acid content was higher in organisms reared in the Sado estuary, when compared to the individuals reared in the Mondego estuary. Carbohydrate analysis showed a significant influence of the rearing site on free sugar and polysaccharide content in fish of both species. Differences in fatty acids and carbohydrates content among juvenile and adult stages were found for all groups studied. The present work supported the existing evidence that semi-intensive rearing systems are subjected to the variability of extrinsic factors, namely the different anthropogenic pressures these systems are subjected to, causing fluctuations in water quality and composition, which may influence the final nutritional value of the same species produced in different sites. From a consumer's perspective, such differences may come as a disadvantage, as it is expected for a product to provide equal nutritional properties and benefits regardless its origin, especially within the same country. From the farmer's perspective, such vulnerability requires constant monitoring of the extrinsic parameters within the facilities, in order to avoid great losses of the product. Further studies may be conducted to define the parameters that should be regulated and monitored in semi-intensive systems to obtain the best product with the highest food quality.

WE200

Effects of aquaculture antibiotics on marine biofilms and on the amphipod Gammarus aequicauda

B. Gonzalez-Gaya, IMDEA Water (G84912732) / Environmental Chemistry; N. García Bueno, I. Gomez, J. Martinez, C. Marin, B. Martinez-Lopez, A. Marin, University of Murcia / Ecology and Hydrology; A. Rico, IMDEA Water Institute / Aquatic Ecotoxicology

Intensive aquaculture is considered to be an important source of antibiotics into the marine environment. Antibiotics may cause adverse effects on potentially sensitive organisms such as non-target microorganisms, primary producers or benthic invertebrates. The objective of this study was to evaluate the potential side effects of two antibiotics used in aquaculture (oxytetracycline and flumequine) on the community composition of marine biofilms exposed to these substances and on the marine crustacean *Gammarus aequicauda*. Marine biofilms incubated in the sea were exposed to 1, 10, 100 and 1000 µg/L of oxytetracycline and flumequine for one week under laboratory conditions. Subsequently, the exposed biofilms were used to feed *G. aequicauda* organisms for two weeks. The *G. aequicauda* aquaria set up was run with two treatments in parallel: (1) with input of antibiotics only from the biofilm and (2) with antibiotics administered via biofilm and also spiked into the water. This was done to test different bioaccumulation routes (i.e., ingestion only and water exposure combined with ingestion). All the treatments for biofilm and crustaceans test were run in triplicate. Preliminary results show a marginally enhanced biomass growth of the biofilm with increasing dose of both antibiotics,

being this slightly higher in the oxytetracycline test. No correlation was found between antibiotics concentration and elemental composition (analyzed carbon, sulfur and total phosphorous), although nitrogen content was slightly higher in the medicated biofilm. Biofilm arborescence (vertical structures observed through optical microscopy) coverage was statistically different among treatments, showing a non-linear response. Experimental results show that low exposure concentrations contributed to a higher arborescence up until 100 µg/L, while the highest tested concentration contributed to a decrease of the biofilm's coverage. Ongoing work includes the evaluation of antibiotic's bioaccumulation in the biofilms, bacterial genetic characterization (microbiome and resistome), diatoms identification, and photosynthetic activity assessment. Regarding the *G. aequicauda* test, the experimental results do not show an effect over their size or weight by the feeding on antibiotic exposed biofilms. Reproduction and survival were not compromised either. Also bioaccumulation will be analyzed to describe the relevance of each of the evaluated antibiotic exposure routes.

WE201

Shifts in the diatom assemblage structure and biological traits of marine biofilms exposed to antibiotics used in aquaculture

N. García Bueno, C. Marin, A. Marin, University of Murcia / Ecology and Hydrology; B. Gonzalez-Gaya, IMDEA Water (G84912732) / Environmental Chemistry; A. Rico, IMDEA Water Institute / Aquatic Ecotoxicology

The use of antibiotics, anti-fouling paints, and fish food are all potential sources of chemical contaminants from commercial aquaculture operations such as fish farms. Intensive aquaculture has led to growing problems with bacterial diseases, the treatment of which requires the intensive use of antibiotics. The objective of this study was to evaluate the potential side effect of two antibiotics used in aquaculture (oxytetracycline and flumequine) on the diatom assemblage structure and biological traits of marine biofilms exposed to these substances. Biofilms were grown during two weeks under natural conditions on glass slides. Then two experiments were carried out under laboratory conditions. In the first experiment field-grown marine biofilms were exposed to 1, 10, 100 and 1000 µg/L of each single antibiotic compound for one week and then changes on chemical and biological composition were analyzed. In the second experiment, biofilms exposed to the same antibiotic concentration range were transported to field conditions after two weeks of exposure in order to evaluate their recovery capacity. In both experiments, diatoms were determined to the lowest possible taxonomic level under the microscope (Nikon Eclipse TE2000-U). The taxon abundance of the sampled quadrants of each replicate was averaged and referred to the area sampled to obtain the taxon density per replicate. The fine structure of diatoms was analyzed under a scanning electron microscope (JEOL-6100). The diatom composition, the relative abundance of species (%), the Shannon-Wiener diversity index (H´) and species richness were calculated for each sample and then summarized per treatment. The growth forms (biological traits) of species were analyzed before detachment and were grouped according to the literature. The biofilms were dominated by a reduced number of taxa, including the diatoms *Brachysira aponina* and *Cocconeis placentula*. High exposure concentrations of oxytetracycline and flumequine (100 and 1000 µg/L) resulted in an abundance decrease of the genera *Navicula*, *Hyalosynedra* and *Licmophora*. The global architecture and traits of the biofilms were also influenced by the high antibiotic exposure concentrations.

WE202

Assessing the oxidizing effects of hydrogen peroxide using flow cytometry as a high throughput method

A. Almeida, Norwegian Institute for Water Research NIVA; A. Lillicrap, NIVA / Ecotoxicology and Risk Assessment

Hydrogen peroxide (H_2O_2) is widely used in commercial, industrial, medical, environmental and hygiene applications. It is applied in aquaculture for controlling biological problems such as salmon lice. H_2O_2 produces highly oxidizing radicals that can cause paralysis, peroxidation in organelle membranes and inhibition of enzymes that replicate DNA in biological organisms. The release of H_2O_2 as an effluent into the marine environment is therefore a cause for concern particularly to primary producers such as algae. With the use of flow cytometry, single cells of algae with different features and physiological states, can be examined based on the quantification of scattered and fluorescent light signals. The use of specific probes enables fluorometric determinations of several parameters such as the ability of reactive oxygen species (ROS) to oxidize non-fluorescent probes to fluorescent products. In the present study, the effects of H_2O_2 on *Skeletonema pseudocostatum* were analysed. The method provided a rapid assessment of several endpoints in the same exposed samples. Effects on growth, photopigments and the detection of *in vivo* ROS production, using 3 molecular probes, were measured over 72 hours. H_2DFFDA was used for determining the oxidative burst, DHR 123 for mitochondrial oxidation, and BODIPY$^{581/591}$ to determine lipid peroxidation (LPO). Exposure concentrations were selected to cover the overall concentration response curve and a short-term exposure was also made to discern initial high reactivity of H_2O_2. Chemical analyses were performed to verify the stability of the concentrations during the exposure duration. The short-term exposures demonstrated rapid high toxicity of H_2O_2 to algae, where ROS production and the response to the photopigments were the observed endpoints. Over the 72 h, the response of the algae at the different test concentration clearly differed. The accessory photopigments actively responded when the main natural pigments declined. The ROS protective system seemed to be active at medium concentrations, whereas at higher concentrations damage on membranes lipids and mitochondria possibly instigated cell failure. This high throughput approach demonstrated a great potential to study the oxidizing effects of hazardous compounds in algae. While growth inhibition allowed to discriminate the overall toxicity, thehigh throughput methods, using flow cytometry, helped to screen and characterize the Mode of Action of H_2O_2.

WE203

An updated version of the SEPA BathAuto tool for assessing anti-parasitic chemical treatments in marine fish farms

J. Carnall, Cambridge Environmental Assessments; A. Berkeley, Scottish Environment Protection Agency; F. Ericher, CEA; G. Hughes, Cambridge Environmental Assessments

Marine fish farms operators in Scotland wishing to use anti-parasitic chemicals as bath treatments must first obtain a discharge licence from the Scottish Environment Protection Agency (SEPA). Discharge licences are granted by SEPA on a per-site basis, with the quantity of chemical that can be released from a particular fish farm determined by computer modelling. This computer modelling considers the location and composition of the fish farm, along with hydrographic data measured at the site and the toxicity and environmental fate of the chemicals concerned. Discharge quantities are typically calculated for three anti-parasitic chemicals: azamethiphos, cypermethrin and deltamethrin. Of these, cypermethrin and deltamethrin are rapidly removed from the aqueous phase *via* binding to particles, and are therefore assessed using SEPA's short-term model, which calculates projected concentrations in the chemical patch up to 6 hours after its release from the fish farm. Azamethiphos, however, remains in the aqueous phase for several days until it is broken down, and is therefore also assessed using a longer-term model, originally developed by Gillibrand and Turrell (1999; MLA Report No 2/99) and recently extended by Carnall, Ericher and Hughes (2017; poster presentation at SETAC Europe 2017). The SEPA tool BathAuto integrates both the short and long-term models, iteratively performing calculations of chemical concentrations in the water in order to arrive at safe discharge limits for a fish farm. In this poster we present an updated version of BathAuto in which the longer-term model is now fully integrated into the BathAuto tool. The standalone executable required previously for the long term calculations (opendisp.exe) is no longer used, improving the compatibility of the revised tool with modern operating systems. While the standard modelling parameters required by SEPA for discharge licensing in Scotland are included as default, BathAuto could readily be modified to include alternative parameters or chemical data, potentially making this tool available for use in other jurisdictions and regulatory frameworks. The revised version of BathAuto also offers improved graphical outputs, and the ability to calculate several options for compliant cage treatment regimes.

WE204

State-of-the-art on the use of models for the ERA of chemicals used in aquaculture

A. Rico, IMDEA Water Institute / Aquatic Ecotoxicology; M. Vighi, IMDEA Water Institute / Earth and Environmental Sciences; T.P. Teixeira, AquaBio Tech Group; A. Lillicrap, NIVA Norwegian Institute for Water Research / Ecotoxicology; A.L. Macken, Norwegian Institute for Water Research, NIVA; P. van den Brink, Alterra/Wageningen UR / Aquatic Ecology and Water Quality Management Group b Alterra; M. ter Horst, Alterra, WUR / Environmental Risk Assessment Team; M. Tsapakis, Hellenic Centre for Marine Science / Institute of Oceanography; I. Kalantzi, Hellenic Centre for Marine Research / Institute of Oceanography; R. Torres, Plymouth Marine Laboratory; N. García Bueno, A. Marin, University of Murcia / Ecology and Hydrology; L. Falconer, Institute of Aquaculture; T.C. Telfer, University of Stirling

As aquaculture is expanding and becoming a more diverse industry in terms of species and production systems, there is an increasing demand to generate improved modelling tools to assess its environmental impacts. Although important developments have been made in disease prevention and treatment, the use of veterinary medicines and other potentially toxic substances (e.g. antifoulants, metals) still raise environmental concerns and need to be adequately assessed. This study presents an overview on the use veterinary medicines and other potentially toxic substances used in EU aquaculture, and the environmental standards and regulatory procedures available for their Ecological Risk Assessment (ERA). Furthermore, it describes the state-of-the-art on the development of models capable of assessing the fate, dispersal, exposure, ecological effects and associated ecotoxicological risks of veterinary medicines applied in aquaculture production. This study shows that a varied range of models has been developed during the last 30 years. Their effective implementation in regulatory ERA is, however, somewhat limited in many state members. Some recommendations are provided as to improve the chemical exposure assessments and the ecological realism of the modelling outcomes, paying a special attention to the protection goals set for the regulatory ERA of veterinary medicines.

WE205

Effects of an aquaculture parasiticide (diflubenzuron) on non-target shrimp

populations: from lab experiments to population-level endpoints

J. Moe, Norwegian Institute for Water Research (NIVA) / Section for Catchment Processes; D. Hjermann, NIVA Norwegian Institute for Water Research; E. Ravagnan, R. Bechmann, International Research Institute of Stavanger

The continued growth of marine aquaculture production has presented the industry with environmental and production concerns, of which the ectoparastic salmon lice (*Lepeophtheirus salmonis*) has gradually become a major problem. A commonly used parasiticide against this crustacean is diflubenzuron (DFB), which acts as a chitin synthesis inhibitor and thereby interfere with the moulting stages during sea lice development. However, DFB from the fish feed may also affect non-target crustaceans such as the Northern shrimp (*Pandalus borealis*), which is an economically and ecologically important species in Norwegian fjords. Laboratory experiments have demonstrated that shrimp exposed to DFB through fish feed have reduced survival (ca. 60%) compared to control, in both the larval and the adult stages. Moreover, the effects of DFB exposure is more severe under future climate conditions (higher temperature). The aim of this study is to make the information on these mechanistic effects more relevant for risk assessment at the population level. We have developed an age-structured population model representing a Northern shrimp population located in a hypothetical Norwegian fjord containing a fish farm, under both ambient and future climates. Our model is based on thorough knowledge of shrimp biology and clear results on toxicological effects from the laboratory experiments. Nevertheless, extrapolating the individual-level effects to the population level poses several challenges. Relevant information on shrimp populations in fjords is sparse (such as abundances, survival and reproductive rates, and density-dependent processes). The degree of exposure to medicine feed at different distances from aquaculture farms is also uncertain. We have therefore developed a set model scenarios representing different medicine application schemes and different degrees of exposure for the shrimp populations. The purpose of the model is to predict effects of DFB exposure on population-level endpoints such as long-term abundance and age structure, and to assess the risk of population decline below threshold abundances.

WE206

Contamination and bioaccumulation of heavy metals in the wild and marine farmed milkfish (Chanos chanos) and mullet (Mugil cephalous) and associated health risk along the coasts of Tanzania

E.B. Mwakalapa, Norwegian University of Life Sciences / Department of Food Safety and Infectious Biology; C.K. Simukoko, University of Zambia; J.L. Lyche, Norwegian University of Life Sciences; M.H. Müller, Norwegian University of Life Sciences / Department of Food Safety and Infection Biology; A.J. Mmochi, Institute of Marine Sciences University of Dar es salaam; R.H. Mdegela, Sokoine University of Agriculture; A. Polder, Norwegian University of Life Sciences / Department of Food Safety and Infection Biology

Concentration of heavy metals Cu, Pb, Fe, Zn, Co, Cr, Cd, Ni, Al and As were analysed in the muscles and livers of farmed and wild milkfish and mullets from Tanzanian coast. Fish samples were collected from January 2016 to April 2016 and analysed for heavy metals by using Atomic Absorption Spectrophotometer while health risk to consumer was estimated using Target Hazard Quotient (THQ). With disregard to fish species and in order of decreasing dominance, the overall range of concentrations in (mg/kg ww) of heavy metals were; Fe (< LOD-11.96), Pb (0.54-1.96), Zn (< LOD-2.81), Cu (< LOD-2.31), Ni (0.015-0.098), Co (< LOD-0.086), Cd (< LOD-0.024, Cr (< LOD-0.079) and Fe (< LOD-147.9), Pb (0.92-47.37), Cu (< LOD-15.08), Zn (< LOD-12.24), Ni (0.027-0.094), Co (< LOD-0.034), Cd (< LOD-0.013) in muscles and livers respectively. Al and As were not detected above limit of detection in any of the analysed samples. Metals showed different affinity to muscles and livers of milkfish and mullets, whereas Co and Cd had higher levels in the muscles than in the livers, Cu, Pb, Fe, Zn and Ni were higher in livers than in the muscles in milkfish. In Mullet, Fe, Co, Cr, and Cd levels were higher in the muscles than in the livers and Cu, Pb, Zn, and Ni levels were higher in the livers than in the muscles. The concentration of Pb in the muscles of all analysed fish were above the Maximum Recommended Levels (MRLs) by FAO/WHO, EU and USFDA for human consumption. Other metals were below the MRLs. The THQ for all analysed metals were below 1 indicating that the metals are likely to present insignificant health risk to fish consumers. However, based on MRLs recommended for human consumption, the Pb metal may pose health risk to human as fish consumer and threatens fish biodiversity. Therefore, this research calls for regular monitoring of heavy metals in fish for consumption and strengthening pollution control measures.

WE207

Potential Toxic and Phototoxic Effects of Benzobicyclon on Crayfish

E.N. Vebrosky, Louisiana State University / Department of Environmental Sciences; L.M. Basirico, Louisiana State Univeristy; W. Xu, C.G. Lutz, Louisiana State University AgCenter / Renewable Natural Resources; K. Armbrust, Louisiana State University / Environmental Sciences School of the Coast and Environment

Benzobicyclon is the active ingredient in the herbicide, BUTTE®. In 2001, various formulations using benzobicyclon as the active ingredient were approved for use in Japan. It was approved for use in the United States and registered in California as of 2016, and was first applied to rice fields in the 2017 growing season. Benzobicyclon is a proherbicide that acts as a HPPD inhibitor, leading to the bleaching of weed species to ultimately kill them. With an increase in herbicide-resistant weed species, new formulations of herbicides to combat this in rice fields is advantageous. California is the second largest producer of rice in the United States, with Louisiana as the third largest producer; Arkansas leads the country in rice production. Unlike California, Louisiana has a unique system where rice fields not only grow rice but also grow crayfish for harvest and consumption. The flooded rice fields are prime habitat for crayfish to reside; therefore, crayfish are at risk for exposure to pesticides used in the rice production. Benzobicyclon readily hydrolyzes to benzobicyclon hydrolysate, and therefore it likely undergoes photolysis as well. The potential for toxic or phototoxic impacts of benzobicyclon or its intermediate degradation products to crayfish is important to know for the possible future registration of BUTTE® in Louisiana.

WE208

Effects of the isoflavones, genistein and daidzein, on Acetylcholinesterase from head of Solea Senegalensis.

G. Albendín, Universidad de Cádiz (Spain) / Toxicology Area; V. Aranda, University of Cadiz / Toxicology Area; M. Manuel, University of Cadiz / Analytical Chemistry; J. Ortiz, C. Sarasquete, CSIC Spanish National Research Council; J. Arellano, University of Cadiz / Toxicology Area

The aquaculture is an important economic activity in our area, and one of the main concerns of this sector is fish feed. One of the proposed alternatives to fish meal in feeds are vegetable proteins, and among plant protein sources, soybean is noteworthy. This plant has flavonoidsincluding the isoflavones daidzein and genistein. *Solea senegalensis* is a flatfish of high commercial importance both in aquaculture and fisheries in Southern Europe with a wide geographic distribution and availability, and its biological cycle is well known. The aim of this work was to study the effects of the isoflavones, genistein and daidzein, on juveniles (weigth 1,23 ±0,41 g) of *Solea senegalensis*. The 96-h toxicity tests were conducted with continuous aeration and water renewal every 24 hours, at a temperature of 19-20ºC and a photoperiod of 12h light/12h dark. Juveniles were exposed at five nominal concentrations of genistein (range 1.25-20 mg/l) and daidzein (range 0.625-10 mg/l), plus an untreated control and a solvent control (DMSO). Mortality was recorded and fish head acetylcholinesterase (AChE) was measured with acetylthiocholine as a substrate after inhibiting butyrylcholinesterase with iso-OMPA. No mortality was observed within the period of the test when the fishes were exposed up to 20 mg/l genistein and 10 mg/l daidzein. Besides, head AChE activity was not altered in fish exposed to genistein, but daidzein was found to enhance AChE activity at a concentration equal or higher than 2.5 mg/l after 4 days of exposure. Acknowledgements: Authors are grateful to the Laboratory of Marine Culture at University of Cádiz for providing the experimental fish used in this study. This work was supported by National R&D&i Plan Ministry of Economy, Industry and Competitiveness (Project: AGL2014-52906-R) and in part by the Andalusian Plan for Research, Development and Innovation (PAIDI group: RNM-345).

Luminescent biomonitoring via bioassays of different complexity - from cells trough enzyme reactions to proteins (P)

WE209

Comparison between results of LumiMARA and Microtox tests

M. LOT, CEHTRA; P. Thomas, CEHTRA SAS; P. Baldoni-Andrey, C. GELBER, F. Mounède, TOTAL SA

In a regulatory context of ever increasing environmental responsibility (OSPAR convention, BREF CWW), there is a need to have biomonitoring tools to evaluate waste water quality. To date, and for several decades, the standard toxicity testing tool used for rapid analysis of waste water has been Microtox®. However, recently a new tool has become available: LumiMARA®, an acute ecotoxicity bioassay which measures the inhibition of luminescence on bacteria in a similar way to Microtox®. Its main advantage is that it performs a simultaneous assay on numerous bacterial species (all in all 11 species, 9 of which are marine and include Vibrio fischeri, plus 2 freshwater bacteria) against only one for Microtox® (Vibrio fischeri). Using a set of narcotic substances with different hydrophobicities and two mixtures (one home-made formulation comprising equimolar concentrations of four of the narcotics tested and one petroleum based complex substance) a comparison of both tools was realised with the aim to determine which of the two is the better tool to evaluate effects on bacterial species. While it was recognised that the substances tested would not be directly comparable to whole effluents, this method was felt to be appropriate to compare the two assays. The in-built EC50 calculation software were also verified. Some errors were detected with the LumiMARA® effects calculation software, and these were recalculated by hand using Regtox software, when possible. We observed that Microtox® is more sensitive than LumiMARA® and that in the latter test, freshwater bacteria toxicity is generally lower than that of marine bacteria. This suggests that there may be a small but real difference between freshwater and marine bacteria toxicity however this cannot be concluded on the basis of this research. Moreover, Vibrio fischeri is the bacterium that usually has the lowest EC50 compared to other bacteria. Thus, the marine bacterium Vibrio fischeri classically used to determine the biotic effect in a freshwater environment may be overestimating toxicity of effluents to the freshwater compartment.

WE210

Bioluminescent assays as tools for studying antioxidant activity and toxicity of bioactive compounds

A.S. Sachkova, Tomsk Polytechnic University / School of Nuclear Science & Engineering; E. Kovel, Siberian Federal University; N. Kudryasheva, Institute of Biophysics SB RAS

This study promotes application of the bacteria-based and enzyme-based bioluminescent assays to evaluate the antioxidant activity of bioactive compounds in oxidizers solutions. Artificial and natural carbon nanostructures – fullerenol derivatives ($?_{60}?_{2–4}(??)_{20–24}+?_{70}?_{2–4}(??)_{20–24}$ and $?_{60}?_{2–4}(??)_{20–24}+?_{70}?_{2–4}(??)_{20–24}$) and humic substances (HS) are used here as bioactive compounds. Fullerenols are polyhydroxylated water-soluble derivatives of fullerenes, HS are products of natural transformation of organic matter in soil. Bioluminescent assays allow studying toxicity and antioxidant properties of substances. We studied the toxicity effect of organic (1,4-benzoquinone) and inorganic ($K_3[Fe(CN)_6]$) oxidizers on bioluminescent tests. We found the effective concentrations (EC_{50}) of these oxidizers decreasing bioluminescent intensity by 50%. The EC_{50} values of 1,4-benzoquinone were 2.5?10^{-7}M and 10^{-4}M for bacterial and enzymatic assays, respectively, while the EC_{50} values of $K_3[Fe(CN)_6]$ - 4?10^{-2}M and $2\cdot10^{-4}$M. Also we studied the influence of bioactive compounds on the assays. They suppressed bioluminescence of the bacterial and enzymatic systems at concentrations >10^{-2}g/L and >5?10^{-3}g/L, respectively. Detoxification coefficients can be calculated to characterize changes in toxicity under the action of bioactive compounds. The values of coefficients >1 and So, the bacteria- and enzyme-based assays showed similar peculiarities of detoxification of oxidizer solutions by bioactive compounds. Results show, that low concentrations of bioactive compounds were active. Our work demonstrates a high potential of the bioluminescent assay systems, bacterial and enzymatic, to characterize and compare antioxidant activity of physiologically active compounds.

WE211

Effect of low-dose gamma-radiation on luminous marine bacteria Photobacterium Phosphoreum.

A.S. Petrova, Krasnoyarsk State Agrarian University / Institute of Agroecological Technologies; D.V. Dementyev, Institute of Biophysics SB RAS / Radioecology Lab; N. Kudryasheva, Institute of Biophysics SB RAS

The study addresses biological effects of low-dose gamma-radiation. Radioactive 137Cs-containing particles were used as model sources of gamma-radiation. Luminous marine bacterium Photobacterium phosphoreum was used as a bioassay with the bioluminescent intensity as the physiological parameter tested. To investigate the sensitivity of the bacteria to the low-dose gamma-radiation exposure (≤250 mGy), the irradiation conditions were varied as follows: bioluminescence intensity was measured at 5, 10, and 20°? for 175, 100, and 47 h, respectively, at different dose rates (up to 4100 mGy/h). There was no noticeable effect of gamma-radiation at 5 and 10°?, while the 20°? exposure revealed authentic bioluminescence inhibition. The 20°? results of gamma-radiation exposure were compared to those for low-dose alpha- and beta-radiation exposures studied previously under comparable experimental conditions. In contrast to ionizing radiation of alpha and beta types, gamma-emission did not initiate bacterial bioluminescence activation (adaptive response). As with alpha- and beta-radiation, gamma emission did not demonstrate monotonic dose-effect dependencies; the bioluminescence inhibition efficiency was found to be related to the exposure time, while no dose rate dependence was found. The sequence analysis of 16S ribosomal RNA gene did not reveal a mutagenic effect of low-dose gamma radiation. The exposure time that caused 50% bioluminescence inhibition was suggested as a test parameter for radiotoxicity evaluation under conditions of chronic low-dose gamma irradiation[1]. The reported study was funded by Krasnoyarsk Regional Fund of Science according to the participation in the event: «28th Annual Meeting of the European Society for Environmental Toxicology and Chemistry / SETAC Europe 28th Annual Meeting. International» References: [1] Kudryasheva N.S., Petrova A.S., Dementyev D.V., Bondar A.A. 2017. Exposure of luminous marine bacteria to low-dose gamma-radiation. Journal of Environmental Radioactivity 169-170:64-69. \n

WE212

Bioluminescent Assay for Toxicological Assessment of Nanomaterials

E. Esimbekova, Institute of Biophysics SB RAS; E. Nemtseva, Siberian Federal University / Institute of Fundamental Biology and Biotechnology; V. Kratasyuk, Siberian Federal University / Biophysical

Due to the increasing scale of production and usage of a vast number of new materials in industrial and economic activities, society is faced with problems associated with a lack of materials safety assessment regarding humans, ecosystems and the biosphere as a whole. Nowadays, numerous toxicological investigations using living organisms, cell lines, etc. are carried out in laboratories in order to assess the potential risks of using these materials and their biological effects on human health and the environment. A rapid bioluminescent enzyme inhibition-based assay was applied to predict the potential toxicity of carbon nanomaterials (CNM) presented by single- and multi-walled nanotubes (SWCNT and MWCNT) and aqueous solutions of hydrated fullerene $?_{60}$ (C_{60}HyFn). This assay specifically detects the influence of substances on parameters of the soluble or immobilised coupled enzyme system of luminescent bacteria: NAD(P)?:FMN-oxidoreductase + luciferase (Red + Luc). A protocol based on the optical properties of CNM for correcting the results of the bioluminescent assay was also developed. If the value of optical density of the nanomaterial solution was greater than 0.1 in the range of 400–600 nm, the light emission intensity was multiplied by the correction factors. It was shown that the inhibitory activity of CNM on Red + Luc decreased in the following order: MWCNT > SWCNT > C_{60}HyFn. The soluble enzyme system Red + Luc had high sensitivity to MWCNT and SWCNT, with values of the inhibition parameter IC_{50} equal to 0.012 and 0.16 mg/L, respectively. The immobilised enzyme system was more vulnerable to C_{60}HyFn than its soluble form, with an IC_{50} equal to 1.4 mg/L. According to EC Directive 93/67/EEC for aquatic organisms, chemicals are classified by their degree of toxicity based on EC_{50} values. We hypothesised that this classification was correlated with IC_{50} values and revealed that MWCNT and SWCNT samples might be characterised as extremely toxic and very toxic, respectively. Due to its technical simplicity, rapid response time and high sensitivity, this bioluminescent method has the potential to be developed as a general enzyme inhibition-based assay for a wide variety of nanomaterials. This study was supported by the Russian Science Foundation (project no. 16-14-10115).

WE213

Delayed chlorophyll fluorescence in biomonitoring of environmental pollution

Y.S. Grigorev, Siberian Federal University / Department of Ecology and Environmental Study; E. Stravinskene, O. Kryuchkova, N. Pakharkova, Siberian Federal University

Plants have important role in biomonitoring of environmental pollution because of their high sensitivity to various pollutants which often disturb photosynthesis. The photosynthetic function of plants is therefore a good indicator to reveal the pollution effects. Fluorescence of chlorophyll is widely used for quick assessment of photosynthesis condition. A prompt fluorescence that can be registered by PAM fluorimeters is most commonly used for these purposes. Recently, we have developed a more sensitive and fast indicator of the state of the photosynthetic apparatus of plants, which is based on the measurement of delayed fluorescence (DF) of chlorophyll. Delayed fluorescence is registered in red spectrum area after exciting light flashes in the form of time-attenuated radiation. We found out that the ratio of DF excited by high intensity flashes to DF excited by low intensity flashes decreases more than tenfold when there is a toxic effect on photosynthesis. This indicator can be measured within a few seconds. Due to the relative nature of this indicator, which characterizes the photosynthetic activity of the plant test organism, it does not depend on the size or volume of analyzed sample. To implement this method, we developed the Photon 10 fluorimeter which automatically measures the relative indicator of delayed fluorescence (RIDF) in 24 plant samples. Simultaneously with RIDF, the device registers the variable part of prompt fluorescence of these samples. Applying this device we carried out transplanted lichen indication of air pollution in Krasnoyarsk. We found that the values of DF of several species of lichen differed up to 20 times between ecologically clean and industrial areas of the city after three weeks of the exposure. The study of the condition of pine needles in these areas showed a difference in the value of the RIDF up to 5 times. Our method also showed a high sensitivity in bioassay of toxicity of natural and waste waters where *Chlorella vulgaris* alga was used as a test-organism. RIDF of the algal cell suspension decreased by the factor of 2 (EC_{50}) in solutions of copper and zinc ions at a concentration of these heavy metals of 0.005 and 0.5 mg / dm^3 respectively.

WE214

Chlorophyll fluorescence temperature curve to estimate changes of the photosynthetic apparatus of coniferous trees during the transition to a state of winter dormancy in urban ecosystems

N. Pakharkova, Y.S. Grigorev, Siberian Federal University / Department of Ecology and Environmental Study; N. Gaevsky, Siberian Federal University

The main regulating factor for the transition of plants from active vegetation to winter dormancy is the change in the duration of daylight. However, the temperature factor and air pollution also have a significant influence both during the autumn photoperiodic reaction and at different phases of winter dormancy. This research aims towards a better understanding of the responses of the Scots pine and the Siberian spruce to air pollution stress in urban and semi-urban conditions of Southern Siberia. It is well-established that during the transition from the phase of active vegetation into the phase of winter dormancy the cells of photosynthesizing parenchyma of needles undergo a number of changes. Changes in the assembly of the photosynthetic apparatus are mirrored in changes of fluorescent signals emitted at different temperatures. Chlorophyll fluorescence temperature curve (FTC) is a dependence of chlorophyll fluorescence intensity on linearly increasing temperature. This curve is used for determination of the stability of PS2 and for evaluation of the structural arrangement of chloroplasts in vegetating plants. Also, based on the changes in the shape of the FTC it can be deduced whether the plant is in the state of winter dormancy or it is vegetating. The calculated ratio of the low- and high-temperature peaks (50°? and 70°?) of zero level fluorescence may be used as an indicator of the degree of the depth of dormancy. FTC was measured with the needles' segments using fluorometer Junior-PAM (Walz, Germany). The needles were linearly heated from 25 to 70° C at a rate of 2° C/min using a computer-controlled heating device. In climate conditions of Southern Siberia, disturbance of winter dormancy under air pollution stress represents a major threat to the health status of *Pinus sylvestris* and *Picea obovata*. Our data demonstrate that regardless the age of needles, the depth of winter dormancy of both species clearly correlates with air pollution levels, and the trees growing in industrial areas are easier to release from dormancy and to be affected by late winter or spring frost. In urban environments the risk of frost injuries is even higher due to early spring warm spells associated with heat island effect. Scots pine is less susceptible to air pollution and temperature fluctuations than Siberian spruce, and therefore represents a better choice for urban forestry projects.

WE215

Luminescent microscopy in the bioindication of the Baikal pollution with oil products and polyaromatic hydrocarbons

M.N. Saksonov, A.E. Balayan, Irkutsk State University / Research Institute of Biology of Irkutsk State University; O.A. Barkhatova, Irkutsk State University / Faculty of Geography; A.D. Stom, Irkutsk State University / Research Institute of Biology of Irkutsk State University

Against the backdrop of the growing diversity of toxic substances that degrade the natural environment, the creation of new analytical systems for assessing the state of the environment is of paramount importance. Among such systems, bioindication methods are increasingly important. Among the many pollutants of water bodies, it is necessary to isolate oil products and polyaromatic compounds (PAHs), the flow of which in the valley is constantly increasing. On Lake Baikal, this is due to the development of tourism, the increase in the number of passenger ships and tourist bases, often not equipped with treatment facilities. The necessary system for monitoring the quality of the aquatic environment, including methods for biotesting and bioindication, not only on generally accepted test facilities, but also on representative hydrobionts for this reservoir. Crustaceans of the order *Copepoda*, which have well-expressed fatty inclusions in the form of drops, there are several hundred species. *Epischura baicalensis Sars* (*Copepoda, Crustacea*) - endemic of Lake Baikal - dwells practically in the entire water column of the pelagic lake. *E.baicalensis* accounts for up to 70% of the total biomass of zooplankton. Crustaceans *Copepoda*, having fat inclusions, accumulate in them oil products. This can, in particular, be observed by the blue-violet glow in a luminescent microscope. The accumulation of diesel fuel by fatty inclusions of *Copepoda* crustaceans and the high sensitivity of luminescent microscopy make it possible to detect the microquantities of this pollutant in the water under investigation. It is noted that in the presence of unicellular algae that are absorbed by the crustaceans, this process is activated and the accumulation time of oil products and PAHs decreases. Another representative of the *Copepoda Cyclops Colensis* also has chaotically scattered fatty inclusions in which the accumulation of oil products can be seen in a fluorescent microscope. It has been experimentally revealed that if *E. baicalensis* with oil products or PAHs accumulated in fatty inclusions is added to pure water to *C. Colensis*, then the staining of fat inclusions also occurs in the latter, that is, this process is transmitted along the trophic chain. Based on these experiments, a method of bioindication on the accumulation of oil products and PAHs in *?opepoda* crustaceans in fat drops was proposed.

WE216

The correlation between fluorescent properties of water extract from soil and its effect on bioluminescent enzymatic bioassay

E. Nemtseva, O. Chmurina, Siberian Federal University / Laboratory of Bioluminescent Biotechnologies; M. Gerasimova, Siberian Federal University / School of Engineering Physics and Radio Electronics; V. Kratasyuk, Siberian Federal University / Biophysical

The work is devoted to the development of the bioluminescent enzymatic bioassay of the soil contamination.The problem of relation of bioassay results with intrinsic properties of the soils or/and the level of their contamination was under consideration. The study aimed at assessing of the correlation between the luminescent characteristics of the various soil samples and the results of their bioluminescent bioassay as well as their physico-chemical characteristics. Water extracts from 56 soils (medium and heavy loams, with humus content 1.2-11.3% and detected arsenic 3.05–15.39 mg/kg) were studied by the method of excitation-emission matrix (EEm) fluorescence spectroscopy. The luminescence in the range 290-600 nm under excitation at 250-450 nm was measured for each extract as well as absorption spectra in the range 200-800 nm. The physico-chemical characteristics of the soils (particle size distributions, pH, humus content, *etc.*) were compared with the inhibitory effect of water soil extracts on enzymatic bioassay based on the coupled bioluminescent reaction of bacterial luciferase and NAD(P)H:FMN-oxidoreductase. It was revealed that the spectral-luminescent characteristics of water extracts are similar for all soils and featured by three types of fluorophores with excitation maxima at about 270, 330 and 360 nm and emission maxima at about 330, 425 and 470 nm, respectively. The residual activity of the bioluminescent bioassay enzymes in the presence of soil extracts was found to correlate with intensity of two first bands that is the measure of the component content. Poor correlation was found between EEm characteristics and remaining chemical parameters of the soils including amount of detected arsenic. The conclusion was derived about relation of bioassay signal from studied extracts with the amount of humic substances in soils. The research was supported by the Russian Science Foundation (project no. 16-14-10115).

WE217

The comparison of enzyme systems for soil contamination bioassay

E. Kolosova, Siberian Federal University / Biophisical; D. Gulnov, Siberian Federal University; N. Rimatskaia, Siberian Federal University / Biophysical; A. Lisitsa, O. Sutormin, V. Kratasyuk, Siberian Federal University

Design of simple, quick and highly sensitive bioassays is extremely necessary for ecological soil monitoring. Enzyme systems may be a perspective basis for the development of modern methods of bioassay. With sets of enzymes, it is possible to simulate the effect of toxic substances present in natural environments on living organisms. Moreover, coupling enzyme-target with bacterial luciferase provides advantages in the signal detection. The purpose of this study is to evaluate the possibility of using various enzymatic systems for the analysis of soil contamination. In this work NADH: FMN-oxidoreductase, alcohol dehydrogenase (ADH), NADH: FMN-oxidoreductase + bacterial luciferase (two-enzyme system), NADH: FMN-oxidoreductase + bacterial luciferase + alcohol dehydrogenase (three-enzyme system) were examined. The enzyme activities were measured by addition of the model soil pollutants such as a blue copperas, the insect powder "Decis Profi" (Bayer CropScience) and diesel fuel. The values of the toxicological parameters $??_{20}$ and $??_{50}$ (concentrations of the pollutants causing the system inhibition by 20% and 50% respectively) were determined. The sensitivity of each enzymatic test system to the aqueous extract of soil was also analysed. The blue copperas (II) –water solution shows an inhibitory effect on all enzymatic systems. The value of EC_{50} is from 0.088 to 8.75 µM/L. The insect powder-water solution also shows an inhibitory effect on all enzymatic systems except ADH enzyme system. In contrast, the diesel fuel impacts only on enzyme systems coupled with bacterial luciferase. As a result the blue copperas (II) –water solution decreases catalytic activities all enzyme systems; the insect powder-water solution decreases only NADH: FMN-oxidoreductase catalytic activity; the diesel fuel decreases luciferase activity. The addition of aqueous extract of soil leads to activation of catalytic activity of NADH: FMN-oxidoreductase; for the two- and three-enzyme systems the addition of the aqueous extract of soil leads to inhibition of catalytic activities of the enzyme systems (more than 50%). The NADH: FMN-oxidoreductase + bacterial luciferase enzyme system showed the greater sensitivity to the soil pollutants than other systems. This fact is confirmed the prospect of using this system for environmental monitoring. *The study was supported by a grant from the Russian Science Foundation (project No 16-14-10115)

WE218

Are changes in bioluminescence kinetics of Photobacterium phosphoreum exposed to low-dose radiation connected with genetic mutations?

O. Guseynov, V. Guseynova, Siberian Federal University; T. Rozhko, Krasnoyarsk State Medical University. prof. VF Voyno-Yasenetsky; A. Bondar, Institute of Chemical Biology and Fundamental Medicine SB RAS; N. Kudryasheva, Institute of Biophysics SB RAS

Luminous bacteria of marine origin are widely employed as biological sensors for monitoring environmental toxicity including radiation toxicity. Due to growing use of radioactive elements and related concerns about the increase of background radiation, special attention is lately paid to the effects of low-dose radiation on the environment. The response of bioluminescence reaction to exposures of bacteria to low-dose alpha, beta and gamma radiation has been examined in previous research. The purpose of the current study was to determine whether bacterial genetic alteration is related to bioluminescence kinetics change under low-dose exposure with alpha-emitting (^{241}Am) and beta-emitting (^{3}H) radionuclides as sources of ionizing radiation. Bioluminescence kinetics of *Photobacterium phosphoreum* in solutions of $^{241}Am(NO_3)_3$, 7 kBq/L, and tritiated water, 100 MBq/L, were recorded and their stages were determined as follows – absence of effect, activation, and inhibition. For genetic analysis bacterial suspension was sampled at different stages of the bioluminescent kinetics ensuring that the doses accumulated by the samples were close or a little higher than a tentative limit of a low-dose interval: 0.10 and 0.85 Gy for ^{241}Am, or 0.11 and 0.18 Gy for ^{3}H. The 16S ribosomal RNA gene was chosen as a target one for sequence analysis aimed to test whether low dose radiation triggers any alterations in this universal throughout bacterial world and primarily important gene and can be a cause of toxicity at the genetic level. Nucleotide sequences of target DNA fragments were determined and compared in bacteria exposed to ^{241}Am or HTO and control bacterial suspension not exposed to radiation. All compared gene sequences were discovered to be identical which does not indicate any occurrences of mutation events in the analyzed gene under the applied conditions of low-dose alpha and beta radiation inducing changes in bacterial luminescence. Previous results on bacterial DNA exposed to low-dose gamma radiation (0.25 Gy) were analyzed and compared to those for alpha and beta irradiation. It is concluded that bioluminescence activation and/or inhibition under the applied conditions of low-dose alpha, beta and gamma radioactive exposure is not associated with DNA mutations in the gene sequences tested.

LCA and beyond - integrating sustainability and/or other dimensions to improve decision support (P)

WE219

Meet the Framework Regulation and Supply Chain secondary standards in wheat cultivation for sustainable pasta production. An example of broadleaf weed control: halauxifen-methyl (Arylex™ active)

C. Vaj, S. Cavanna, Dow AgroSciences Italia srl; A. Haley, Dow AgroSciences Ltd; W. Jones, DuPont

The need and the awareness of sustainable food production has increased in recent years, driven by the growing awareness of global population increase and its burden on the limited agricultural land available to sustain the required food production. This is being reflected in the choices made at all the steps of food production from field to table. The sustainable production of pasta starts by optimising agricultural practices, which includes the key component of Plant Protection Products (PPP) applied to wheat crops. The development of new Plant Protection Products in Europe is governed by the strictest regulatory framework in the world: Regulation (EC) 1107/2009 concerning the placing of plant production products on the EU market; Directive 2009/128/EC on Sustainable Use of pesticides and its national implementations (National Action Plans), and Regulation (EC) 396/2005 concerning the Maximum Residue Levels of plant protection products allowed in food. In addition, the Secondary standards coming from Food Processors and Retailers regarding chemical residues in food place increasing standards which have to be considered. The Plant Protection Industry is increasing its focus on sustainable food production not only for complying with the evolving Regulations, but also for helping the farmer to meet the Food Supply Chain needs. A good example of this new emphasis is brought by an innovative auxinic herbicide, halauxifen-methyl (Arylex™ active), for the control of broadleaf weeds in spring and winter soft and durum wheat. This highly efficacious herbicide requires low use rates and its inherent properties (rapid degradation in the soil and plant) mean a low environmental and human health impact. Utilising halauxifen-methyl according to its label offers wheat growers a key tool for optimizing production, while producing a commodity with no detectable residues in the grain, in the processed product (flour, bran, bread, wheat germ, malt), and in pasta. Results will be presented and discussed. Therefore, the properties of halauxifen-methyl are fully aligned with increasingly strict environmental requirements from regulatory authorities and the Food Chain Secondary standards. ™ Trademark of Dow AgroSciences

WE220

Cradle to grave Life Cycle Assessment of Traditional and Vegetative roofs

J. Koura, University of Balamand / Chemical engineering department; R. Manneh, University of Balamand / Department of Chemical Engineering; R. Belarbi, University of La Rochelle / Laboratory of Engineering Science for Environment LaSIE; V. El Khoury, University of Balamand / Chemical engineering department; H. El Zakhem, University of Balamand / Department of Chemical Engineering; M. EL Bachawati, University of Balamand / Chemical engineering department

The aims of this study are to (1) assess the environmental performance of an extensive green roof (EGR) mock-up installed on the rooftop of the Chemical Engineering Department at the University of Balamand, in the region of El Kurah, North Lebanon (34º31’N, 35º50’E) from the raw material phase until the end-of-life phase through a cradle to grave Life Cycle Assessment (LCA) study; and (2) compare the environmental impacts of an EGR mock-up to a traditional gravel ballasted roof (TGBR) mock-up. In this research, the Life Cycle Inventory was modelled using the SimaPro 8.3.0 software and the Ecoinvent database, and the IMPACT 2002+ methodology was selected as the Life Cycle Impact Assessment method. Vegetative roofs seem like a possible solution for the environmental issues in Lebanon since this small Mediterranean country lacks a clear sustainability plan as well as an infrastructure update and only 13.4% of the total surface area (10,452km^2) are forested area. Vegetative roofs embellish the unused roof surface available in most urban areas, increase the roof lifetime, reduce the need for a heating/cooling system as a result the building energy consumption is decreased, etc. Vegetative roofs capture a fraction of the rainwater through their growing medium and the vegetation could remove airborne pollutants. The findings of this research seem to be very promising, the extensive green roof mock-up has the least environmental impacts for all impact categories except for the “land occupation” impact category due to the vegetation layer. Moreover, a real-time monitoring of temperature was done to assess the electricity consumption or both TGBR and EGR mock-ups within the use phase. Furthermore, the sensitivity and uncertainty analyses will be performed to check the robustness of the results.

WE221

Filling whole building life cycle assessment gaps for conceptual building design

V. Hasik, University of Pittsburgh; J. Chhabra, G. Warn, Pennsylvania State University; M. Bilec, University of Pittsburgh / Civil and Environmental Engineering

Resource consumption, harmful emissions, climate change, and hazard events have triggered increased interest in sustainable and resilient buildings over the past 20 years. The sustainability and resilience performance of buildings has been covered in numerous rating systems and building codes; however, these are typically prescriptive methods focused on setting minimum performance requirements rather than helping us understand and optimize buildings. Life cycle assessment (LCA) and performance-based methods (e.g., energy modeling, seismic loss assessment) can be more beneficial from this standpoint but can be complex and isolated from each other. Most building LCA studies to date have limited their scope to embodied and operational energy use, due to their overwhelming impacts in conventional buildings, while some aspects, such as repair from damages (e.g., seismic) and water use have been largely missing. This study uses LCA, energy modeling, water modeling, and seismic loss assessment simultaneously to obtain a comprehensive estimate of the costs and life cycle environmental impacts of various building design alternatives. Preliminary analysis of a hypothetical building shows that in some metrics, such as cost and eutrophication, repair and water phases, respectively, can contribute more than 10% over the lifetime of the building. One design parameter expected to influence all of the mentioned performance aspects is the building form-factor, and its effect on the results will be presented. Based on the typical approach to building LCA and the shift in the sources of impacts away from operational energy as buildings transition to net-zero energy, this study investigates the potential contribution from missing phases in building LCA. This research brings together knowledge from multiple disciplines and links them through life cycle thinking, investigating the effects of building design decisions in multiple metrics. This approach can be crucial in identifying optimal solutions early on during the conceptual building design phase.

WE222

Prospects for multidimensional assessment of sustainability in urban environments

F. García-Guaita, L. Lijó, Universidade de Santiago de Compostela / Chemical Engineering; A. Núñez, L. González Louro, FEGAMP - Santiago de Compostela; E. Andrade, Universidade de Santiago de Compostela; M. Moreira, G. Feijoo, University of Santiago de Compostela / Chemical Engineering; S. González-García, University of Santiago de Compostela CIF Q1518001A / Chemical Engineering

Urban systems can be considered as living organisms driven by materials and energy flows (urban metabolism). Once these flows are computed, the environmental profile of the city can be analysed. However, when only assessing the environmental aspects of cities, a limited view of their performance is possible, as they are complex systems in which social and economic aspects are at least as important as environmental ones. This fact raises a dilemma, since today’s developed society bases its social and economic well-being on the high consumption of resources and, therefore, on causing a large impact on the environment. The definition of sustainability includes three main pillars: society, economy and environment. However, considering the social and economic aspects of urban metabolism, two main gaps emerge. The first one is the lack of standardisation. Indicators are used to evaluate these aspects, but different specialized organisms propose different sets of indicators. The second gap is the difficulty of comparing indicators to each other in order to evaluate which city is the most sustainable in a sample. This study aims to fill both gaps by systematically assess the sustainability of several cities in Galicia (NW Spain). To select an appropriate set of indicators, including environmental, social and economic criteria, a Leopold matrix has been constructed considering: i) the data available for the system under study, ii) the frequency of occurence in the data sets of specialized agencies (United Nations, European Commission, OECD and The World Bank) and iii) the relevance for the case study. The selected indicators do not have a significant common unit of measurement; therefore, to obtain a common scale for comparisons, all indicators should be normalized. In this study, this have been done by considering unsustainable and sustainable values as reference (Phillis et al., 2017). Finally, a composite indicator, i. e. a sustainability index, is obtained for each city based on the three composite sub-indicators of the sustainability dimensions (environmental, social and economic criteria). *Acknowledgements* This work was financially supported by the Spanish Ministry of Economy and Competitiveness (project ref. CTQ2016-75136- P) and by Xunta de Galicia (project ref. ED431F 2016/001). Dr. S. González-Garcia would like to express her gratitude to the Spanish Ministry of Economy and Competitivity for financial support (Grant reference RYC-2014- 14984).

WE223

Life Cycle Analysis of remediation solutions in railways and surrounding areas

M. Riera, Leitat Technological Center

An important environmental problem is the pollution associated with trains on external or underground railways. Despite is an issue less studied, a real contamination by heavy metals and hydrocarbons exists in railways, affecting also the surrounding areas. Following the strategic line of Horizon 2020, **RECOVER** project aims to develop new technologies to reduce the environmental impact of transport systems. The project is executed by COMSA, CETIM and LEITAT, and is within the framework of the call "Challenges of Collaboration" in 2015, with partial funding from the Ministry of Economy and Competitiveness of Spain. The main aim of the project is to develop systems for the collection and elimination of pollutants (hydrocarbons and heavy metals) present in the ballast and on the ground adjacent to the railway tracks through the use of different technologies: Ballast modified by sol-gel coating based on silicon oxide to capture heavy metals and titanium oxide for the degradation of hydrocarbons. Phytoremediation processes (use of plants to decontaminate soils) and bioremediation (bioaugmentation of the microbial population of the soil) for the uptake of heavy metals and hydrocarbons in the soil adjacent to the roads. The solutions are first performed at the laboratory scale, and subsequently they are located in a real area to evaluate their effectiveness. The remediation procedures are assessed through a comprehensive **Life Cycle Assessment** (LCA) to identify the environmental benefits obtained with the introduction of the solutions in railways. The environmental analysis includes the life cycle stages of raw materials, application and end of life scenarios, including transport and other related aspects. The LCA is performed based on the methodology which is standardized by the ISO 14040 and ISO 14044. Calculations are done using the SIMAPRO software and taking as a basis the Ecoinvent3.1 database. Besides the direct environmental benefits of the pollutants reduction in the soil, the LCA allows the identification of other environmental aspects. The impact categories included in the analysis are: Climate Change, Ozone Depletion, Photochemical Ozone Formation, Acidification, Terrestrial and Freshwater Eutrophication, and Freshwater Ecotoxicity. The pollution due to rail transport is a problem identified in Member state of the European Union, the solutions proposed in RECOVER project could an important contribution to the current railway legislations.

WE224

Life Cycle Assessment of Asphalt Mixtures vs Road Pavements

D. Lo Presti, The University of Nottingham / Nottingham Transportation Engineering Centre NTEC; A. Jimenez del Barco Carrion, The University of Nottingham

Road infrastructures are one of the major assets all over the world. The appropriate construction and maintenance of roads promote economic growth and development of countries. Within the field of road infrastructures, road pavements construction and maintenance require particularly high energy and raw materials consumption and generate elevated GHG emissions. For this reason, great efforts are being made in order to move toward the implementation of sustainable technologies and operations, and decision-making tools are essential to help authorities to accept them. In this regard, LCA has become popular in pavement engineering but there is still a lack of detailing, consensus (especially in terms of system boundaries) and reliable data. Beyond ISO 14040:2006, ISO 14044:2006, ISO/TS 14067:2013, EN 15804:2012 and GHG Protocol 2013, there is no specific methodology for selecting the processes and activities that should be included in either road pavement or asphalt mixtures LCA. LCA of road pavements is a complex process which needs the collaboration of the different partners involved, such as road authorities or contractors, to provide information, and therefore there exists a common tendency to simplify the LCA of pavements and individually analyse their components - usually asphalt mixtures. This process is not always clarified and may lead to erroneous conclusions or incoherent procedures. In order to use LCA for decision-making in asset management, it must be more related to road pavements rather than asphalt mixtures. In other words, the LCA of the pavement components (e.g. asphalt mixes) should be a mere input and the overall methodology should focus mainly on dealing with data such as road geometry, maintenance strategies, traffic, pavement conditions and statistical parameters to account for data changing over the analysis period. The study presented here aims at highlighting the differences between the LCA of asphalt mixtures and road pavements. For this, the different phases of LCA are defined and analysed for the systems respectively. Finally, system boundaries are proposed for each system in order to move towards LCA of road pavements and standardised methodologies.

WE225

Sustainability assessment of an integrated innovative wastewater and greywater system for an optimal and safe closed water cycle in Mediterranean tourist facilities: DemEAUmed solution

A. Claret, C. Hidalgo, Leitat Technological Center / Sustainability Division; S. Vazquez, ACONDICIONAMIENTO TARRASENSE (LEITAT TECHNOLOGICAL CENTER) / Sustainability Division

The main objective of demEAUmed is to tackle water scarcity in the Mediterranean area, especially in places with high tourist activity. Also, the project wants to foster the incorporation of sustainability aspects in the tourism sector. To achieve both challenges, demEAUmed has demonstrated the integration of innovative wastewater/greywater treatment technologies to achieve an optimal and safe closed water cycle in Mediterranean tourist facilities. Water resources are limited and unequally distributed geographically and among the year seasons, with higher pressure during summer, in Mediterranean regions. For instance, water consumption per guest has been estimated at 222 L/day in hotels in Spain. So, it is of great importance to achieve a holistic water resource management. DemEAUmed affords the reuse of greywater and wastewater generated in touristic facilities with an integrated approach bringing environmental benefits such as water savings and water management carbon footprint reduction. 8 different innovative technologies with an advanced monitoring, control and decision support system have been integrated and implemented on the demonstration site: Samba Hotel-Lloret de Mar, Catalunya, Spain. These technologies have been assessed through a comprehensive LCA, assessing the impacts for each individual technology and for the demo-site integration (7 different configurations). Besides the LCA, a Life Cycle Costing (LCC) is being performed in order to analyse the economic costs. A social LCA (S-LCA) is also conducted in order to assess the social impacts generated by demEAUmed. Life cycle stages of construction and operation of technologies have been studied. Final results determined that the technologies and combined configurations have achieved important environmental impact savings thanks to the greywater/wastewater recovery and water reuse. As an example, for demEAUmed combined strategies, the carbon footprint is reduced up to 136% (greywater scenario) or up to 62% (wastewater scenario) thanks to water reuse. Focusing on the technologies, main environmental impacts are localized on the operation stage, due to electricity consumption. Concerning the LCC, the overall cost of treating one cubic meter ($1m^3$) of greywater or wastewater by the demEAUmed technologies along their life cycle are being determined. Finally, S-LCA has presented some indicators and the quantification of the socioeconomic impacts and benefits provided by demEAUmed solution.

WE226

Integrating Life Cycle Assessment and Risk Assessment to support decision making in the framework of Enhanced Landfill Mining

G. Sauve, KU Leuven Research & Development / Department of Materials Engineering; K. Van Acker, KU Leuven / Materials Engineering

Life Cycle Assessment (LCA) is a commonly used tool to assess the environmental impacts of product systems throughout their life cycle. However, when addressing waste management strategies, and in particular landfills, this tool could lead to some limitations. Landfills are in fact highly complex systems and their impacts are affected by several site- and time-dependent parameters. When assessing the potential for enhanced landfill mining (ELFM), the relative perspective of LCA both in time and space could lead to under- or over-estimations of landfill impacts and to inaccurate results. The aim of the research is to support decision making by defining a consistent “Do Nothing” scenario for landfills for the evaluation of the environmental potential of ELFM. This can be achieved by understanding the processes underlying the emissions of different compounds and estimating the long term emission potential of landfills. In fact, landfill leachate emissions are, on the long term, the major environmental concern and the risks to human health and the environment (HHE) are dependent on environmental and time-dependent conditions. In this context, metal speciation is considered an important aspect to include in the estimation of the emission potential, as the emissions and eco-toxicological impacts of metals depend on the variation of site-specific conditions in time. In light of these considerations, a more consistent evaluation of impacts on a global and local scale and considering a long-term perspective could be achieved by integrating LCA with risk assessment (RA), which is a more site-specific tool. In fact, the evaluation of the long-term emission potential of landfills would include the definition of a fate, transport and exposure model for leachate emissions that would then be integrated in the impact assessment stage of LCA. The eco-toxicological impacts would then be assessed by integrating the variation of pollutants’ concentrations in time and under specific conditions, and by including the variation of background concentrations in the receptor. Literature studies with focus on the integration of spatial differentiation (regionalization) and time-dependency (Dynamic LCA) will be used as references for the study.

WE227

Comparative environmental sustainability analysis of waste-to-energy techniques for municipal solid waste

A.I. Ramos, INEGI / INEGI; A.I. Rouboa, University of Pennsylvania / Mechanic Engineering and Applied Mechanics

Progressively advancing societies generate increasingly complex mixtures of residues which led waste thermal treatment methods to evolve greatly in the last decades [1]. Incineration is among the most waste-to-energy (WtE) techniques used for solid residues treatment [2], still gasification is gaining notoriety due to its proven benefits namely concerning efficiency indicators and environmental outputs [3, 4]. Three waste-to-energy techniques for the treatment of municipal solid wastes were assessed through a life cycle analysis (LCA) perspective and compared so as to evaluate their environmental impacts: incineration, regular gasification and two-stage plasma gasification. The functional unit chosen was one tonne of residues within similar boundary conditions for each technique. Incineration has shown a sustainable profile, 1tonne of debris saving up to 1.3 million kg of resources and materials, while environmental indicators such as global warming potential (GWP), eutrophication potential (EP) and terrestrial ecotoxicity potential (TETP) depicted enhanced results. Regular gasification uses higher temperatures, thermally decomposing waste and originating syngas which may be utilised as feedstock for further applications. Its environmental impacts revealed poor results for some of the evaluated impact categories, performing worse than incineration. In its turn, two-stage plasma gasification sets an upgrade to more common waste-to-energy techniques, adding an extra cleaning step to the raw syngas produced by gasification, where even higher temperatures are applied through the use of a plasma torch that literally “melts” the residues. This technique grants environmental benefits such as lower levels of pollutant emissions, less landfilling needs and also higher conversion efficiencies and syngas quality. All the environmental categories depict negative values, meaning environmental savings as well as each tonne of treated residues redeeming more than 500 tons of emissions. Two-stage plasma gasification proved to be the most effective method, within the assessed techniques. Keywords: waste-to-energy, sustainability, life cycle assessment, solid residues References\n 1. Chen H, Jiang W, Yang Y, Yang Y, Man Y. 2015. J Air Waste Manage 65:1161-1170. 2. Brunner PH, Rechberger H. 2015. Waste Manage 37:3-12. 3. Ramos A, Monteiro E, Silva V, Rouboa A. 2018. Renew Sust Energ Rev 81:380-398. 4. Lopes EJ, Okamura LA, Yamamoto CI. 2015. Braz J Chem Eng 32: 87-97. \n

WE228

Life Cycle Assessment of Pharmaceutical Waste Disposal in the UK

S. Mohamed Yunus, University of York / Environment; A. Boxall, University of York / Environment Department; E. Igos, Luxembourg Institute of Science and Technology / Environmental Research and Innovation

Unused or expired medicines from the hospital and household waste can ultimately end up in landfills or be released to the wastewater system. Therefore, there is the potential for active pharmaceutical ingredients (APIs), from a range of medicinal products, to be present in landfill leachate and sewage effluents. Unused medicines may also be returned to the pharmacist and then be incinerated as hazardous waste. In this project, a household survey was performed to understand the typical waste generation patterns for medicines and the most disposal routes for these substances in the UK. The results show that rubbish disposal (34%) is the common disposal methods by the UK residents with highest estimated emission of APIs to the environment being estimated for paracetamol within the range 7.63 mg/person/day (sewerage after sink and toilet disposal) to 76.52 mg/person/day (wastewater after excretion). Based on the survey data, a life cycle assessment study was performed to assess the broader environmental impacts of typical medicinal waste disposal management practices in the UK. The functional unit of this study was 1 tonne of pharmaceutical waste generated in the UK. Three scenarios and treatments explored: incineration after returning back to the pharmacy; landfill treatment after rubbish disposal; and wastewater treatment after toilet and sink disposal. For the life cycle inventory (LCI) phase, two types of databases were used in the study: primary data from the survey study and secondary data from the ecoinvent database. Most of the datasets used in this study are from the whole of Europe (RER) or the United Kingdom (GB). To carry out the life cycle impact assessment (LCIA) phase, the method that has been considered is ILCD 1.0.8 2016 midpoint with APIs. Meanwhile, USETox 2.0 was also used in this study to calculate characterisation factors for the APIs that were not commonly used in the previous study. Furthermore, the impact categories that were considered for this study are climate change, ecosystem quality, human health and resources. The study is still ongoing and the results will be presented at the event.

WE230

Streamlined life cycle assessment of emerging batteries in early design phases using CCaLC tool

C. Tomasini Montenegro, KIT, Karlsruhe Institute for Technology; M. Weil, Karlsruhe Institute of Technology KIT / Institute for Technology Assessment and Systems Analysis ITAS

Tomasini-Montenegro, C [a,c] , Weil, M [a, b, c] **a** HIU, Helmholtz-Institute Ulm, Helmholtztr 11, 89081 Ulm, Germany **b** ITAS, Institute for Technology Assessment and Systems Analysis, Karlsruhe, Germany **c** KIT, Karlsruhe Institute for Technology, P.O. Box 3640, 76021 Karlsruhe, Germany In our modern and globalized society, meeting energy needs in a sustainable way pose one the biggest challenges for the scientific, political and regulatory bodies around the world. Therefore, in the context of the United Nations Development Goals, affordable and clean energy access has been defined as a reachable goal for 2030. In addition to the social impacts associated with this action plan, both tackling climate change and defining regulatory and market frameworks are common elements to identify global solutions for a low carbon energy market. Although it is recognized that geopolitical factors will shape a tailored solution for each geographical region, a transformation of the energy system with a high share in renewable energy sources is necessary to reach a decarbonized energy supply. In particular, considering an energy system with a high share in solar and wind power, energy storages technologies are required to level fluctuating energy production and demand. However, even though when it is recognized that the energy storage technologies exhibit different maturity stages, information about their associated environmental impacts is required to evaluate the sustainability trade-offs inherent to a technology decision-making process. In order to avoid environmental burdens shifting, a life cycle approach is proposed to develop a model for the preliminary evaluation of emerging batteries or components of these batteries using CCaLC as an assessment tool. The outcome of this work is aimed at contributing to understanding the environmental impacts associated with batteries from a life cycle perspective, while evaluating the advantages and disadvantages of using CCaLC as an assessment tool.

WE232

Development of Environment Hotspots of Analysis and the consideration of availability to eco-labeling program

Y. KURAHARA, N. Itsubo, Tokyo City University

In 2014, the UNEP/ SETAC life cycle initiative published a guidebook regarding hotspots analysis which enables to extract important elements from the life cycle. They defined this method as “a methodological framework that allows for the rapid assimilation and analysis of a range of information sources, including life cycle based studies, market, and scientific research, expert opinion and stakeholder concerns. The outputs can be used to identify potential solutions and prioritize actions around the most significant economic, environmental, ethical and social sustainability impacts”. Therefore, the scope of hotspots analysis covers environmental and social aspects. The methodology of environmental hotspots analysis has been developed in Japan using latest Japanese inventory database IDEA2 and WIO (Waste Input Output table) and the environmental impact assessment method (LIME). About 100products and services are evaluated by using this calculation tool. However, the case number of studies using hotspots analysis tool are few, advantages and limitations are unknown. In this research, the usefulness of environmental hotspots analysis through a variety of case studies for Japanese products.

WE233

Environmental burden reduction in the FTA framework using network analysis

S. Tokito, Kyushu University

The CO_2 emissions embedded in international trade have rapidly increased in countries with lax environmental regulations with expansion of trade and the international fragmentation of productions (Peters *et al*., 2011). In addition, with the importance of the improvement of environmental efficiency at industry level of a specific country, it is important to corporate within well-specified industrial clusters through supply chain engagement over developing and developed countries (e.g., Kagawa *et al*., 2015). Moreover, with expansion of trade and the international fragmentation of productions, the promotion of free trade has been increasingly required. In this circumstance, “mega-regional” Free Trade Agreement have been a significant trade policy issues. For countries participating the FTA framework, it is important to promote economic growth and reduction in various environmental burden simultaneously through the cooperation between the participating countries. To decide the cooperation policy for reducing environmental burden, it needs to understand the structure of complicated supply chain network. However, to the best of our knowledge, previous studies did not analyze the supply chain network induced by the production and consumption of FTA member countries. This study used the network centrality analysis, especially, the applied structural path betweenness (Liang et al., 2016; Hanaka et al., 2017) to EORA database (Lenzen et al., 2012, 2013) and analyzed the critical sectors for the cooperation policy in the global supply chain network. From the centrality analysis, I identified the critical sectors and transmitters. In the case of TPP framework, the largest CO2 emitter are “JPN_Electricity, Gas and Water” and “CHN_Electricity, Gas and Water.” On the other hand, the largest CO2 transmitter are “RUS_Mining and Quarrying→JPN_Petroleum, Chemical and Non-Metallic Mineral Products” in the sector level and “CHN→JPN” in the country level. We can see the large CO2 emission from China and Russia which are not TPP member is induced by consumption of TPP members. In the cases of other FTA framework, similarly, mining and qualifying sectors in Russia and China tend to be critical sectors. Replacement of the mining goods suppliers is not easy. Therefore, FTA member countries should make a guideline for acquisition of greener materials and parts produced in the critical sectors.

WE234

Developing life cycle assessment to fight climate change

P. Goglio, Carnfield University / School of Water, Energy and Environment; A.G. Williams, N. Balta-Ozkan, N. Harris, Cranfield University / School of Water, Energy and Environment; P. Williamson, University of East Anglia / School of Environmental Sciences; A. Marvuglia, Luxembourg Institute of Science and Technology (LIST) / Environmental Research and Innovation (ERIN); P. Baustert, Luxembourg Institute of Science and Technology LIST / Resource Centre for Environmental Technologies (CRTE)

Climate change targets could only be achieved with the contribution of greenhouse gas removal technologies (GGRT). Several GGRT have been proposed: direct carbon capture and storage (CCS), the use of wet raw materials in cement production, enhance weathering, enhancing soil C; forest management; bioenergy with CCS. Life Cycle Assessment (LCA), has been widely adopted to assess GGRT. However, there is no consensus on the methodology to assess GGRT, causing poor understanding of their implications. Therefore a new methodological framework is necessary. This study i) presents some methodological approaches for LCA of GGR technologies and ii) discusses their strengths and weaknesses. The preliminary comparison and development of approaches was based on a bibliographic review combined with expert discussions. The approaches have been classified according to their completeness, uncertainty and complexity**.** Several approaches were discussed: combining LCA with agent based modelling; combining LCA with climate scenarios from integrated assessment model IAM (IAM) used to assess future trajectories for GHG and climate change; combining socio-political factors with consequential LCA or using agent-based modelling for the socio-political factors affecting the choice of results from the equilibrium models in consequential LCA. None of the approaches presented resulted fully comprehensive, with limited uncertainty and complexity. Both approaches based on agent based modelling require a large amount of data and expertise to be used, not often available to the LCA practictioner. The weighted consequential approach has a level of subjectivity higher than other approaches. Instead, the scenario based approach using IAM has a medium level of completeness, uncertainty and complexity. However, scenario based LCA should have assumptions fully in agreement with the IAM assumptions to be used and this constitutes a major limit. The choice among the approaches depends on the objectives of the LCA and should be as complete and comprehensive as possible when climate change claims are made. Future perspectives include a comparative testing of these approaches for selected GGRT and future research should develop and assess potential alternative approaches to those presented. Further research is necessary to develop the appropriate LCA methodology for GGRT.

WE235

HYBRID FULFILMENT-IMPORTANCE MATRIX FOR ASSESSING SOCIOECONOMIC IMPACT

J. Espí Gallart, Fundacio CTM Centre Tecnologic; J. Berzosa, L. Vendrell, Fundació CTM Centre; F. Clarens, Fundacio CTM Centre Tecnologic

More often, methodologies to assess socioeconomic impact are focused just on determining just a few indicators instead impacts, which don’t use to cover the whole spectrum of socioeconomic insights. In this sense, the hybrid fulfilment-importance matrix emerges with the aim to solve these limitations and to calculate the socioeconomic impacts of a new process, service or project. This methodology is not only based in technical performance of the studied system, but also in the holistic approach offered by the LCA, LCC and sLCA methodologies. This semi-quantitative system is based on the scores of the relationship between indicators and impacts. In this way, this methodology allows calculating in which degree the objectives have been achieved, and how the impacts and indicators affect the system. One of the main strong points is its integrated approach which allows to consider the impacts of the project during different dimensions of the project. The indicators, placed in rows, are organized in four categories: technical indicators, environmental indicators, economic indicators, and social indicators. For technical indicators, data coming from performance of the system is implemented. The indicators for the environmental category are taken from LCA studied impact categories, which are supposed to be relevant for the project. In the case of the economic category, indicators studied in the LCC study are considered. Regarding the social indicators, those listed come from sLCA study. The distribution of the columns shows two parts: the fulfilment part, and the relevance part. In the fulfilment part, three columns are deployed: Baseline status, expected results set with the goals of the new process, and current or final results. On the importance part, the added columns represent the impacts of the project, which entail technical, sustainability, economic and social insights. When the impacts are selected, its importance in reference to the studied indicators must be defined based on expert know how and opinion. This importance is set by applying a value between 0 and 3. The socio-economic scores are calculated combining the importance values with the fulfilment factors. As a case study, this methodology has been applied to LIFE RELEACH project, which is aimed at managing leachates coming from landfill by concentrating technologies. In this way, the methodology has allowed to determine which socio-economic impacts have the higher contribution.

WE236

SETAC Sustainability Interest Group

D.L. Carr, Texas Tech University / Biological Sciences

WE237

SETAC LCA Interest Group (Europe)

H. Stichnothe, Thünen Institute / Agricultural Technology

WE238

Life cycle assessment of a thermoplastic starch obtained from mango kernel

A. Cardoso, Embrapa - Brazilian Agricultural Research Corporation / Embrapa Agroenergy; P. Marques, F. Freire, University of Coimbra / ADAI-LAETA, Mechanical Engineering; P. Melo, Federal University of Ceará; M. Figueiredo, Brazilian Agricultural Research Corporation Embrapa / Embrapa Tropical Agroindustry

Agrifood industry generates large amounts of residues with potential to be used as feedstock for bio-based products. Mango fruit annual production in Brazil is expected to increase to 1.4 million tons by 2024, and processing residues can account for more than 40 %. The mango pulp is the main product, and mango kernel, a so-called residue, is disposed of at landfill, but containing starch, oil and phenolic substances. This study assesses the environmental life cycle impacts of thermoplastic starch produced from mango kernel (MK-TPS), and compare it with fossil-based low-density polyethylene (LDPE). The system boundaries of the MK-TPS start with transportation of mango kernel residue, followed by extraction of starch from mango kernel (together with oil and phenolic substances), and production of thermoplastic. The functional unit adopted was 1 kg of thermoplastic. A life cycle inventory for MK-TPS was implemented based on primary data gathered at a laboratory scale. Six environmental impact categories were assessed based on the ReCipe Life cycle impact assessment method**.** A sensitivity analysis to the allocation approach for the starch extraction process will be performed comparing mass allocation (56 % starch, 28 % phenolic compounds and 16% oil) with economic allocation (using a range of expected market prices). Impacts based on mass-allocation for MK-TPS show lower climate change, fossil depletion and ozone depletion, but higher impacts on terrestrial acidification, freshwater eutrophication and marine eutrophication, comparatively to LDPE. The most important contributor to impacts is starch extraction (due to hexane and methanol), except for marine eutrophication, for which the main contributor is glycerin used to produce the thermoplastic. The paper may contribute to the eco-design of a new bio-based product using an important residue from the mango agro-industry as feedstock, through a Life cycle assessment based on laboratory scale data. Future studies shall take into consideration critical aspects and improvement opportunities identified through the study on a larger scale extraction process.

Environmental monitoring of contaminants using terrestrial ecological biomonitors (P)

WE239

Geostatistically estimating spatial structures of heavy metals and nitrogen accumulation in mosses sampled between 1990 and 2015 throughout Germany

S. Nickel, W. Schröder, University of Vechta / 2

Mosses are used to spatially complement the collection of atmospheric deposition by technical samplers and to validate deposition modelling results. Since 1990, the European Moss Surveye have been providing data on element concentrations in moss every five years at up to 7300 sampling sites. In the moss specimens, heavy metals (since 1990), nitrogen (since 2005) and persistent organic pollutants (since 2010) were determined. Germany participated in all surveys with the exception of that in 2010. In this study, the spatial structures of element concentrations in moss collected between 1990 and 2015 in Germany were comparatively investigated by use of Moran´s I statistics and Variogram Analysis and mapped by use of Kriging interpolation. This is the precondition to spatially join the moss survey data with data collected at other locations within different environmental networks. The calculated maps reveal a clear and statistical significant decrease of concentrations of most heavy metals in moss but not for nitrogen. Due to decreasing element concentrations and the unchanged application of the element concentration classification for the mapping, the heavy metals maps for the survey 2015 do not any longer depict much spatial variation. Therefore, in an upcoming study, this analysis needs to be complemented for the heavy metals by mapping percentile statistics for the whole period 1990-2015 with maps depicting the spatial structure of survey-specific percentile statistics 1990, 1995, 2000, 2005, and 2015. **Keywords:** Atmospheric Deposition, European Moss Survey, Geostatistics, Kriging Interpolation, Mapping, Variogram Analysis. **References** [1] Nickel S, Schröder W (2017) Reorganisation of a long-term monitoring network using moss as biomonitor for atmospheric deposition in Germany. Ecological Indicators 76:194-206. [2] Schröder W, Nickel S, Völksen B, Dreyer A. (2017) Nutzung von Bioindikationsmethoden zur Bestimmung und Regionalisierung von Schadstoffeinträgen für eine Abschätzung des atmosphärischen Beitrags zu aktuellen Belastungen von Ökosystemen. 4. Zwber., F&E UFOPLAN 3715632120, i.A. UBA, Dessau. Text: 82 S, 4 Anh.: 212 S. **Keywords**: Bioaccumulation of atmospheric deposition, European moss survey, heavy metals, nitrogen. *Acknowledgement* - The authors thank the German Environment Agency for funding.

WE240

Semi-volatile organic contaminants (SVOCs) in pine needles from Iceland

M. Moeenfard, J.A. Silva, S. Ramos, LEPABE University of Porto; H. Indriðason, T. Eysteinsson, T. Jónsson, A. Sigurgeirsson, Icelandic Forest Research; N. Ratola, Faculty of Engineering - University of Porto / Laboratory for Process Engineering, Environment, Biotechnology and Energy

Iceland is famous for a great number of things, but vegetation (particularly forestry) is not one of them. However, trees do exist in this country and pine stands and reasonably widespread along the coastal areas of the island, allowing the biomonitoring of compounds such as the semi-volatile organic contaminants (SVOCs). The most common species are somewhat different from those used more frequently in these kind of studies, but offer nevertheless the same performance and possibilities. *Pinus contorta*, *Pinus mugo*, *Pinus sylvestris* and *Pinus cembra* needles were collected upon availability in 24 sampling sites that included remote and rural areas but also some urban settlements like Reykjavík or Selfoss. In seven of these sites it was possible to collect needles from more than one species, allowing a comparison between their respective uptake abilities for SVOCs. In this work the levels of polycyclic aromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs), polybrominated diphenyl ethers (PBDEs), organochlorine pesticides (OCPs) and musks were analysed and allowed an original description of the state-of-the-art of these compounds in this remote location. Acknowledgements: This work was the result of the project: (i) POCI-01-0145-FEDER-006939 (LEPABE – UID/EQU/00511/2013) funded by the European Regional

Development Fund (ERDF), through COMPETE2020 - Programa Operacional Competitividade e Internacionalização (POCI) and by national funds, through FCT - Fundaçao para a Ciencia e a Tecnologia; (ii) NORTE-01-0145-FEDER-000005-LEPABE-2-ECO-INNOVATION, supported by North Portugal Regional Operational Programme (NORTE 2020), under the Portugal 2020 Partnership Agreement, through the ERDF; (iii) Investigador FCT contract IF/01101/2014 (Nuno Ratola).

WE241

Study of global diffuse pollution levels in remote high mountain areas and their impact on the organisms from these ecosystems

R.M. Prats, Institute of Environmental Assessment and Water Research (IDAEA-CSIC) / Environmental Chemistry; B.L. van Drooge, IDAEA-CSIC / Department of Environmental Chemistry; P. Fernández, B. Piña, J. Grimalt, Institute of Environmental Assessment and Water Research IDAEA CSIC / Department of Environmental Chemistry

Global diffuse pollution results from the emission of multiple sources and long-range transport. Effects of this background contamination have been observed in the recent past in fish from remote high mountain lakes through mRNA measurements in which showed feminization effects and oxidative stress (S. Jarque *et al.* 2015). Although some of these effects were related to persistent organic pollutants, there is still a gap of knowledge on their toxicological mechanisms and possible influence of other chemical pollutants. Persistent organic pollutants are incorporated to remote high mountain areas through atmospheric transport and deposition, where they may be bioavailable for organisms in these ecosystems and, depending on the physical-chemical properties of the pollutants, they may bioaccumulate. A comprehensive study of the pollutants in the atmosphere, lake waters and fish is presented in the present work. The six remote high mountain lakes investigated were located in the National Park of Aigüestortes i Estany de Sant Maurici (Pyrenees). They encompassed an altitudinal gradient from 1600m to 2500m asl. Active and passive monitoring devices were used for air and water sampling in order to get insight on the presence and environmental fate of organic contaminants over an extended time period. Moreover, brown trout (*Salmo trutta*) specimens were captured in each lake for bio-monitoring. They were analyzed for contaminants in the muscle, hepatopancreas and gallbladder tissue. The transcriptomic and epigenetic analysis of mRNA was also performed to link the levels of pollutants found in these remote high mountains to the effects in these organisms. The observed concentrations will also be compared to past measurements in other high mountain environments for assessment of temporal trends of this background contamination. S. Jarque *et al.* (2015). Background fish feminization effects in European remote sites. *Sci. Rep.*, *5*, 11292.

WE242

Spatial distribution of mercury and trace metals in epiphytic lichens in Nova Scotia, Canada

S. Klapstein, Acadia University / Earth & Environmental Science; I. Carvahlo, Técnico Lisboa; R. Cameron, Nova Scotia Provincial Government / Department of Enviroment; A. Walker, Acadia University / Biology department; J. Murimboh, Acadia University / Chemistry; C.H. Saunders, Acadia University / Biology department; J. Canario, Instituto Superior Técnico / Centro de Química Estrutural; R. Keenan, Environment and Climate Change Canada; N.J. O'Driscoll, Acadia University / Department of Earth and Enviromental Science

Mercury is a persistent pollutant present in all ecosystems. The prevalence and speciation of mercury will determine its movement in the atmosphere and potential to bioaccumulate and biomagnify through food webs leading to mercury contamination in top predator organisms. Monitoring of mercury and other trace metals can be costly, whereas the use of naturally occurring epiphytic lichens can be an effective tool for these types of studies. Nova Scotia, Canada is a hotspot for mercury and other trace metal accumulation in ecosystems, partially attributed to long-range transport of anthropogenic air pollution. The region also contains a number of historic gold mining sites that are known to have persistent high levels of mercury and arsenic in sediment. The relative contribution of local and national sources of mercury to local air is unknown. This work aimed to address which elements can be effectively biomonitored through lichens. Trace metals in lichens other than mercury may also help elucidate the potential sources of these elements: whether from geological, re-emission, or long-range transport. Over 300 lichen (*Usnea* spp.) samples were collected across Nova Scotia and analyzed for total mercury (THg); a subset of these samples were analyzed for other trace metals, including arsenic, nickel, copper, cadmium, lead, and selenium (n=163). Average THg concentrations were 365 ± 391 ppb (n=340). Significant variation in mercury content was observed across sampling sites and GIS analysis was used to display and model these regional trends. While broad spatial resolution was the initial focus for these collections, a few target areas (biological mercury hotspot Kejimkujik National Park and historic gold mining areas) were also sampled in more intensively to confirm spatial patterns. Lichens were also collected from one old growth forest site weekly for a one year period to investigate if there were detectable seasonal patterns in the mercury accumulation on lichens. We show that the association between mercury and lichens is stable over a one year period with minimal variability due to abiotic climate factors (solar radiation and temperature). The use of lichens as biomonitors of air quality is inexpensive and effective.

WE243

Biological monitoring of environmental quality near a solid waste incinerator in central Lithuania

G. Sujetoviene, P. Smilgaitis, Vytautas Magnus University

Waste disposal has huge environmental impacts including toxins, leachate and greenhouse gases. Lichens *Evernia prunastri* (L.) Ach. and *Ramalina farinacea* (L.) Ach. were used for biomonitoring the effect of air pollution in an area of one of the largest solid waste landfill in central Lithuania. Lichen transplants were exposed for 3 months. Chlorophyll content increased in both transplanted lichens with increase in distance from the landfill. Chlorophyll content in lichens was significantly lower in the nearest study site in comparison with the control. Potential quantum yield expressed as F_v/F_m in thalli was lower under the influence of solid waste incinerator in comparison with the reference. Higher chlorophyll degradation was characteristic to the transplanted lichens under the influence of landfill. The conductivity of leachate and content of thiobarbituric acid reactive substances (TBARS) increased in lichen material transplanted at sites facing the landfill. The results showed that biological monitoring can be useful tool for indication of environmental quality.

WE244

Nothing is what it seems: Levels of PCDD/Fs in the surroundings of a hazardous waste incinerator

M. Marques, Rovira i Virgili University / Chemical Engineering; M. Mari, Universitat Rovira i Virgili / Chemical Engineering; M. Nadal, Universitat Rovira i Virgili / School of Medicine, IISPV; M. Schuhmacher, Rovira i Virgili University / Departament d Enginyeria Química; J.L. Domingo, Universitat Rovira i Virgili / Laboratory of Toxicology and Environmental Health

Soil and vegetation were used as environmental monitors to assess the occurrence of dibenzo-*p*-dioxins and polychlorinated dibenzofurans (PCDD/Fs) in the vicinity of a hazardous waste incinerator (HWI) located in Tarragona (Catalonia, Spain). Results belonging to 2015 and 2016 were compared to a previous study conducted in 1998, before the plant started operating, to evaluate the potential impact of the facility after several years of regular operation. The median concentrations of PCDD/Fs in soil samples collected around the HWI were 0.46 pg I-TEQ/g (dw) (range: 0.14 to 1.96 pg I-TEQ/g (dw)) and 0.44 pg I-TEQ/g (dw) (range: 0.13 to 1.34 pg I-TEQ/g (dw)) in 2015 and 2016, respectively. No statistical differences were found between 2015 and 2016 campaigns. Comparing the study of 1998 (median: 0.75 pg I-TEQ/g (dw)) with these carried out in 2015 and 2016, the concentration of PCDD/Fs statistically decreased by 41 and 55%, respectively. Median concentrations of PCDD/Fs in samples of vegetation collected in the vicinity of the incinerator were 0.23 pg I-TEQ/g (dw) (range: 0.11 to 0.68 pg I-TEQ/g(dw)) in 2015 and 0.17 pg I-TEQ/g(dw) (range: 0.09 to 0.36 pg I-TEQ/g(dw)) in 2016.The temporal trend of PCDD/Fs in vegetation was very similar to that of soil, with overall reductions of 4%, 30% and 27% over the period 1998-2015, 1998-2016, and 2015-2016, respectively, being statistically significant in the two latter periods. Although the concentrations of PCDD/Fs in both soil and vegetation samples collected in urban areas showed higher levels than those from rural areas, there was no direct relationship between the levels of PCDD/Fs and the direction or proximity to the plant. In addition, the comparison of PCDD/Fs profile of chimney emissions and the corresponding samples of soil and vegetation denotes noteworthy differences in the contribution of some congeners. Consequently, there is a low potential impact of the plant on the environment, regarding to the emission of PCDD/Fs. Finally, concentrations of PCDD/Fs in soils and vegetation here reported are similar and/or below those observed in the scientific literature for similar areas.

WE245

The use of land snail Cornu aspersum as sentinel organism to monitor air pollution

L. Sturba, M. Vannuccini, G. Liberatori, F. Nannoni, G. Protano, University of Siena / Department of Physical, Earth and Environmental Sciences; N. Fattorini, University of Siena / Department of Life Sciences; I. Corsi, University of Siena / Physical, Earth and Environmental Sciences

The use of bioindicator organisms for monitoring air pollution allow to assess real hazardous effects of airborne contamination over a geographical and temporal scale depending on selected species and scientific approach. The present study aimed to validate the use of the land snail *Cornu aspersum* as bioindicator of airborne pollutants effects by transplanting snails in plastic cages positioned in un urban area strongly impacted by several industrial activities nearby. Ten sites were selected based on winds directions and the distance from the main industrial area in order to assess pollutant distribution in terms of bioavailability and biological responses in a relatively short period of time. After four weeks trace metals levels in soft tissues of whole organism and several biochemical responses were investigated in different tissues and organs as: lysosomal membrane stability (LMS) and Micronuclei (MN) in hemocytes and antioxidant enzymes Catalase (CAT) and Glutatione-S-transferase (GST), lipid peroxidation (MDA) and total Metallothionein proteins content (MTs) in midgut gland. Results obtained by generalized linear mixed models (GLMMs) revealed significant correlations among trace metals levels and biological responses investigated and with the distance from the main industrial site. Based on such findings and previous evidences of the ability of this species to respond to vaporized metals as cadmium in laboratory controlled condition, the present study support the suitability of *C. aspersum* as bioindicator for heavy metals exposure in air pollution monitoring studies.

WE246

The relationship between lead exposure on dogs and their behavior around Pb mining area, Kabwe, Zambia

H. Toyomaki, Hokkaido University / Graduate School of Veterinary Medicine; J. Yabe, The University of Zambia / Veterinary Medicine, Paraclinical Studies; S.M. Nakayama, Hokkaido University / Graduate School of Veterinary Medicine; Y.B. Yohannes, Hokkaido Univesity / Laboratory of Txicology; K.M. Muzandu, University of Zambia; H. Mizukawa, Hokkaido University / Environmental Veterinary Sciences; Y. Ikenaka, Hokkaido University / Graduate School of Veterinary Medicine; H. Nakata, Hokkaido University; R. Dowling, J. Caravanos, Pure Earth; M. Ishizuka, Hokkaido University / Graduate School of Veterinary Medicine

Lead (Pb) toxicity on both of human and animals has been known and caused neurological symptoms and even death in the worst cases. Our previous study has revealed Pb exposure on domestic dogs around a Pb mining area, Kabwe, Zambia. There was a trend that Blood Lead Levels (BLLs) in dogs in sites near to the mining area were higher than those in sites far from the mining area. Moreover, the difference of BLLs in the same area among individuals was found. This difference may be attributed to some factors, and behavior of dogs could be one of the important factors. The present study was undertaken to determine a relationship between Pb exposure on domestic dogs and their behavior recorded using GPS machines around the mining area in Kabwe, Zambia. Blood samples of domestic dogs which were freely roaming in the area were collected twice before putting GPS and after a week. BLLs were analyzed by LeadCare II. GPS devices were set to log every 1 minute and 30 seconds and attached to dog collars. In total, 53 male and 48 female domestic dogs were sampled. The overall mean of BLLs before and after a week were 24.2 μg/dL and 24.8 μg/dL, respectively. There was no significant difference between BLLs before and after a week. GPS log data was averagely collected for 4.4 days and the mean of distances of dog movements per day was 17.6 km. There was no significant relationship between distances of dog movements per day and the gap of BLLs in dogs between before and after a week. The distance between the mining area and dogs' home was significantly negatively correlated with BLLs ($p < 0.05$). The previous study revealed the concentrations of Pb in soils were negatively correlated with distance from the mine. These trends suggest that the distance from the mine is a key factor of Pb exposure on dogs. In the present study, there was a significantly negative correlation between BLLs in dogs and the distance between the mining area and their home. In contrast to our hypothesis, there was no significant relationship between BLLs in dogs and their behavior. It suggested that mature dogs in the study area could be highly exposed to Pb regardless their behavior. An additional study focusing on the behavior of young dogs which are more vulnerable to Pb could get a different result of the relationship with their Pb exposure.

WE247

Monitoring and impact assessment of terrestrial ecosystem using Eisenia fetida affected by chemical incidents

K. Kim, H. Jeon, H. Kim, Y. Kim, Y. Choi, S. Lee, Kyungpook National University

Many chemicals can be accidently spilled in the environment and it is important to know their presumable toxicities on the living organisms to determine risk assessments. There are no information on the terrestrial organisms of six chemicals possibly spilled into the environment, containing sulfuric acid, methanol, methylethylketone, nitric acid, formic acid, and toluene. In this study, we conducted acute toxicities of these six chemicals on *Eisenia fetida* in an artificial soil according to the OECD guideline 207. We used *E. fetida* adults grown in our laboratory for 10 generations in soil consisting of pig manure composts fortified with steamed sweet potatoes at 25°C. The earthworms used in this study were sexually well developed with an average body weight of 100 to 200 mg. The artificial soils were composed of industrial sand (70%, 50 to 100 micron particle), kaolin (20%, pH 4.5 to 7.0), and peat (10%). After mixing the components, pH was set in the range of 6.0 to 6.5. At least five diluted serial solutions were used to determine LC_{50} values, whereas pure acetone was used in the control group. LC_{50} values of sulfuric acid, methanol, methylethylketone, nitric acid, formic acid, and toluene were 1.41, 5.71, 2.16, 1.76, 1.24, and 2.86 g/kg soil, respectively. These results are very different from the acute toxicities using filter papers, which toluene exhibited 26-fold lower acute toxicity than sulfuric acid, the strongest toxic chemical among the tested chemicals. Using the filter papers, methanol and methylethylketone did not possess a negative effects on the earthworm. With these results, earthworms may act differently to the chemical incidents in relation to their residential condition when they expose to the chemicals.

WE248

Biochemical and behavioural responses in two endogeic earthworm species exposed to parathion

F. JOUNI, UAPV/IMBE; J. Sanchez-Hernandez, University of Castilla La Mancha; C. Mazzia, University of Avignon / Biologie; M. Jobin, University of Avignon; Y. Capowiez, INRA Avignon; M. Rault, University of Avignon

The earthworm species *Eisenia fetida* is a common organism in the soil toxicity testing framework, however, recent studies have point out endogeic species are more sensitive to pesticide than *E. fetida*. Moreover, interspecific differences in the response of this ecological group of earthworms to agrochemicals should be investigated for a better understanding of pesticide impact at population level. Herein, two endogeic and abundant species in the agroecosystem (*Allolobophora chlorotica* and *Aporrectodea caliginosa*) were incubated in Oleabladan® (ethyl parathion)-contaminated soils. Behavioural (burrowing, casting and feeding, this latter assessed through earthworm mass changes) and biochemical (acetylcholinesterase [AChE] and carboxylesterase [CbE] activities) were measured after 7 days of pesticide exposure. Our results clearly showed species-specific differences in behavioural and biochemical biomarkers, indicating *A. caliginosa* the most sensitive species to this pesticide under the exposure conditions of our study. Although CbE activity was determined in an attempt to account for these interspecific differences because the implication of this esterase activity in organophosphate detoxification, we found that CbE activity of both species had the OP sensibility. However, an *in vitro* inhibition trial with ethyl paraoxon evidenced a higher sensitivity of *A. caliginosa* AChE activity compared with that of *A. chlorotica*, which suggested that this toxicological endpoint may contribute to the interspecific differences of behavioural responses such as cast production rate. Our findings suggest the use of more than one endogeic earthworm species to assess toxicity from organophosphate insecticides, overall when these earthworms have a beneficial impact on soil fertility.

WE249

Cr transport in sweet peppers plants cultivated with vermicomposted tannery wastes

M. Rezende, Universidade de Sao Paulo / Chemistry; R.R. Rachide Nunes, Federal Rural University of Pernambuco / Chmistry; T. Oliveira, University of São Paulo / Chemistry; R.M. Bontempi, USP - Universidade de São Paulo / IQSC Instituto de Química de São Carlos

Untreated waste water and solid waste generated by the tanning industry can cause serious environment damage, particularly to surface and groundwater. In order to avoid pollution, specific chemical, physical and biological treatments have been adopted to reduce waste toxicity, caused mainly by the presence of the chromium. Chromium exists in oxidation states of Cr (III) and Cr (VI). As it is well known, the trivalent oxidation state is the most stable form of chromium and it is essential to plants in trace concentrations. In other hand, the hexavalent is toxic and carcinogenic to mammals, even in small concentrations. Thus, the aim of this work was to investigate the Cr transport in sweet peppers cultivated with vermicomposted tannery wastes. In order to investigate the Cr transport from the vermicomposts and its possible transportation through the plant, the content of Cr (III) and Cr (VI) were evaluated in all the binary soil-plant: soil (at begging and post-harvest), roots, stalk, leaves, and mature fruits. Cr (III) and Cr (VI) were determined through graphite furnace atomic absorption spectroscopy (GF AAS). Values of Cr (VI) were below the detectable level (LOQ) in all the analysis. In general, all treatments showed a decrease on their Cr (III) content during the sweet pepper cultivation. The concentration of Cr (III) varied in leaves < stalks < roots < fruits. Values of chromium were in accordance with the maximum permitted in the Brazilian legislation for food security; Cr (VI) was not reported in any sample. A significant quantity of Cr (III) decreased between the concentration at the beginning and at final experiments (post-harvest). Assessing the dynamics of the Cr (III), root and fruits showed a higher concentration, followed by the stalks and the leaves. Differences in the chromium contents were not observed between samples and treatments which received vermicomposted tannery wastes with others, without addition of chromium residues. Keywords: vermicomposting; tannery wastes; chromium; sweet pepper

WE250

Insecticide resistance in the natural enemy F. auricularia: detoxification pathways and sensitivity of acetylcholinesterase to organophosphate insecticide.

A. Le Navenant, UAPV/IMBE/INRA; M. Siegwart, INRA Avignon / Unité PSH, Equipe Ecologie de la Production Intégrée, Site Agroparc; Y. Capowiez, INRA Avignon; M. Rault, UAPV/IMBE / IMBE UAPV UMR 7263, Pôle Agrosciences

Apple orchards are highly treated crops, in which organophosphorus (OP), neonicotinoid and synthetic pyrethroid compounds were heavily sprayed insecticides. These compounds are toxic to non-target arthropods and increase the risk of resistance making apple orchards an interesting case to study the deleterious effects of insecticides on non-target species. In the European context of reduced pesticide use, the development of biocontrol agents has to be promoted. This work focuses on the assessment of the resistance/tolerance to OP insecticide of the earwig *Forficula auricularia,* an effective generalist predator. The threshold activities of enzymes that belong to detoxification pathways involved in insecticide resistance were estimated depending on the origin of earwigs. Then, variations in those activities were assessed under environmental conditions prior and after exposure to normal application rate of chlorpyrifos. Adult earwigs were sampled in apple orchards conducted under different management strategies: conventional, Integrated Pest Management (IPM, reduced pesticide use thanks to mating

disruption practice), and organic ones. Two frequently involved in pesticides resistance enzyme families: Glutathion-S-transferases (GST) and Carboxylesterases (CbEs) were studied, by measuring their activities on earwig extracts. Acetylcholinesterase (AchE) activity, the molecular target of OP insecticides, was monitored as toxicological endpoint. We observed that the mortality rate of adult earwigs exposed to the authorized dose of chlorpyrifos depends on their origin, with lower mortality in individuals sampled in conventional orchards. AChE activity appears to be highly inhibited in earwigs from organic or IPM compared to conventional orchards. Moreover AChE inhibition increased when earwigs were exposed to both chlorpyrifos combined to a specific inhibitor of CbEs. Moreover, we observed that basal-activities of CbEs and GST of unexposed individuals are higher in conventional orchards compared to IPM and organic ones. All these observations support the hypothesis of a molecular target modification in AChE conducing to a decrease of affinity with the insecticide, and highlight the role of CbEs ensuring effective protection of AChE. Our findings suggest the acquisition of resistance to chlorpyrifos in earwigs caught in conventional orchards and point out the necessity to understand these mechanisms in order to evaluate their relevance as biocontrol agents.

WE251

Bioaccumulation of persistent halogenated organic pollutants in insects: Common alterations to the pollutant pattern for different insects during metamorphosis

l. yu, Guangzhou Institute of Geochemistry / State Key Laboratory of Organic Geochemistry and Guangdong Key Laboratory of Environmental Resources Utilization and Protection; X. Luo, Guangzhou Institute of Geochemistry / State Key Lab. Organic Geochem; L. Tao, Guangzhou institute of Geochemistry Chinese Academy of Sciences; Y. Zeng, B. Mai, Guangzhou Institute of Geochemistry

Ubiquitous use of halogenated organic pollutants (HOPs), such as polychlorinated biphenyls (PCBs), polybrominated diphenyl ethers (PBDEs), and dichlorodiphenyltrichloroethanes (DDTs), can accumulate in organisms and become magnified along the food chain. Insects play an important role in the transformation of pollutants from abiotic to biotic media. However, few studies have been discussed the accumulation and fate of contaminants in insects. Furthermore, metamorphosis effects on the stable isotope signatures and enrichment characteristic of contaminants in insects, but the bioaccumulation pattern and mechanism during metamorphosis is not well understood. Therefore, we detected the concentrations of halogenated organic pollutants in four taxonomic insects (dragonfly, butterfly and moth, grasshopper, and litchi stinkbug), including three kinds of metamorphosis type, collected from an e-waste pollution region in South China. Stable isotopic analysis showed grasshopper have the highest $\delta^{13}C$ value, indicating a C_4-plant-based food source. In contrast, the butterfly, moth, and litchi stinkbug all represent a C_3-based diet preference characterized by lower $\delta^{13}C$ values. Moreover, enrichment of the heavy N isotope during metamorphosis is observed in the dragonfly and litchi stinkbug, but the other species (grasshopper, butterfly and moth) did not show significant increases in the values of $\delta^{15}N$ from larvae to adults. Principal component analysis (PCA) was conducted using the fraction composition of HOPs were performed to evaluate the species-specific bioaccumulation. Different species of insects exhibited different contaminant patterns, which could be attributed to their habitats and feeding strategies. For example PBDEs were predominant in the dragonfly collected from the pond, which has been seriously contaminated by electronic waste; however, DDTs significantly contributed to the total HOPs in the butterfly and moth, and in the litchi stinkbug, and that their host plants also have a high DDTs concentration. In addition, common multi-linear correlations between ln adult/larva and log K_{OW} of the compound was observed for the four taxonomic insects. The ratio of larva to adult decreased with increasing values of log K_{OW} (log K_{OW} < 6-6.5), then increased (6 < log K_{OW} < 8) and decreased again (log K_{OW} > 8). The results of this study demonstrated that a common mechanism is responsible for the fate of HOPs during metamorphosis in those insects.

WE252

Glyphosate: toxic or not toxic, this is the question.

M. Verderame, R. Scudiero, University Federico II / Department of Biology

In the recent years the potential toxicity of Glyphosate-based herbicides (GBH), a broad spectrum herbicide widely used in agricultural, industrial and urban areas, is a great matter of debate. Although classified by the EPA as "non-toxic and not an irritant" and by the EFSA as "no carcinogenic to humans", converging evidence suggests that GBHs, such as Roundup (Monsanto), pose serious health risk on non-target wildlife. Many studies demonstrate that GBHs threaten the reproduction interfering with the activity of aromatase, an important enzyme involved in the production of testosterone. Moreover, in vertebrates such fish and mammals GBHs cause the raise of oxidative stress markers and alterations in liver and kidney. Hence, the question about the real glyphosate (Gly) toxicity is still open. To concur to an answer to this question, we decided to investigate the effects of Gly exposure on the Italian wall lizard *Podarcis sicula*, a suitable bioindicator of terrestrial environmental pollution. Adult *P. sicula* specimens were divided in 3 groups (n=6): group 1 and 2 were exposed to pure Gly 0.1 and 1 μg/L, respectively, via gavage every other day for 3 weeks; group 3 received by gavage the same dose of tap water (100μl). The results demonstrate that both Gly doses are toxic for the liver that shows an increase of melanocytes degranulation and the appearance of nodular/cystic formations mainly consisting of collagen fibers, typical of hepatic fibrosis. The liver of Gly-treated males also displays the biosynthetic alterations typical of an estrogenic contamination: hepatocytes, in fact, contain transcripts for both vitellogenin and estrogen receptors. At reproductive level, male gonad is affected by the treatment. Spermatogenesis is slightly slower, at low dose of Gly scattered spermatocytes II fuse to form rosette-shaped arrangement, at high dose the amount of rosettes increases; spermatids are damaged and cells in degeneration are evident in the lumen of the tubules. Alterations in the expression of estrogen and androgen receptors and aromatase are also detected. Interestingly, in females, the ovary is not affected by Gly exposure, no matter the dose. Our results suggest that Gly exposure in a terrestrial vertebrate commonly inhabiting the fields potentially exposed to GBHs causes tissue toxicity, with possible serious health implications for wild and breeding animals as well human populations.

WE253

Concentration of perfluoroalkyl substances decreases according to the laying order in the yolk of yellow-legged gull eggs

C.D. Possenti, Università degli Studi di Milano; M. Mazzoni, University of Insubria, DISTA / Water Research Institute; B. De Felice, Università degli Studi di Milano; F. Cappelli, Water Research Institute - Italian National Research Council IRSA-CNR; S. Polesello, Water Research Institute- CNR / Water Research Institute; N. Saino, University of Milano; M. Parolini, University of Milan / Department of Environmental Science and Policy; S. Valsecchi, Water Research Institute - Italian National Research Council IRSA-CNR

Perfluoroalkyl substances (PFAS) are chemicals used as surface-active agents in diverse industrial applications. Because of their incessant disposal and release to the environment, these molecules caused the contamination of both fresh and seawaters, entailing their accumulation in the biota. Seabirds are highly exposed to environmental contamination because of their ecological habits, high trophic position in the marine food webs and relative long life-span. Contaminants accumulated in birds can be transferred to the offspring via their eggs, which are considered as good bioindicators of environmental pollution for a plethora of contaminants. However, the information concerning the maternal transfer of PFAS in bird eggs and the variation of their concentration according to the laying order is still inadequate. Thus, the aim of the present study was to determine the levels of PFAS in three-egg clutches of the yellow-legged gull (*Larus michahellis*) breeding in a colony located at the Comacchio lagoon (Northeastern Italy) and their variation according to the position in the laying sequence. Eleven perfluoroalkyl acids (PFAA) were analyzed in the yolk of eggs sampled at the time of deposition from 15 three-egg clutches. Independently of the laying order, perfluorooctane sulfonate (PFOS) was the main compound detected in the egg yolk, followed by perfluorooctanoic acid (PFOA) and perfluorododecanoic acid (PFDoDa). Overall, the ΣPFAA decreased according to the position in the laying sequence, with first- and second-laid eggs showing higher concentrations compared to last-laid eggs. A similar decreasing trend was also noticed for single compounds, namely PFOS, perfluorononanoic acid (PFNA), perfluorodecanoic acid (PFDA), perfluoroundecanoic acid (PFUnA) and PFDoDa, with concentrations measured in the last-laid eggs that were significantly lower compared to those from the first- and second-laid eggs.

WE254

First assessment of metal concentration in the crab Goniopsis cruentata (Latreille, 1803) (Decapoda, Grapsidae) from two brazilian mangroves areas with different levels of contamination

M. Vedolin, University of São Paulo USP; T.H. Trevizani, Universidade de Sao Paulo / Instituto Oceanográfico; M. Petti, University of São Paulo USP; R.C. Figueira, University of São Paulo USP / Institute of Oceanography

The crab *Goniopsis cruentata* is a common semi-terrestrial species in brazilian mangroves. Its geographical range includes the western Atlantic Ocean from Bermuda to Brazil, and the eastern Atlantic Ocean from Senegal to Angola. The species is an importante fishery resource for traditional communities in the some regions of Brazilian coast. These ecosystems are located in regions of intense anthropic activity and have been proved to accumulate heavy metals. The use of crustacean as bioindicators of metal bioavailability in coastal environment allows to outline comparisons over space and time and provides significant ecotoxicological integrated measures of the selected metals within the studied system. This study aimed to assess the levels of metals (As, Cd, Cu, Cr, Ni, Pb e Zn) in different tissues (muscle, hepatopancreas and gills) of *G. cruentata* and compare populations from contaminated and noncontaminated areas. Samples were collected in two mangrove areas of São Paulo State with different levels of contamination during a period of one year, to assess the bioaccumulation of metals associated with seasonality. A two-way ANOVA was carried to analyze interactions between season and sites. The results showed high concentrations of metals in the gills, which was considered a strong reflection of high exposure of *G. cruentata* to these contaminants. In general, the hierarchical pattern of metals concentration in organs was represented by gills> hepatopancreas≥ muscle, except for Zn. There were significant differences in metal levels between seasons and sites ($p < 0,05$). The highest concentrations were observed in the summer, which corresponds to the period of greatest metabolic activity of the organisms. Surprisingly, organisms from unpolluted regions, accumulated more metals than from polluted areas. Thus, we concluded that there are external factors (grain size, pH, salinity) that reduce the mobilization of these chemicals to the tissues and, consequently, their bioavailability to the local biota. Therefore, studies of metal concentrations in mangrove areas are relevant and useful for monitoring the health of environment, maintenance of biodiversity, and for assuring the quality of life, mainly for human when consumed.

WE255

Maternal Transfer of persistent halogenated organic pollutants in Watersnakes (Enhydris chinensis)

X. Luo, Guangzhou Institute of Geochemistry / State Key Lab. Organic Geochem; l. yu, Guangzhou Institute of Geochemistry, Chinese Academy of Sciences / State Key Laboratory of Organic Geochemistry; B. Mai, Guangzhou Institute of Geochemistry

Halogenated organic pollutants (HOPs) such as polychlorinated biphenyls (PCBs), polybrominated diphenyl ethers (PBDEs) and dichlorodiphenyltrichloroethanes (DDTs) are ubiquitous contaminants in the environment. Maternal transfer of HOPs to the offspring has been observed in oviparous species, i.e. fish, bird and frog. Few studies are focus on viviparous species, but ovoviviparous species have not yet been studied. It is known that watersnake (*Enhydris chinensis*) was ovoviviparous species. Their fertilized eggs develop into new individuals in the maternal body before producing offspring. The source of contaminants in watersnake eggs mainly derived from maternal tissues. In order to fully elucidate the deposition of contaminants in eggs, Firstly, the lipid-normalized concentration ratios of egg to muscle (EMR_L) were usually used to assess maternal transfer efficiency of contaminants in oviparous organisms. Secondly, due to relatively high lipid and weight of egg in watersnake, the ratios of contaminant burden in egg over the sum in muscle and egg (EMER) was used to evaluate the tissue distribution of contaminants in watersnake eggs. The values of EMR_L and EMER were respectively 2.93 and 95% for PCBs and 0.35 and 68% for PBDEs. Meanwhile, DDTs, PCBs, PBEB, HBB, PBB 153 and lower-brominated BDE congeners showed the ratios of EMR_L and EMER higher than 1 and 88% (the lipid percentage of egg to egg plus musle), respectively. The results indicated that these chemicals were readily transferred from muscle to egg or preferential accumulation in egg compared with muscle. Other chemicals, such as higher-brominated BDE congeners, DP, PBB209, and DBDPE, showed ratios of EMR_L and EMER lower than 1 and 88%, respectively, indicating less readily maternal transferred or a preference for muscle. A multi-linear relationship exists between EMER and log K_{OW} of the chemicals for the watersnake. For compounds with high hydrophobicity (log $K_{OW} > 8$), a negative relationship between EMER and log K_{OW} is observed ($p < 0.01$). While for compounds with log $K_{OW} < 8$, the values of EMER were no significant variety ($p = 0.19$), all greater than 90%. Maternal transfer potential and the deposition of contaminants in watersnake eggs are obviously different with other species in previous study, which implied potentially high inter-species differences in the maternal transfer mechanism.

WE256

Development of a Multi-compound Multi-matrix Method for Analysis of Halogenated Flame Retardants Comprising a Multi-step Cleanup and Use of GC-API-MS/MS and GC-EI-MS

F. Neugebauer, Eurofins GfA Lab Service GmbH / R&D; A. Dreyer, Eurofins GfA GmbH; N. Lohmann, Eurofins GfA Lab Service GmbH; J. Koschorreck, Umweltbundesamt

The ultra-trace analysis of halogenated flame retardants (HFR) leads more and more to the question of analyzing substances of actual or future relevance such as e.g. Dechlorane Plus and other dechloranes or novel brominated flame retardents together with legacy compounds as PBDEs. In order to address this issue, comprehensive analytical methods covering at the same time compound groups of different chemo-physical properties are more and more required, especially for monitoring purposes like analyses within environmental specimen banks. The presented method is validated for a broad range of different environmental matrices (spruce shoots as representatives for plant materials, bream fillet as representative for animal tissue, herring gull eggs as representatives for bird eggs and riverine suspended particulate matter as representatives for organic matter rich in solids) and presently capable of analysing 21 alternative HFRs and 24 PBDEs. The analytes cover different chemical substance groups from Dechloran Plus and other dechloranes to brominated benzenes and alkyl benzenes, ethers and esters (TBA, ATE, BATE, PBT, PBEB, HBBz, DPTE, BEHTBP, EHTeBB, BTBPE, Dec602, Dec603, Dec604, DPMA, Cl10-antiDP, Cl11-antiDP, syn-DP, anti-DP, DBDPE). In this way, it gives an analytical basis for further extension towards other compounds. We will show details of different analytical aspects of the method, especially regarding different column chromatographic clean-up steps and use of modern analytical equipment as e.g. a GC-API-MS/MS-system, pointing out possibilities and limitations of such a broad scope of analytes.

Product benefits and positive outcomes: valuation and beyond (P)

WE257

A method to calculate carbon handprint

T.K. Pajula, S. Vatanen, VTT Technical Research Centre of Finland Ltd; K. Grönman, R. Soukka, Lappeenranta University of Technology

Environmental impacts are typically assessed by measuring and modelling the negative effects that products, services and companies cause to the environment. In practise this means evaluating the used resources and energy and the emissions caused. However, many companies do good causing also positive environmental impacts. The strong need for communicating the positive environmental impacts has been identified e.g. by Pihkola et al. (2010). Only, we lack systematic methods to quantify and communicate these impacts that are also called handprints. This presentation proposes a concept to assess and communicate the carbon handprint of a product. The method is in line with life cycle assessment and footprint methods and is built on the principle that reducing one's own footprint is not a handprint. Instead, the handprint comes through improvements caused in the performance of another actor. The most fundamental parts of defining the carbon handprint are to recognize the mechanisms of forming the handprint and to determine the baseline. The carbon handprint can be created via more efficient material or energy use, by replacing or avoiding unwanted materials, waste reduction or extended service life and reuse. Also carbon capture and storage is a way to contribute to carbon handprint. The paper demonstrates through case studies situations where different approach for the determination of the handprint is required. The quantification of the carbon handprint requires several carbon footprints calculated in order to find out if the new solution or product actually reduces the carbon footprint of another actor and how much. The footprints are needed from the baseline solution, the target actor using the baseline solution, the new solution and the target actor using the new solution.

WE258

Associating regionalised Life Cycle Assessment (LCA) and economic values of ecosystem goods and services: Impacts of upstream natural land transformations on ecosystem quality

A. Ajayebi, University of Exeter / Renewable Energy

Setting up operational and spatially-explicit sustainability assessment models with practical levels of data requirement is becoming more essential as the trend of globalised economy is surging and accounting for impacts of human activities is becoming more complicated. Here we developed a model based on regionalisation of Life Cycle Assessment (LCA) that is capable of employing a holistic perspective while taking into account natural land transformations that are related to the life cycle processes. Furthermore, our model can interpret the impacts of land transformations on the ecosystem quality. Economic values of Ecosystem Services (ES) are used as an indicator and the difference between the value of land before and after transformations is representing the damages to the ecosystem quality. We performed a case study for the deployment of a 10 MW photovoltaic solar farm in the UK. The results demonstrated that the upstream life cycle processes transform 6354 m^2 of natural land into artificial land covers. We also estimated that these transformations reduce the ecosystem service value of the transformed natural land in its life cycle upstream processes from \$2364/year to \$1587/year, resulting in a decrease of \$765/year. The majority of ecosystem service value decreases are in China, where the manufacturing processes take place. The rest of Asia, Australia, Russia and Africa also suffer from noticeable ecosystem service value decreases because of upstream production industries. Europe, where the solar farm is deployed, only suffers modest ecosystem service value decreases due to upstream processes in comparison to the whole life cycle impacts. This implies that despite the apparent advantages of 'clean' renewable electricity production at the deployment location, the majority of ecosystem service damages might be relocated to other parts of the world.

WE259

Recent advances in natural capital accounting

S. Deacon, Ramboll Environment & Health Limited; A.E. Bartram, Ramboll Environ / Product Safety, Ecology and Sediment Management; L. Alvarez, Ramboll Environ

At the recent World Forum on Natural Capital (27-28 November 2017) a wide range of corporates, researchers, investors and policy makers came together to discuss progress on accounting for Earth's natural capital - the challenges, the innovations and the actions still needed. This poster will bring some personal reflections of the conference, including key findings from related recent literature, and elaborate on how scientists at SETAC Europe might engage with developments in natural capital learning. Recent publications, such as "Can we stop depleting natural capital?" (Cohen et al, 2017) have highlighted global financial prosperity yet scientific research shows that some natural capital is in a poor state, and declining further. The report finds political and economic systems are unprepared for responding to the risk of natural capital degradation for three reasons: (i) natural capital is not being accurately measured or valued in the context of ecological tipping points; (ii) aggregate economic models are ill-equipped for seeing the dependencies between 'capitals' as most cost-benefit analyses used in everyday decisions assume that natural capital can be easily substituted by manmade capital, when in fact it cannot; and (iii) we lack appropriate political and economic institutions to manage natural capital effectively. Two opportunities emerge including all natural capital could support greater prosperity if it were more appropriately valued and hence more efficiently used. The second is governance regimes based on scientifically informed political decisions should protect natural capital. Governance of natural capital stocks should be informed by biophysical limits, potential irreversibility, thresholds and risks to essential function. At the global level, the UN Sustainable Development Goals apply to all countries and provide a foundation for such a governance framework. We are all consumers and beneficiaries of natural capital, but it seems clear that SETAC members can also inform the debate. As environmental scientists we can contribute to developing an understanding of criticality – the tipping points for ecosystems, in particular where these relate to chemical pressures. There are opportunities to work with economists to ensure natural capital accounting and valuation is accurate and measurable, preferably quantitative. One of the strongest themes was that we all need to improve our story telling across multiple disciplines and institutions.

WE260

A Life Cycle Costing and Analysis of a Hybrid-Electric Engine

G. Bailey, KU Leuven / Material Sciences; W. Dewulf, KU Leuven; K. Van Acker, KU Leuven / Materials Engineering

The sustainability of hybrid-electric vehicles (HEVs) has been called into question (Bailey et al., 2017, Hickman, 2012, Publishers, 2008, Biello, 2016). With the recent onslaught of HEVs to the motor vehicle market, there is a need for the internalization of the costs—both environmental and economic—associated with one of its most valuable parts, the HEV motor. For this purpose, a screening Life Cycle Assessment (LCA) and Life Cycle Costing (LCC) study has been carried out on the entire value chain of a dismantled HEV motor. The results of this study are analyzed quantitatively. The aim is to identify bottlenecks for such a quantitative analysis as well as to identify the hotspots from both an environmental and economic point of view. Furthermore, the results will serve as one of the important inputs in laying the foundation for the development of the quantitative sustainability assessments for the HEV materials market. This study computes total life cycle costs of a HEV motor. The analysis considered capital and operating costs in order to define the total vehicle cost of ownership over 10, 15, and 20 year life expectancies. The development of an integrated life cycle costing and life cycle analysis will allow any individual to evaluate properly tradeoffs of a hybrid-electric vehicle. References\nBAILEY, G., MANCHERI, N. & VAN ACKER, K. 2017. Sustainability of Permanent Rare Earth Magnet Motors in (H)EV Industry. Journal of Sustainable Metallurgy, 3, 611-626.\nBIELLO, D. 2016. Electric Cars Are Not Necessarily Clean. Scientific American. Scientific American, a division of Nature America, Inc.\nHICKMAN, L. 2012. Are electric cars bad for the environment. The Guardian. \nPUBLISHERS, I. 2008. Hybrid Electric Vehicles Not As Green As They Are Painted, Analysts Contend [Online]. Inderscience Publishers\nAvailable: www.sciencedaily.com/releases/2008/02/080207094314.htm [Accessed November 27 2017].

WE261

Developing a National Food Inventory to estimate the Carbon Footprint of the diet of an average Spanish. Future requirements and policy recommendations

L. Batlle-Bayer, Universitat Pompeu Fabra UPF / UNESCO Chair in Life Cycle and Climate Change Escola Superior de Comerç Internacional ESCI; A. Bala, UNESCO Chair in Life Cycle and Climate Change (ESCI-UPF) / UNESCO Chair in Life Cycle and Climate Change. Escola Superior de Comerç Internacional ESCI; P. Fullana, Universitat Pompeu Fabra UPF / UNESCO Chair in Life Cycle and Climate Change Escola Superior de Comerç Internacional ESCI; R. Aldaco, Universidad de Cantabria / Department of Chemical and Biomolecular Engineering

Dietary patterns have a significant impact on greenhouse gases (GHGs), and diet choices can increase or reduce the Carbon Footprint (CFP) of consumers. Recently, more research has focused on estimating and comparing the CFP of different diets; however, high uncertainty is caused by the lack of reliable, available or representative data. The current study discusses this issue on data availability, and it results from our previous study on the CFP of the annual food consumption of an average Spanish citizen. To calculate the CFP of the average Spanish dietary pattern, a list of food categories with its representative food products was developed, and an extensive literature review was done in order to build up an inventory (inputs, outputs and emissions) per each food product. The system boundaries of this study are from cradle-to-consumer, and data for all life cycle stages (crop cultivation, farming systems, fisheries, industrial processing, manufacturing, distribution and consumer use) were gathered. Furthermore, food losses and food waste along the whole supply chain were also considered. While total annual emissions, about 1.4 Tn per Spanish citizen, were considered within the usual range, this result remain uncertain due to lack of representative data at the National level. The proxies used to fill the data gaps were considered of good quality. However there is a need to develop inventories for production of certain food products for which there is no inventory data available, as well as other life cycle stages, such as the wholesale & retail and the consumer phase. Furthermore, we suggest the inclusion of environmental data in food policy, for example, adding the CFP of food products and dietary patterns within the national dietary guidelines.

WE262

Life Cycle Air Emissions External Costs Assessment for comparing Electric and traditional passenger cars

P. Girardi, P.C. Brambilla, RSE SpA / SFE

The scope of this study is to compare the externalalities of electric, gasoline and diesel motorisations of an average passenger car (aVW Golf) giving a complementary reading of the results of an LCA. Starting from the results of the NEEDS project, authors present a methodolgy taking into account: the year of emission; the geographical area where the pollutant emissions take place; the height of release; the population density of the area where the emission take place; the average level of income of the country in which the emission take place. A complete LCA of an electric, gasoline and petrol VW Golf has been carried out considering city cycle real consumptions from EPA (fueleconomy.gov) and real emissions from national inventory on transport air emission factors (http://www.sinanet.isprambiente.it/it/sia-ispra/fetransp/). The use phase of the vehicles occurs in Italy, the energy used for battery charging is the Italian marginal mix, the vehicles assembly occurs in Germany while batteries are assembled in Austria. The upstream of fossil fuel is consistent with the nowadays actual national import mix. Emissions of PM_{10}, $PM_{2.5}$, NO_x, SO_x, NH_3 NMVOC, CO_{2eq} have been taken into account for externalities evaluation. Considering that more than ten tousand processes where involved, for each LCA phase accounting for more than 2 % of the weighed emission of PM_{10}, $PM_{2.5}$, NO_x, SO_x, NH_3 NMVOC, a specific height of realise and geographica area have been assinged distinguished by: Italy (wher car are used and most of electricity produced), Germany (where car are produced), Austria (where battery are assembled) Lybia, Algeria, Holland, Russia (mainly for upstream of fossil fuels), EU 27 an Rest Of the World (for materials production). The damage factor used in NEEDS project have been modified taking into account the real per capita income growth rate from year 2000 to 2015 instead of the foreseen rate and the per capita GDP of considered countries. Moreover, the damage factor of PM 2.5 have been divided in three different damage factors taking into account the different population density of urban, suburban and rural areas . The external costs evaluation for the analysed vehicles shows that Electric Golf performes better in terms of external costs, mainly thanks to the minor costs due to Climate Change. As regard regional externalities, the external costs due to emissions in Italy make the electric vehicle even more competitve than considering the overall regional externalities.

WE263

Life Cycle Costing: methodological description and implementation

B. De Caevel, J. Dulbecco, RDC Environment; A. Ciroth, GreenDelta; T. Huppertz, I. Descos, RDC Environment; J. Garcia, SCORE LCA

The complexity of production processes and products combined with an increasingly dynamic competitive environment has created the need to monitor and analyze not only the production phase, but also all upstream and downstream costs. Thus, producers are seeking to minimize the overall cost of their product generated throughout the life cycle. The private consumer, on the other hand, seeks to compare the different investment or purchase options by trying to integrate, in a holistic way, their present costs (acquisition prices and associated taxes) and future costs (use, maintenance and end-of-life). Life Cycle Costing (LCC) is the typical tool to meet these objectives as it allows to analyze the cost structure of a specific object throughout its life cycle. Due to their convergent approach over life cycle, there are some complementarities between LCC and Life Cycle Assessment (LCA). A combined application of the two approaches could help economic actors and decision makers to find cost-effective solutions, while minimizing their environmental impacts. However, combining both approaches requests monetizing the environmental impacts, to be able to aggregate them with the economic costs. Responding to SCORE LCA's enquiring about LCC and its complementarity with LCA, the purpose of this study is to provide the keys to understanding LCC, its concepts, its scope of use and its theoretical and conceptual limits. It also presents operational implementation elements such as: current methods of applying the LCC, means available for its implementation and possibilities of coupling it with LCA. In a first part, we present the theoretical bases and the state of LCC practice, i.e. definition and history, usefulness and reasons why the method is still not widely used. The second part presents the different types of LCC and the cost perimeters considered for each of them. The third part presents how to implement LCC, with detailed recommendations and guidelines. The fourth part discusses the possibilities of coupling LCC and LCA in practice by first showing the usefulness of such a combination, then by presenting the software that allows coupling methods. We conclude by identifying the necessary work to improve the practice of LCC and LCC coupled with LCA.

WE264

Pizza: it is dangerously delicious!

K. Stylianou, University of Michigan - School of Public Health / Environmental Health Sciences; V. Nguyen, University of Michigan / Department of Computational Medicine and Bioinformatics; V.L. Fulgoni III, Nutrition Impact, LLC; O. Jolliet, University of Michigan

The saying, we are what we eat, is true! Diets are key for human health, more than 10 million deaths/year worldwide are attributable to dietary risk factors. A challenge food Life Cycle Assessment (LCA) faces is that nutrition, a dominant impact pathway for health, is often neglected. At the same time, food LCA primarily focuses on single ingredient items while a large portion of modern diets is comprised of mixed dishes, a mixture of ingredients, which often remains unexplored. We propose a framework for evaluating mixed dishes in LCA that considers nutritional health impacts and benefits and demonstrate its application on pizza. We develop 14 marginal nutritional characterization factors (CFs) that cover major food groups and nutrients and allow the assessment of nutritional health effects in LCA. CFs are estimated by coupling age- and gender-adjusted outcome-specific incidence rates with risk ratios and severity factors from the Global Burden of Disease, measuring benefits (+) and impacts (-) in avoided μDALY/g. To evaluate the environmental impacts, we deconstruct pizza into "basic ingredients" using the USDA Standard Reference 28 database with a resolution of 3,200 single- and multi-ingredients that we further deconstruct. Ingredients are then linked to life cycle inventory (LCI) datasets from the Ecoinvent v3.2, the World Food LCA Database v3.1, and the ESU World food database. We evaluate impacts using Impact World+. Nutritional CFs for food group and nutrient range between -8 (sodium) and 57 (omega-3 from seafood) avoided μDALY/g. Human health scores for pizzas range from -35 avoided μDALY/serving pizza with extra meat to 2 avoided μDALY/serving pizza with no cheese. For the environmental impact assessment, global warming estimates vary from 0.06 (pizza with no cheese) to 0.20 (pizza with extra meat) kg CO2 eq/serving, corresponding to -0.04 and -0.17 avoided μDALY/serving, respectively. When it comes to pizza, environmental emissions further enhance nutritional health impacts. Nutrition can dominate the human health and should be considered in food LCA. We have developed an approach bringing together environmental and nutritional health effects in a common metric that could be used as a benchmark for a comprehensive assessment of all food items and diets in LCA. Expanding this approach to various food items could help decision-makers and consumers not only make better comparisons but also identify sustainable food items and adopt sustainable diets.

WE265

The impact of supplemented amino acids in animal feed - a new Life Cycle Assessment approach using the Protein Quality Index as functional unit for comparing protein sources

A. Wojciechowski, Evonik Technology & Infrastructure; M. Binder, Evonik Nutrition & Care GmbH

Proteins, which are made up of amino acids (AA), are essential for human health. Most of AA can be synthesized by the body but 8 of them are called „Essential Amino acids" (EAA) because they cannot be produced by human or animal and it is crucial to get them through food or feed intake. AA are also used as supplements in animal feed, providing the option to reduce the protein content of feed. Protein production has a major impact on the environment: it is responsible of ~14,5% of all human caused greenhouse gas emissions but also requires large amount of land and water due to the high quantity of crops required for feeding animal. Consequently, comparing protein sources using Life cycle assessment (LCA) is important for decision-making. However, choosing the functional unit (FU) is often a critical issue for food systems. The quantity of food (i.e. 1 kg) is the most used FU currently. Nevertheless, this FU does not represent the function of food that is to provide proteins. A more precise FU is to compare protein sources based on their protein content (i.e. 1 kg protein). To have a more holistic approach, nutritional and qualitative aspects should also be included in the FU. Actually, most plant protein sources do not bring all the EAA required. In this study, the Protein Quality Index (PQI) developed by Sonesson (Sonesson et al., 2016) was applied as a FU. It takes into account aspects such as EAA, AA digestibility, AA requirements but also food habits. In our study, an LCA was conducted on several protein sources: conventional (pork, chicken meat, salmon and tofu) and non-conventional sources (insects and algae). The role of the AA supplementation in animal feed was also investigated. On the one side, the analysis has shown that non-conventional protein sources perform better in all environmental categories, independently of the choice of the functional unit. Tofu performs better than animal protein but the difference between animal and vegetable based proteins becomes much lower when a more elaborated FU is used. On the other side, the supplementation in AA allows a reduction of the environmental impact of chicken and pork. Using the PQI as a FU, the impact of chicken and pork with AA supplementation is even lower than the one of tofu in some categories. Using the PQI as a FU is a step toward a more holistic assessment. A next step might be to include other nutrients such as iron and vitamins in the FU.

WE266

The ISO/DIS 14008 standard: Monetary valuation of environmental impacts and related environmental aspects -- Principles, requirements and guidelines - an overview

J. Serre, VERI; T. Bachmann, EIFER - European Institute for Energy Research / Urban systems group

Key words: monetary valuation, framework, standard, ISO 14008 The use of monetised environmental impacts and associated aspects has substantially increased in the last couple of years. This trend is mirrored since 2016 by efforts at ISO level to develop standards on environmental costs and benefits. This contribution will present the result of the work achieved in ISO/TC 207/SC 1/WG 7, developing ISO 14008 whose current title is "Monetary valuation of environmental impacts and related environmental aspects — Principles, requirements and guidelines". The work started in February 2016. After five WG meetings, ISO 14008 has reached the Draft International Standard (DIS) stage in fall 2017. The comments and ballot results of this DIS will be discussed during a WG 7 meeting in June 2018. Many organizations have experience in assessing environmental aspects and related environmental impacts resulting from their activities in physical units (e.g. tons of CO_2 emitted or numbers of disability adjusted life years, DALYs). To further integrate this information into the decision making process, it is useful to determine the monetary values of these environmental impacts and also of related environmental aspects. Monetary valuations enable comparisons and trade-offs between different environmental issues. The aim of this standard is to increase the awareness, understanding, comparability and transparency of monetary valuation of environmental impacts and related environmental aspects. To achieve this purpose, standardised and transparent documentation of the methods used to derive monetary values is essential. The multiplicity of monetary values, methods to determine monetary values, and ethical perspectives on money, requires careful consideration and prudent communication. ISO/DIS 14008 provides a framework that includes principles, requirements and guidance for monetary valuation of environmental impacts and related environmental aspects. Many methodological requirements or recommendations are intended for persons assessing monetary values. Following these requirements and recommendations enables good practice. The requirements in the reporting clause assist the user of monetary values in assessing the quality of the monetary valuation study. The presentation will give an overview of the ISO/DIS 14008 document.

WE267

The safe and sustainable loops framework for assessing residual material flows

J.T. Quik, RIVM / DMG; J. Spijker, A. van Drongelen, E. Van der Grinten, RIVM; A. Hollander, RIVM / DMG; R. de Jonge, J. van Leuken, L. de Poorter, H. Schmitt, B. Venhuis, RIVM; P.v. Vlaardingen, RIVM / Expert Centre for Substances; S. Waaijers-van der Loop, National Institute for Public Health and the Environment (RIVM) / Centre for Safety of Substances and Products; P. Wassenaar, National Institute for Public Health and the Environment (RIVM); M.C. Zijp, RIVM / Centre for Sustainability Environment and Health; J. Lijzen, RIVM

The circular economic system was developed to foster an industrial system that is restorative or regenerative by intention and design. An obstacle in the transition to such a system is that restoration of materials by reuse or recycling is subjected to safety legislation with an origin in the linear economy. In order to combat this obstacle, a shift is required from a purely safety based assessment to a more holistic assessment focused on sustainable development. Such a holistic assessment would integrate the absolute safety assessment, a requirement by law, with an assessment of the relative benefit that reuse of material flows have on all aspects of sustainability. However, assessing all aspects of sustainability is not practical for final decision making or feasible, considering the state of development of the tools, methods and data availability. Assessments of current recycling options are mainly focused on safety risks towards the environment and human health. Here we propose a first step in including environmental impacts or benefits related to closing material loops and increasing material value. This step is part of a bottom up approach to a more holistic methodology. It holds a novel framework (Safe and Sustainable Loops, SSL) aimed at assessing the safety as well as the sustainability changes of residual material flows within a clearly defined scope. In the Netherlands specific end of waste criteria can be applied to make the use of residual materials flows as a resource possible. The SSL framework in essence is a selection of themes that are important in the choices regarding derogation of residual material flows for new applications. These themes are the building blocks of the framework, the modules. In theory, the framework itself is the back bone that connects these modules together. The current themes which are developed into modules are: Substances of very high concern (SVHCs), Pharmaceutical residues, Pesticides, Pathogens, Antimicrobial resistance, Circularity and Environmental Sustainability. These were selected for the first iteration of the framework because of their relevance for assessing risks and benefits of residual material flows during the past few years. The aim of this approach is to allow a level playing field using a generic framework with modules based on lessons learned from earlier cases.

WE268

Who is being served? Considering the values stakeholders wish to sustain in decision making

S.E. Apitz, SEA Environmental Decisions Ltd

If we want our science to be part of the environmental decision process, we need to engage with stakeholders of all types about what they value to ensure that the science we generate is relevant to and translated in terms of these values. This requires a consideration of as diverse a range of affected stakeholders as possible. Unengaged subjects, due to a lack of resources, interest, or awareness, may not have their needs and values addressed unless a special effort is made to identify and consider them. One can view the concept of social equity as all-encompassing, under the premise that all impacts (positive and negative) of decisions can be seen as social impacts; and stakeholders must decide what services they envision for their land- and water-scapes - what values they wish to sustain. In selecting indicators to represent stakeholder values, the challenge is to build a conceptual framework which links measurable metrics of impact to value terms that resonate with the public, and reflect value statements made by the community. When the trade-offs are considered, it is important to consider the needs, demographics and vulnerabilities of a diverse population. Sustainability and ES concepts can and should be support environmental decision making; the application of threshold criteria ensures 'strong' sustainability in which environmental considerations are not compromised in trade-off consideration. The use of a framework which guides stakeholders to consider the extent to which they prioritize impacts to all (rather than just a narrow sub-set) of their values provides for a balanced public comment process, less subject to single- or narrow-issue lobbying. Identification of the risks and benefits of most interest to stakeholders also can support negotiation and optimization of alternatives under consideration, support collaborative design of more sustainable options and help inform the design of a long-term monitoring plan that addresses community values. The goal should be to envision a sustainable approach from the beginning of a project with collaborative input from a large group of stakeholders, supporting informed, transparent, and balanced decision making that protects services of importance to the community. Tools and approaches, and the path forward, will be discussed.

Salt of the earth - causes, consequences and management of salinization of surface freshwaters, groundwaters and soils (P)

WE269

Effects of long-term exposure to increased salinity in the amphibian skin bacterium Erwinia toletana

A.C. Gabriel, University of Aveiro / Biology; I. Lopes, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro; S. Costa, Universidade de Aveiro / Department of Biology & CESAM, 3810-193 Aveiro; I. Henriques, Universidade de Aveiro / Departamento de Biologia   CESAM

Amphibians constitute the class of vertebrates with the highest proportion of endangered species; chemical contamination being a main factor for its populations and species decline. The skin bacterial community of amphibians may help them to cope with such chemical contamination. If skin bacteria may increase its tolerance to chemicals, then they could be used for bioaugmentation in amphibians to help them cope with contamination. This work aimed at assess if an amphibian skin bacterium could increase its tolerance to NaCl after long-term exposure to low levels of salinity. *Erwinia toletana*, isolated from the skin of *Pelophylax perezi*, was selected as the model species. Clonal populations of this bacterium (5 replicates) were exposed for 46 days to LB medium (Et-LB) or to the effective concentration of NaCl causing 10% of growth inhibition (Et-NaCl; 18g/L). To assess the capacity of recovery from long-term exposure to NaCl, after the 46 d period, Et-NaCl was transferred to LB medium and cultured for a period of 16 d (Et-R). The Et-LB isolate also continued to be cultured in LB medium for further 16 d. The tolerance of ancestral and evolved populations to NaCl was assessed by exposing them to 6 NaCl concentrations (5, 10, 15, 20, 25 and 35 g/L) plus a control (LB medium). Effects of NaCl on bacteria growth and metabolic mechanisms (as degradation of carbon compounds) was monitored. Genotypic alterations were assessed using a PCR-based molecular typing method (BOX-PCR). Results of growth shown that long-term exposure to NaCl slightly increased the tolerance of *E. toletana* to this salt, EC_{50} for growth were: 22.5g/L (8.64-36.4) for Et-LB; 30.3g/L (23.2-37.4) for Et-NaCl, and 26.1g/L (19.3-32.9) for Et-R. Though, as confidence limits overlapped, tolerance increase was not considered significant. Furthermore, differences in metabolic processes were observed between Et-LB and Et-NaCl, suggesting the use of different carbon sources. This could be associated with the activation of detoxification mechanisms or energetic demanding mechanisms to cope with osmotic stress. Genotypic alterations were not observed, indicating that *E. toletana* increased tolerance to NaCl could be due to membrane plasticity mechanisms to cope with osmotic stress. The tendency shown by *E. toletana* to acquire increased tolerance to low levels of salinity could constitute a promising bioaugmentation tool in amphibian's skin, aiming the improvement of these organisms tolerance to chemicals.

WE270

Impacts of agriculture brackish effluents in saline ecosystems: when the low salinity cannot be an advantage but an impact

J. Alvarez-Rogel, Escuela Técnica Superior de Ingeniería Agronómica. Universidad Politécnica de Cartagena / Ciencia y Tecnología Agraria; F. Jiménez-Cárceles, BIOCYMA, Consulting in Environment and Quality, S.L. Murcia, Spain.; M. González-Alcaraz, Department of Biology & CESAM - University of Aveiro / Biology & CESAM

Salinity is usually considered one of the main factors of soil and water degradation worldwide. Anthropogenic salinization due to the use of low quality water and/or over-fertilization leads the existence of saline effluents that degrade water quality and constrain plant growth and crop production. However, in saline wetlands salinity is not undesirable, but a proxy parameter to maintain the singularity of the ecosystem. In these environments the existence of a diversity of habitats is closely related to gradients of soil salinity and moisture, with extremely saline sites, brackish sites, wetter sites and drier sites. While agricultural saline effluents may salinize normal soils, they can degrade saline wetlands by decreasing the native soil salinity. This work reports changes in ecosystem structure and diversity in a saline wetland adjacent to the Mar Menor saline lagoon (SE Spain). Species cover, soil salinity, and the groundwater level were monitored in two 2-years periods with a difference of 13 years between them. The results indicated an elevation of the water table throughout the 13-year period, which was attributable to brackish water flowing from areas with intensive agriculture. The latter led to an increase of flooding periods, a decrease of soil salinity in the most saline sites and increased in the least saline ones. Following these changes, damages in protected habitats were observed, due to the proliferation and increase of biomass of several species. *Sarcocornia fruticosa*, *Phragmites australis* and *Juncus maritimus* strongly expanded at the wettest sites, which led to the disappearance of the original zonation pattern and the homogenization of the ecosystem. Bare areas, necessary for nesting and feeding of several bird species, disappeared. According to the results obtained, a decrease of soil and water salinity was one of the main factors contributing to the degradation of the saline wetland leading to an exacerbated growth of some species and a decrease of habitat diversity. In this case, the low salinity of the effluents reaching the wetland was a problem, not an advantage.

WE271

Context dependent toxicity - do ecological interactions alter the effects of salinity on stream macroinvertebrate communities?

B.J. Kefford, J. Reich, J. Bray, University of Canberra / Institute for Applied Ecology

The effect chemicals on populations and communities have long been noted to vary between different studies although the mechanism(s) for this variation is unclear. Research has examined variability associated with chemical and physical environments (e.g. bioavailability, co-occurring contaminants) and ecotoxicological and physiological (e.g. temporal and spatial variability in species' sensitivity). Less consideration has been given to ecological mechanisms including those mediated via indirect effects, such as competition and predation. For example, a given population of a species may be able to persist in the presence of a particular level of contamination, but this persistence may be dependent on competition and predator prey interactions, and the relative fitness of these taxa at that level of contamination. Here we report the results of a mesocosm experiment that examined the effects of biotic interactions on salinity effects. We examined effects across a broad salinity gradient using 'sensitive' communities collected from a low salinity site (~80 µS/cm) and 'tolerant' communities (collected from a high salinity site ~1600 µS/cm). This was examined using a mesocosm experiment consisting of 32 independent re-circulating 1000 L mesocosms. Controls (100 µS/cm) and salinity treatments (500, 1000, 2500 and 5000 µS/cm) these were replicated 4 fold and were crossed in an orthogonal design with the source biota (stream macroinvertebrates and microbe) either from: (1) a low salinity site only or (2) both low and high salinity sites. The experiment is based on the logic that if salinity increases at a site, organisms have the potential to migrate from higher salinity sites within the same region. Thus the organisms from the (previously) low salinity site would have to be able to tolerate both the increase in salinity and ecological interactions with organisms from higher salinity sites. We observed differing effects of salinity on the macroinvertebrate community from the low salinity site depending whether these biota were co-inhabiting with biota from a high salinity site. Such context dependent toxicity deserves greater consideration in studies of the effects of chemicals on populations and communities.

WE272

Challenges in developing a water quality guideline for water hardness

S. Bogart, University of Lethbridge / Department of Biological Sciences; E. Stock, University of Lethbridge; A. Manek, University of Saskatchewan; A. Tillmanns, C. Meays, British Columbia Ministry of Environment & Climate Change Strategy; G.G. Pyle, University of Lethbridge / Biological Sciences

Increases in salinity, or the total ionic content, of water can be toxic to freshwater species, as can shifts in the balance of major ions (Ca^{2+}, Mg^{2+}, Na^{+}, K^{+}, HCO_3^{-}, SO_4^{2-}, Cl^{-}), or increases in the Ca^{2+} and Mg^{2+} content of water alone (i.e. water hardness). Although anthropogenic salinization of freshwaters is increasing, virtually no water quality guidelines (WQG) exist for regulating these individual ions or ion mixtures. Water hardness in receiving waters can be increased to potentially toxic concentrations via effluent, produced waters, and saline run-off from various human activities, e.g. coal mining, oil and gas extraction, the use of Ca- and Mg-based road de-icers, and agriculture. Thus, developing a water hardness based WQG for the protection of aquatic life is warranted. Using Canadian protocols for WQG development, we attempted to derive a WQG for water hardness. Relevant literature was collected and reviewed for potential inclusion in the WQG, however, current literature offered several challenges and major data gaps that hindered WQG derivation. Moreover, the background variation of water hardness throughout an exemplar regulatory region of interest also did not support WQG development using traditional methods. These challenges and limitations will be discussed in the context of similar regulations from other jurisdictions, the need to consider additional, practical limitations of regulating water hardness, or major ions in general, recommendations for improved data consistency, and potential regulatory options.

WE273

Prioritization of water quality stressors according to their relative impact on ecological quality of rivers using large-scale field data: salinity first?

E. Berger, Senckenberg Gesellschaft / Department Quantitative Landscape Ecology; R. Schäfer, University Koblenz-Landau; P. Haase, A. Sundermann, Senckenberg

The political aim of achieving good ecological quality of all European water bodies requires knowledge on how to prioritize stressors and human pressures for management based on their relative impact. A challenge thereby is the frequent co-occurrence of multiple stressors. We applied eco-epidemiological approaches to large scale monitoring data from Saxony, Germany, to investigate the relative contribution of different water quality and land-use gradients to ecological change. Two approaches were applied: First, water quality gradients (e.g. oxygen, conductivity, phosphorous and micropollutants) and land-use gradients (e.g. % arable and urban catchment land cover, position of wastewater treatment plants) were used as predictor variables in multiple linear regression analysis and hierarchical partitioning with ecological quality indices based on invertebrates (% EPT, MMI, ASPT, BMWP, GSI, SPEAR %) as response variables. Secondly, individual taxon responses with respect to different water quality gradients (including also major ions such as potassium, sodium, chloride etc.) were assessed using Threshold Taxa Indicator Analysis (TITAN). The method is based on change point and indicator species analysis and allows the identification of ecological change points that may be used to derive environmental quality criteria. Both regression analysis and TITAN results indicate a high impact of oxygen and salinity, which were associated with arable and urban catchment land cover. Although observed associations may not be direct causes of ecological impairment, it may be worthwhile to implement legally binding quality standards for these variables. Of the 324 analyzed taxa 23% had change points far below the German orientation value for chloride (200 mg/L) that should not be exceeded to achieve good ecological status according to the water framework directive. Thus, lowering of orientation values for salinity and associated ions should be considered to protect and restore stream biodiversity. Moreover, the results suggest that preventing release of poorly treated wastewater should be prioritized over up-grading of well-functioning treatment plants.

WE274

Estimating protective potassium concentrations for freshwater mussels, a taxon of global conservation concern

T. Augspurger, U.S. Fish and Wildlife Service / Ecological Services

Globally, there are about 620 species of freshwater mussels (Family Unionidae), and IUCN lists 28 species as extinct and 106 as endangered or critically endangered. Mussels are among the most sensitive freshwater water organisms to toxicity from chloride and potassium, and the environmental relevance of these is increasing with sea level rise and brine discharges. Average potassium concentrations in relatively unpolluted streams of North Carolina (USA) range from 0.2 to 2 mg/L. An industrial effluent with potassium averaging 504 mg/L and proposed for discharge to a stream with endangered mussels necessitated derivation of protective potassium limits (because there are no State standards or USA water quality criteria for potassium). From the literature, we compiled potassium 96-h EC50s (with endpoints of lethality or immobilization) for mussels and retained those with > 90% control survival, measured test chemical concentrations, and acceptable test water quality. Five EC50s ranged from 31 to 48 mg/L at a water hardness of 100 mg/L as $CaCO_3$, and we applied the North Carolina guidance of one-third of the lowest EC50 to define an acceptable concentration to avoid acute toxicity. We adjusted the 10 mg/L acute limit to a water hardness of 18 mg/L which is the 5th percentile of the proposed receiving stream (protective most of time because potassium is less toxic as hardness increases). The hardness-adjusted acute water quality guideline of 7 mg/L potassium was recommended as an instantaneous concentration not to be exceeded. Chronic toxicity data for potassium and mussels were available for two studies from 28 to 300-d. Because mussels can live for decades, we used the 300-d test. The geometric mean of the test NOEC (1 mg/L) and LOEC (7 mg/L) yield a chronic value of 2.6 mg/L potassium which was recommended as a monthly average guideline not to be exceeded more than once every three years. We used 32 years of receiving stream flow data to derive estimates of instream waste concentration and effluent limits. We identify uncertainties in guideline derivation and discuss recommendations for quarterly mussel toxicity tests, instream monitoring, and research to narrow uncertainties. There are several means by which stream-specific and mussel-specific potassium guidelines could be derived. This method tracks North Carolina water quality standards and definitions and is reasonable with available data.

WE275

LIFE LAGOON REFRESH - Coastal lagoon habitat (1150*) and species recovery by restoring the salt gradient increasing fresh water input. Management measures in the northern Venice Lagoon (NE, Italy)

F. Cacciatore, ISPRA-Institute for Environmental Protection and Research / Mitigation and Prevention of Impacts; A. Bonometto, A. Feola, E. Ponis, ISPRA Institute for Environmental Protection and Research; A. Sfriso, University Ca Foscari of Venice; B. Matticchio, IPROS; M. Lizier, Regione del Veneto; V. Volpe, Provveditorato OO. PP. Veneto, Trentino Alto Adige e Friuli Venezia Giulia; M. Ferla, R. Boscolo Brusà, ISPRA - Institute for Environmental Protection and Research

The northern Venice Lagoon (SCI IT3250031) holds several Annex I-listed habitats of the Habitats Directive, such as the habitat type 1150* (Coastal lagoons). Recent monitoring activities showed that conservation status of the habitat 1150* is improving within SCI IT3250031, but it is still unfavourable in the inner landward areas, due to lack of ecotonal buffer areas, favouring self-regulation processes, between lagoon and mainland. In the past, the project area was occupied by reedbeds in large amounts, now significantly receded due to increasing of lagoon water salinity, caused by historical human activities (e.g. diversion of rivers with reduction of freshwater supply, inlet and channel excavation). With reduction or disappearance of reedbeds, their contribution to ecosystem services, like supporting numerous biological communities and species, are minimised. The LIFE LAGOON REFRESH project, started on Sept 2017, foresees the restoration of favourable conservation status of habitat 1150* in the northern Venice Lagoon and the recreation of favourable habitats for faunal species of community interest. The project actions involve: diversion of a freshwater flow from the Sile river into the lagoon (necessary for the recreation of the typical salt gradient of buffer areas between lagoon and mainland); restoration of intertidal morphology through the implementation of structures properly arranged to slow down the freshwater dispersion and to favour reed development; planting of *Phragmites australis* to accelerate the development of the reedbeds; transplantation of small dumps of seagrass species of the habitat 1150*, suitable to accelerate the recolonization by aquatic plants of low-salinity environments. The project aims to exploit the ecosystem services resulting from the recreation of a typical estuarine system to: counteract the depletion of lagoon bottom and fish communities; reduce eutrophication through reedbed phytoremediation function, favouring the presence of sensitive species and high ecological value aquatic plants; improve conservation status of bird species, including those listed in Annex I of the Birds Directive; increase the presence of fish species, listed in Annex II of the Habitats Directive. The restoration of salinity gradients will also contribute to increase biodiversity in the Natura 2000 network site, and to help achieving targets defined by the 2020 Biodiversity Strategy.

WE276

Comparing the growth of fescue and clover plants in petroleum industrial effluents and solutions of similar salinity

P. Srikhumsuk, Uiversity of Strathclyde / Department of Civil & Environmental Engineering; C. Knapp, J. Renshaw, University of Strathclyde / Civil and Environmental Engineering

Effluents (produced and flow-back waters) from the petroleum industry have been investigated for their potential toxicity to the environment, particularly in regards to chemical composition and salinity. The purpose of this study was to investigate whether their toxicity is any greater than exposure to solutions of similar salinity. The tolerance of *Festuca rubra* L. (fescue) and *Trifolium pratense* L. (clover) were investigated for 8 weeks under hydroponic conditions to compare their growth in brine effluents from tertiary recovery operations. Experiments further compared serially-diluted effluents and synthetic solutions (e.g., $NaCl/CaCl_2$) of similar salinities. There were different growth responses to the wastewater and saline solution among both plant species. *F. rubra* was exhibited a significant higher survival percentage than *T. pratense*. After four weeks of exposure, *T. pratense* exhibited greater sensitivity and lethality. Interestingly, biomasses of both plants were greater from industrial wastewater than the comparable brine solution. Although salinity limited plant growth, the tertiary wastewaters contained abundant inorganic and organic substances that may have triggered plant survival and salt-tolerance. *F. rubra* grew under salts stress, and presented a mechanism to crystalize salt on their leaves. Hence, plant uptake, under certain conditions, may be promoted as an alternative treatment for high salt concentrations.

WE277

Contribution to the salinization risk assessment, under drought conditions, in the Alqueva irrigation area (South Portugal)

A. Tomaz, S. Fialho, A. Lima, Instituto Politécnico de Beja; A. Penha, H. Novais, M. Potes, M. Iakunin, G. Rodrigues, Instituto de Ciências da Terra; P. Alvarenga, LEAF Centro de Investigação em Agronomia, Alimentos, Ambiente e Paisagem, Instituto Superior de Agronomia, Universidade de Lisboa; M. Costa, M. Morais, R. Salgado, Instituto de Ciências da Terra; P. Palma, Instituto Politécnico de Beja / Department of Tecnologies and Applied Sciences

In Mediterranean regions, climate changes have enlarged water limitation for crops, leading to an increased demand for irrigation water. During the hydrological years of 2016 and 2017, Portugal experienced a drought season that has extended throughout almost the entire mainland territory reaching a severe drought level. Under water scarcity conditions and high atmosphere evaporative demand, the risk of land salinization is one of the major threats to the sustainability of irrigated agriculture. Therefore, it is very important to assess the quality of irrigation water and the risks of salinity for crop production, in order to adopt appropriate management practices in irrigated areas. This study is focused on the salinity risks for the production of the most representative crops grown in the Alqueva irrigation area. This is a large irrigation scheme with a total area of 120 000 ha centered in the Alqueva reservoir. For the purpose of the study, a chemical assessment of some major inorganic ions (Na^+, Ca^{2+}, Mg^{2+}, K^+, SO_4^{2-} and Cl^-), pH and electric conductivity (EC_W), was conducted throughout 2017, on water samples collected on four platforms sited in the reservoir. Water quality for irrigation was evaluated considering both the Portuguese regulations and the FAO guidelines. Sodium adsorption ratio (SAR) and soil salinity (ECe) were estimated, in order to assess potential sodium-related soil permeability and crusting problems, as well as, potential yield reductions in the most significant crops of the Alqueva perimeter. Higher ion concentrations and water salinity were quantified with the increase of atmosphere evaporative demand. Sodium hazard assessment showed slight to moderate risk of reduced infiltration rates, a result that should be taken into account when surface or sprinkler irrigation systems are used. Furthermore, relative yield reductions may be mainly found in horticultural crops, classified as moderately sensitive to sensitive in the salt tolerance scale.

Systems ecotoxicology: application of OMICS data across multiple level of biological organization in research and risk assessment (P)

WE279

Investigating wildlife diets using high-tech DNA sequencing

J. Ludwigs, Rifcon GmbH; I. Katzschner, RIFCON GmbH Goldbeckstr Hirschberg Germany; G. Weyman, ADAMA; A. Winkler, J. Kalinowski, Center for Biotechnology (CeBiTec) Universität Bielefeld

In wildlife risk assessments according to EFSA (2009), the ingested diet is one of the core factors to define exposure, using default diet compositions in the first tier risk assessment. The so-called PD factor (composition or portions of diet) is one of the standard refinement parameters which intend to add realism to higher tier risk assessments. Publically available dietary data are often used to refine PD in wildlife risk assessments; however, such data are often variable and/or not representative for the specific risk assessment scenario. Besides such literature data, specifically registration-relevant PD field studies can be conducted. PD values gathered from such studies are based on collected faeces, stomach samples, or stomach flushing. In these studies samples (or parts thereof), are investigated visually by microscope and food items are identified based on comparison with comprehensive reference data libraries and collections of potential diet items. For plant material, the results are mainly presented on a fairly basic taxonomic level and are often related to wildlife risk assessment defined diet fractions which have different default residue levels (i.e. dicotyledonous plants or monocotyledonous plants only). However, this is rather time-consuming and imprecise. Recently, DNA sequencing techniques are increasingly applied for diet composition analyses in ecological science. We initiated an approach using ‘next-generation’ DNA amplicon sequencing to quantitatively assess the diet composition of wild herbivorous mammals, taken from faeces samples collected on arable fields. Data on the relative abundance of each plant species were derived by enrichment and sequencing of a specific DNA region (ITS2 region of the ribosomal DNA) and by comparison to comprehensive plant species DNA databases. The approach has proved to be very useful on identification of relative abundances of plant species from faecal samples. This new genomics approach, its needs and limitations for refined risk assessment will be presented and discussed.

WE280

Design of a Real-Time PCR array to analyze the gene expression in Physella acuta (Gastropoda) in chemical stress and starvation

M. Novo, J. Martinez-Guitarte, UNED / Fisica Matematica y de Fluidos

Molecular endpoints are nowadays under study for their inclusion in test toxicity tests. While vertebrate species are usually well-known; there is a lack of information on invertebrates. The study of the latter is complex since their body shape, behavior, and ecology are very diverse, and great differences can be found even within the same animal group. In order to improve our knowledge in putative molecular endpoints and to evaluate some genes as biomarkers, a Real-Time PCR array has been designed for *Physella acuta*. This species of freshwater snail is used in environmental toxicology studies and it has been proposed as an adequate species for toxicity tests because of its sensitivity to different toxicants and ease of culture. A transcriptome for this species was assembled, by sequencing cDNA libraries from individuals of different developmental stages and exposed to different toxicants. Comparison with database allowed the identification of genes involved in pathways related with the response to toxicants. We selected 42 of these genes plus six genes used as reference to design an array for Real-Time PCR analysis. Stress response, detoxification mechanisms, endocrine system, or epigenetics were some of the pathways analyzed in the array. In order to validate the toxicological and ecological interest of this approach, individuals were treated with an antibiotic, tetracycline, for seven days or were left starving for 7 and 10 days. The results obtained for these experiments are presented, showing the interest of designing specific arrays to perform more detailed analysis of molecular endpoints that can be related with toxicant mode of action and stress situation. We hope that the methodology presented here can serve as an example for the study of other species in order to improve our knowledge of their biology. This work has been funded by the *Ministerio de Economía y Competitividad*, CICYT (SPAIN), CTM2015-64913-R.

WE281

Effects of temperature on the transcriptome of the marine copepod Temora longicornis

I. Semmouri, Ghent University (UGent) / Animal Sciences and Aquatic Ecology; J. Asselman, Ghent University / Laboratory for Environmental Toxicology and Aquatic Ecology GhEnToxLab unit; C.R. Janssen, Ghent University / Applied Ecology and Environmental Biology; K. De Schamphelaere, Ghent University (UGent) / Applied Ecology and Environmental Biology

Over the past decades, the world's oceans and seas have been influenced by several human induced impacts, including climate change. In the North Sea region, the average sea surface temperature of the water has already risen with 1-2 °C over a time-period of twenty-five years and is likely to rise further. Understanding the impacts of this changing environmental condition in zooplankton communities is crucial, as alterations in the zooplankton communities can affect entire marine ecosystems. Here, we focus on the potential effects of an increase in temperature on the calanoid copepod species, *Temora longicornis*, the dominant zooplankton species of the southern part of the North Sea. Since responses to environmental stress are genome-driven, a genetic study on the physiological responses to thermic stress can provide an increased mechanistic understanding and help predict potential responses to climate change in this copepod species. Therefore, we sequenced the whole transcriptome (using RNA-sequencing technology) in *T. longicornis*, after being exposed to thermal stress, to investigate gene expression differences as a response to temperature fluctuations. As such, this dataset will provide us with new insights on how exposure to increased sea water temperatures may affect the fitness of the most dominant zooplankton species of the southern part of the North Sea.

WE282

A traditional approach to modern endpoints - quantitative assessment of stress gene expression response to a range of copper concentrations in the freshwater mussel Anodonta anatina

G.M. Ekelund Ugge, Lund University / Biology; A. Jonsson, University of Skövde / Department of Bioscience; O. Berglund, Lund University / Dept of Biology

In the field of ecotoxicology, modern *transcriptomics* technologies have the potential to improve and complement current toxicity assessment methods and biomonitoring protocols. Early warnings of general stress and specific toxic modes of action could in theory be used as biomarkers of pollutant exposure or adverse effects. However, necessary base level understanding is currently lacking considering how gene expression may vary under realistic exposure scenarios. Therefore, we adopted an approach of quantitative assessment as an alternative to more descriptive methods. We chose the freshwater mussel *Anodonta anatina* as our model organism. Being a stationary filter feeder, it shows promise for use in exposure studies under both laboratory and field conditions. Furthermore, it is the most abundant freshwater mussel species in Sweden, and occurs in freshwater ecosystems over most of Europe. For the present study, mussels were collected locally in Vinne å (southern Sweden), on a location free from point source pollution. After two weeks of acclimatization to laboratory conditions, mussels were exposed for 96 h to one of three copper treatments (nominal concentrations of 1, 10 and 100 µg/l Cu^{2+}), or a control treatment (n= 5 per treatment). Using RT-qPCR, relative expression of a selection of general stress genes will be quantified in extracted digestive gland and gill tissue. Preliminary data will be presented, testing the hypotheses that the amplitude (fold-change) of relative expression differs (i) between treatments of the same gene, and (ii) between genes in the same treatment. Results from this initial experiment will be used to design a follow-up experiment, in order to test the dose-dependence of gene expression responses. Gradually, successively increased exposure scenario complexity (e.g. duration, chemical composition) will help us to better understand how expression patterns potentially vary under environmental exposure. By subsequent incorporation of biochemical and physiological biomarkers, we also aim to link stress gene expression patterns to effects at higher biological levels. Ultimately, a more thorough understanding of natural and pollution-induced variation in gene expression may allow *transcriptomics* to be usefully and successfully incorporated into various ecotoxicological assessment protocols.

WE283

Validating a contamination assessment tool from lab to the field: Folsomia candida exposed to a fungicide-based formulation

T.F. Simoes, S.C. Novais, Polytechnic Institute of Leiria / MARE IPLeiria; T. Natal da Luz, University of Coimbra / Department of Life Sciences, University of Coimbra; J. Renaud, CFE - Centre for Functional Ecology; J. Sousa, University of Coimbra / Department of Life Sciences; J. Römbke, ECT Oekotoxikologie GmbH; D. Roelofs, Vrije Universiteit / Department of Ecological Science; N. van Straalen, Association of Retired Environmental Scientists ARES / Department of Ecological Sciences; M.F. Lemos, Instituto Politécnico de Leiria / MARE IPLeiria

Folsomia candida is a widespread arthropod that occurs in soils throughout the world and has been used as a standard test organism in past decades for estimating the effects of pesticides and environmental pollutants on non-target soil arthropods. This species is among the most sensitive representatives of its taxon, being selected as a genomic model organism for soil toxicology studies. Although laboratory

experiments with a transcriptomics approach are essential to unravel modes of action of chemical compounds, higher-tier studies (e.g., field studies) are crucial as a validation criterion in environmental risk assessment trials, while their ecological relevance is increased when complemented by pertinent information at lower-tier studies (molecular level).Therefore, the main goal of the present study was to validate the mode of action of a commercial fungicide formulation in *F. candida* under a more realistic field exposure scenario, by targeting specific molecular biomarkers retrieved from a previous laboratory experiment. Based on previous data of survival and reproduction effects in *F. candida* exposed to a commercial formulation of the fungicide chlorothalonil (40% Bravo®500) in a natural agricultural soil under laboratorial conditions, organisms were now exposed under field conditions for 4 days to the same concentration as for laboratorial exposure (causing a 75% reduction on reproduction) and the Predicted Environmental Concentration (5 mg a.i./kg). Invertebrates were previously cultured in laboratory and simultaneously 12 replicate soil cores per treatment (including control) were collected from the field and defaunated. The cores were placed back in the field and 220 organisms (10-12 days old) were added per replicate core. Field contamination was made by spraying after a 3 hours acclimation period to the field by the organisms. RNA was extracted from each pool of organisms using the TRIZOL® methodology. According to previous laboratorial "omics" results with the same formulation, a set of specific genes were selected for a targeted gene expression analysis by qRT-PCR, corresponding to key genes of affected biological pathways including glutathione metabolism, oxidation-reduction, body morphogenesis and reproduction. This work contributes with a set of molecular biomarkers which can be used to develop a more effective set of tools to assess the early effects of such fungicide formulations in a real scenario of soil contamination.

WE284

Proteome response of Chironomus riparius under exposure to the neurotoxic insecticides Spinosad and Indoxacarb

H.R. Monteiro, University of Aveiro / Department of Biology and CESAM; J. Pestana, CESAM & University of Aveiro / Biology; A.M. Soares, University of Aveiro / department of Biology & CESAM; B. Devreese, Ghent University / Laboratory for Protein Biochemistry and Biomolecular Engineering; M.F. Lemos, Instituto Politécnico de Leiria / MARE IPLeiria

The development of quantitative methodologies in proteomics opened new doors for their potential applications within environmental sciences. Since proteins are the functional units of cells, the proteome of an organism at a given time and at a given condition reflects its current state. In this sense, any protein profile changes in response to toxicants may reveal their molecular targets and/or specific stress-response mechanisms, and thus can be used as potential early warning biomarkers of toxicant exposure. In this study, the potential of proteome changes as an early warning indicator for pesticide exposure in *Chironomus riparius* (Meigen) was evaluated using as model compounds two neurotoxic pesticides with distinct modes of action, spinosad and indoxacarb. *Chironomus riparius* third-instar larvae were exposed to three concentrations of each pesticide and iTRAQ methodology was performed to relatively quantify protein expression changes between exposed and non-exposed organisms. As expected, the pesticides exposure triggered different responses at the proteome level. Changes caused by spinosad were more noticeable than for indoxacarb exposure. Our results revealed a general decrease in the expression of globin proteins with the increase of spinosad concentration. Additionally, for spinosad, a significant decrease in the expression of an actin and a cuticle protein were also observed. Moreover, correlations between proteomics data and previously determined biochemical biomarkers responses were found for both pesticides. Our results suggest that protein profile changes have the potential to be used as early warning biomarkers of pesticide exposure, providing an interpretation of molecular pathways of toxicity behind the organismal response, therefore supporting the risk assessment of pollutants. This work contributes to the growing knowledge of sub-lethal effects of pesticides in invertebrates and their molecular targets. *Chironomus riparius*, a model organism in aquatic toxicology, is also presented as a promising model organism for environmental proteomics.
Aknowledgements: This study had the support of the Fundação para a Ciência e a Tecnologia through project PROTEOME (PTDC/AAG-MAA/1302/2014), co-financed by COMPETE (POCI-01-0145-FEDER-016773).

WE286

Assessing Cu impacts on freshwater diatoms: biochemical and metabolomic responses of Tabellaria flocculosa (Roth) Kützing

S.I. Gonçalves, Universidade de Aveiro / Biology; M. Kahlert, SLU Swedish University of Agricultural Sciences / Department of Aquatic Sciences and Assessment; S.F. Almeida, University of Aveiro / Department of Biology and GeoBioTec; E. Figueira, University of Aveiro / Biology CESAM

Metals are a recognised threat to aquatic organisms but the impact of metals such as copper (Cu) on benthic freshwater diatoms is poorly understood, even if diatoms are commonly used as water quality indicators. Our study aimed to elucidate the cellular responses of diatoms to Cu toxicity. The freshwater diatom *Tabellaria flocculosa* (TFLO), isolated from a Cu contaminated stream, was exposed to 0.3, 6 and 10 µg Cu/L, and the tolerance level and the cellular targets were studied using biochemical, physiological and metabolomic approaches. Cu was already toxic to *T. flocculosa* at concentrations common in environments which are usually not considered to be contaminated (0.3 µg Cu/L), and toxicity increased with Cu concentration. Strategies to cope with Cu varied with the level of Cu stress. Under Cu impact, the metabolome of *T. flocculosa* changed significantly, especially at high concentrations (6 and 10 µg Cu/L). Cu toxicity was counteracted by increasing extracellular immobilization (EPS, frustulins), antioxidant (SOD, CAT) and detoxifying (GSTs) enzymes activity and low molecular weight antioxidants (GSH). These mechanisms are fuelled by a higher energy production (ETS activity, use of sugars and lipids). At the highest Cu concentration (10 µg/L), these metabolic processes were specially enhanced in an attempt to restrain the oxidative stress generated by high intracellular Cu concentrations. However, these mechanisms were not able to fully protect cells, and damage in membranes and proteins occurred. Moreover, the decrease of hydroxylamine and unsaturated FA and the increase of saturated FA, 2-palmitoylglycerol, glycerol and diterpenoid compounds should be tested as new specific markers of Cu toxicity in future studies. This information can support the prediction of diatom behaviour in different Cu contamination levels, including highly impacted environments, such as mining scenarios, and may assist in environmental risk assessment policies. https://www.dropbox.com/s/8b1jsezdiqx3rmw/graph%20abs%20Copper.tif?dl=0

WE287

Non-targeted approach to identify metabolic perturbations in gilt-head bream liver and brain exposed to benzophenone-3

H. Ziarrusta, L. Mijangos, University of the Basque Country UPV/EHU / Department of Analytical Chemistry; S. Picart, A. Perera, Polytechnic university of Catalonia UPC / ESAII department; A. Usobiaga, A. Prieto, N. Etxebarria, M. Olivares, O. Zuloaga, University of the Basque Country UPV/EHU / Plentzia Marine Station (PiE-UPV/EHU) & Dep Analytical Chemistry

Benzophenone-3 (BP-3) is a widely used organic UV filter to protect humans and materials from damage by UV irradiation. The extensive use of BP-3 has led to its ubiquitous occurrence in the aquatic environment, causing an ecotoxicological risk to biota. Although some studies reported adverse effects, such as reproductive toxicity, further research needs to be done in order to assess its molecular and physiological effects, and modes of action. Therefore, in the present work, we investigated metabolic perturbations in juvenile gilt-head bream (*Sparus aurata*) exposed over 14 days via the water to BP-3 (50 mg/L). Liver and brain were collected prior to dosing and on exposure days 2, 4, 7 and 14 from control (n=10) and exposed (n=10) animals. Samples were flash frozen and then stored at -80ºC until analysis. Methanol:chloroform (80:20, v/v) mixture was used for non-selective extraction of fish tissues and subsequent non-target analysis was performed by means of UHPLC-Orbitrap MS in positive and negative modes with both C18 and HILIC separation. Metabolites were identified using Compound Discoverer (Thermo) interfaced to MZmine and the statistical data treatment was carried out with R software. Mortality was not observed during the experiment, and no statistical changes in fish weight, fish length, condition factor and hepatic somatic index were observed regardless of tank or dosing period. First of all, using sequence quality control samples, data had to be corrected to remove the effect of injection order. On the other hand, since some metabolites were significantly altered in both exposed and control tanks over the course of the experiment, time-series statistical analysis was carried out to identify the major trends (adjusted p-value < 0.05) associated with the interaction between exposure day and animal group (exposed or control). Metabolites driving group separation were further investigated using the Kyoto Encyclopedia for Genese and Genomes (KEGG) in order to determine affected pathways. Overall, these data demonstrate the usefulness of metabolomics to assess molecular-level effects of emerging contaminants. *Keywords:* Benzophenone-3, gilt-head bream, non-target metabolomics *Acknowledgements* - This work was financially supported by the Ministry of Economy and Competitiveness through the project CTM2014-56628-C3-1-R. H. Ziarrusta is grateful to the Spanish Ministry and L. Mijangos to the Basque Government for their predoctoral fellowships.

WE288

EFFLUENTS FROM PULP AND PAPER MILLS PROMOTE METABOLIC ALTERATIONS IN LIVER AND GONADS OF FISH

M. Barbosa Xavier, Universidade Federal de Santa Catarina / Biochemestry; C.H. Soares, Universidade Federal de Santa Catarina / Biochemistry Department; I. Baptista, Universidade Federal de Santa Catarina

Effluents from pulp and paper mills are internationally recognized as containing toxic substances with properties to alter the reproductive capacity of fish. The objective of this study was to evaluate the metabolic modifications (using metabolomics tools) of fish gonads and liver (*Danio rerio*) exposed to effluent from the pulp and paper industry, as well as the embryonic development of embryos of respective fish. The effluent was diluted by a factor of 1/25. The fish (10 males and females) were exposed to the effluent, and males and females were kept separated for 7 days at 28 ºC and a light / dark cycle of 12/12 h in 3.5 L tanks. After the pre-exposure period, males and females were mated (1 couple per Becker containing a net at the bottom) and monitored for 6 days with daily water/pollutant renewal and fed twice a day. The eggs were collected and placed on plates containing the test agent, kept protected from light. The fish were sacrificed for liver and gonads removal, from which a homogenate was prepared. Then, extraction was performed with chloroform/ methanol/water (3: 2: 1). The extract obtained was prepared for gas chromatography. For the exposed fish, there was a significant reduction in egg production, 65%. A high percentage of the eggs presented dark staining, which are not viable. Chromatography analysis revealed significant changes in the amino acid, sterol and fatty acid profile in both tissues, liver and gonads. The results showed a strong impact on the metabolism, egg production and embryo development for the studied fish, which point to the alteration of their reproductive capacity.

WE289

Developing biomarkers of sewage effluent exposure in the freshwater amphipod Gammarus fossarum

D.R. Caputo, University of Portsmouth / Biological Sciences; I. Werner, Ecotox Centre Eawag-EPFL / Department of Anatomy Physiology and Cell Biology; S. Robson, University of Portsmouth / School of Pharmacy & Biomedical Science; A. Ford, University of Portsmouth / Biological Sciences

Pesticides, pharmaceuticals, industrial chemicals and complexing agents coming from sewage effluents, can be detected in the aquatic environment in very low concentrations. They are able to impact ecological communities, causing biological alterations in many species. Although biomarkers in vertebrates have long been applied, attempts to monitor dysfunctions in invertebrates using orthologous genes have produced inconsistent results. Gammarids are a group of amphipods that have been shown as very sensitive to pollution, having been used in various studies for toxicity evaluation of river waters through exposure to many different chemicals. However, to date most studies have been focusing on specific life-cycle stages, potentially missing complex interactions among expressed genes not involved in development. The aim of this study is to provide a set of new transcriptomic and metabolomic markers in *Gammarus fossarum*. After validations in further studies, the new biomarkers found in this project could be used to evaluate the state of aquatic ecological niches and the efficiency of wastewater treatment plants (WWTPs). Amphipods were sampled from a freshwater stream located in Elgg, Switzerland in September 2017, using standard kick-net method. This stream flows through an industrial WWTP, steadily exposing the whole fauna and flora to a range of pollutants. Sampling was performed 50 m downstream of the WWTP and 50 m upstream, as reference site. Five biological replicates and five technical replicates for both males and females, sampled upstream and downstream were used to get 20 samples containing total RNA. A subsample of amphipods was collected for metabolomic analysis and additional samples were fixed to record population markers, such as sex ratio, fecundity rate and potential intersexuality phenotypes. The RNA samples have been sequenced by *Illumina Genome Analyzer*. A differential expression analysis will be conducted to identify significantly different genes between upstream and downstream populations. Subsequently, a comparison between transcriptomic and metabolomic data will be carried out, for a better understanding of the biological functions impaired after amphipods wastewater exposure. A biological pathway analysis will be also performed on the differentially expressed genes, allowing a correlation of the impaired molecular pathways after chronic exposure to water pollutants with the chemical mixture found in the wastewater effluent.

WE290

Optimising the algal toxicity test towards generation of multi-omics data and adverse outcome pathway discovery

S. Schade, Birmingham University / Biosciences; N. Taylor, Cambridge Environmental Assessments (CEA) / Ecotoxicology; J. Zhou, S. He, University of Birmingham / Computer Science; E. Butler, Unilever; G. Hodges, Unilever / Safety and Environmental Assurance Centre SEAC; J. Colbourne, M.R. Viant, University of Birmingham / School of Biosciences

The adverse outcome pathway (AOP) concept represents a framework to organize mechanistic understanding of toxicological interactions by causally linking critical molecular key events (KE) to apical endpoints relevant for chemical risk assessment. Currently, only few methodologies can be considered for an accurate and reliable discovery and quantification of KEs in an exhaustive approach, commonly requiring sustained research effort. In this context, the objective of our presented proof-of-concept study was to showcase the identification and characterisation of molecular KEs from the molecular stress response of *Chlamydomonas reinhardtii* to toxic insult, applying a novel setup of high-throughput genome-wide scaling multi-*omics* technologies.The approach towards achieving this end was a suite of untargeted (direct-infusion mass-spec, DIMS; RNA sequencing) and targeted (LC-MS/MS, -UV, qRT-PCR) metabolomics, lipidomics and transcriptomics technologies. This methodology enabled us to profile the concentration- and time-response profiles of molecular signatures from algae exposed to non-specific mechanism (baseline toxicity) and target-specific mechanism (carotenoid biosynthesis inhibition) toxicants. To enable this work, a rigorously controlled algal culturing and testing system was optimised regarding growth rate, final cell density, pH stability, cell cycle synchronisation, reproducible exposure to volatile chemicals, and rapid quenching and harvesting of biomass for *omics* data collection. Furthermore, a unique multi-phase experimental design was developed for rapid identification (untargeted), characterisation and verification (targeted) of putative KEs over a time-course design. Multi-*omics* data from toxicant-exposed *C. reinhardtii* were collected and initial progress made towards computational analysis, putative KE designation, and targeted verification of identified biomarkers. With this study, a powerful experimental approach for hypothesis-free KE discovery and AOP hypothesis is being developed, employing *omics*-driven algal phenotyping to advance the integration of *omics* data into AOP development and ultimately, to provide mechanism-based support for regulatory decision-making in environmental risk assessment.

WE291

Elucidating interactive toxic effects of copper and lead on marine mussels: molecular to physiological consequences

C. Crowther, V. Sharma, Plymouth University; A. Turner, Plymouth University / Food Safety; A.N. Jha, Plymouth University / Biological Sciences

It is increasingly recognised that anthropogenic contaminants are not isolated in their threats to the aquatic environment. Recently there has been a shift towards measuring the effects of exposure to low-concentrations chemical mixtures under chronic conditions to predict outcomes on the ecosystem. Adopting an integrated approach the aim of this study was to assess the interactive effects of copper (Cu) and lead (Pb) either alone or as a mixture at various levels of biological organisation, ranging from molecular to individual levels. The combination of proteomics, molecular and physiological measures with bioinformatics adopted in this study will allow a model of mixture exposures to be created which can be translated to early warning indicators within the marine environment. *Mytilus galloprovincialis* were exposed to a range of concentrations of Cu (5, 32 µg/L) and Pb (5, 25 µg/L) both individually and in a binary mixture. After a 14 day exposure, a number of physiological and molecular parameters were assessed. This included: measurements of 'clearance rate', acetylcholinesterase activity, inductions of micronuclei (MN), DNA strand breaks (i.e. Comet assay) and measurement of protein profile (i.e. proteomics using LC-MS). The clearance rate shows that the mussels exposed to all treatments containing 32 µg/L of Cu have a significantly decreased feeding capability compared to controls, regardless of singular or binary exposure. This was in contrast to the comet assay results which suggests a synergistic relationship between singular treatments and the highest binary treatment (32, 25 µg/L Cu and Pb, respectively). Mussels exposed to 25 µg/L of lead showed an increase in acetylcholinesterase activity, this treatment was significantly higher than both the highest copper binary mixtures. The analysis of protein profile is in process which should provide the potential functional effects of exposures of these two environmentally relevant metals, either alone or in combination. Furthermore, once this study has determined the chemical interactions between binary metals and the mussel's proteome this could pave the way for further omics being performed and adapted for the ability to create early warning environmental indicators, not only for the environmental health but also for human health.

WE292

The Identification of Toxicological Markers in Adverse Outcome Pathway Discovery in Chlamydomonas reinhardtii

G. Reynolds, Unilever / Safety and Environmental Assurance Centre SEAC; S. Schade, Birmingham University / Biosciences; N. Taylor, Cambridge Environmental Assessments (CEA) / Ecotoxicology; G. Hodges, Unilever / Safety and Environmental Assurance Centre SEAC; E. Butler, Unilever; M.R. Viant, University of Birmingham / School of Biosciences

Current regulatory toxicity testing methods have become unsuitable for the thorough assessment of chemicals for commercial use, as lack of insight into toxicological mechanism prevents accurate predictive risk assessment. Adverse outcome pathways (AOPs) offer a framework for collating mechanistic data from a diverse range of methodologies, including *in silico* and *in vitro* approaches, for use in regulatory decision making. The aim of this work was to develop and test a more comprehensive experimental design, for the targeted characterisation of key events in the toxicological response of *Chlamydomonas reinhardtii* upon herbicide exposure, thereby contributing to the development of a quantitative AOP. Here we present how an initial hypothesis for an AOP was created from available literary evidence, with focus on 'omics and multiple-endpoint assay data, for the selected herbicide, norflurazon. This hypothesised AOP allowed development of targeted assays for investigation of predicted key events in a time- and concentration-response methodology. LC-UV was used to monitor suspected toxicological markers of the carotenoid biosynthesis pathway (phytoene, phytofluene, b-carotene). qPCR was used to identify differential mRNA expression of chloroplast-specific thioredoxin PRX1, and a lipid peroxidation assay was applied for determining downstream effects of non-specific oxidative stress. A concentration- and time-dependent response in phytoene accumulation was observed, whilst concentration dependent b-carotene depletion was shown at later time-points (24 hours post exposure). Increased PRX1 expression was identified within an hour of exposure, whilst lipid peroxidation occurred between 4 and 24 hours post-exposure. Significant ($p = < 0.01$) effects on cell number, an adverse outcome, were observed at 2000µg/L after 24 hours. This study highlighted the necessity for use of synchronous algal cultures for accurately understanding mechanism, as this would enable more accurate determination of time- and concentration- responses due to diurnal algal life cycles. Ultimately, this work has shown proof-of-concept and laid the foundation for development of a quantitative AOP for phytoene desaturase inhibition leading to growth inhibition and population decline.

WE293
Effects of water-borne benzo[a]pyrene on early-life stages of the fathead minnow (Pimephales promelas)
M.T. Schmitz, RWTH Aachen University; M. Brinkmann, University of Saskatchewan / School of Environment and Sustainability and Toxicology Centre; N. Apenova, C. Grimard, University of Saskatchewan / Toxicology Centre; A. Alcaraz, University of Saskatchewan - Toxicology Centre / Toxicology Centre; D. Green, University of Saskatchewan - Toxicology Centre / Toxicology; K. Bluhm, University of Saskatchewan / School of Environment and Sustainability; T. Lane, University of Saskatchewan; N. Baldwin, J. Taghavimehr, A. Masse, University of Saskatchewan / Toxicology Centre; D. Crump, Environment and Climate Change Canada / National Wildlife Research Centre; N. Basu, McGill University / Faculty of Agricultural and Environmental Sciences; H. Hollert, RWTH Aachen University / Institute for Environmental Research; N. Hogan, University of Saskatchewan / Toxicology Centre and Department of Animal and Poultry Science, College of Agriculture and Bioresources; M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre
Polycyclic aromatic hydrocarbons (PAHs) are a class of ubiquitously distributed environmental pollutants that mainly originate from petrogenic and pyrogenic sources such as combustion of fossil fuels and other organic material. Various PAHs, including benzo[*a*]pyrene (BaP), have been demonstrated to cause a wide range of effects in exposed wildlife, including alterations of immune responses, impaired development and reproduction, as well as mutagenesis and carcinogenesis. Most studies to date, however, have used comparably high exposure concentrations, dietary routes of exposure or intraperitoneal injection to administer BaP, and knowledge of low-dose effects at concentrations around water solubility (approx. 4 µg/L) is generally limited. This route of exposure, however, must be considered highly relevant in light of the distribution of PAHs even into remote aquatic systems. To bridge this knowledge gap, early-life stages of the fathead minnow (*Pimephales promelas*) will be exposed to waterborne BaP as a model compound to characterize toxicity pathways that drive the sensitivity of early life-stage fish to PAHs. Molecular responses at the whole transcriptome, proteome and metabolome level will be investigated at the swim-up stage, and quantitatively correlated with effects on apical (growth, survival, development), histopathological, and biochemical endpoints 28 d post-hatch. The data generated within this experiment will help to better understand the relevance of aqueous exposure to BaP specifically, and PAHs in general, and provide important insights into the relevance of molecular responses in early-life stages as early-warning biomarkers for apical outcomes in juvenile and/or adult fish. *This study is part of the EcoToxChip project (@ecotoxchip).*

WE294
SETAC OMICS Interest Group
B. Campos, Unilever R&D / Environmental Chemistry

Epigenetic and evolutionary toxicology: from mechanisms to risk assessment (P)

WE296
Epigenetic effects in Daphnia magna by characterizing quantified abundance of global methylation, gene expression and histone modifications
J. Thaulow, NIVA - Norwegian Institute for Water Research / Freshwater Ecology; L.C. Lindeman, Norwegian University of Life Sciences / Dept. for Basic Science and Aquatic Medicine (BasAm); Y. Song, NIVA Norwegian Institute for Water Research / Ecotoxicology and Risk Management; J. Kamstra, NMBU / BaSam; L. Xie, NIVA - Norwegian Institute for Water Research; J. Asselman, Ghent University / Laboratory for Environmental Toxicology and Aquatic Ecology GhEnToxLab unit; P. Aleström, Norwegian University of Life Sciences; K. Tollefsen, NIVA / Ecotoxicology and Risk Assessment
Daphnia magna is used in toxicology and environmental science as a monitor for ecosystem health. Epigenetic analysis is enabled by the genome of the closely related *D. pulex*. Epigenetic mechanisms allow gene regulation in a developmental context and as response to environmental stimuli. The best studied epigenetic mechanisms are methylation forms on cytosines in a CpG context and post translational modifications (PTMs) on histone proteins attached to DNA. The global abundance or change of 5-methyl-cytosine (5mC) may indicate epigenetic reactions to environmental stimuli, since these methylation forms facilitate in the regulation of gene expression the change in expression can detected and compared. The elucidating histone PTM code give insight on how these proteins modifications regulate gene expression and crosstalk with each other and with DNA methylation. Chromatin immunoprecipitation (ChIP) is the standard assay of choice for analyzing the genomic localization of histone modifications. Exposure to the well-known epigenetic modulator, the DNA methylation inhibiting agent 5-Azacytidine, resulted in a global reduction of DNA methylation in *Daphnia magna* over one generation, while H3K4me3 and H3K27me3 remains unchanged on the investigated loci. The unchanged response in ChIP was contradictive to significant gene expression responses and to what was expected of this epigenetic modulator. The present study therefore demonstrates differentiated response of LC-MS/MS, ChIP-PCR and gene expression to 5-Azacytidine exposure when charactering epigenetic stress response in *D. magna*. **Acknowledgements**: funding from the Norwegian Research Council (NRC) project 223268 (CERAD).

WE297
Role of microRNAs in the response of the European eel Anguilla anguilla to water pollution
A. Bertucci, F. Pierron, Université de Bordeaux / UMR EPOC CNRS 5805; T. Ye, T. Christelle, IGBMC / CNRS UMR 7104 - Inserm U 964; P. Gonzalez, University of Bordeaux / UMR EPOC CNRS 5805; M. Baudrimont, Université de Bordeaux / UMR EPOC CNRS 5805
MicroRNAs (miRNAs) are a class of small non-coding RNA. These 20-24 nucleotides-long sequences associate with the 3'-untranslated region (3'-UTR) of target messenger RNAs (mRNAs), and post-transcriptionally regulate the expression of numerous genes by mediating translational repression or mRNA degradation. In mammals, more than 50% of mRNAs are predicted to be the subject of miRNA-mediated control. One miRNA may regulate hundreds of target mRNAs, and one mRNA may contain multiple binding sites for multiples miRNAs, thus resulting in a complex regulatory network. Although miRNAs are involved in regulation of almost all cellular processes, such as development, growth, apoptosis, immunity and maintenance of tissue-specific function, mechanistic aspects of this regulation are not fully understood. In Human, the aberrant expression of miRNAs has been linked to various diseases and toxic environmental factors such as nanoparticles, organic pollutants and metals can alter miRNA expression. The first aim of the present study was to identify miRNAs in the European eel *Anguilla anguilla* by using next generation sequencing. We identified 230 evolutionary conserved and 145 novel miRNAs. Amongst these 375 miRNAs, 242 were predicted to be able to interact with 3,637 transcripts in the previously described *A. Anguilla*'s transcriptome. No gene ontology, nor metabolic pathway, was significantly enriched in the list of target genes, suggesting that miRNAs might affect any biological process. Our second aim was to compare the differential expression of miRNAs between a pristine site located in Arcachon bay and a polluted site in the Gironde estuary (France). Nineteen miRNAs were up-regulated and 22 were down-regulated depending on the pollution profile. This approach may provide innovative molecular markers and the comparison of miRNAs regulation with classical transcriptomic studies are likely to reveal new aspects of the toxicology mechanisms involved between environmental factors and diseases aetiology.

WE298
Exposure to copper during embryogenesis caused temporary increased tolerance in two subsequent generations in the three-spined stickleback (Gasterosteus aculeatus)
L.V. Laing, University of Exeter / Biological Sciences; H. Littler, J. Fitzgerald, University of Exeter / Biosciences College of Life and Environmental Sciences; N. Bury, Kings College London; R. van Aerle, Centre for Environment Fisheries and Aquaculture Science / Biosciences College of Life and Environmental Sciences; R. Wilson, University of Exeter / Biosciences; J. Mill, University of Exeter / Exeter Medical School; E.M. Santos, University of Exeter / Biosciences College of Life and Environmental Sciences
The sustainability of fish populations in our increasingly polluted environment is critically dependent on their ability to adapt via (epi)genetic mechanisms. Copper is an essential element but when present at high concentrations in the water it can become toxic to aquatic organisms. Recent studies in the UK suggest that copper is the most significant metal pollutant threatening fish in UK freshwaters. We conducted a series of copper exposures in stickleback to investigate whether prior exposure can result in altered susceptibility in subsequent generations. Stickleback embryos were exposed to 0.015mg/L copper during early life (0-9dpf), causing ~1.2% mortality, ensuring that selection for a tolerant genotype did not occur. They were then kept under control conditions until sexual maturity. Copper pre-exposed fish were shown to have a significantly higher basal copper tissue burden as adults; and upon re-exposure, they showed a differential response compared to control fish. Mortality curves on F1 embryos revealed that embryos originating from parents who were exposed to copper during embryogenesis were significantly more tolerant to copper when compared to controls. This copper tolerance was shown to be still present in the F2 generation, but not the F3 generation. Our data supports the hypothesis that exposure to low levels of copper during early life has the potential to reduce the susceptibility of a vertebrate model across generations. We hypothesise that the multigenerational tolerant phenotype observed was caused by parental effects, owing to altered copper handling and increased copper tissue burden in F0 adults, resulting in secondary exposure of F1 embryos and their germ cells which gave rise to the F2 generation.

Emergence and multidimensional interactions of engineered nanoparticles in toxicology (P)

WE299
Do global warming increase bioaccumulation of copper nanoparticle in

tilapia?

J. kuo, Kaohsiung Medical University; L. Li, W. Chen, Department of Biomedical Science and Environmental Biology, Kaohsiung Medical University, Kaohsiung

Abstract Nanomaterial technology was well developed in recent years, and it lead to the nanomaterials accumulation in the aquatic organism. Otherwise, increased atmospheric carbon dioxide causing the phenomenon of global warming. However, less is known about the effect of warming whether increase the bioaccumulation of copper nanoparticles in freshwater fish. The purpose of this study is to assess whether warming synergistically increase the bioaccumulation of copper nanoparticles in tilapia (*Oreochromis niloticus*). Tilapia were randomly exposure to 25 nm of copper nanoparticle (0.3 mg/L) under different temperature (26, 28 and 30?) for periods of 7 days for uptake and 7 days for depuration, to analyze the accumulation of copper nanoparticle on muscle. Results showed that the copper accumulation of muscle in the high temperature group was higher than that of control group on day 7 of uptake phase, but there were not significant difference. For one day of depuration phase, the copper accumulation of the 30? group was significantly higher than of 26 and 28? groups (p< 0.01). However, they are similar accumulation concentration in the end of depuration period. This study concluded that global warming could increase bioaccumulation of copper nanoparticle in tilapia.

WE300

Environmental mixtures of nanomaterials and chemicals: proposal for a consistent nomenclature of mixture effects in environmental organisms

D. Kühnel, Helmholtz-Centre for Environmental Research / Bioanalytical Ecotoxicology; R. Altenburger, UFC Centre for Environmental Research / Department Bioanalytical Ecotoxicology; S. Naasz, UFZ Helmholtz Centre for Environmental Research / Bioanalytical Ecotoxicology

A review of the existing literature on mixture effects of nanomaterials (NM) and chemicals in environmental organisms was conducted in order to evaluate the current state of knowledge. More than 120 studies were assessed to explore the relationship between changes in contaminant and NM uptake, bioconcentration, and toxicity. The specific aim of the review was to describe more specifically the interactions that have been observed and to classify the most common mechanisms. As the literature evaluation demonstrated, the existing evidence for interference of NM-chemical mixture exposure with uptake and toxicity is rather diverse. Based on the observations made, we could discriminate at least 7 different categories to capture the evidence ranging from no changes in uptake and toxicity to an increase in uptake and toxicity upon mixture exposure to chemicals and NM: (1) increase in accumulation and toxicity (2) increase in accumulation and no change in toxicity (3) increase in accumulation and decrease in toxicity (4) no change in accumulation and toxicity (5) no change in accumulation and decrease in toxicity (6) decrease in accumulation and toxicity (7) decrease in accumulation and increase in toxicity However, we assume that these observations were caused by different underlying mechanisms and processes, hence we developed a process oriented, tiered approach considering (1) Adsorption / Interaction between NM and chemicals, (2) Uptake of NM by organisms, (3) Desorption of chemical inside / outside the organism, and (4) Toxicity. By sorting the individual datasets from the studies according to these processes, 6 groups were build. Based on these 6 groups, a consistent nomenclature is proposed: (1) Trojan-horse (+) (2) Trojan-horse (-) (3) Surface enrichment (4) Retention (5) Inertism (6) Coalism The poster will present in detail the characteristics of the 6 groups and the criteria that were used for the assignment of datasets. All in all, this in-depth analysis of mixture datasets underline the importance of a process-oriented approach in the elucidation of specific mixture effects. The tiered approach results in a consistent terminology to unambiguously describe the different mechanisms of mixture effects that may occur in environmental organisms. *Acknowledgement* - DK was partially funded by the German Federal Ministry for Education and Research (BMBF) in the frame of the DaNa2.0 project (Data and knowledge on nanomaterials), grant no. 03X0131.

WE301

Investigating the Trojan horse effect of nanoparticles on an aquatic community - An outdoor mesocosm study

T. Strauss, Research Institute gaiac / gaiac - Research Institute for Ecosystem Analysis and Assessment; S. Classen, Research Institute gaiac; T. Knautz, M. Hammers-Wirtz, Research Institute gaiac / gaiac - Research Institute for Ecosystem Analysis and Assessment

Carbon based manufactured nanomaterials (C-MNMs) are promising materials in nanotechnology. Although both fullerenes and carbon nanotubes have been detected in aquatic organisms, there is a lack of data on their bioaccumulation, potential toxic effects, transfer in the food chain, and interaction with other anthropogenic pollutants. Most effect studies performed until now dealt with waterborne exposure of single species for short time periods in the laboratory. Here, we present a long-term experiment under environmentally relevant conditions. In particular, the Trojan horse effect has been investigated in this study, in order to obtain more data on the interaction between nanoparticles, other pollutants and biota. In principle, pollutants can become more bioavailable by adsorption to carbon-based nanomaterials. In addition, a spatial transfer of contaminated nanoparticles from the water phase to the sediment could increase the exposure to benthic macroinvertebrates but might also reduce the effect on the planktonic organisms. An outdoor freshwater mesocosm study was conducted with C60 fullerenes and the biocide triclocarban (TCC) using twelve outdoor ponds with a water volume of 3 m^3. In addition to uncontaminated controls, both substances were tested alone and in combination. The aim of this mesocosm study is to investigate long-term effects of C60 fullerenes on the community level and to assess their potential to affect the toxicity of TCC. In this outdoor mesocosm study direct and indirect effects on single species as well as on community level endpoints like diversity were evaluated. The taxonomic groups of interest are phytoplankton, zooplankton (e.g. *Daphnia* species), and macroinvertebrate species (e.g. chironomids, mayflies, oligochaetes, leeches). Different sampling techniques were used in order to include macroinvertebrates living on and within the sediment as well as hatching insects. In this presentation the results of the mesocosm study will be presented. This work has been supported by the German Federal Ministry of Education and Research (BMBF) as part of the NANO-transfer project.

WE302

Nano silver based products and environmental challenges: toxicity and accumulation in a marine sentinel species

G. Liberatori, University of Siena / Department of Physical, Earth and Environmental Sciences; A. Ale, Inali-Conicet; C. Jimena, Instituto Nacional de Limnología (CONICET-UNL); S. Ancora, University of Siena / Physical sciences, Earth and environment; N. Bianchi, University of Siena / Department of Physical, Earth and Environmental Sciences; I. Corsi, University of Siena / Physical, Earth and Environmental Sciences

The use of nanotechnology-based consumer products is constantly increasing worldwide and their release into the environment is thus expected, especially in aquatic ecosystems, which are considered the ultimate sink. The use of nano silver (AgNP) based products as antimicrobial agents is undergoing a rapid increase in terms of production due to its biocidal properties. Here we studied the impact of AgNP-based commercial products named NanArgen (Nanotek S.A.) on a common marine bivalve sentinel species as the *Mytilus galloprovincialis* in terms of biological responses and Ag accumulation. Animals were *in vivo* exposed for 96h to NanArgen product containing 20-40nm AgNP, according to the manufacturer, at two different concentration (1 μg and 10 μg/L) using natural sea water (NSW) as exposure media. Lysosomal membranes stability (NRRT) and micronuclei frequency (MN) were measured in mussel's hemocytes. Catalase (CAT) and glutathione-s-transferase (GST) activities were measured in digestive gland as well as the content of malondialdehyde (MDA) and metallothioneins (MT). Effect on multi xenobiotic resistance (MXR) phenotype was assessed by measuring efflux ABC transporters also *in vitro* using mussel's gills. Total Ag was analyzed in exposure waters after 24h and in mussel's soft tissue after 96h. DLS analysis as well as TEM have been also performed on NanArgen formulation in NSW. A significant increase in lysosomal destabilization and MN frequency was observed in hemocytes of mussels exposed to both concentrations of NanArgen. Furthermore, MT content was significantly higher in digestive gland of mussels exposed only to 10 μg/L while oxidative stress parameters did not show any change compared to controls. A slight negative effect on MXR functionality is observed *in vitro* but not *in vivo*. Chemical analysis confirm Ag exposure and showed a dose-dependent increase of Ag in exposed mussels. In conclusion we can state that this nano silver-based commercial product can induce toxicity even at low concentrations and in short-term exposure scenarios. The observed toxicity of NanArgen underlines the need to further test commercial formulations of nanotechnology-based consumer products instead of bare nanoparticles in order to properly address any risk associated to their use and release into aquatic environment and in non-target aquatic species.

WE303

Effect of gold nanoparticles on feeding, growth and enzymes activity of amphibians

B. Rabelo Costa, Department of Biology & CESAM - University of Aveiro / Biology; C. Quintaneiro, Department of Biology & CESAM - University of Aveiro; A.M. Soares, University of Aveiro / department of Biology & CESAM; I. Lopes, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro

The gold nanoparticles are widely used in medical therapy and cosmetics. However, the study of this kind of nanoparticles represent only 3% of the relative abundance of papers focusing on engineered nanoparticles, including both freshwater and saltwater species. The knowledge about the potential ecotoxicity of these nanoparticles is essential before their use by society at a large scale, since they will ultimately be released in to the environment. Thus, the aim of this study was to determine the effect of gold nanorods (Au-NR; 45nm) in the feeding rate, growth and enzymatic activity of tadpoles of the amphibian species Xenopus laevis. A significant decrease in feeding rate and snout to vent length (SVL) of tadpoles was observed at concentration equal or higher than 0.004 μg/ml. For biomass a significant effect was observed at concentration 0.007 μg/ml or higher, though, a decrease in weight gain rate was observed at a lower concentration (0.004 μg/ml). At the biochemical level, the activity of enzyme lactate dehydrogenase (LDH) increased at 0.002 μg/ml of Au- NR, that of catalase (CAT) was significantly reduced at 0.005 μg/ml or higher, and glutathione S-transferase (GST) and acethylcholinesterase activity (AChE) was significantly higher, relatively to the control, in the two highest tested concentrations 0.007 and 0.01 μg/ml. The observed reduction in SVL, added to decreased feeding rates, in tadpoles exposed to Au-NR, are important effects that may compromise the fitness of the organisms, since they may cause a delay in the metamorphosis, leading to a longer exposure period of tadpoles to the chemical and to an increase in the time to reach adult and reproductive stage. The higher activity of LDH, at 0.002 µg/ml, may suggest that tadpoles activate first (at low concentrations of Au-NR) a detoxification pathway involving anaerobic metabolism. Furthermore, an excess of reactive oxygen species (ROS) may have led to the inactivation of catalase and the induction of AChE related with the mechanisms of cell apoptosis. Though, the induction of GST at the two highest Au-NP concentrations, suggest that the cells are inactivating the Au-NP by its conjugation with reduced glutathione. The results obtained in the present work indicates that Au-NR may induce several sublethal effects in tadpoles of *X. laevis*, that compromise their fitness. Furthermore, since these effects occur at very low concentrations (as low as 0.002µg/ml) it should be classified as "extremely toxic" (EC20 < 0.1 µg/mL; CEC, 1996), suggesting a high environmental risk.

WE304

Interaction of the biocide triclocarban and weathered multiwalled carbon nanotubes (wMWCNT) in freshwater algae: chronic effects & bioaccumulation

I. Politowski, M.P. Hennig, H. Hollert, RWTH Aachen University / Institute for Environmental Research; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics

The disposal of various plastic nanocomposites containing multiwalled carbon nanotubes (MWCNT) will occur more frequently in the next decades. Thus, MWCNT release into the aquatic environment due to degradation of the polymers is inevitable. Changes in their properties might happen by several abiotic influences, like weathering by sunlight radiation. MWCNT undergo thereby structural changes compared to their pristine state, like formation of hydroxyl and carboxyl groups on the surface of the MWCNT. As a consequence, weathered MWCNT (wMWCNT) have an altered agglomeration- and sorption behavior to other contaminants like triclocarban (TCC). This might lead to a different environmental fate of both the wMWCNT and the contaminants in aquatic ecosystems and eventually an enhanced chronic or mixture effect on organisms like freshwater algae. In our studies we examine the chronic effects of wMWCNT and the 'Trojan horse' effects of TCC in presence and absence of wMWCNT on *Pseudokirchneriella subcapitata* and *Chlamydomonas reinhardtii*. Growth inhibition tests were performed according to OECD test guideline 201. In a first part of experiments, the growth inhibition of both species was tested in the range of 10 – 60 µg/L for TCC and 0.1 – 16.0 mg/L for wMWCNT. The mixture toxicity of a TCC test series (10 – 60 µg/L) and 100 µg/L wMWCNT was additionally investigated on *P. subcapitata*. A second series of experiments was carried out by adding the highest TCC concentration (60 µg/L) to variable wMWCNT concentrations to figure out, which wMWCNT amounts are necessary to reduce the toxicity of TCC. We determined a concentration dependent growth inhibition of *P. subcapitata* for TCC and TCC + 100 µg wMWCNT/L with an EC_{50} of 37 and 36 µg TCC/L, respectively. This amount of wMWCNT appears to be not sufficient to adsorb the entire free TCC from the water phase, which eventually leads to very similar EC_{50} values in both scenarios. Only in a second experiment it could be shown, that wMWCNT amounts > 1 mg/L reduce algae growth inhibition completely caused by 60 µg TCC/L. Regarding to mixture effects of TCC and wMWCNTs to algae, bioaccumulation of wMWCNTs by freshwater algae needs to be investigated, especially in respect to long term incubation times and low wMWCNT amounts. **Acknowledgements** The work is supported by the European Project NANO-Transfer that receives funding from the Bundesministerium für Bildung und Forschung (BMBF) under agreement with the FP7 ERA-NET SIINN.

WE305

Comparative assessment of the interactive effects of Carbon-based nanomaterials and Benzo(α)pyrene on zebrafish embryos

C. Della Torre, State University of Milano / Biosciences; A. Ghilardi, S. Magni, University of Milan; N. Santo, University of Milan / Bioscieces; D. Maggioni, University of Milan; C. Landi, University of Siena; M. Parolini, University of Milan / Department of Environmental Science and Policy; L. Madaschi, University of Milan; C.C. Parenti, M. Ascagni, University of Milan / Biosciences; L. Bini, University of Siena; L. Del Giacco, University of Milan; A. Binelli, University of Milan / Department of Biosciences

This study aimed to assess the ecotoxicological consequences related to the interaction of Benzo(α)pyrene (B(α)P) with two CNMs with different physico-chemical properties, namely carbon nanopowder (CNPW) and fullerene (C_{60}), on zebrafish embryos. To this aim CNMs were contaminated with B(α)P and the effective sorption of the hydrocarbon on CNMs was quantified. A thorough evaluation of chemico-physical interactions between the two CNMs and B(α)P has been performed. Embryos were exposed to CNPW, C_{60} and B(α)P alone and their combination. The uptake of CNMs and B(α)P and their localization in embryos were assessed by immunofluorescence and electron microscopy. To evaluate the toxic effects due to interaction of B(α)P with CNMs, a set of biomarkers of genotoxicity and oxidative stress was applied. Proteomics analysis allowed also the identification of molecular events involved in the responses to pollutants alone and in co-exposure. Overall results showed that the different physico-chemical properties of the two CNMs influenced their interactions with B(α)P and generated distinct toxic effects. Indeed the adsorption on CNPW modified the accumulation of B(α)P, which followed the distribution of the physical pollutant instead of its natural bioaccumulation. On the contrary the co-exposure with C_{60} did not affect the uptake/distribution of B(α)P. Instead, C_{60} doped with B(α)P is more prone to sedimentation and less bioavailable for the embryos compared to C_{60} alone. The integrated results from biomarkers and proteomics showed that different stress response pathways were induced by the pollutants alone respect to their combination. The CNPW doped with B(α)P mainly mirrored the effects shown by the physical contaminant rather than by the hydrocarbon, while C_{60} doped with B(α)P seems to induce a cellular response similar to B(α)P alone. The study highlighted that in the aquatic ecosystems complex interactions are established between pollutants and CNMs which could elicit unexpected ecotoxicological effects.

WE306

IN VITRO TOXICITY OF MODEL ZnO NANOPARTICLES ON HEMOCYTES OF MUSSEL *Mytilus galloprovincialis*

I. Efthimiou, University of Patras / Department of Environmental and Natural resources Management; N. Anastasi-Papathanasi, University of Patras / Department of Biology; E. Mouzourakis, Y. Georgiou, University of Ioannina / Department of Physics; S. Dailianis, University of Patras / Department of Biology; Y. Deligiannakis, University of Ioannina / Department of Physics; D. Vlastos, University of Patras / Department of Environmental and Natural resources Management

Zinc oxide nanoparticles (ZnO NPs, size 58 nm, as calculated using X-Ray diffraction data) were manufactured through Flame Spray Pyrolysis, and their effects were subsequently investigated on hemocytes of mussels *Mytilus galloprovincialis*. Following the collection and preparation of cell suspensions, mussel hemocytes were treated for 1 h with different concentrations of ZnO NPs (5, 10, 25 and 50 µg mL^{-1}, from a stock solution of ZnO NPs in Hepes/2dH_2O, in 1:1 ratio, dispersed with a probe sonicator), as well as $ZnCl_2$ (10 and 25 µg mL^{-1}: positive control, from a stock solution of $ZnCl_2$ in 2dH_2O, in 1:1 ratio). Afterwards, stress indices such as (a) cell viability (in terms of Neutral Red Retention Time/NRRT assay) (b) the generation of superoxide radicals ($O_2^{\cdot -}$), using Nitro blue tetrazolium/NBT, (c) the production of nitrogen oxides (NO, in terms of nitrites), and (d) lipid peroxidation (in terms of malondialdehyde/MDA equivalents) were measured. The results demonstrated a significant increase of cell death after treatment with ZnO NPs at concentrations higher than 5 µg mL^{-1}, with maximum values (>50%) of cell death after exposure to ZnO NPs 50 µg mL^{-1}. Furthermore, hemocytes treated with sub-lethal concentrations of ZnO NPs (5-25 µg mL^{-1}), showed a significant increase of $O_2^{\cdot -}$, NO and MDA, compared to those values observed in control cells in each case. Finally, the results of the exposure to ZnO NPs were compared with the respective results after exposure to $ZnCl_2$, showing a similar pattern. Those effects of ZnO NPs on mussel hemocytes confirm the cytotoxic and oxidative potential of well-promised nanomaterials, such as ZnO NPs, widely used in a variety of new cutting-edge applications.

WE307

Toxico-transcriptomics as tool to identify nano-specifc toxicity profiles

M. Burkard, Eawag Aquatic Water Science / Southern Ocean Persistent Organoic Pollution Program; A. Betz, Eawag / UTOX; K. Schirmer, Eawag / Environmental Toxicology; A. Zupanic, Eawag Swiss Federal Institute of Aquatic Science and Technology / Environmental Toxicology

The use of omics is rapidly increasing in the field of nano-ecotoxicology; an increasing number of studies are aiming to investigate the effects and mode of action of engineered nanomaterials (ENM) in this way. However, a systematic synthesis of the outcome of these studies in order to identify common responses between ENM and organism groups has not yet been performed. We therefore established a computational analysis pipeline with the aim to re-analyze relevant transcriptomic datasets in a consistent manner. The pipeline allows a re-mapping of array probe sequences, followed by established statistical analysis and thus improves data set comparability. Differentially expressed genes (DEGs) are determined by comparison between treatment and untreated samples (pFDR

WE308

Zinc toxicity to A549 cells and Daphnia magna changes after incubation with iron oxide nanoparticles

J. Cabellos, LEITAT Technological Centre; V. Gonzalez, Leitat Technological Center; M. Almira-Casellas, Leitat Technological Center / HEHS; M. Diez-Ortiz, G. Janer, Leitat Technological Center

The use of iron oxide nanoparticles (IONPs) as an environment remediation tool is based on their ability to adsorb and immobilize metals and decrease their bioavailability. However, the adsorption of metal contaminants by nanoparticles can also potentially increase the toxicity of either NPs or the metals, for example due the *Trojan horse effect*. In this study, we evaluated the acute effect of zinc (Zn) as zinc sulfate heptahydrate ($ZnSO_4 \cdot 7H_2O$) after an incubation period with a fixed concentration of humic acid (ha) coated IONPs (ha-IONPs), on the *in vitro* toxicity to human A549 cells and on the toxicity to *Daphnia magna* as a model freshwater invertebrate species. Non-toxic concentrations of ha-IONPs were selected for the assays taking also into account the predicted adsorption of Zn. The ha-IONPs concentrations used were 0.45g/L and 0.52 g/L for the A549 and the Daphnia magna experiments, respectively. In A549 cells, the incubation of Zn with ha-IONPs did not change the Zn effects on cell viability after 24h in terms of IC_{50} (0.006 g/L vs. 0.010 g/L with and without ha-IONPs, respectively). However, the shape of the dose-response curve became shallower (e.g., the IC_{90} for Zn was 0.070 g/L and 0.019 g/L with and without ha-IONPs, respectively). This indicates a potential protective effect of IONPs at high metal concentrations and a synergistic effect at low metal concentrations. These experiments were also conducted in the presence of serum proteins, and despite the toxicity of Zn decreased, the same effect of co-incubation with ha-IONPs was observed. Optical microscope images showed that ha-IONP aggregates were uptaken by the cells during the experiments. Therefore, even if adsorbed on ha-IONPs, a fraction of Zn would also reach intracellular compartments. Differences in the relative uptake of free vs. ha-IONP adsorbed zinc as well as intracellular bioavailability of Zn in these two forms would be explaining the changes in the dose-response curve that were observed. Acute studies (up to 48-hours) with *Daphnia magna* showed a protective effect of the ha-IONPs on the toxicity of Zn. The EC_{50} value for Zn increased from 0.23 mg Zn/L to 11.30 mg Zn/L in presence of ha-IONP. According to DLS data, the adsorption of Zn to NPs decreased their stability and subsequently increased their co-precipitation in the exposure media. This settling process would decrease the bioavailable zinc concentration in the exposure medium and therefore its toxicity in *Daphnia magna*.

WE309

Internalization of graphene-related nanomaterials in fish cell lines

J. Kalman, Instituto Nacional de Investigación y Tecnología Agraria y Alimentaria / Environment; C. Merino, Grupo Antolin Ingenieria SA; M. Fernandez-Cruz, J. Navas, INIA - National Institute for Agricultural and Food Research and Technology / Environment

The H2020 NanoReg2 project aims to develop and implement grouping and Safe by Design strategies for nanomaterials (NMs). For this purpose, toxicity of selected carbon-based NMs have been assessed in order to refine Safe by Design approaches considering three pillars: safer product, safer use, and safer production process. Graphene-related nanomaterials (GRMs) are among the newest and most important NMs. Their extraordinary physicochemical properties have attracted great interest in most areas of science and industry. Nevertheless, the incorporation of these NMs into products inevitably leads to their release into the aquatic environments. In a previous study we assessed the cytotoxicity of tubular-shaped carbon nanofibers (CNFs) and graphene oxide (GO) on fish hepatocytes (derived from topminnow fish, *Poeciliopsis lucida*) and macrophages (derived from carp leukocytes, *Cyprinus carpio*). In general, the observed IC50 values after 72h exposure were higher than 100 µg/ml with some exceptions in the case of CLC cells. In the present work we focussed on the uptake and intracellular fate of these NMs. Cells were exposed to three different concentrations (non-toxic, low toxic and relatively toxic) of each NM for 72 h. Transmission electron microscopy was used to investigate possible internalization and intracellular fate of these NMs in hepatocytes and macrophages. All GRMs were visualized in both cells even at the lowest exposure concentrations. Carbon nanofibers were taken up into vesicles of hepatocyte cells in a size-independent manner, whereas in macrophages, longer CNFs were encountered free in the cytoplasm and only the shorter CNFs were localized in membrane-bound compartments. GO sheets were present within vesicles as well as free in the cytoplasm of both cell types. Understanding the behaviour of these NMs in living systems aid in designing safer materials for the environment. This research is supported by the EU's Horizon 2020 research and innovation programmes (NanoReg2, Grant Agreement nº 646221 and MSCA-IF-2016, Grant Agreement nº 746876).

WE310

Molecular mechanism and physicochemical properties of Cadmium-TiO2 nanoparticle mixtures when co-exposed to the nematode Caenorhabditis elegans

L. Kleene, Hamburg University of Applied Sciences (HAW) / Life Sciences; A. Hursthouse, University of the West of Scotland / School of Science; S. Heise, Hamburg University of Applied Sciences / Life Sciences

The number of engineered nanomaterials (ENM) is rising continuously in consumer products and industrial fields. Therefore, knowledge about their ecotoxicity in aquatic and soil systems is very important but rare. Nanoscale titanium dioxide ($nTiO_2$) is probably among the most relevant ENMs with a projected accumulation rate in European river sediments of 1.4 $mg*kg^{-1}*yr^{-1}$ (Gottschalk et al., 2009). Investigations of Angelstorf *et al.* (2014) have shown that $nTiO_2$ is far more toxic to the nematode *Caenorhabditis elegans* than bulk TiO_2, especially under simulated solar radiation (SSR), probably a consequence of its photocatalytic property. Further experiments by Samet (2017) focused on the interaction of $nTiO_2$ with cadmium (Cd), another environmental contaminant. *C. elegans* was exposed to $nTiO_2$ (P25, primary particle size of 21 nm) and Cd in single and co-exposure for 72 h under dark conditions and SSR. Choosing growth and reproduction as toxic endpoints, co-exposure with 40 $mg*L^{-1}$ $nTiO_2$ and 50 $µg*L^{-1}$ Cd under SSR led to a synergistic inhibitory effect of 80 % of reproduction, twice as high compared to $nTiO_2$ alone. As Cd is known to induce intracellular calcium signaling as part of protective cell processes (Thévenod, 2009), in the study presented here, the effect of the mixture on intracellular calcium release will be investigated applying the following methods: 1) The molecular mechanism of $nTiO_2$ and Cd will be investigated with NS8593, a known human TRPM7 ion channel blocker. Because of high reproduction inhibition, the TRPM-like channel gene *gon-2* could interact with the mixture. *Gon-2* is responsible for gonadal cell division in *C. elegans*. If Cd is a Ca-channel-blocker, the combination of $nTiO_2$ and NS8593 should show the same effects under SSR. 2) The mode of action of $nTiO_2$-Cd-agglomerates is still not identified. They could interact if Cd is bound to $nTiO_2$ or if Cd and $nTiO_2$ are in close proximity. The impact of $nTiO_2$-Cd-agglomerates will be examined using calcium as a potential competitive ligand. 3) The photocatalytic activity of nTiO2 could damage cell membranes under SSR and Cd could enter the cell. Measurements of membrane integrity with propidiumiodid and hexokinase will be tested. First results will be presented. Angelstorf et. al., 2014. Environ. Toxicol. Chem., 33, 2288-2296. Gottschalk et. al., 2009. Environ. Sci. Technol., 43, 9216-9222. Samet, Abstract SETAC Brussels 2017. Thévenod 2009. Toxicol. Appl. Pharmacol., 238, 221-39.

WE311

Influence of temperature and salinity on toxicity of zinc oxide nanoparticle on the marine copepod Tigriopus japonicus

W. Lai, The University of Hong Kong; M.M. Yung, The University of Hong Kong / The Swire Institute of Marine Science and School of Biological Sciences; K.M. Leung, The University of Hong Kong / The Swire Institute of Marine Science

Zinc oxide nanoparticles (ZnO-NP) are ranked as the 5th most produced and the 7th most prevalent nanomaterials (NMs) in commercial products. About 93% of ZnO-NP-containing products are paints, cleaning and personal-care products, from which ZnO-NP can be easily leached. Annually, around 250 tonnes of ZnO-NP were estimated to be released from sunscreens alone into the marine environment. However, there are no comprehensive regulations of NMs, including ZnO-NP, in any countries due to the diverse physicochemical properties of NMs and their complicated interactions with the environment. In the marine environment, ZnO-NPs are exposed to various environmental factors, such as temperature and salinity, but influences of these factors on the physicochemical properties and toxicity of ZnO-NP are often tested individually. As both factors co-exist in the environment and may influence one another, it is vital to study their effects concurrently to tease out any potential interactions. This study, therefore, investigates the interacting effects of temperature and salinity on ZnO-NP to a common marine copepod (*Tigriopus japonicus*) along the Western Pacific coast. Physicochemical properties (aggregate size & ion dissolution) of three zinc-associated compounds, including ZnO-NP, ZnO bulk-particles (ZnO-Bulk) and $ZnSO_4 \times 7H_2O$ ($ZnSO_4$), were characterized to compare their toxic mechanisms. Acute toxicity was determined with 96-h standard toxicity tests under nine different combinations of temperature (15, 25 & 35 °C) and salinity (12, 22 & 32 PSU), i.e., a 3 x 3 factorial design. Preliminary results showed that increase in temperature and salinity could increase aggregate size of ZnO-NP and ZnO-Bulk, but reduce their ion dissoution rate. At 25 °C, similar to previous studies which suggest that dissolved zinc ions (Zn^{2+}) is the major contributor of toxicity, ZnO-NP was the least toxic at 22 PSU, where the dissolution rate of Zn^{2+} was the smallest. $ZnSO_4$ was the least toxic compound, implying that Zn^{2+} were not the only contributor to the observed toxicity. Higher toxicity of ZnO-NP and ZnO-Bulk might be due to particle and animal interactions such as physical damage by larger aggregate at larger salinity. The results will advance our understanding of interactions of ZnO-NPs with temperature and salinity, from which it will be able to determine effect threshold concentrations for regulation of products of ZnO-NP under different combinations of these two environmental factors.

WE312

Multigenerational effects of gold nanorods to Raphidocelis subcapitata and Chlorella vulgaris

C. Monteiro, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro; C. Venâncio, Department of Biology / Biology; A.L. Daniel-da-Silva, S.F. Soares, University of Aveiro / Department of Chemistry & CICECO, 3810-193 Aveiro; A.M. Soares, University of Aveiro / department of Biology & CESAM; T. Trindade, University of Aveiro / Department of Chemistry CICECO; I. Lopes, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro

In real scenarios, exposure to nanoparticles may occur over several generations, which may exhibit a higher sensitivity (due to the accumulation of adverse effects) or tolerance (due to phenotypic plasticity mechanisms) to the nanoparticle. The aim of this study was to evaluate the multigenerational effects of gold nanorods (Au-NR) in two freshwater microalgae: *Chlorella vulgaris* and *Raphidocelis subcapitata*. These specieswere exposed, for four generations (F1 to F4), to the respective Au-NR concentration causing 10% reduction in growth rate ($EC_{10,72h}$, computed for F0). The sublethal sensitivity of each species to Au-NR was then quantified and compared among generations, i.e. generations not exposed to (F0) and exposed to Au-NR (F1 to F4). For this, algae were exposed to concentrations of Au-NR ranging from 8 to 90 µg/L, for 72h. At the end of the assays, growth rate was computed for all generations of each alga. The following physic-chemical parameters of Au-NR concentrations were monitored: size, morphology and total concentration of Au. In addition, the toxicity of the capping agent cetyltrimethylammonium bromide (CTAB) was quantified by exposing both algae to the concentration of CTAB present in the highest tested concentration of Au-NR: 90 µg/L for *C. vulgaris* corresponding to 0.257 mM of CTABand 53 µg/L for *R. subcapitata* corresponding to 0.152 mM of CTAB. *Chlorella vulgaris* exhibited a higher tolerance to Au-NR than *R. subcapitata*: $EC_{20,72h}$ for F0 was 79 µg/L and 39 µg/L, respectively). For *C. vulgaris*, a gradual increase of its tolerance to Au-NR was observed over generations; after being exposed for four generation to this chemical no significant effects on growth rate were observed between all concentrations and the control. A different pattern of response was observed for *R. subcapitata*. This speciessignificantly increased its sensitivity to Au-NR from F0 to F2 generation, but recovered a similar sensitivity to that quantified for F0 from F2 to F4. CTAB significantly reduced the growth of microalgae by 42%. Over generations the sensitivity of the two algae species to CTAB was not changed. The results obtained in the present work reveal that traditional standard assays with short-term exposure may over- or underestimate the real risk posed by Au-NR to freshwater microalgae. Therefore, it is suggested that long-term exposures should be included in the ecological risk assessment.

WE313

Effects of climate change combined with copper nanoparticle on early development of Japanese medaka (Oryzias latipes)

I. Meng Ian, Y. Zhang, W. Chen, Department of Biomedical Science and Environmental Biology, Kaohsiung Medical University, Kaohsiung

Nowadays, global warming and aquaitc acidification were occured by rising carbon dioxide (CO_2). The factory have been continuously emit copper nanoparticle into ocean and river. They probably induced harmful biological effect on organisms. However, the combined effects of three environmental stressors on aquatic species have not been well studied. The purpose of this study is to assess the effects of aquatic acidification and warming combined with copper nanoparticle on survival and hatchability of early development of Japanese medaka (*Oryzias latipes*). Firstly, the embryos were exposed to 25 nm copper nanoparticle (30 µg/L) and without copper nanoparticle under nine temperature and pH conditions (26°C/7.5, 26°C/6.5, 26°C/5.5, 28°C/7.5, 28°C/6.5, 26°C/5.5, 30°C/7.5, 30°C/6.5 and 30°C/5.5) for 14 days to observe hatchability and mortality. Then, the newly hatched fry were exposed to same condition for 14 days to observe the survival. Results showed that the mortality of embryos had a upward trend, and the hatching rate had a downward trend in exposure group of pH 5.5 compared with that of others pH groups. However, only 28°C/5.5 combined with copper nanoparticle group was significantly increased mortality and inhibited hatchability than that of 28°C/6.5 combined with copper nanoparticle group. While the survival rate of larvae had a significantly downward trend in exposure groups of lowest pH level and highest temperature on days 10. In conclusions, medaka larvae have more sensitive than embryos by survival ability. Moreover, aquatic acidification and warming were synergistic effect to increase copper nanoparticle toxicity to larvae. Therefore, we found that three environmental stressors caused a potential hazards to fish population.

WE314

The use of the marine mussels Mytilus hemocytes as a model for studying the impact of NPs on innate immunity

M. Auguste, Unversity of Genova / DISTAV; T. Balbi, L. Canesi, university of genoa / DISTAV

Nanoparticles (NPs) are widespread used in consumer products and industry; they are of growing interest due to their interaction with biota and especially their potential impact on environmental species. Recent studies have demonstrated that NPs could affect a number of marine species, and interact with their immune system. Within the PANDORA project (Probing safety of nano-objects by defining immune responses of environmental organisms), a European Training Network (ETN) funded in the framework of H2020 Marie Sklodowska- Curie ITN programme*, objectives are to identify immunological mechanisms triggered by nano-objects, and predictive markers of risk vs. safety, with a collaborative cross-species comparison. The use of mussel hemocytes, from *Mytilus galloprovincialis*, for in vitro testing is a valuable tool in the screening of the toxicity of NPs as the tests are inexpensive and use alternative animals in experimental science. In vitro analysis are performed to determine functional immune parameters (e.g. lysosomal membrane stability, superoxide and NO production, phagocytic activity) and particle internalisation by hemocyte upon short-term exposure to NPs (different concentrations and times of exposure from 30 min to 1h). Once entering the organism, NPs are in contact with other type of media e.g. hemolymph serum for mussels. For some type of NPs, the response is affected by the presence of protein components of hemolymph serum involved in the formation of a NP-protein corona. In order to have a wider view of the interactions and mechanisms of actions of NPs, the same parameters are measured with NPs suspensions in artificial seawater (ASW) and serum. The results obtained with *Mytilus* hemocytes will be compared with those obtained in immune cells of other model organisms within the PANDORA project. According to the special properties of every NPs, the aim is to understand the main mode of action at the cell level that will help designing predictive in vitro assays to measure the immuno-risk of NPs to the environment in the future. *Funded within the European Union's Horizon 2020 research and innovation programme under the Marie Sklodowska-Curie grant agreement PANDORA No 671881.

WE315

Influence of warming and acidification on copper nanoparticle bioaccumulation in medaka (Oryzias latipes) embryo

Y. Zhang, I. Meng Ian, Department of Biomedical Science and Environmental Biology, Kaohsiung Medical University, Kaohsiung; Y. Chen, Kaohsiung Medical University / Biomedical Science and Environmental Biology; W. Chen, Department of Biomedical Science and Environmental Biology, Kaohsiung Medical University, Kaohsiung

The level of atmospheric CO_2 has elevated significantly since the Industrial Revolution, leading to global warming and ocean acidification. With the development of industry and technology, many emerging contaminants such as copper nanoparticle (CuNPs) may be exposed to environment. However, it is unclear whether the accumulation of copper nanoparticles in organism will increase under the warming and acidification scenarios. Therefore, the purpose of this study is to investigate whether CuNPs (25 nm, 0.03 mg/L) will accumulate in Japanese medaka (*Oryzias latipes*) embryo under the condition of elevated temperature combine with acidification. The medaka embryo was followed four consecutive days at nine temperature and pH conditions (26°C/7.5, 26°C/6.5, 26°C/5.5, 28°C/7.5, 28°C/6.5, 26°C/5.5, 30°C/7.5, 30°C/6.5 and 30°C/5.5). Results showed that the temperature and pH did not affect the accumulation of CuNPs, respectively. When combined with temperature and acidification factors, Cu accumulation of group 30°C/6.5 was increased significantly than that of groups of 26°C/6.5, 26°C/5.5, and 30°C/5.5. However, it revealed that there was no significant evidence of warming and acidification on increased Cu accumulation of medaka embryo. Therefore, we concluded that there was no influence of Cu accumulation in medaka embryo when warming and acidification occurs in the future.

WE316

Assessing the combined toxicity of metals and metal-oxide nanoparticles in a benthic estuarine microalgae

R.B. Ogunjemilusi, M. Yallop, G. Barker, University of Bristol

Coastal aquatic ecosystems such as estuaries are at risk of metal pollution due to anthropogenic inputs from acid mine drainage, industrial and agricultural run-off. In addition, advances in nanotechnology in the last decade have increased the production engineered nanomaterials (ENMs) used widely in fields such as medicine, energy, agriculture and consumer goods production. As a result, there are increasing concerns about the release of engineered nanomaterials such as metal oxide nanoparticles into the environment. While the effect of metals and ENMs as single contaminants have been extensively studied, much research is needed to account for potential mixture effects due to heavy metal-ENMs interactions. Studies suggests that the toxicity of ENMs such as metal-oxide nanoparticles is mainly due to the release of dissolved metal ions. However, majority of these studies have mainly focused on impact of ENMs in freshwater environment and results are extrapolated for other types of environmental systems (marine, soil, sediment). Evidence is accumulating that the dissolution of ENMs is dependent on the characteristics of the exposure medium. Metal-oxide nanoparticles tend to form aggregates in seawater medium, reducing the amount of released metal ions. In this research, the combined toxicity of heavy metals (Cu and Zn) and their metallic oxide nanoparticles will be evaluated in a benthic estuarine microalgae - *Cylindrotheca closterium*. We hypothesize that the aggregates formed by the metal-oxide nanoparticles may adsorb the free metal ions, reducing the bioavailable fraction of heavy metal ions in the exposure medium. Because of the bottom dwelling nature of the algae, we also investigate whether reduced light intensity (shading effects) as a result these aggregates may be a potential mechanism of ENMs toxicity in benthic aquatic organisms.

WE317

Comparative toxicity of silver nanocolloids and titanium dioxide nanoparticles using medaka

Y. Kato, Toyo University / Faculty of Life Sciense; T. Ariyoshi, C. Kataoka, S. Kashiwada, Toyo University / Graduate School of Life Sciences

Silver nanoparticles and titanium dioxide nanoparticles are representative nanomaterials and massively used for multiple purpose to improve human life. Hence, environmental fate and unintentional ecological effects and/or toxicities have been concerned and many studies are reported using model organisms. We have been investigating fish toxicity and ecological risk of silver nanocolloids (SNCs, Φ40 nm) using medaka model. SNCs have embryonic (at 0.5 mg/L of SNC) and larvae (at 5 mg/L of SNC) toxicities including lethality, inhibition of embryo developments, shortened body length, small eye development, ischemia, reduced heart beating, and caused some oxidative stresses such as GSH reduction and lipid peroxidation. To adults, SNCs exposure (at 5 mg/L) did not exhibited significant lethality; however, it was observed that SNCs exposure (at 0.05 mg/L) disrupted immune system and reduced tolerance to infective bacterial disease (*Edwardsiella tarda*). In SNCs exposure, we resulted that silver chloro-complexes, which were made of dissociated silver ion from SNCs, should be essential toxicants of SNCs exposure. On the other hands, titanium dioxide nanoparticles (TiO_2-NP, Φ< 90 nm) are well-known causing oxidative stress by UV radiation; however, there are some reports that TiO_2-NP does not have significant toxic effect to fish other than hypertrophy of gill mucus. We have assumed that ion dissociation will be a key to understand nano-toxicity depended on materials. TiO_2-NP which does not dissociate ions, was employed as a reference to ion dissociation NP (*i.e.* SNCs). In exposure of TiO_2-NP (at 10 mg/L) to embryo and larvae, there was no biological toxic effect mentioned above at all. In our presentation, we will discuss comparative toxicity of SNCs and TiO_2-NP regarding general toxicity, oxidative stress, cytotoxicity (apoptosis and necrosis), immuno-toxicity, and tolerance to infective bacterial disease. Through this study, we will figure out that dissociated ions should be toxic essential of nanomaterials, and not always nano-sized material will be toxicant in aquatic environment.

WE318

Genotoxicity assessment of aluminium oxide nanoparticles in relation to Escherichia coli and Aeromonas hydrophila

n. doskocz, M. Załęska-Radziwiłł, K. Affek, Warsaw University of Technology, Faculty of Building Services, Hydro and Environmental Engineering / Department of Biology

Increasing production and use of nanoparticles contributes to their widespread dissemination in the environment and their unique physical and chemical properties lead to unlimited distribution in environmental compartments. Migration ability of nanoparticles can have very dangerous consequences, as they can be transferred to potable water. Genotoxicity biomarkers are regarded as useful tools for the assessment of chemical hazards in aquatic ecosystems, because chemicals which damage DNA can significantly alter the functioning of ecosystems. Recent advances in molecular biology have led to the development of several techniques, which can be used for DNA analysis in the field of genetic ecotoxicology. The randomly amplified polymorphic DNA (RAPD) method is a PCR-based technique that amplifies random DNA fragments with the use of single short primers of arbitrary nucleotide sequence under low annealing conditions. RAPD-PCR test has been used successfully for detection of genetic damage in animals and plants. The SOS-Chromotest is one of the most commonly used bacterial tests. It is based on the induction, by genotoxic compounds, of a bacterial SOS repair system conjugated to the b-galactosidase gene and the subsequent measurement of the enzyme expression. In this work, genotoxicity studies on the basis of the RAPD-PCR and SOS-Chromotest assay were performed for aluminium oxide nanoparticles (nano-Al_2O_3). Little information is available on the fate, transport, and effects of nanomaterials, including metal based particles such as nano-Al_2O_3, in the environment. The interest in nano-Al_2O_3 is due to the fact that their influence on genetic material of bacteria is practically unknown. Results obtained for the nanocompound were compared with those for Al_2O_3 macro form. The nanocompound caused changes in the genetic material of bacteria *A. hydrophila.* Degree of genetic similarity of obtained profiles bands for primer OPA2 differed from the results obtained for the negative control by more than 27,3%, while from positive control - only by 15,6%. Furthermore, the largest decrease in genetic stability was 89,3%. The values of genotoxicity induction coefficient (I) in the SOS-Chromotest showed strong genotoxicity for nano-Al_2O_3, in the presence of S9 fraction and slight genotoxicity in the absence of S9 fraction in mutants of *Escherichia coli*. The results showed also that the nano-Al_2O_3 can induce genotoxicity a greater extent than the same compounds in their macro form

WE319

Effects of Copper Oxide Nanoparticles and Arsenic on the Whole-Life Cycle of Rice (Oryza sativa japonica)

J. Liu, M. Simms, G.P. Cobb, Baylor University / Department of Environmental Science

Copper oxide nanoparticles (nCuO) and arsenic (As) phytotoxicity to rice plants (*Oryza sativa japonica*) was evaluated in a factorial study using (0, 0.1, 1.0, 10, 50, and 100 mg/L) in daily watering and As (0 and 10 mg/kg) in soil. Experiments were conducted in a greenhouse during 130 day interval. Two rice plants were grown in 3L plastic pots without drainage. Toxicants were quantified in soils and water by inductively coupled plasma-mass spectrometry before, during and after testing. nCuO particle sizes were determined by differential light scattering before addition to the test system. Exposures began at planting, continued through germination, and to seed production. Thus, our study is the first to examine the influence of nCuO in combination with As on the whole life cycle growth of rice plants. No significant effect was observed on the seed germination, but both nCuO and As had a significant main effects on the fresh weight (FW) of rice straw and the number of rice panicles (NRP). The interaction of the two toxicants was also significant on both FW and NRP. A decrease in the FW was observed for rice exposed to As, only. The midlevel concentrations (1.0 and 10 mg/L) of nCuO decreased the FW with no As addition, while higher concentrations (50 and 100 mg/L) significantly increased the NRP. Low and high nCuO concentrations (0.1, 50, and 100 mg/L) decreased the FW with the As addition, and NRP decreased in treatments receiving As along with either lower or higher nCuO concentrations. More data from this greenhouse study are currently being compiled and analyzed to determine the effect of As and nCuO on grain yield as well as Cu and As uptake, distribution, and speciation in rice plants and the grain.

WE320

Behavior of cerium oxide nanoparticles in presence of pharmaceuticals compounds on aquatic specimens

G. AMARIEI, Universidad de Alcala; K. Boltes, University of Alcala / Chemical Enginnering; P. Letón, University of Alcala

Nanoparticles, in particular metal oxide nanoparticles, have found extensive usage in a wide range of services and industries. Subsequently, they can be released into environment and finally end up in water bodies. That may suppose a potential risk to aquatic environment, exerting toxic effects at the level of cells, tissues or the whole organisms[1,2]. The present study, evaluate the toxicity behavior of cerium oxide nanoparticles (CeO_2NPs) on three aquatic specimens- algae *Selenastrum capricornutum*, bacteria *Vibrio fischeri*, and activated suldge, by exploring concentration-dependent effect and changes induced due to the presence of Ibuprofen (Ibu) or Levofloxacin (Levo). 72h algae growth-inhibition, marine bacteria-luminiscence reduction and 24h sludge enzymatic activity and oxidative stress were used as endpoints. Nanoparticles concentration ranged from 0.6 to 160 mg L^{-1}. The particle size and the ζ-potential of NPs in the culture media were measured to analize the relation between stability profile and the observed toxicity behavior. The obtained results reveal toxic effects of CeO_2 nanoparticles leading to growth inhibition in algae. The presence of Ibu did not produced significant changes, while Levo showed drastic negative effect in algae growth. Short-term exposure produced significant reduction of luminescence intensity in marine bacteria. The presence of both, Ibu or Levo, reduced the negative effects of single nanoparticles in *Vibrio fisheri*. Exposure produced significant oxidative stress in bacteria forming activated sludge, with lower damage to enzymatic activity. The presence of pharmaceuticals compounds did not produced significant changes on nanoparticles behavior for activated sludge. Results indicated that algae was more strongly affected than the marine bacteria and activated sludge, respectively. These can be attributed to the culture media and organisms structural characteristics, respectively. [1] Neale PA, Jamting AK, O'Malley E, Herrmann J, Escher BI. 2015. Behaviour of titanium dioxide and zinc oxide nanoparticles in the presence of wastewater-derived organic matter and implications for algal toxicity. Environmental Science: Nano *2*:86-93. [2] Sahle-Demessie E, Changseok H, Amy Z, Bill H, Heidi G. 2016. Interaction of engineered nanomaterials with hydrophobic organic pollutants. Nanotechnology 27:284003. *Acknowledgement* - The research was funded by the Comunidad de Madrid, grants S2013/MAE_2716 REMTAVARES.

WE321

Toxicity of nanoparticles of titanium dioxide to Daphnia longispina: waterborne versus dietary exposure

E. Padilla, Institute for Environmental Sciences / University Koblenz-Landau; C. Venâncio, Department of Biology / Biology; I. Lopes, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro

Nanoparticles of TiO_2 (n-TiO_2) are extensively used in many commercial products. Maybe for this reason, this nanoparticle is amongst the most studied in ecotoxicology. This study intended to discriminate the toxicity caused by n-TiO2 to the daphnid species *Daphnia longispina*, either through waterborne or dietary exposure routes. For this, neonates of *D. longispina* were exposed to a control and to the following n-TiO_2 treatments: i) medium spiked with a concentration of 0.625 mgL^{-1} n-TiO_2 (corresponding to the EC_{20} for *D. longispina*), ii) food (microalgae) spiked with nano-TiO_2 (after being exposed for 3 days to a concentration of 0.615 mgL^{-1} n-TiO_2), and iii) water and food spiked with n-TiO_2. The effects of the nanoparticle were monitored on the feeding rate (after a 24h exposure period), somatic growth rate (after a 10-day exposure) and in reproduction (after 21-dasy of exposure) of *D. longispina*. A significant reduction on feeding rate was only observed when daphnids were exposed to n-TiO_2 simultaneously through the water and food items. In this same treatment a significant increase in the somatic growth rate was observed relatively to the control. No significant effects were observed for time to release the first brood and for length of females at first brood. However, neonates from females exposed to the nanoparticle through dietary were smaller (2.52±0.32 mm) than the ones in the control (3.04±0.11 mm) and the total number of neonates released per female was significantly higher for females exposed to n-TiO_2 both in dietary and waterborne (6.1 ±1.37 neonates/female versus 3.8 ±1.619 in the control). The obtained results report higher effect on somatic growth and reproduction of n-TiO2 when exposure occurs via the two routes: waterborne and dietary. However, the tested concentrations seemed to increase the fitness of D. longispina, which could be due to hormesis effects.

WE322

Dynamics of Cu accumulation in charophyte cell compartments after its exposure to nCuO

L. Manusadzianas, NatureResearch Centre / Institute of Botany, Laboratory of Aquatic Ecotoxicology; B. Gylyte, S. JURKONIENE, R. Vitkus, Nature Research Centre / Institute of Botany

In plant and bacterial cells, prior to be internalized, NPs have to pass semipermeable cell wall and plasma membrane barrier. Metal nanoparticles may interact with the cell directly or induce toxicity through the release of metal ions. Internodal cell of characean green alga posses features such as big size and clear separation of the main cell compartments, which enable mechanical separation of the compartments, namely cell wall, cytoplasm and vacuole. In this study, fractionation procedure was verified by cytoplasm and vacuole specific biomarkers, malate dehydrogenase and α Mannosidase, respectively. A high-purity vacuolar (99.5%) and cytoplasm (86.7%) fractions of the cells of *Nitellopsis obtusa* were obtained. The cell wall fraction contained approximately 1.8 and 13.4% of cytoplasm and vacuole. By additional washing of the cell wall it was possible to diminish contamination with cytoplasm. The data on Cu accumulation dynamics within the compartments after cell exposure to nCuO suspensions will be presented and the role of the cell wall in the accumulation process will be discussed.

WE323

Are graphene nanomaterials "Trojan horse" carriers for oil compounds in mussel hemocytes in vitro?

G. Nicolussi, University of the Basque Country / CBET Research Group, Dept. Zoology and Animal Cell Biology, Science and Technology Faculty and Plentzia Marine Station, University of the Basque Country (UPV/EHU). Basque Country, Spain; A. Katsumiti, M.P. Cajaraville, University of the Basque Country / CBET Research Group, Dept. Zoology and Animal Cell Biology Faculty of Science and Technology and Research Centre for Experimental Marine Biology and Biotechnology PIE

In the aquatic environment, complex mixtures of pollutants are usually found. Polycyclic aromatic hydrocarbons (PAHs) are prioritary pollutants and main constituents of the water accommodated fraction (WAF) of petroleum. Graphene nanoplatelets can adsorb organic compounds thus being potentially useful in oil spill remediation. However, they could also act as vehicles of organic contaminants to aquatic organisms (“Trojan horse” effect). This study aimed to evaluate the potential “Trojan horse” effect of graphene nanoplatelets (graphene oxide (GO), GO-polyvinylpyrrolidone (GO-PVP) and reduced GO-PVP (rGO-PVP)) with adsorbed oil compounds from naphthenic North Sea crude oil WAF using *in vitro* toxicity assays in hemocytes of marine mussels. Two approaches were tested to obtain graphene nanoplatelets with adsorbed oil compounds: filtration and centrifugation. Hemocytes were exposed to a wide range of concentrations of GO, GO-PVP and rGO-PVP with and without adsorbed oil compounds and to a series of WAF dilutions. After 24 h exposure, cell viability (MTT assay) and ROS production were assessed. Centrifugation (270*g* for 30 min) successfully separated WAF solution from graphene nanoplatelets with adsorbed oil compounds. This procedure was thus used for *in vitro* toxicity testing. WAF decreased cell viability and increased ROS production in hemocytes starting at 25% WAF. GO, GO-PVP and rGO-PVP nanoplatelets were moderately toxic to mussel hemocytes and produced a significant increase in ROS production. In exposures to graphene with adsorbed oil compounds, hemocytes viability decreased at similar concentrations as in exposures to nanoplatelets alone. However, ROS production increased in hemocytes exposed to lower concentrations of graphene with adsorbed oil compounds (10 mg/L) compared to nanoplatelets alone (25 mg/L), indicating that adsorbed oil compounds increase nanoplatelets toxicity. In conclusion, a protocol to obtain graphene nanoplatelets with adsorbed oil compounds was established. Nanoplatelets with and without adsorbed oil compounds showed similar cytotoxicity to hemocytes but the ones with adsorbed oil compounds increased ROS production earlier, indicating that graphene nanoplatelets may act as “Trojan horse” carriers of oil compounds. This work was funded by the EU H2020 GRACE project (grant 679266), Spanish MINECO (project NACE, CTM2016-81130-R), Basque Government (consolidated research group IT810-13) and University of the Basque Country (UFI 11/37).

WE324

Multigenerational effects of titanium dioxide and silver nanoparticles on Daphnia magna: gene expression and morphological changes in the presence or absence of aged nanomaterials

L.A. Ellis, The University of Birmingham / GEES; E. Valsami-Jones, University of Birmingham / School of Geography Earth and Environmental Sciences; I. Lynch, University of Birmingham / Geography Earth Environmental Science

Recent studies have investigated nanoparticle (NP) physicochemical properties and interactions with biological systems. *Daphnia magna* was chosen as the model organism, as they are well characterized and reproduce parthenogenetically, which is well suited for experimental genetic studies and monitoring stress/adaptive change to their environments. Herein, we investigated key biological endpoints, such as survival, growth, reproduction, and gene expression of related molecular pathways in response to exposure to silver (AgNPs) and titanium dioxide (TiO_2) nanoparticles (NPs). Particles were either pristine or aged, uncoated or stabilized with either PVP or sulphide (AgNPs only). Our aims were to identify specific stress responses from NPs which could lead to molecular defects in order to understand if: (1) different NP compositions induce the same pathways and effects; (2) exposure in the presence of macromolecules such as natural organic matter changes the pathways and/or severity of changes observed; (3) if the aging of particles make them more or less toxic; (4) if long-term low dose exposure (25 days, EC_{20} concentrations) leads to developmental and reproductive changes, and (5) whether these NP-exposure induced changes to the F0 generation are passed onto subsequent generations, who themselves are not exposed directly. In all cases, both morphological changes and expression of key biomarkers were analysed in order to identify whether chronic exposure to NPs induces stress responses. We observed morphological changes, including eyes and tail defects, to each of the subsequent F1-F3 generations. We also observed differences in gene expressions compared the control populations, supporting that AgNP and TiO_2 do have toxicological impacts from chronic exposure irrespective of particle aging. TEM observations of consequent histological accumulation of the TiO_2 and AgNPs supported the assumption that NPs manifest themselves as particulates. We were also able to see some recovery in the F3 generations that had their subsequent parent generations removed from exposure. The influence of biomolecules secreted by the organisms in response to the presence of NPs, and the influence of humic acid containing medium during the exposure phase were also investigated, providing important insights regarding the need for realistic exposure scenarios during chronic exposure scenarios.

Ecological risks under complex, multiple-stressor threat scenarios: integrating chemical effects with environmental drivers (P)

WE325

Evaluating the contribution of environmental stressors to sediment concentrations of PAHs in the northern Gulf of Mexico

L.M. Basirico, Louisiana State Univeristy; H. Rockett, R.J. Portier, Louisiana State University / Environmental Sciences

Toxicity associated with organic pollutants in aquatic sediments has not been fully characterized for the major estuarine and marine systems of coastal Louisiana. As such, five inshore and three offshore transects of the Mississippi River delta were sampled for sediment concentrations of polycyclic aromatic hydrocarbons (PAHs) and environmental variables. From 2012 to 2014, five environmental variables were measured in the northern Gulf of Mexico including: 1) the salinity 2) dissolved oxygen and 3) temperature of the overlaying water column, and 4) the percent sediment moisture and 5) percent organic matter of the aquatic sediment. A main effects-model was implemented in order to assess the impact of environmental variables on the concentrations of ten PAHs and three toxicity indicators in freshwater and marine sediments. A backwards, step-wise linear regression analysis of variance (ANOVA), generalized linear model (GLM) was performed to determine significant effects of measured environmental parameters. The most important environmental variables affecting the concentrations of the measured compounds were those describing the characteristics of the aquatic sediments. The percent moisture of the sediment was the most important environmental parameter, significantly affecting eight of the ten organic pollutants and all three toxicity indicators. Percent organic matter of the sediments was the second most significant parameter, accounting for the variability in concentration for five of the measured pollutants. Temperature was significant for three of the PAHs and TEQ, and in every instance it had a negative effect on concentration. Dissolved oxygen of the water column was a significant variable on the concentration a single organic compound. Water column depth and salinity did not have a significant effect on the concentration on any of the constituents. The most significant environmental variables accounting for the variability of sediment PAH concentrations, included sediment moisture and organic matter. The physiochemical properties of the overlaying water column had little to no significant effect on the concentrations of the majority of the measured pollutants. The current research suggests that sediment organic matter and percent moisture measurements at each sampling location should be incorporated into monitoring study design in order to more completely interpret the sediment burden of organic pollutants in aquatic sediments.

WE326

Microbial resistance to chemical pollution by urban effluents might be triggered by desiccation events.

F. Romero, S. Sabater, ICRA Catalan Institute for Water Research; O. Pereda, University of the Basque Country; l. sabater, CSIC-IDAEA / Department of Environmental Chemistry; C. Font, V. Acuña, ICRA Catalan Institute for Water Research

Freshwater ecosystems are subjected to different anthropogenic pressures. Among them, wastewater treatment plant (WWTP) effluents can represent a significant proportion of total discharge, mostly in regions suffering from water scarcity. WWTP effluents contain a mixture of assimilable and toxic compounds, which concentration ultimately determines the effect of the mixture on freshwater biota. Moreover, overexploitation of water resources together with climate change-derived pressures is causing drought events leading to desiccation to increase in number and intensity. To date, little is known about how desiccation events shape the response of river microbial communities to WWTP effluents. The present study used 24 experimental channels in a replicated regressional design to evaluate how an acute desiccation event shapes the response of a complex microbial community (i.e. a river biofilm) exposed to a dilution of a WWTP effluent. We found that desiccation and wastewater effluent significantly affected bacterial community and key biofilm processes such as photosynthesis, denitrification and methanogenesis. After the desiccation event, the biofilm associated to coarse sediment showed a stimulatory effect even at low dilution factors, which was not observed under control (i.e. no desiccation) conditions. Our results seem to indicate a simplification of the biofilm community after the desiccation event and a subsequent co-tolerance phenomenon. We argue that the acute desiccation event reduced diversity, selecting for resistant species. These resistant species could benefit from low dilution factors of wastewater effluent. Given that microbial metabolism powers biogeochemical cycling in ecosystems, we argue that functioning of freshwater ecosystems may be shaped as a result of the combined action of climate change-related stressors such as desiccation and chemical pollution.

WE327

Synergy effects of fluoxetine and variability in temperature lead to proportionally greater fitness costs: A multigenerational test

M. Oliveira, University of Aveiro; N. Inocentes, Department of Biology CESAM University of Aveiro / Biolo; A.M. Soares, University of Aveiro / department of Biology & CESAM; M. Barbosa, Department of Biology & CESAM - University of Aveiro / Biology

Increased variability in water temperature is predicted to impose disproportionally greater fitness costs than mean increase in temperature. Additionally, water contaminants are currently a major source of human induced stress likely to produce fitness costs. Global change models forecast an increase in these two human induced stressors. Yet, in spite the growing interest in understanding how organisms respond to global change, the joint fitness effects of water pollution and increased variability in temperature remain unclear. Here, using a multigenerational design, we test the hypothesis that exposure to high concentrations of fluoxetine, a human medicine commonly found in freshwater systems, causes greater lifetime fitness costs when associated with increase variability in temperature. Although fluoxetine and variability in temperature elicited some fitness cost when tested alone, when both stressors acted together the costs were disproportionally greater. The combined effect of fluoxetine and variability in temperature led to a reduction of 37% in lifetime reproductive success and a 17.9 % decrease in population growth rate. Interestingly, fluoxetine and variability in temperature had no effect on the probability of survival. Freshwater systems are of the most imperilled ecosystems, often exposed to multiple human induced stressors. Our results indicate that organisms face greater fitness risk when exposed to multiple stressors at the same time than when each stress acts on alone. Our study highlights the importance of using a multi-generational approach to fully understand individual environmental tolerance and its responses to a global change scenario in aquatic systems.

WE328

Influence of extreme heat events in the recovery capability of Mytilus galloprovincialis exposed to mercury contamination

F. Coppola, Department of Biology & CESAM - University of Aveiro / Biology; B.M. Henriques, CESAM University of Aveiro and CIIMAR University of Porto / Department of Chemistry; A.M. Soares, University of Aveiro / department of Biology & CESAM; R. Freitas, University of Aveiro / Departamento de Biologia CESAM; E. Figueira, University of Aveiro / Biology CESAM; M.E. Pereira, CESAM University of Aveiro / Department of Chemistry

Several studies already described the impacts caused by metals in estuarine species, including mussels, but very scarce information is available regarding their effects in a global warming context. Moreover, increased temperatures can not only affect the response of organisms but their capacity to recover from pollution events. In this way, the present study aimed to understand the impact of warming in the capacity of *Mytilus galloprovincialis* to recover their biochemical performance after being pre-exposed to Hg. For this, mussels were exposed during 14 days at 17 ºC in the absence or presence of Hg (17 ºC; 17 ºC Hg), after which mussels were exposed during 28 days at 21 ºC in the absence or presence of Hg (21 ºC; 21 ºC Hg) or kept for the same period at 17 ºC in the absence or presence of Hg (17 ºC; 17 ºC Hg), and biomarkers related to mussels' metabolic and oxidative stress status were evaluated as well as Hg bioconcentration. Our findings revealed that independently on the temperature regime, organisms previously exposed to Hg followed by a 28 days period in the absence of Hg were able to significantly decrease their metal concentration. Furthermore, energy-related and oxidative stress markers in mussels exposed for 28 days in the absence of Hg demonstrated no differences between mussels exposed to warming conditions (21 ºC) and control temperature (17 ºC), with a tendency to reach control values (observed in mussels exposed the entire experiment to 17 ºC in the absence of Hg).

WE329

Impacts of ocean warming and BDE-209 contamination on the energy budget of juvenile white seabream (Diplodus sargus)

P. Anacleto, Instituto Português do Mar e da Atmosfera (IPMA) / Division of Aquaculture and Seafood Upgrading; C. Figueiredo, M. Baptista, MARE Marine and Environmental Sciences Centre; A. Maulvault, Instituto Português do Mar e da Atmosfera / Division of Aquaculture and Seafood Upgrading.; C. Camacho, IPMA, I.P.; M. Santos, Instituto Português do Mar e da Atmosfera / Division of Aquaculture and Upgrading; P. Pousão-Ferreira, Portuguese Institute for the Sea and Atmosphere; L. Valente, Interdisciplinary Centre of Marine and Environmental Research; A. Marques, Portuguese Institute of Sea and Atmosphere IPMA / Division of Aquaculture and Upgrading; R. Rosa, Faculdade de Ciências da Universidade de Lisboa / MARE Marine and Environmental Sciences Centre

Climate change and chemical contamination are global environmental threats of growing concern for the scientific community and regulatory authorities. Yet, the impacts and interactions of both stressors (particularly ocean warming and emerging chemical contaminants) on the physiological responses of marine organisms remain unclear and require further research. In this context, our main goal was to study, for the first time, the effects of warming (+5 °C, i.e. 24 °C) and accumulation of a polybrominated diphenyl ether congener (BDE-209, brominated flame retardant) by dietary exposure on the energy budget of juvenile white seabream (*Diplodus sargus*; 3.9±1.2 g total weight), used as a model. Specifically, growth (G), routine metabolism (R), excretion (faecal, F and nitrogenous losses, U) and food consumption (C) were calculated to obtain the energy budget. The results demonstrated that the energy proportion spent for growth dominated the mode of the energy allocation of juvenile white seabream (56.0-67.8%), including even under the synergistic effect of warming and BDE-209 exposure. On the other hand, energy loss via faeces was significantly higher under control temperature and BDE-209 exposure (16.0%). In all treatments, the energy channelled for metabolism was around 26% and a smaller percentage was channelled for excretion (faeces: 4.3-16.0% and ammonia: 2.3-3.3%). In general, the parameters were significantly affected by increased temperature and exposure to the flame retardant, with higher levels found under warming conditions (for wet weight, relative growth rate, protein and ash contents), BDE-209 exposure (only for O:N ratio) or under both stressors (for ammonia excretion rate and routine metabolic rate). On the other hand, lower viscerosomatic index (VSI) was observed under warming and lower fat content was observed under both stressors. Overall, with such extreme and expected conditions of warming and contamination, the energy budget of marine fish species is expected to be greatly affected, leading to impacts on fish communities and ecosystems. Further research combining other climate change stressors and emerging contaminants are needed to better understand and forecast their ecological effects, in order to implement potencial mitigation measures.

WE330

Transgenerational effects of pesticide on vector mosquito Culex pipiens under global warming

T. Tran, L. Janssens, KULeuven; K.V. Dinh, DTU Technical University of Denmark; R. Stoks, University of Leuven / Department of Biology

Recent transgenerational studies have showed that some species could acclimate to warming and pesticide separately. Transgenerational plasticity is even being considered as a powerful mechanism to enhance species resilience to projected warming. However, it is unknown how exposure to pesticide under warming in the parental generation will shape the offspring susceptibility to these stressors, specifically in vector species. We studied the transgenerational effects of single and combined exposure to warming (4°C increase) and the pesticide chlorpyrifos on life history traits and antipredator behaviors of the vector mosquito *Culex pipiens* using a bifactorial transgenerational experiment. Parental exposure to a single stressor, either warming or the pesticide, had negative effects on the offspring: both parental exposure to warming and to the pesticide resulted in an overall lower offspring survival. Parental warming impaired the anti-predator behaviors of the offspring by decreasing the diving proportion and diving time off the offspring. Within both the parental and the offspring generations, warming made the pesticide more toxic in terms of survival. However, this synergism disappeared in the offspring of parents who had been exposed to both stressors simultaneously because in this condition the pesticide was already more lethal at the lower temperature. For anti-predator behaviors, in both generations the two stressors reduced diving time in a synergetic way. In the parental generation, the effect of pesticide were stronger at 20°C than at 24°C. In the offspring generation, this synergetic effect depended on parental temperature. Pesticide induced stronger reduction in diving time at 20°C than at 24°C but only in the offspring of parents exposed to 20°C. Our results indicate that transgenerational effects will not increase the ability of this vector species to deal with pesticides in a warming world. This study highlights the importance of using bifactorial transgenerational experiment to understand the combined impact of pesticide and warming across generations, hence to assess the efficacy of vector control in a warming world.

WE331

1 + 1 ≠ 2: Heritage-dependent synergistic development responses in copepods exposed to predator cues and copper

T. Lode, J.D. Heuschele, T. Andersen, J. Titelman, University of Oslo / Department of Biosciences; K. Hylland, Department of Biosciences, University of Oslo, Oslo, Norway / Department of Biosciences; K. Borga, Department of Bioscences, University of Oslo / Department of Biosciences

This study examines sub-lethal developmental effects of combinations of predator cues (kairomones, threespine stickleback) and copper (20 µg Cu L^{-1}) on the marine copepod *Tigriopus brevicornis*. Focusing on nauplii development, the aim was to examine effects of treatments on: 1) age at maturity; and 2) stage-dependent sensitivity. Potential importance of pedigree was also tested by comparing offspring from different females. Individual nauplii from egg-bearing females (8 randomly picked individuals per females' clutch) were incubated individually and exposed to one of the four treatments: control, kairomone, copper or kairomone + copper. The experiment ran for 13 days (313 hours) with daily exchange of exposure solutions and simultaneous registration of survival (activity) and development (counting shed exuviae). Food (*Rhodomonas salina*) was added daily *ad libitum*. All individuals in control were mature by the end of the experiment. A

2-parametric non-linear mixed effect model was used to describe nauplii development over time (*Instar = K / (1 + (K – 1) * exp (- exp (log.mu) * age))),* where *K* = the asymptotic development stage and *mu* = the average stage transition rate). Effects of treatment and pedigree on the two model parameters were examined by comparison of models with likelihood ratio tests and Akaike's Information Criterion (AIC). This analysis finds that treatment influenced developmental stage at the end of the experiment, while pedigree affected the time to reach it. Developmental effects were further examined by comparing development stages of surviving individuals. When all individuals in control had reached maturity (288 hours), individuals exposed to the combined treatment kairomone + copper were significantly delayed compared to all other treatments. Effects on individuals in the combined treatment were greater than expected based on the two stressors alone. An adverse effect on development was already evident at the time of the first emerging copepodites (138 hours). These results indicate a synergistic relationship between risk of predation and copper by increased age at maturity in developing individuals of *T. brevicornis*. The results also show the significant role of pedigree in determining development rate. This highlights the need to consider both natural stressors and individual heterogeneity when conducting ecotoxicological studies.

WE332

Functional and structural soil-vegetation indicators of ecosystem functioning in metal-contaminated environments: a case study in SE Spain

J. Alvarez-Rogel, A. Peñalver Alcalá, M. Tercero Gómez, H. Conesa Alcaraz, O. Martínez Oró, Escuela Técnica Superior de Ingeniería Agronómica. Universidad Politécnica de Cartagena / Ciencia y Tecnología Agraria; F. Jiménez-Cárceles, BIOCYMA, Consulting in Environment and Quality, S.L. Murcia, Spain.; C. van Gestel, Vrije Universiteit Amsterdam / Ecological Science; M. González-Alcaraz, Department of Biology & CESAM - University of Aveiro / Biology & CESAM

Functional and structural soil parameters of six environments defined by stages of vegetation colonization were studied in abandoned mine tailings area. The environments were: A) Within the mine tailings: 1. Bare soils (S); 2. Small groups of Pinus halepensis trees (2-5) ≤≈2.5 m high, growing scattered (P); 3. Isolated P. halepensis trees >≈4 m high with shrubs and herbs under the canopy (P+MS); 4. Dense patches with several P. halepensis trees (>5) >≈4 m high and shrubs and herbs under the canopy (DP+MS). B) Outside the mine tailings: 5. Polluted forest with P. halepensis trees > 5 m high and shrubs and herbs under the canopy (PF); 6. Control mature forest not contaminated with P. halepensis trees > 5 m high and shrubs and herbs under the canopy (CF). Ecological indexes of vegetation were evaluated and soils analyzed for physical, chemical, and biological parameters. Soil temperature, feeding activity of invertebrates and decomposition were measured in situ. P+MS, DP+MS and PF showed the highest diversity of plant species and P the lowest. The organic C/N ratio was ≈20 in P+MS, DP+MS, PF and CF and ≈13 in S and P, which was in accordance with larger accumulation of litter in the first four environments. Cation exchange capacity (CEC), an indicator of the buffer capacity of the soil and the stability of the organic matter, was largely higher in CF (≈32) followed by PF (≈20), P+MS and DP+MS (≈12) and finally P and S (≈5). Water soluble organic carbon (the most labile fraction of soil organic C) and C from microbial biomass (indicator of micro-organisms population) followed the same pattern than CEC. Total metal(oid)s concentrations (mg kg-1) widely varied within the tailings, without a clear pattern related with plant colonization (e.g. Pb: 5400-14600; Zn: 8600-18000; As 200-1200). Water soluble metal(-oid)s (µg kg^{-1}), the most toxic fraction, were largely higher in S (e.g. Pb≈4600; Zn≈210000). Tea bags decomposition showed two different tendencies: DP+MS, P+MS and S had a higher weight loss than PF, CF and P. Feeding activity was (% of holes fed upon): CF =42%, P =39%, S= 31%, P+MS =21%, AF =8%, DP+MS =7%. Total and soluble/available metals concentrations cannot to be considered the only factors related with the activity of biota in polluted sites. Field studies including physical, chemical, and biological parameters must be considered together to obtain realistic information for understanding soil ecosystem functioning and recovering.

WE333

Effects of imidacloprid and a neonicotinoid mixture on aquatic invertebrate communities under Mediterranean conditions

A. Rico, A.A. Sanchez, IMDEA Water Institute / Aquatic Ecotoxicology; J. Pasqualini, A. García-Astillero, L. Cherta, L. Nozal, IMDEA Water Institute; M. Vighi, IMDEA Water Institute / Earth and Environmental Sciences

Neonicotinoids are a group of insecticides that are used worldwide in agriculture production to treat piercing-sucking and chewing insect pests. These insecticides are considered contaminants of emerging concern due to their high toxicity potential to non-target terrestrial and aquatic organisms. Furthermore, they are candidates to be included as part of the EU monitoring watch list. In this study we evaluated the effects of imidacloprid (the most used neonicotinoid) and a mixture of five neonicotinoids (imidacloprid, acetamiprid, thiacloprid, thiamethoxam, and clothianidin) on freshwater macroinvertebrate and zooplankton communities. The experiment was performed using lenthic mesocosms in Central Spain under Mediterranean conditions. This study demostrated that the Concentration Addition (CA) model for the prediction of the toxic effect of mixtures of chemicals with the same mode of action may be applied to describe the short term effects of complex communities and not only individual organisms. However, some of the recorded indirect effects and the recovery of some populations showed slight differences between the imidacloprid and the neonicotinoid mixture treatments. This was attributed to the different dissipation rates of some of the test compounds included in the mixture as compared to imidacloprid. Therefore, it may be concluded that the CA model provides an accurate prediction for short-term effects at the population and community levels but requires the inclusion of other lines of evidence (e.g. ecological modelling results) to predict long-term effects and recovery. Some aquatic insect taxa (Chironomini, *Cloeon dipterum*) were found to be highly sensitive to neonicotinoid concentrations under Mediterranean conditions.The lowest calculated NOECs from this study are below 0.2 µg/L for imidacloprid and for the neonicotinoid mixture, indicating that the current water quality criteria proposed by regulatory agencies and recent scientific publications (0.2 µg/L) may be underprotective for lentic aquatic ecosystems under Mediterranean conditions. However, due to the high variability of the results at the low test concentrations, the hypothesis must be confirmed by performing more specific tests.

WE334

Multiple stressor effects of ionising (γ) radiation and non-ionising (UV) radiation in duckweed (Lemna minor)

L. Xie, NIVA - Norwegian Institute for Water Research; Y. Song, NIVA Norwegian Institute for Water Research / Ecotoxicology and Risk Management; O.C. Lind, K.A. Solhaug, B. Salbu, Norwegian University of Life Sciences; K. Tollefsen, NIVA / Ecotoxicology and Risk Assessment

In nature environment, aquatic biota is facing to the ionizing radiation emitted from natural occurring radionuclides where phenomenal of effects may also be enhanced by radiation from human activity such as nuclear power plants accident, nuclear medicine and weapon tests. (Van Hoeck et al.2015). Among different ionizing radiation types, the toxicity of high dose gamma (γ) radiation is frequently studied in different aquatic organisms such as mammals, fish, crustaceans, higher plants, and algae. However, there is still lack of knowledge about the toxic effects of low dose ionizing radiation and how other stressors such as ultraviolet radiation (UVR) cause multiple stressor effects in aquatic organisms. The objective of the present work was to study the combined effect of low-dose γ-radiation (13.2, 20.3 and 47.1 mGy/h) and UVR (UVB 0.5 w/m2) in the aquatic plant duckweed (*Lemna minor*) using a combination of genomic, functional and adverse toxicity endpoints. The results indicate that single g-radiation reduced *L. minor* reproductive rate at a high dose (47.1 mGy/h, 7.9 Gy) after 7 days' exposure. At the cellular level, γ-radiation inhibited photosystem II (PS II) maximal efficiency (F_v/F_m) and oxidative phosphorylation (OXPHOS) and enhanced the non-photochemical quenching (NPQ), light-saturated PS II operating efficiency (F_v'/F_m'), electron transport rate (ETR) and reactive oxygen species (ROS) formation. Single UVR caused similar effects as IR and additionally induced morphological change (size and colony disconnection) in the plant. When exposed in combination, enhanced reproductive inhibition, OXPHOS reduction, PSII inhibition, NPQ and ROS formation were observed for the high γ-radiation dose (47.1 mGy/h). Antagonistic effects on F_v'/F_m', pigments content, photochemical quenching (qP) and ROS formation were observed at low to intermediate γ-radiation doses (13.2 and 20.3 mGy/h). Mechanistic studies to elucidate the mode of action (MOA) are will be used to identify the most relevant toxic pathways being perturbed by the single and multiple stressors tested. Combine study with radiations and chemicals are currently on going.

WE335

Natural organic matter determines the potential of titanium dioxide nanoparticles to mitigate pesticide toxicity in presence of UV light

S. Lüderwald, Universität Koblenz-Landau / Institute for Environmental Sciences; V. Gerstle, F. Meyer, R. Schulz, University of Koblenz-Landau / Institute for Environmental Sciences; R. Bundschuh, SETAC Europe Office / Department of Aquatic Sciences and Assessment

Nanoparticle-based technology has evolved to a global industry with a tremendous economic potential. Since 2006, the investment in nanotechnology increased from estimated $11.8 billion to more than $2.5 trillion in 2015. Among all manufactured nanoparticles (NPs), titanium dioxide NPs ($nTiO_2$) belong to the most frequently produced and applied NPs. As a consequence of their incremental use, $nTiO_2$ will end up in surface waters, e.g. via wastewater treatment plant effluents. There, UV light triggers the photocatalytic potential of $nTiO_2$ to form reactive oxygen species (ROS). ROS have the ability to reduce the toxicity of co-occuring pesticides on aquatic invertebrates. The role of ubiquitous natural organic matter (NOM) for this interaction is, however, not well understood. Therefore, this study systematically assessed the influence of ambient UV-A radiation (0.00, 0.40-0.60, 1.00-1.40, and 2.20-2.60 W UV/m^2) and NOM (seaweed extract, 0.00 or 4.00 mg TOC/L) on the potential of $nTiO_2$ (P25, 0.00 or 0.05 mg/L) to reduce the acute toxicity (96-h) of three selected pesticides (Azoxystrobin, Dimethoate, and Pirimicarb) towards the waterflea *Daphnia magna*. Azoxystrobin toxicity was up to 1.6-fold reduced in the presence of $nTiO_2$ with increasing UV intensity (0.00 vs. 2.20-2.60 W/m^2). The combination of $nTiO_2$ and NOM enhanced the toxicity of Azoxystrobin 2-fold (0.00 vs. 1.00-1.40 W/m^2). Dimethoate toxicity was 3-fold decreased with increasing UV (0.00 vs. 2.20-2.60 W/m^2). NOM generally decreased the toxicity of Dimethoate by a factor of ~3, whereas the combination of $nTiO_2$ and NOM revealed the highest toxicity reduction with increasing UV (4-fold, 0.00 vs. 2.20-2.60 W/m^2). The toxicity of Pirimicarb was reduced 1.7-fold with increasing UV (0.00 vs. 2.20-2.60 W/m^2). In presence of NOM, Pirimicarb toxicity was generally decreased (up to 2.3-fold, e.g. 0.00 W UV/m^2). Depending on pesticide type and factor combinations we observed both positive and negative effects of UV radiation on the toxicity of the selected pesticides. A general prediction on the combined effects of $nTiO_2$, NOM, and UV on the toxicity of pesticides seems currently difficult. Rather, physiochemical properties like pesticide structure, solubility, adsorb- or decomposability seem to be crucial for their interaction with $nTiO_2$, NOM, and UV-A, and the ultimate pesticide toxicity.

WE336

Effects of inorganic sunscreen formulations on the algal symbionts of reef-building corals, Symbiodinium spp., and their combined toxicity with ocean warming.

A. Tagliati, Heriot Watt University / Institute of Life and Earth Sciences; S.J. Hennige, The University of Edinburgh / School of Geosciences; T.F. Fernandes, Heriot-Watt University / Institute of Life and Earth Sciences

Between 4,000 and 6,000 tons of sunscreens annually are washed from the skin by swimming or bathing, and directly released into reef waters, posing a potential threat to coral reef ecosystems. Titanium dioxide nanoparticles ($nTiO_2$) are common UV filters in cosmetic products, and in typical sunscreen formulations they are dispersed in an oil phase, a mixture of emulsifiers and emollients which can reach up to 20% of the product content by weight. In order to assess the impact of inorganic sunscreen on corals' endosymbiotic algae (*Symbiodinium* spp.), three $nTiO_2$ with different sizes and surface coatings were dispersed in an oil phase to mimic commercial available sunscreen formulations. Two *Symbiodinium* phylotypes, known for their different tolerance to environmental change and stress, were exposed to oil:$nTiO_2$ dispersions at both ambient temperature (26°C) and thermal stress condition (32°C). Growth rates, maximum photosynthetic activity and reactive oxygen species (ROS) production were evaluated. *Symbiodinium* spp. exposed to sunscreen ingredients show negative effects on all endpoints studied, and the toxicity is enhanced with simultaneous heat stress. Results indicate toxicity is not dependent on the type of nanoparticle and it is likely driven by the oil carrier, a major ingredient in all cosmetic sunscreens. Released oil, and the nanoparticles in the oil droplets, accumulate in the water surface microlayer and sediments due to its lipophilic characteristics and resistance against degradation, potentially constituting a major risk to marine organisms. Corals rely on the photosynthate provided by the endosymbiotic algae for their nutrition, and the production of excess ROS by *Symbiodinium* cells as a consequence of heat stress, is considered to be a trigger of coral bleaching (the loss of *Symbiodinium* from the coral host). The significant decrease of maximum photosynthetic activity at 32°C coupled with the algal growth decline suggest serious damage of the photosynthetic apparatus of the algae. The increased ROS production following sunscreen exposure, in addition to the reduction of photosynthetic activity, provide evidence that exposure to these types of sunscreens may exacerbate bleaching response in corals and pose a risk to coral reef ecosystems in a changing ocean.

WE337

Metallothioneins as an indicator of metal exposure in a naturally mineral enriched aquatic environment

H. Pienaar, C. Wolmarans, G. Van Niekerk, NorthWest University School of Biological Sciences / Zoology; V. Wepener, North-West University - School of Biological Sciences / School of Biological Sciences

The Marico River, in the North-West province of South Africa, is relatively unaffected by anthropogenic activities. However, metal concentrations – mainly from natural sources – occasionally exceed environmental quality guidelines or toxic concentrations. Macroinvertebrates are capable to react to these metals through processes such as the induction of metallothioneins (MTs). The aims of this study were to determine whether the induction of MTs can be used as indicator of natural metal exposure in anthropogenically unimpacted systems and whether there are relationships between metal concentrations in water, sediment and macroinvertebrates and concomitant MT levels. This was done by sampling macroinvertebrates, water and sediment from eight sites in the Marico River and tributaries. Water and sediment samples were prepared and analysed with an ICP-MS to determine selected metal concentrations. Seven families including Notonectidae, Coenagrionidae, Atyidae, Libellulidae, Baetidae, Caenidae and Chironomidae were selected and digested for metal analyses. Four families including Gerridae, Aeshnidae, Atyidae and Coenagrionidae were also analysed to determine their MT concentration. To test for possible relationships between metal concentrations in the macroinvertebrate families, MTs and metal concentrations in the water and sediment, Spearmans' non-parametric correlation tests were done among sites. Positive correlations were found between metals in sediment and macroinvertebrates, while there were no correlations between metal concentrations in water and macroinvertebrates. Even in freshwater river systems with a relatively low human impact and no mining activities, a positive correlation existed between trace metal bioaccumulation (e.g. Ni, Pb, Zn) in macroinvertebrates and the induction of MTs. There were, however, no correlations between MTs and bioaccumulation of earth metals (e.g. Al, Fe, Mn, Ti). These data clearly demonstrate the application value of MTs as biomarkers for metal exposure in freshwater systems.

WE338

Mollusks as indicators of environmental pollution (case studies in marine mussel Mytilus galloprovincialis Lam. and terrestrial snail Bradybaena fruticum Mull.)

T. Kuznetsova, Saint-Petersburg Scientific Research Center for Ecological Safety Russian Academy of Sciences / Lab.Bioelectronic Methods for Geoecological Monitoring; S. Kholodkevich, Saint-Petersburg Scientific Research Center for Ecological Safety; V.M. Makeeva, Earth science Museum of Lomonosov Moscow State University; N.N. Kamardin, Saint-Petersburg Scientific Research Center for Ecological Safety Russian Academy of Sciences; A.V. Smurov, Earth science Museum of Lomonosov Moscow State University

Snails are widely used as sentinel species in marine and terrestrial biomonitoring. Their wide distribution in different biotopes, high sensitivity to pollution makes them good bioindicators in environmental quality assessment. The comparability of mollusks in pure and polluted sites was performed in field studies round Moscow Region and in Moscow city as well as for marine bioindication in a few locations round Sevastopol city. Cardiac activity registration in selected mollusks was carried out as a tool for measuring deterioration of general health of local biota in the sites of the concern. Methodology with standard testing of heart rate (HR) and HR variability using functional loads (short-term temperature, salinity change, some mechanical stimuli, etc.) were carried out. In land biodiagnostics a 3-4 years old genetically modified morphs of bush snails *Bradybaena fruticum* (Mull.) were used collected from Moscow city (Kartmazovo, Kuz'minki,Izmailov Park) with reference site out of the Moscow city (Mozhaiskiy Region, Gorodok).Paired comparison was done based on cardiac activity monitoring in mentioned groups of snails under thermic treatment (20-50min, 50±0,5°?). It was revealed that snails of the same genotype (stripped and without strips) from chemically polluted sites (Kartmazovo, Kuz'minky) differed in low thermoresistance from those of the reference side and Ismailovsky Park demonstarted in dynamics of HRs. The analogous HR-monitoring using marine mussel *Mytilus galloprovincialis* Lam. was peformed in Sevastopol Bays using salinity change test. Possible links between chemical pollution by dioxin (in terrestrial snails) and heavy metals contents in mollusk's tissues (in marine mussels) and peculiarities of HR recovery after thermal or salinity loads were discussed.

WE339

The effect of temperature on toxicity of cypermethrin on Daphnia magna

P.T. Kajankari, University of Helsinki / Department of Environmental Sciences; V. Junttila, University of Helsinki; A. Rantalainen, Univeristy of Helsinki; O. Penttinen, University of Helsinki / Faculty of Biological and Environmental Sciences

Cypermethrin is an insecticide which European Parliament of the Council has classified as a priority substance in Directive 2013/39/EU in the field of Water Policy. The Annex II of the Water Policy sets the environmental quality standards (EQS) for the priority substances. Cypermethrin's annual average and maximum allowable concentration in inland surface waters are 0.08 ng/L and 0.6 ng/L respectively and annual average and maximum allowable concentration in other surface waters are 0.008 ng/L and 0.06 ng/L respectively, one of the lowest annual average and maximum allowable concentrations in environmental quality standard. The abiotic stressor like the temperature can have an effect on toxicity of the chemical. Previous studies have shown that the temperature of the environment affect the toxicity of pesticides belonging to the pyrethroids which cypermethrin is part of. The 48 hour half maximal concentration (EC50) and median effective time (ET50) values were tested with custracean *Daphnia magna* immobilization at the temperatures 10 ºC, 16 ºC and 20 ºC in laboratory experiments. Cypermethrin was almost twice as toxic at 10 ºC (2.17 ± 0.20 µg/L) compared with 20 ºC (4.10 ± 0.30 µg/L). The EC50 value of 16 ºC was 2.64 ± 0.21 µg/L which was closer to EC50 value of 10 ºC than 20 ºC. The temperature had statistically significant effect in the EC50 experiment. The temperature did have a slight effect in the ET50 experiment. The ET50 value at 10 ºC was 30.60 ± 0.74 hours compared with 33.12 ± 0.79 and 32.86 ± 0.83 hours respectively at 16 ºC and 20 ºC. The only statistically significant difference between the temperatures was between 10 ºC and 16 ºC. The temperature dependent behavior of cypermethrin was not taken into account when the environmental quality standard for cypermethrin was set by European Union and the United States Environmental protection agency's reregistration eligibility decision for cypermethrin. With pyrethtroids deltamethrin and permethrin the Canadian guidelines do not take account effects of temperature. When guidelines and legislations are prepared to certain chemicals some abiotic factors like the temperature should be considered. Key to changing guidelines and legislation is to take account the possible effects of temperature in test standards.

WE340

Pattern oriented food web modelling of metal mesocosm datasets

K. Viaene, Ghent University / GhEnToxLab; F. De Laender, Université de Namur ASBL / Research Unit in Environmental and Evolutionary Ecology; K. De Schamphelaere, Ghent University (UGent) / Applied Ecology and Environmental Biology; F. Van Assche, IZA; S. Baken, European Copper Institute; P. Van Sprang, ARCHE

The risk assessment of metals has a long history and over time a large collection of

ecotoxicity data has been accumulated. The most informative tests performed for the ecological effects of toxicants are mesocosm studies: controlled experiments where the effects of toxicants on model communities are studied for extended periods of time. Mesocosm studies are cost- and labor-intensive but offer a unique insight into realistic ecological effects of toxicants: they address not only direct effects on sensitive species, but also indirect effects resulting from ecological interactions (e.g. competition, predation) between sensitive and less sensitive species. Typically, the effects occurring in mesocosm studies are however complex and difficult to interpret. A study has been set up to investigate whether food web modelling can reveal additional patterns, trends, or interactions in existing copper and zinc mesocosm datasets. Pattern oriented food web modelling – an ecological modelling technique – is used to reveal the mechanisms underlying metal effects. With pattern oriented modelling, multiple characteristic patterns in the datasets will be identified and described. Models are then evaluated for their ability to reproduce these patterns. In the case of mesocosm studies, food web models can be applied to understand the mechanism behind observed patterns. A large diversity of food web models exists with large differences in their complexity and underlying theories. Food web models based on ordinary differential equations are relatively simple in structure while they can still account for the interactions between species in the food web. They are therefore ideally suited to study mesocosm data. Additionally, the effects of environmental parameters such as temperature and pH, often key determinants of metal toxicity, can be included. Finally, the potential Pathway of Effects – explaining how metal toxicity leads to the observed patterns – can be evaluated to identify the most important drivers of metal toxicity in the mesocosm food webs and derive community-based threshold levels.

WE341

Bioaccumulation and physiological conditions in Ruditapes philippinarum from the Vallona lagoon (northern Adriatic Sea, NE Italy): Application of Contaminant/shell weight indices

F. Cacciatore, ISPRA-Institute for Environmental Protection and Research / Mitigation and Prevention of Impacts; V. Bernarello, R. Boscolo Brusà, G. Franceschini, G. Sesta, C. Maggi, D. Berto, M. Gabellini, C. Virno Lamberti, ISPRA Institute for Environmental Protection and Research

Ruditapes philippinarum (Adams & Reeve, 1850)is a soft-bottom dweller bivalve commonly used in biomonitoring programs, especially in bioaccumulation assessment, owing to its high tolerance to toxic compounds. However, bioavailability and accumulation of contaminants in the soft tissue of molluscs could be affected by abiotic factors, such as food availability, pH and temperature, and also by biotic factors, such as the seasonal changes of flesh weight in molluscs. In this context, some issues could arise especially when comparing different sites in a long-term biomonitoring with data obtained from different periods of the year. In this study, bioaccumulation of metals, Polycyclic Aromatic Hydrocarbons (PAHs) and Butyltins (BTs) in Manila clams was monitored twice a year, from November 2010 to June 2015, in order to assess impact of human activities on *R. philippinarum* population from the Vallona Lagoon, a transitional area located in the delta of the Po River (NE, ITALY) which is the largest and most important Italian watercourse and one of the main sources of contaminants to Adriatic environments. Although levels were quite consistent with those reported from other geographical areas with low to medium pollution, seasonal trends were showed for each contaminant with higher concentrations on autumn rather than on spring surveys. The physiological condition of clams was also examined through two indices (condition index and survival in air) and they both exhibited seasonal variations connected to natural endogenous and exogenous factors. Physiochemical parameters of water (temperature, pH, salinity and dissolved oxygen) were also considered. To ensure that bioavailability assessment was not affected by seasonal variation of soft tissues of molluscs, Contaminant/shell weight indices, which consist of normalization achieved multiplying each contaminant concentrations by the condition index, were applied. Accordingly, the normalization enabled us to highlight the contaminant uptake from clams in some particular periods and to compare different sites in a long-term biomonitoring with data obtained from different periods of the year. Indeed, some contaminants showed quite a steady state all over the monitoring period and at the different sites, whilst others, such as Arsenic, Chrome, Nickel, Lead, Copper, Zinc and BTs, showed different patterns of bioaccumulation with some periods presenting enhanced concentrations probably related to anthropogenic activities.

WE342

Biomonitoring of Singapore mangroves using biomarker expression and contaminant burden in caged green mussels, Perna viridis.

G. Juhel, National University of Singapore / Tropical Marine Science Institute; S. Bayen, McGill University / Singapore-Delft Water Alliance; E. Segovia, C. Goh, W. Lee, National University of Singapore; B.C. Kelly, National University of Singapore / Civil & Environmental Engineering

Mangroves are fragile coastal ecosystems whose ecological and socioeconomic importance for adjacent ecosystems and local populations is being clearly recognized nowadays. Mangroves are one of the most threatened tropical environments and our understanding of the impact of chemical pollution on these ecosystems is still at its infancy. In this study, nine mangroves sites were selected around Singapore coastline to cover various contamination profiles, and green mussels, *Perna viridis* were used as bioindicators. Bivalves were deployed in cages at each of the nine mangrove patches for 28 days on two consecutive years (one year during the monsoon period and the other year during a dry-weather period), and collected for subsequent analysis. A series of biochemical and cellular biomarkers were developed and measured using various organs. Metallothioneins (MTs), Glutathione-S-Transferase (GST), Ethoxyresorufin O-deethylase activity (EROD), Vitellin-like proteins (Vn), inhibition of Acetylcholinesterase (AchE) were measured in the bivalves' soft tissues. Mussel’s haemolymph was also used to evaluate various immunological parameters (Total Haemocyte counts, phagocytosis and lysozyme levels) and the level of haemocytes’ DNA damage, using the Comet assay. Results of this study revealed different profiles of biomarker expression between the various sites. Most notably, metallothionein induction was observed at some the sites, indicating potential exposure to heavy metals while higher levels of DNA damage and EROD were also recorded at some of the mangrove patches pointing towards possible exposure to organic contaminants. Some biomarkers appeared to be subject to seasonal variations while others were very stable. Possible correlation between biomarker expression and the level of various contaminants (metals, PAHs, pharmaceuticals, endocrine disruptive chemicals, personal care products) in caged mussels were also studied. Using an Integrated Biomarker response index, the various mangrove sites were eventually ranked amongst each other. Our findings ultimately indicated a clear segregation of mangrove sites, indicating that some mangroves patches were potentially more at risk than others towards chemical contamination.

WE343

Impacts of climate change on mercury bioaccumulation in large ocean predators

E.M. Sunderland, Harvard University / School of Engineering and Applied Sciences; A.T. Schartup, Harvard T.H. Chan School of Public Health / Department of Environmental Health; C. Thackray, Harvard University / School of Engineering and Applied Sciences; A. Qureshi, IIT Hyderabad / Civil Engineering; C. Dassuncao, Harvard University

Many marine ecosystems are being affected by climate driven changes in freshwater discharge, circulation, productivity and seawater temperature. Large ocean predators such as tunas account for a large proportion of methylmercury exposure in many countries (e.g., almost 40% of U.S. population wide intake between 2010-2012). Methylmercury is a potent neurotoxicant, particularly for children, and has been associated with impaired cardiovascular health in adults. We have developed a new mechanistic model for methylmercury bioaccumulation in marine food webs (BAM^3). The model is driven by methylmercury concentrations in seawater and ocean biogeochemical conditions (temperature, dissolved organic carbon: DOC, and trophic status). Here we apply this model to investigate the magnitude of changes in methylmercury concentrations expected from decadal oscillations in seawater temperature in the North Atlantic. We compare the magnitude of these changes to those occurring in the Pacific Ocean and discuss how climate related variability is likely to affects exposures of humans and wildlife to methylmercury. Our results suggest changes in tissue burdens driven by oscillations in seawater temperature are similar in magnitude to those that have been achieved by reductions in emissions in the North Atlantic.

WE344

Chemical stress on aquatic communities under semi-arid conditions: towards an improved multimetric approach

A.A. Sanchez, IMDEA Water Institute / Aquatic Ecotoxicology; C. Alonso, P. García, A. Romero, A. Castaño, I. López, L. Nozal, IMDEA Water Institute; M. Vighi, IMDEA Water Institute / Earth and Environmental Sciences; A. Rico, IMDEA Water Institute / Aquatic Ecotoxicology

Combined effects of chemical pollution and water scarcity are one of the most important threats to aquatic ecosystems in (semi-)arid regions of the world. The low dilution potential of chemicals (urban, industrial and agricultural) discharged in aquatic ecosystems under water scarcity conditions could lead to devastating toxic effects. Moreover, a large proportion of aquatic bodies in these regions are known to have a high hydrological variability in a temporal scale. In this sense, communities naturally adapted to this condition are expected to respond differently to chemical stress than those adapted to more constant water flows. The aim of this study was to evaluate the combined effects of multiple stressors on aquatic ecosystems in scenarios that are characteristic of (semi-)arid regions. In particular, this study focused on identifying the main stress factors that are influencing aquatic communities in the semi-arid region of Madrid (central Spain). Sixteen sites were selected in the watershed of the Tagus River (Madrid, Spain) and sampled in three seasonal periods (spring, summer and fall). Hydrological and physico-chemical parameters of aquatic ecosystems were monitored, together with concentrations of metals and organic contaminants (pharmaceuticals, home-care products, pesticides). With respect to organic contaminants, the results from a screening analysis revealed the presence, at detectable levels, of 100 compounds in water samples. A group of 42 contaminants were selected for quantification due to their high toxicity potential to aquatic organisms and frequent detection. Complex mixtures of pharmaceuticals, as well as highly toxic pesticides were identified. Through a multivariate analysis including pollution data, flow variability and related physico-chemical parameters, the main stressors and possible differences at a temporal and spatial scale were evaluated at a taxonomic and at a biological trait level. Significant responses to multiple stressors from some invertebrate taxa and functional traits (feeding habits, reproduction and respiration) were determined. Based on these results, suggestions for a biological vulnerability multimetric index, which considers more site-specific conditions, will be presented.

WE345

Long- term effects on transplanted caged- freshwater bivalves Diplodon chilensis to the assessment of water quality in a Patagonian river

M.S. Yusseppone, Facultad de Ciencias Exactas y Naturales Universidad de Buenos Aires / Department of Biochemistry, IQUIBICEN-CONICET, FCEN-UBA; S.E. Sabatini, IQUIBICEN CONICET Universidad de Buenos Aires / Department of Biochemistry; V.A. Bianchi, Universidad Nacional del Comahue, Neuquén / INIBIOMA-CONICET, CEAN; J.M. Castro, INIBIOMA CONICET - CEAN / Laboratorio de Ecotoxicología Acuática INIBIOMA; C.M. Luquet, CONICET / Laboratoy of Aquatic Ecotoxicology, INIBIOMA; I. Rocchetta, IQUIBICEN CONICET Universidad de Buenos Aires / Department of Chemistry Biochemistry; M.d. Rios de Molina, Universidad de Buenos Aires, Facultad de Ciencias Exactas y Naturales / Departamento de Química Biológica, CONICET-UBA, Instituto de Química Biológica - Ciencias Exactas y Naturales (IQUIBICEN)

Population growth and human activity are contributing to habitat deterioration in Patagonian rivers with the consequent threat to water quality and biodiversity. The bivalve *Diplodon chilensis* is a key species in the macroinvertebrate fauna of Patagonian lakes and rivers which has been proposed as a sentinel organism in the area. We evaluated the long-term effects of transplantation of caged *D. chilensis* to different sites in the Chimehuin river (reference site (S1), downstream from an aquaculture facility (S2), and downstream from an open dump and from the sewage treatment plant (S3)) after 3, 6, 9 and 12 months of exposure. We combined the antioxidant response, oxidative damage, ROS production and energetic status, with water and sediment analysis (physic-chemical and biological variables, and organic matter content). Physic-chemical variables varied according to site and time of exposure. Sites S2 and S3 showed generally higher chlorophyll a concentration and total coliform bacteria values compared to site S1, whereas organic matter content in the sediment was elevated only at site S2. In *D. chilensis*, gill SOD and GST activity was higher in both S2 and S3 than in S1 by the end of the exposure time. During the last month of exposure (month 12), GSH levels dropped dramatically and lipid peroxidation levels increased in individuals from S2 and S3 sites. Digestive gland factor (DGF) and energy values in digestive gland were increased at sites S2 and S3, from 6 to 9 months of exposure. Our results indicate that despite the large flow rate of Chimehuin river water quality is deteriorated in areas impacted by anthropic activities (aquaculture, solid waste disposal and sewage). This effect is reflected by a physiological response of *D. chilensis*, which is especially significant during period of their highest metabolic activity (austral fall/ winter).

WE346

The influence of selected seasonal and anthropogenic phenomena on a perennial river in South Africa.

G. Van Niekerk, North West University (Potchefstroom Campus) / Zoology-School of Biological Sciences; C. Wolmarans, H. Pienaar, North-West University - School of Biological Sciences / Zoology

The quality of surface waters worldwide is declining fast. This is due to anthropogenic impacts, climate change and natural occurring floods and droughts which are predominant abiotic agents of disturbance in intermittent streams. The objective of this study was to establish whether seasonal fluctuations can reduce the effect of anthropogenic impacts on the river and whether this is reflected by macroinvertebrate assemblages, physico-chemical water parameters and metal concentrations in the water and sediment. Water quality and macroinvertebrate community data were collected in the Crocodile River (South Africa). Four surveys were conducted, two during the low-flow and two during the high-flow season. Spatial and temporal variations were found with regard to suspended solids and total organic material. The low flow surveys had a slight increase in suspended solids and total organic material, whereas a substantial increase occurred during the high flow seasons. The highest concentrations of metals in the water column were in contrast to the sediment concentrations observed during the high-flow seasons at the majority of the sites. Only minor differences in sediment metal concentrations were noted between high and low flow seasons. The pH values increased from the origin of the river downstream. However this did not result in a clear trend with regard to either an increase in sediment metals or a decrease in the concentration of dissolved metals. With regard to the electrical conductivity, the highest values were obtained during the low flow seasons, when compared to the high flow seasons. A combination of biological indices (Taxa Richness, Shannon-Wiener diversity index and Pielou's evenness index) were calculated for each site and season, and no significant differences were found between the high and low flow seasons for any of the indices at each of the sites (P>0.05). The highest percentage of families at all the sites and seasons were classified as highly tolerant and tolerant to organic enrichment. It can be concluded that the high flow seasons (associated with rain and floods) did not have a rejuvenating effect on the river, as mentioned in previous studies. This phenomenon is substantiated by the metals concentrations, total organic matter, total suspended solids, electrical conductivity and the fact that all the taxa collected in large numbers during both seasons were tolerant to highly tolerant.

WE347

Growth, Photosynthetic and Antioxidative Defense System Response of Hordeum vulgare to Combined stress of Heat wave and Drought

A. Diksaityte, Vytautas Magnus University; R. Dagiliute, Vytautas Magnus University / Environmental Science Department; L. Kubilė, D. Miškelytė, G. Juozapaitienė, Vytautas Magnus University

Extreme climatic events such as heat waves and drought periods are predicted to increase in frequency and severity in many regions under future climate scenarios, and in natural environment these two abiotic stresses often occur simultaneously. The short-term (3 day-long) impact of +10 °C heat wave treatment 6.5 h per day was investigated on *Hordeum vulgare* under well-watered and water deficit conditions in Closed-top chambers under controlled environment. The decreases in shoots dry weight, shoots length and leaves area were observed in the water deficit treatment after exposure to heat wave, while all these parameters in the well-watered treatment were not affected significantly. The decline in shoots growth under water deficit conditions was most likely caused by a considerably greater reduction in photosynthetic rate as well as far stronger oxidative stress caused by combined impact of heat weave and drought than that from single heat wave treatment as revealed by higher level of malondialdehide content and considerably stronger stimulation of antioxidative enzymes. Full recovery of physiological processes and membrane damage were observed in well-watered treatment after one-day regeneration period. In contrast, neither shoots dry weight nor leaves area as well as most physiological processes analyzed, membrane damage, and catalase activity in water deficit treatment were not recovered to the control value. The obtained results showed that drought-stressed *Hordeum vulgare* plants suffered markedly stronger physiological and oxidative stress caused by interactions of two abiotic stressors, as compared to single stress treatment, and revealed an importance of soil water availability even during the short-term heat wave period. **Keywords:** *Hordeum vulgare*, heat wave, drought stress, antioxidant system,photosynthesis, growth

WE348

Does elevated CO2 protects plants against heat waves damage?

J. Zaltauskaite, Vytautas Magnus University / Department of Enviromental Sciences; G. Sujetoviene, I. Januskaitiene, A. Diksaityte, D. Miškelytė, G. Kacienė, G. Juozapaitienė, R. Juknys, Vytautas Magnus University

The frequency and severity of heat waves is increasing as a result of climate change. These extreme events may lead to decreased crop productivity and financial incomes. Differential responses of crops and weeds to heat waves and CO_2 may also cause shifts in their competitive interactions. The aim of the study was to examine the influence of extreme events (heat wave plus drought) and CO_2 on the growth of spring barley (*Hordeum vulgare* L.) and wild mustard (*Sinapis arvensis* L.). Barley and wild mustard, growing together in the microcosms at the combination 2:1, were subjected to short-term heat wave in combination with drought (35°C vs. 21°C) under ambient CO_2 (400 ppm) and elevated CO_2 (800 ppm). The growth and response of photosynthesis system of both plant species were evaluated.

WE349

Combined effects of increasing temperatures, drought and an insecticide on freshwater zooplankton communities: a microcosm study

A.A. Sanchez, IMDEA Water Institute / Aquatic Ecotoxicology; I. López, L. Nozal, IMDEA Water Institute; M. Vighi, IMDEA Water Institute / Earth and Environmental Sciences; A. Rico, IMDEA Water Institute / Aquatic Ecotoxicology

Water scarcity and chemical pollution are two of the main groups of stressors causing ecological impairment in freshwater aquatic ecosystems of (semi-)arid regions. Predictions about human and climatic pressure on water resources in these regions reveal that the interaction between these two groups of stressors will increase in the nearby future. In line with that, advances in ecological risk assessment recognise that stress factors harming aquatic ecosystems rarely operate individually. Therefore, new approaches to assess interactions between multiple stressors are needed. In this study we evaluated the combined effects of the insecticide lufenuron and two additional stress factors: increasing water temperatures and droughts. Twenty-seven microcosms were stocked with pond water, sediment, and a homogeneous plankton assemblage. Three environmental scenarios were simulated: 20°C and 28°C without desiccation, and 28°C with desiccation. The experiment was performed in triplicate with three insecticide treatments (Control, Low and High Concentration) in each environmental scenario. The insecticide was applied twice, with a 10 day interval between applications. Test units without desiccation were refilled twice per week with distilled water. Test units exposed to drought stress were not refilled and allowed to desiccate. After that, these units were refilled up to the initial level and maintained for two more weeks. Lufenuron concentrations in water and sediments were periodically measured, together with water quality parameters (DO, pH, T, EC and nutrients). Zooplankton composition was determined on a weekly basis, and the isolated and interactive effects of temperature, drought and lufenuron were evaluated using suitable statistic techniques. Effects were assessed at the community and at the

population level. Lufenuron was the main stressor in all the environmental scenarios, with a significant decrease of Cladocera and Copepoda, and an increase of Rotifera. Temperature and drought had slight effects on community composition and accelerated insecticide dissipation, influencing community recovery capacity. Interaction between factors at community and population level was mainly observed at the beginning of the experiment. Direct and indirect responses at population level varied between environmental scenarios. The results of this study contribute to understand differences in vulnerability of aquatic ecosystems to multiple stressors in (semi-)arid regions.

WE350

Toxicity of phenoxy herbicide: the effects of elevated temperature and CO2 concentration

J. Zaltauskaite, Vytautas Magnus University / Department of Enviromental Sciences; G. Sujetoviene, A. Diksaityte, I. Januskaitiene, G. Kacienė, G. Juozapaitienė, D. Miškelytė, Vytautas Magnus University; S. Sakalauskienė, J. Miliauskienė, Institute of Horticulture, Lithuanian Research Centre for Agriculture and Forestry; R. Juknys, Vytautas Magnus University

Climate change is a major concern for agriculture and crop productivity. Crop productivity strongly depends on crop protection measures such as use of herbicides. Climate change will influence the fate and ecotoxicity of herbicides by altering their environmental partitioning and degradation, distribution and abundance of weeds and growth and development of weeds and crops. Differential responses of crops and weeds to elevated temperature and CO_2 may also cause shifts in their competitive interactions. The aim of the study was to examine the influence of elevated temperature and CO_2 on the effects of phenoxy herbicide to spring barley (*Hordeum vulgare* L.) and common lambsquarter (*Chenopodium album* L.). Two climate scenarios were investigated: current climate (21 °C, 400 ppm CO_2) and future climate (25 °C, 800 ppm CO_2). The terrestrial target *Ch. album* and non-target *H. vulgare* plants, growing together in the microcosms at the combination 2:1, were sprayed with herbicide sprays solutions equivalent to 0.5-2 of field application rate. The plants were sprayed at the four- to five-leaf stage with herbicide solutions prepared with 4-Chloro-2-methylphenoxyacetic acid (MCPA, CAS No. 94-74-6, Sigma-Aldrich). The growth and response of antioxidative defence system of both species were evaluated.

WE351

Combined effects of insecticide exposure and predation risk on freshwater detritivores

A. Rodrigues, University of Aveiro / Biology Department and CESAM; M.D. Bordalo, University of Aveiro; O. Golovko, O. Koba, University of South Bohemia in Ceske Budejovice / South Bohemian Research Center of Aquaculture and Biodiversity of Hydrocenoses; C. Barata, CSIC / Environmental Chemistry; A.M. Soares, University of Aveiro / department of Biology & CESAM; J. Pestana, CESAM & University of Aveiro / Biology

Exposure to sub-lethal concentrations of insecticides are known to pose at risk non-target insects due to effects on physiology and behaviour. Under natural conditions, the negative effects of pesticides may be magnified by the presence of natural stressors, such as predation. Perception of predation risk alone may change the behaviour and physiology of prey species, impacting their fitness and, thus alter their susceptibility to chemical exposure. There are thus growing efforts to understand how the combined effects of toxicants and biotic stressors may affect populations, food web dynamics and ecosystem functioning. Chlorantraniliprole (CAP) used here as a model compound is an anthranilic diamide largely applied due to its specificity for insect ryanodine receptors of target species. So, to assess the combined effects of insecticide exposure under predation risk on freshwater detritivores we studied the behavioural and developmental responses of *Chironomus riparius.* Plus, we tested whether the responses of the *C. riparius*, a collector, would change in the presence of a shredder species and the associated production of fine particulate organic matter (FPOM). For that, trials were performed using a simplified trophic chain: *Alnus glutinosa* leaves as food resource, the shredder *Sericostoma vittatum*, the collector *C. riparius* and their natural predator the dragonfly *Cordulegaster boltonii*. A full factorial design tested the effects of the CAP (0 or 2 µg/L), presence/absence of the predator *C. boltonii* and of the shredder *S. vittatum* on leaf mass loss and *C. riparius* growth. Results show that exposure to an environmentally relevant concentration of CAP decreased leaf decomposition in all treatments. Predation risk marginally reduced shredder effects on leaf decomposition. Considering detritivores interaction, an interspecific competition is suggested since the presence of shredders impaired chironomids performance despite the increased in leaf fragmentation. *C. riparius* growth rate was thus decreased independently by all factors (CAP exposure, predation risk and shredder presence) and a marginal interaction between CAP and predation risk was also observed. To conclude, this study highlights the need to consider natural biotic stressors and species interactions in risk assessment of chemical pollution, since both vertical and horizontal diversity play their role on response to stress.

WE352

How sugarcane and high temperatures are contributing to amphibian declines in Brazil? Morphological, biochemical and molecular approaches

J. Freitas, University of Sao Paulo - USP / Department of Hydraulic and Sanitation; E.A. de Almeida, Fundação Universidade Regional de Blumenal; D. Schlenk, University of California-Riverside / Department of Environmental Sciences; E. Espindola, University of São Paulo USP / Hydraulics and Sanitation

Sugarcane is the most efficient first-generation source of ethanol in the market, which has contributed to the rapid expansion of its crops and generated concerns related to its environmental impacts to Brazilian territory. In tropical areas, agricultural activities of sugarcane are intensified during the rainy season, which coincides with the period of occurrence and reproduction for most amphibian species. Tadpoles and other aquatic animals from tropical areas of the world also experience large temperature fluctuations in their habitats. So, there is a great concern that amphibians are not only being affected by pesticides in their environments, but also by the combined effects of pesticides and temperature changes. In this study, we evaluated biochemical, morphological and molecular effects caused by the exposure to herbicides used on sugarcane crops in Brazil (diuron and its metabolite, 3,4-DCA, clomazone and sulfentrazone) on tadpoles of different species, using different thermal gradients. Our results showed that temperature is an important factor influencing the toxicity of pesticides in tadpoles. Diuron combined to higher temperatures accelerated metamorphosis process in tadpoles of *Lithobates catesbeianus*. T_3 levels and metamorphosis-genes expression (*dio2, dio3, thibz, trα, trβ* and *klf9*) were mostly upregulated in these groups, showing disrupting effects of diuron for amphibians. 3,4-DCA presented similar responses to diuron on *L. catesbeianus* and its effects were also pronounced at high temperatures. Native tadpoles of *Rhinella schneideri* and *Eupemphix nattereri* had their antioxidant defense system affected by exposure to the herbicides clomazone and sulfentrazone. Sulfentrazone and clomazone altered antioxidant (SOD, CAT, G6PDH) and biotransformation enzymes activities and induced lipid peroxidation with temperature associated responses in tadpoles of both species. Clomazone also increase carboxylesterase activities in tadpoles exposed at higher temperatures. Integrated Biomarker responses (IBR) index showed a synergic effect of temperature and sulfentrazone or clomazone in *R. schneideri* and *E. nattereri*. Our data showed that sugarcane pesticides impair several physiological aspects of tadpoles, and its effects can be pronounced at higher temperatures. These findings imply that the effects of abiotic factors should be taken into account to evaluate the real risks of exposure of amphibians to commonly used pesticides, mainly in tropical areas.

WE353

Adaptation vs. acclimation of natural phytoplankton communities towards herbicide exposure

S. Rizzuto, Lancaster University / Lancaster Environment Centre; D. Baho, NIVA Norwegian Institute for Water Research; K.C. Jones, Lancaster University / Lancaster Environment Centre; E. Leu, Akvaplanniva AS; L. Nizzetto, NIVA

Freshwater ecosystems are subject to natural and anthropogenic disturbances such as climate change, landscape management, natural resources overexploitation and also pollution. Chronic background contamination by pesticides applied in agriculture poses a selective pressure on natural phytoplankton communities, favouring species and strains that can handle herbicide exposure better than others. Concomitantly, there might be a development of resistance towards a specific substance over time. It can be expected, however, that the community resulting from the historic exposure to the stressors is not uniformly well optimized to utilize available resources in the best possible way. The hypothesis behind this study is drawn from the following concept: The history of community exposure to chemical pollution in the environment influences the sensitivity of responses to contemporary stressors. Such trade-off may persist for a variable amount of time, depending on whether it is underpinned by purely ecological or evolutionary processes and on the ability of the ecosystem of recruiting diversity and the structure necessary to cope with new environmental conditions. In order to assess these hypotheses we have studied the effect of long-term adaptation vs. acclimation in a two phase community level experiment with natural phytoplankton communities from a pristine and an agricultural catchment. Using a controlled experimental setup, phytoplankton communities were germinated from sediments with and without herbicide exposure (Isoproturon, nominal concentration – 12 µg/L) in phase 1. Afterwards (Phase 2), we subjected the resulting communities to a stress experiment where we applied 4 different concentrations of the same herbicide (nominal concentrations of 7, 12, 61, 92 µg/L), and followed the algal response with respect to biomass development, photosynthetic yield and resource use efficiency (RUE). First results indicate less pronounced differences between the community responses from the two different locations than anticipated. At the same time, we see indications for a mitigating effect of herbicide exposure during germination (Phase I).

WE354

Impacts of climate change on freshwater pesticide exposure

T. Sinclair, University of Sheffield / Animal and Plant Sciences; A. Boxall, University of York / Environment Department; L. Maltby, The University of Sheffield / Dpt. of Animal & Plant Sciences; S. Beulke, Enviresearch / Food and Environmental Safety Programme; R. Williams, Centre for Ecology & Hydrology Maclean Building

Climate change will modify environmental conditions which will likely have knock-on effects on the usage and environmental fate and behaviour of active

pesticide ingredients. Temperature, rainfall, soil parameters, pest ranges and cropping patterns are all expected to alter under climate change conditions, and all of these parameters could affect pesticide use and environmental exposure patterns which will alter the risk that these compounds pose to the environment. Here, we report the results of a study to evaluate the impacts of climate change on the exposure of aquatic systems in the UK. Concentrations of a number of case study pesticides, covering a range of physico-chemical properties and uses, were modelled in streams in different regions in the UK using two models recommended by the Forum for the Co-ordination of Pesticide Fate Models and their Use, namely MACRO and PRZM. Model predictions were obtained for current conditions and for expected conditions in the 2080s by parameterising the models for current and future predicted weather conditions, cropping patterns, soil properties and pesticide use patterns. The results show that exposure patterns of the APIs are likely to change in the future. These changes in exposure vary by pesticide type and the region modelled with exposure in some chemical-region combinations increasing and in others decreasing.

WE355

Ranking micropollutants in effluent by exposure indices evaluated via suspect/nontarget screening

P. Naree, Changwon National University / Environmental Engineering; c. younghun, Changwon National University / FEED of Eco-Friendly Offshore Structure; J. Jeon, Changwon National University / Environmental Engineering

Information on the occurrence and concentration of micropollutants in effluents from wastewater treatment plants (WWTPs) provides important clues for evaluating the risk and availability of river ecosystems. However, the chemical monitoring work for the numerous trace contaminants is time-consuming, labor-intentive, and cost a lot. To overcome the problems, the effluent monitoring programs have been conducted for limited, but prioritized pollutants. In general, the prioritization has been mainly based on effect/toxicity information rather than exposure relevant indices. Thus, risky pollutants with high occurrence frequency and concentration have often been underestimated when their effect/toxicity are neither considerable nor well defined. In the present study, a list of prior effluent micropollutants is suggested with a exposure-index based scoring/ranking procedure following qualitative chemical analysis. A scoring table with exposure indices such as occurrence frequency and chromatogram peak area was applied for the ranking. WWTP effluent samples were taken in september, 2016 and analyzed via suspect/nontarget screening using LC-HRMS (QExactive+ Orbitrap). Within a suspect list, about 60 compounds were tentatively identified and ranked by the score. After purchasing reference standards for high rankers, about 20 micropollutants were orthogonally confirmed and roughly quantified. The quantified micropollutants were mostly pharmaceuticals and personal care products including 9 groups such as analgesics/anti-inflammatories (acetaminophen, mefenamic acid), antibiotics/antifungal (climbazole, fluconazole, sulfamethoxazole, sulfamethazine), anticonvulsant (carbamazepine, carbamazepine-epoxide, oxcarbazepine), antihistamines (diphenhydramine, fexofenadine), antihypertensive agent (irbesartan, valsartan), antipsychotic (amisulpride), CNS stimulants (caffeine), and antiulcer (cimetidine). The concentrations for the top ranker, acetaminophen detected in all 7 samples, was ranged up to 1,300 ng /L. The 2nd ranking pollutant was caffeine and followed by cimetidine> mefenamic acid>fexofenadine>carbamazepine>irbesartan>fluconazole>dephenhydramine> sulfamethoxazole. Since some tentatively identified pollutants were left unconfirmed, the priortized compound list should be updated along with additional confirmations. Nevertheless the ranked list still include highly exposable micropollutants which are worthy for intensive monitoring in effluents.

WE356

Interspecific effects of temperature shifts on life parameters, oxidative stress, and expression of fatty acid synthesis genes and heat shock protein genes in two congeneric copepods Tigriopus sp.

J. Han, Sungkyunkwan University / Biological Science; J. Lee, Sungkyunkwan University

In this study, we compared the effects of temperature changes on lifecycle parameters, intracellular reactive oxygen species (ROS) levels, glutathione *S*-transferase (GST) enzymatic activity, and gene expression profiles of both the *de novo* lipogenesis (DNL) pathway and heat shock protein (*hsp*) genes in the temperate copepod *Tigriopus japonicus* and the Antarctic copepod *Tigriopus kingsejongensis*. The median lethal temperature (LT50) and no observed effect level (NOEL) in *T. japonicus* were determined to be 35.3ºC and 32ºC, respectively, in response to temperature increase of 2°C per day. In *T. kingsejongensis*, LT50 and NOEL were determined to be 24.8ºC and 12ºC, respectively. Levels of ROS and GST activity were slightly elevated (<em>P

WE357

Effects of water browning on zooplankton physiology and fitness driven by food characteristics in a long-term enclosure experiment

L. Minguez, LIEC (CNRS UMR 7360, Université de Lorraine) / Laboratoire Interdisciplinaire des Environnements Continentaux, CNRS UMR 7360; E. Sperfeld, University of Oslo / Centre for Ecological and Evolutionary Synthesis (CEES), Department of Biosciences; S.A. Berger, J.C. Nejstgaard, Leibniz Institute of Freshwater Ecology and Inland Fisheries / Dept. Experimental Limnology; M.O. Gessner, Leibniz Institute of Freshwater Ecology and Inland Fisheries IGB

Ecotoxicological assays using *Daphnia* species are generally performed under optimal food and light conditions. However, results of such assays may not adequately reflect stress responses in the wild, since the ability of organisms to cope with adverse conditions critically depends on the amount of available energy. One type of potential stressors that is becoming increasingly important is terrestrial-derived dissolved organic carbon (tDOC) that causes browning of lakes and streams, but long-term effects of tDOC on freshwater organisms are not sufficiently known. Using a combination of an *in situ* enclosure experiment and laboratory incubations, we tested whether long-term tDOC exposure affects the physiology and life-history traits of the waterflea *Daphnia longispina*, and whether any observed effects are reversible. Daphnids were collected from a long-term, large-scale enclosure experiment conducted in a clear-water lake in northeastern Germany, where a natural plankton community was exposed to a standard source of tDOC (HuminFeed®). The physiological state of daphnids in the enclosures was followed after addition of the tDOC. In the first week of exposure, daphnids experienced oxidative stress, i.e. an imbalance in favour of oxidative damage, but this response was no longer observed after 36 days. Daphnids and water from the enclosures sampled again after 10 weeks were used to assess survival and reproductive performance under laboratory conditions. Both survival and reproduction were related to seston characteristics and the elemental and biochemical composition of the daphnids. Surprisingly, daphnids kept in brown water (B) showed higher fitness (better survival and reproduction) than daphnids in clear water (A). This unexpected outcome is explained by higher seston quantity and quality in enclosures receiving tDOC, related to a higher abundance and biomass of nutritious food algae. Moreover, transplantation of daphnids from A to B enclosure water and vice versa revealed considerable plasticity, as the daphnids were capable of rapidly adjusting their metabolism to a similar level as that observed in the specimens exposed already previously to the recipient environment. Thus, our study points to the importance of accounting for exposure duration and food quality and quantity when assessing *Daphnia* responses to environmental stressors such as tDOC.

WE358

Interactive effects of multiple stressors on estuarine processes

A. O'Brien, The University of Melbourne; K. Dafforn, Macquarie University / Evolution and Ecology Reserach Centre; M. Mayer-Pinto, University of New South Wales / Evolution   Ecology Research Centre School of Biological Earth and Environmental Sciences; E.L. Johnston, University of New South Wales / Evolution and Ecology Research Centre; A. Chariton, Macquarie University / Molecular Ecology and Toxicology

Natural systems are threatened by a variety of different anthropogenic stressors. These are often considered in isolation, but in reality most are found in combination and have the potential to interact with different outcomes. Urban systems such as estuaries and harbours are commonly exposed to chemical (e.g. contaminants), physical (e.g. built infrastructure) and biological (invasive species) stressors. It is important to understand the interactive threats posed by these stressors. Here we use a systematic literature review to explore selected urban stressors and their potential interactions under current envrionmental conditions. We link the interactive effects to responses in key ecological processes including biogeochemical cycling, primary production and ecological interactions. Using functional endpoints that are common across systems we have the ability to identify relevant patterns and trends. This allows us to go beyond comparisons that rely on community structural endpoints and are potentially only relevant at local scales.

WE359

Ecology or reproducibility crisis? - Lessons from a laboratory scale tri-trophic test system

V. Riedl, Environment Department, University of York / Environment Department; A. Agatz, IBACON GmbH / Environment Department; R. Benstead, Fera Science Ltd / Centre for Chemical Safety and Stewardship; R. Ashauer, University of York / Environment

In recent years, concerns have been raised regarding a lack of reproducibility in scientific research. There is indeed evidence that a number of research findings are not reproducible by others or even within the original laboratories. Yet, while the reproducibility of results might often be difficult, it is essential in the context of regulatory decision making. In the environmental risk assessment of pesticides, for instance, replication, standardization and reproducibility are of great importance to increase the statistical power and the robustness of test findings. For this reason, rapid single-species tests that only allow for the assessment of direct pesticide impacts are still more frequently used than multi-species systems. Although they are ecologically more relevant, micro-/mesocoms often yield lower statistical power due to higher complexity, difficulty of standardization, resource demand and variability among replicates. However, growing evidence suggests that direct effects measured at the individual level do not proportionally translate to impacts at the population and community level. The use of testing procedures that are ecologically more realistic but ideally comply with regulatory needs should thus be a priority to risk assessors and regulators. The tri-trophic aquatic test system

TriCosm (*P. subcapitata*, *Ceriodaphnia*, *Hydra*) was developed as an intermediate link between simple single-species tests and complex multi-species systems, to detect small stressor-induced alterations in ecological interactions. The achievement of standardization, replication and reproducibility was given close attention during the development of the system, yet, the TriCosm was found to be compliant in terms of repeatability and reproducibility only in the short term. Here we present experiments designed to discern effects of variation due to ecologically interacting factors that impact on the community dynamics in this aquatic multi-species system.

WE360

Improving tolerance to natural and chemical stressors by inducing early life stress in the rotifer Brachionus sp. Cayman

L.G. Almeida, MARE - Marine and Environmental Sciences Centre / Instituto Politécnico de Leiria; C. Ferreira, Polytechnic Institute of Leiria / Polytechnic Institute of Leiria; M.F. Lemos, Instituto Politécnico de Leiria / MARE IPLeiria; H.N. Cabral, Faculdade de Ciências da Universidade de Lisboa / MARE-FCUL; P. Bossier, Universiteit Gent / Laboratory of Aquaculture & Artemia Reference Center; S.C. Novais, Polytechnic Institute of Leiria / MARE IPLeiria

Rotifers are widely used as bio-indicators and models for ecotoxicology due to characteristics such as high ingestion rate, rapid growth, ease of culture in small volumes, ease of establishing clone cultures, short generation time, small size, and sensitivity to various toxicants. The monogonont rotifer *Brachionus plicatilis* (Müller, 1786) is a euryhaline species, typically found in salt lakes and coastal brackish waters, presenting high commercial importance and value, since it is commonly used as live feed for several marine species larvae in aquaculture productions. In the last decade it has become commonly accepted that environmental stimuli can induce phenotypic alterations in the organisms. Thereby, the primary objective of this project was to assess if temperature shocks during early life stages of rotifer development could improve their tolerance to different natural and chemical stressors later in life. Firstly, 24h acute toxicity bioassays and 48h chronic toxicity bioassays were conducted with two strains of *Brachionus* sp. Cayman (MRS10 and IBA3), a biotype within the *B. plicatilis* complex, obtained from the Laboratory of Aquaculture and Artemia Reference Center (Ghent University, Belgium). The bioassays were performed in accordance with the International Standards ISO 19820 and ISO 20666, respectively, using 25ºC and 25 psu as control environmental conditions. The organisms were exposed to different concentrations of antibiotics, pesticides, and oxidative stress inducers, as well as to different salinity and temperature conditions. Consequently, to test for possible increased tolerance to these stressors, neonates (0-4h) were exposed to short non-lethal temperature shocks (cold and heat), and after a recovery period, survival tests were performed and sensitivity values compared. Results show that temperature shocks during early life stages result in changes in the sensitivity of rotifers to different stressors. These are very promising results, and their application in an aquaculture context could be advantageous to decrease the mass mortality in rotifer production. Future research within this project will address possible epigenetic mechanisms (DNA methylation or histone modifications) behind those greater tolerances and assess if their maintenance can be achieved through several generations.

WE361

Effects of a mixture of pharmaceuticals in a freshwater model ecosystem

S. JOACHIM, INERIS-UMR SEBIO / CIVS; V. David, INERIS; K. Nott, Société Wallonne des Eaux; a. chaumot, Irstea / UR MALY Laboratoire Ecotoxicologie; H. QUEAU, N. Delorme, Irstea Lyon / UR MALY Laboratoire Ecotoxicologie; K. Sossey, Université de Liège ULg; P. Baudoin, C. TURIES, INERIS / INERIS UMRI SEBIO ECOT; A. Catteau, A. Bado-Nilles, INERIS; M. Fouarge, Unamur; O. Geffard, Irstea / UR MALY Laboratoire Ecotoxicologie; J. Porcher, INERIS / INERIS UMRI SEBIO ECOT; A. Geffard, Université de Reims Champagne Ardenne; F. De Laender, Université de Namur ASBL / Research Unit in Environmental and Evolutionary Ecology; R. Beaudouin, INERIS / Models for Ecotoxicology and Toxicology METO

Owing to their ecological importance, freshwaters provide important services which leads to a strong societal demand concerning the preservation of their quality. They are the receptors of many contaminants emitted by human activities and more specifically by wastewater treatment plants effluents. Water resources of the Meuse watershed are used on both sides of the French-Belgian border which involves a coherent and joint management. In that framework, the DIADeM project suggests developing and spreading out a cross-border multidisciplinary approach to improve the diagnosis and the chemical and biological (biomarkers) monitoring of freshwaters using the Meuse river as a case study. In the past, results from chemical surface water monitoring of the Meuse has revealed the occurrence of numerous substances and more particularly pharmaceuticals. Overall, the project suggests coupling chemical and biomarkers analysis on caged organisms (a crustacean, a mollusk, a moss and a fish species) with predictive mathematical population level models. In order to calibrate and validate these models, a lotic mesocosm experiment was set up. Five substances were chosen: diclofenac, carbamazepine, naproxen, paracetamol and irbesartan. An environmental realistic mixture M of the five substances was tested along with MX10 and MX100. The study was carried out in twelve 20 m long lotic channels. The mesocosms were set up with artificial sediments, macrophytes, periphyton, benthic and pelagic invertebrates, decomposers and one fish species (*Gasterosteus aculeatus). zebra* mussels, *Fontinalis antipyretica* and *Gammarus fossarum* were also caged in the mesocosms. After 3 months of stabilization, treatment lasted 5 months. Periphyton biomass, macrophyte biovolume, zebra mussel biomarkers and growth, *G. fossarum* survival, reproduction and growth, *F. antipyretica* biomarkers and growth, zooplankton and invertebrate abundance and diversity, and fish individual physiological responses along with population dynamics were the main measured biological endpoints. The concentrations of each substance in water was monitored monthly along with some physico-chemical parameters. The overall experimental design will be presented along with the results related to the monitoring of substance concentrations in water, physico-chemical parameters, macrophyte biovolume, invertebrate community response, fish larvae densities. A brief discussion of the direct and/or indirect effects will then be performed.

Improving the Quality of Ecotoxicological Testing and Assessment (P)

WE362

Relationships between aquatic toxicity, chemical hydrophobicity and mode of action: log kow QSARs revisited

M.G. Barron, U.S. EPA / Gulf Ecology Division; D.N. Vivian, C.T. Stevens, U.S. EPA

Quantitative structure toxicity relationships (QSARs) between chemical hydrophobicity and toxicity have been shown for nearly 100 years in both mammals and fish, typically using the log of the octanol:water partition coefficient (kow). The current study reassessed the influence of mode of action (MOA) on aquatic toxicity-log kow relationships using a comprehensive database of curated and standardized acute toxicity and consensus log Kow values, and weight of evidence MOA classifications. Log Kow QSARs were developed as linear regressions of log acute toxicity and log kow for 50 different combinations of taxa (e.g., fish, invertebrates, species-specific) and MOA (6 broad; 3 specific narcosis subtypes). MOA categories included narcosis (non-polar, polar, ester), acetylcholinesterase inhibition, neurotoxicity, electron transport inhibition, iono/osmoregulatory/circulatory impairment, and reactivity. Forty-eight of the 50 MOA-based models were statistically significant ($p< 0.05$; most $p< 0.001$), but r^2 values were generally less than 0.5, particularly for non-narcosis MOAs. The results showed that MOA-based QSAR models can improve the accuracy of aquatic toxicity predictions for a range of taxa, and that incorrect classification of a specific acting chemical can result in toxicity prediction errors greater than 1000 fold.

WE364

Data-mining: Making use of aquatic lower-tier data for higher-tier risk evaluation of agrochemicals

G. Eck, U. Memmert, E. Eschenbach, Eurofins Regulatory AG

Apart from delivering relevant toxicity data, standard lower-tier toxicity studies on aquatic organisms also provide valuable additional information for higher-tier testing strategies for risk assessments for plant protection products. While typically only the standard endpoint (e.g. 96-hour LC_{50}) is used, the thorough analysis of existing studies (i.e. individual tests or combined knowledge from different studies) as well as possible adaptations of standard test designs at the organism group of concern may provide valuable facts like time-dependency or reciprocity of exposure magnitude and exposure duration or information about relevant sensitive life stages. This additional information may be relevant in context with higher-tier testing strategies as revised exposure testing or to justify the deployment of time-weighted average surface water concentrations for risk evaluation. Gaining of this additional information should be taken into consideration for planning of lower-tier studies with the most relevant organisms. For example spacing of the test concentrations or additional assessment dates during the test period can maximize the knowledge that may be retrieved from these tests with regard to potential risk refinement. This poster presentation gives examples on how results of standard ecotoxicity studies can more efficiently be used as basis for higher-tier approaches in the environmental risk assessment of agrochemicals.

WE365

Comparison of models and tools for derivation of species sensitivity distributions (SSDs) for use in pesticide risk assessment

L. Azevedo, BASF SE, Agrarzentrum Limburgerhof / Global Toxicology; S. Charles, University Lyon 1 / Laboratory of Biometry and Evolutionnary Biology; G. Schmidt, BASF SE

EFSA’s guidance document for the risk assessment of edge-of-field aquatic organisms (EFSA, 2013;11(7):3290) recommends the use of species sensitivity distributions (SSD) as a second-tier approach for the aquatic risk assessment of plant protection products. For macrophytes, the hazardous concentration to 5% (HC_5) of tested species can be attained by deriving a species sensitivity distribution (SDD) composed of effective concentrations to a 50% effect on the growth rate of primary producers (ErC_{50}). Various probability distributions are available for the derivation of a SSD (e.g., lognormal, loglogistic) as well as publicly-available tools (RIVM’s ETX, MOSAIC_SSD from the University of Lyon, US EPA’s SSD generator CADDIS, BurrliOZ – hosted by CSIRO and the Australian Government Department of Environment, and ECETOC's hSSD). The extent to which the choice of the different probability distributions and tools influences the risk assessment has thus far not been evaluated. In this study, we compare macrophyte-based HC_5 values derived with different probability distributions and tools using standard toxicity ErC_{50} data from a selective herbicide. This study sheds light into the influence of probability distributions and tools on standard aquatic risk assessments and aims to give recommendations on the choice of the most appropriate combination.

WE366

Effects on NTA communities: HCx vs NOEC designs

F.M. Bakker, Eurofins-Mitox; S. Aldershof, Bioresearch and Evaluation

We discuss two examples of field fauna study designs with non-target arthropods (NTA). In both cases a hay meadow was chosen as a paradigm representative for off-field habitats at risk. One example concerns an HC_x approach where EC_x for various x were estimated from a field experiment and used to derive a Species Sensitivity Distribution. The other example concerns a more "classical" approach where a limited number of rates was tested in a replicated block design and the NOEC endpoint was found via statistical hypothesis testing. The merits and caveats of both approaches will be discussed and a protocol for evaluating and documenting statistical and biological significance of a NOEC study design will be presented. We analyse whether "No Effects" may have statistical or biological causes. In the HC_x-study consistent dose-response curves were obtained within 4 major arthropod taxa (63 out of 776; 8%) and SSD's could be constructed for each of them. Due to full overlap of curves the classes could be combined, resulting in narrow confidence intervals. In the NOEC-study 66 from 596 (11%) taxa were valid for univariate analysis, representing all major taxa. The protocol developed for the classification of results yielded an informative evaluation and allowed results to be classified as inconclusive or conclusive on a confidence scale of 1-4. Both study designs were fit for purpose and yielded biologically and statistically valid results. Where the HCx-design was statistically straightforward, the regulatory implication of the findings was not. On the other hand, where the regulatory implication of the NOEC is clear, the methodological issues related to hypothesis testing hamper a clear-cut presentation. An evaluation summary table, involving MSD-analysis, as presented here may be helpful in this regard.

WE367

α-Dominance versus β-Prominence

F.M. Bakker, Eurofins-Mitox; S. Aldershof, Bioresearch and Evaluation

The NOEC or an equivalent regulatory set EC_x- value are key endpoints to assess safe use of pesticides. These are challenging endpoints, especially when their assessment takes place in a multispecies context, e.g. in non-target arthropod (NTA) field studies. To date most ecotoxicological faunistic NTA field studies follow a hypothesis test design. Few examples can be found that address EC_x-finding by extrapolation from a regression model, such as the SSD-curve. There are two risks associated with hypothesis tests: the producers' risk and the consumers' risk, better known as Type-I and Type-II errors that occur with frequencies α and β, respectively. This contribution challenges the dominance of α and underscores the prominence of β when it comes to consumer safety. After all, a false positive result in a regulatory context implies a potential economic loss (re-testing, lost market share, lack of appropriate protection agents), whereas a false negative result implies a risk to environmental health. Statistical insignificance ($P> \alpha$) is often equated with safety, whereas it only means that the producer's risk is low. Safety stems from rejecting the null hypothesis when false and therefore the compliment of β is known as the power of an experiment. We show how power criteria can be derived and used to construct a biology based confidence profile for studies addressing NTA communities. We also show how relaxing α helps to identify those taxa for which an experiment does not provide sufficient conclusive data to draw meaningful conclusions. In a multi-rate study design, the proposed increase of α to 10% is shown to be off-set by applying expert criteria such as inconsistencies in dose or time.

WE368

Defining simple toxicity values (EC, BMD) is not so simple

E. Billoir, Université de Lorraine, CNRS UMR 7360; F. Larras, Helmholtz Center for Environmental Research - UFZ GmbH; V. BAILLARD, LIEC (CNRS UMR 7360, Université de Lorraine); S. Devin, LIEC, CNRS UMR 7360, Université de Lorraine / LIEC, CNRS; M. Schmitt-Jansen, UFZ - Helmholz Ctre Environm. Research / Department of Bioanalytical Ecotoxicology; M. Delignette-Muller, VetAgro Sup / Laboratory of Biometry and Evolutionnary Biology

Effective Concentrations (ECx) have now largely supplanted No Observed Effect Concentrations (NOEC), after decades of statistical criticisms towards the latter. ECx has a simple definition which sounds unambiguous. However, depending on the concentration-response pattern, its derivation is not trivial and should be paid attention in the context of ecotoxicological risk assessment. We recently developed a workflow for high-throughput concentration-response modelling of omics data (e.g. transcriptomics, metabolomics). Such data often displayed non-monotonic trends (U or Umbrella shape) as well as linear and exponential trends. Regarding our results, sigmoidal concentration-response shape was more the exception than the rule, as also reported in the literature for such omics data. In this context, we will discuss and explain why these non-sigmoidal trends lead to several issues regarding the derivation of toxicity values. In particular, the derivation of EC does make sense only provided an asymptotic response level is observed at high exposure concentrations and, in the case of non-monotonic trends, requires the definition of a maximal amplitude of the response. Alternatively to EC, the Benchmark Dose (BMD) has been proposed in the field of toxicology for setting toxicity values. The BMD approach as mentioned in EFSA guidance proposes two options. The first one considers a x-fold change of the control response which seems hazardously sensitive to the signal level (if the control response is zero, so will be the x-fold change). The second option defines a critical response level accounting for the standard deviation of data (control response +- SD). We will illustrate why this feature makes it more robust and usable whatever the concentration-response model.

WE369

Calculating the true ECx/LCx for non-linear models

Z. Gao, Bayer AG Crop Science Division; A. Solga, Bayer Ag; H. Fremdt, Bayer AG Crop Science Division; V. Ducrot, Bayer Ag / Environmental Safety Ecotoxicology; T. Preuss, Bayer Ag / Environmental Safety

ECx and LC50 are most frequently used endpoints for deriving Predicted No-Effect Concentration (PNEC) or Regulatory Accepted Concentration (RAC). ECx is defined as the concentration that shows x% effect compared to the control and LC50 is the concentration at which 50% mortality was observed. These definitions inherently assume the adverse effect at control is 0%. For example, in terms of mortality, the mortality at control should be 0% and when there is background mortality, Abbot's correction has to be applied. In terms of plant growth, percentage of inhibition in growth compared to control is calculated to feed in the dose-response model. These approaches have bypassed the requirements to use the standardly used probit dose-response model by modifying the data to make the model assumptions valid. However, use of these approaches without caution can cause serious over- or under-estimation of ECx/LCx due to the ignorance of control variability, the improper use of binomial assumption behind the probit model, *etc.* On the other hand, it is nowadays recommended that nonlinear regression models shall be used for dose-response analysis for metric data, which creates another problem in practice because there is inconsistency in the definition of ECx in regulatory context and that in the software implementing these 3 or 4 parameter models procedures. The ECx in programming implementation often assumes the 100% effect is the difference between the baseline response at control and the maximum response at dose level of infinity. EC50 is then defined as the 50% effect between baseline and maximum effect calculated from the model, not with respect to the control as how it is defined. This can lead to misinterpretations in the context of regulatory risk assessment. In this study, simulation examples and real data examples will be presented to illustrate the impact of the misuse of the current standard dose-response analysis procedures.

WE370

Review of Dose-Response Analyses in Regulatory Framework

Z. Gao, Bayer AG Crop Science Division; A. Solga, Bayer Ag; H. Fremdt, Bayer AG Crop Science Division; T. Preuss, Bayer Ag / Environmental Safety

Low effect EC values (EC10, EC20) derived from dose-response models have been recommended to replace NOECs in the pesticide regulatory context (e.g., new Regulation (EC) No. 1107/2009) The use of NOEC is often criticized because the statistical power to detect small effect sizes on the basis of null hypothesis testing can be very low due to high variability and small sample size. However, the concept and the limitations behind the various dose-response models have not been systemically addressed. There are ambiguities in the terminologies used such as linear and nonlinear dose-response models. When to use which model is not clear to practitioners. Practical difficulties in the implementation of the methodology lead to questions like what to do when there are no monotonic dose-response relationships, when ECx is superior to NOEC and when NOEC is more appropriate, why the confidence intervals are very broad in the range of low effect dose levels, and so on. In this study, we provide an in-depth review of the various dose-response models and associated assumptions and indications to answer these questions. Circumstances in which a certain dose-response model is more appropriate than others were described and illustrated using both real and simulated data examples. We show that the type of data, quantal, count or continuous are important to determine the error structure in the statistical model and the data characteristics provide inherently hints in the choice of dose-response model. The shared parameterizations and curve shapes between the so-called linear and non-linear models are clarified and the differences in the model assumptions are emphasized. We also identify a few common mistakes in practice due to wrong interpretation of dose-response analysis or wrong understanding of the software implementations. Potential improvements over the decision tree approaches proposed in the EFSA Guidance are discussed. The knowledge gaps related to non-monotonic dose-response relationships are also tackled. The connection between the multiple comparison procedure to derive NOEC and the model-based dose response analysis are presented and hybrid approaches are discussed.

WE371

Deriving no effect levels using probabilistic approaches: Application to trichloroethylene (TCE) and potential impacts to risk-based exposure concentrations

N.D. Forsberg, Arcadis U.S., Inc. / Environmental and Molecular Toxicology; B. Magee, S. Sager, ARCADIS US Inc

Derived no effect levels (DNELs) are indispensable tools needed to quantitatively evaluate the safety of various chemical exposures to humans and inform decisions related to exposure mitigation and environmental remediation. Typically, DNELs are calculated using deterministic methods that rely on single point estimates of no-effect levels, assessment factors (AFs) that allow extrapolation to human exposure scenarios and account for uncertainties in toxicological information, and allowable risk level. However, the point estimates used to calculate DNELs are by design conservative estimates that when combined lead to a phenomenon termed "compounded conservatism". The consequence of this phenomenon is DNELs that likely overestimate potential risks associated with human exposure to chemicals. Probabilistic risk assessment (PRA) approaches can be used to characterize the level of conservatism in deterministically-derived DNELs and to directly calculate DNELs. The advantage of calculating DNELs using PRA approaches is the ability to incorporate all available data and information associated with a particular input variable (i.e., variability amongst toxicity values, distributional assumptions of AFs, etc.) instead of relying on a single value, as is necessary for deterministic methods. An added benefit of the PRA approach is increased transparency regarding the protectiveness of a chemical's DNEL. This work will demonstrate how PRA is used to calculate DNELs using trichloroethylene (TCE) as an example. The evaluation will focus specifically on TCE's non-carcinogenic effects and incorporate the variability and uncertainties associated with dose-response modeling, physiologically-based pharmacokinetic modeling, assignment of AFs, and the choice of allowable risk level. The potential impacts of using PRA approaches to calculate DNELs will be discussed in relation to resulting risk-based exposure concentrations.

WE372

Aquatic higher-tier exposure testing of pesticides - from complexity to simplicity

G. Eck, E. Eschenbach, Eurofins Regulatory AG

Field exposure of pesticides is usually characterized by time-variable substance entries into water bodies resulting in complex exposure patterns which often significantly deviate from the constant exposure in standard ecotoxicity tests with aquatic organisms. As an appropriate risk refinement option higher-tier exposure testing is proposed in the current EFSA guidance document for aquatic risk assessments providing the possibility to define ecotoxicologically relevant test concentration patterns as more meaningful link to FOCUS exposure modelling outputs. Studies designed to reflect realistic exposure often result in lower effects. However, FOCUS exposure patterns are frequently challenged in regard to their representativeness for the variety of possible field scenarios and hence are generally not accepted as valid refinement option by several EU Member States. Besides, it is often difficult to generalize various critical patterns of different FOCUS scenarios for an efficient testing and to align these generic patterns with sensitive life stages of the organisms of concern. Practical aspects, types of effects and life traits of test organisms add to the complicacy of designing reasonable higher-tier exposure studies. The challenge is to balance representativeness and practicability of test designs. Representativeness can for example be enhanced by multi-year modelling simulations where the variability of treatment and climate constellations are taken into account. Detailed analysis of the multitude of predicted exposure scenarios as well as a detailed analysis of available standard toxicity data is required to develop meaningful test designs and strategies. Generating simplicity from complexity rather than matching exactly the FOCUS modelling exposure pattern is proposed as potential solution to concerns on representativeness as well as practicability for ecotoxicity testing and finally acceptability in a regulatory context. The poster will present examples for the generation of reasonable test designs and strategies that are considered to meet objections towards higher-tier exposure testing as adequate means for refinement of aquatic risk.

WE373

Keeping it real: multidisciplinary approaches to aquatic risk assessment

A.C. Brooks, Cambridge Environmental Assessments / Department of Enviromental Science; A. Lawrence, Cambridge Environmental Assessments / Regulatory Ecotoxicology; M. Hackett, Cambridge Environmental Assessments / Cambridge Environmental Assesssments; G. Hughes, Cambridge Environmental Assessments; T. Pepper, Cambridge Environmental Assessments / Cambridge Environmental Assessments; A. Abu, J. Carnall, M. Franey-Gardiner, F. Pickering, Cambridge Environmental Assessments; H.S. Schuster, Cambridge Environmental Assessments (CEA) / Aquatic Ecotoxicology; F. Joyce, M. Allen, Cambridge Environmental Assessments

Aquatic risk assessments for plant protection products (PPPs) can often be complex, comprising multiple crops, application rates, Member States (MSs) and therefore exposure scenarios. Where scenarios fail, it is critical to consider what that hazard quotient (PEC/PNEC ratio) really represents and whether it is a realistic representation of the true risk. A number of assumptions and worst-case parameters are used within the Tier I risk assessment, both on the exposure (predicted exposure concentration; PEC) and the effect (predicted no effect concentration; PNEC) side of the equation. To maximise the realism within the risk assessment, it is therefore advantageous to take a multidisciplinary approach, involving specialists in environmental fate, exposure modelling, aquatic ecotoxicology testing and regulatory ecotoxicology when developing refinements for the risk assessment. By developing integrated solutions, it is possible to progress from theoretical to more realistic estimations of risk. For example, assessing the relevance of efate parameters used in standard exposure models and designing field fate studies to derive more realistic parameters; analysing the exposure profiles associated with the maximum predicted exposure concentration in surface water (PECsw) compared to the exposure conditions used in standard aquatic ecotoxicology studies and designing modified exposure studies to more accurately mimic these exposure profiles; etc. The aim of this poster is to illustrate how different disciplines can work together to challenge the default assumptions of standard aquatic risk assessment, thus enabling appropriate refinement options to be derived and together design optimal solutions that are closer to addressing the real risks, rather than the theoretical ones.

WE374

Critical aspects of higher-tier laboratory exposure testing with different aquatic organisms

G. Gonsior, Eurofins Agroscience Services Ecotox GmbH; U. Memmert, G. Eck, E. Eschenbach, Eurofins Regulatory AG; C. Hafner, Eurofins Agroscience Services Ecotox GmbH / Aquatic Ecotoxicology

Revised exposure testing is an option for higher-tier risk evaluations proposed in the current EFSA guidance document for aquatic risk assessments for plant protection products. It offers scope for risk refinement by defining ecotoxicologically relevant concentrations that might be less conservative than constant exposure scenarios realised in standard effect studies or to justify time-weighted average concentrations in context with chronic risk assessments. Higher-tier exposure testing comprises challenges on both, the exposure as well as the effect side. To align test designs with predicted exposure, detailed analysis and generalisation of exposure is required. In order to represent worst-case scenarios in refined effect studies the exposure characteristics, life traits of the test organisms and their sensitive life stages are to be considered. Finally, test designs are restrained by practical considerations regarding the handling of test species, the test item dosing methods in order to realise desired exposure patterns and active substance characteristics. A systematic overview on critical aspects with regard to the conduct of studies will be presented to assist in designing higher-tier laboratory exposure tests with aquatic test organisms.

WE375

Repeated pulsed exposure in a partial life cycle test with zebrafish: Keep it realistic!

M. Teigeler, Fraunhofer IME / Ecotoxicology; B. Weber, D. Warnecke, RIFCON GmbH Goldbeckstr Hirschberg Germany

Refined exposure tests can be used to transfer more realism into standardised aquatic ecotoxicity testing. The aim is to achieve a more realistic perspective under consideration of the intended (worst-case) application pattern of a specific plant protection product. This option of risk refinement is also reflected in the recent Aquatic Guidance Document (EFSA, 2013). Here we present a partial life cycle test with zebrafish (*Danio rerio*) performed in a static water sediment system under pulsed exposure conditions. The test design allows addressing effects on different sensitive life stages of fish, subsequently and multiply exposed to the test item within the same environment. In the first part, adult spawning fish (i.e. the parental generation, F0) were exposed to 4 pulsed applications at weekly intervals. The performance of the reproduction in terms of egg numbers and fertilisation rate was assessed. The second part was initiated by placing fertilized eggs from the parental groups into the same water sediment systems. This F_1 generation was also exposed to 4 pulses of the test item at weekly intervals. Survival and growth of the early life stages were assessed. Other endpoints like endocrine-disruptor effects can be covered by measurement of vitellogenin and histopathological analysis of fish gonads. Glass aquaria of a total volume of 30 L were used and filled with a layer of artificial sediment to ensure stability of the test system throughout the 9-weeks test period. After pulse application, the dissipation of the test substance was analytically monitored. The evaluation of biological effects was based on mean measured as well as on area under the curve concentrations (AUC) of the test substance in order to be able to compare it to predicted environmental concentrations (PEC_{sw}, calculated with the FOCUS tools). The analysis of the AUC as well as of the DT_{50} values showed that the dissipation profile in the test systems were in line with the predicted exposure profiles in the field. In contrast to a continuous exposure, the procedure of several pulse applications may have an impact and possible distortion of the static system. However, it was demonstrated that the performance of the parental as well as the filial fish was fine and in line with the quality criteria set by the official test guidelines (OECD, USEPA). The test design was shown to provide a suitable approach to address both complex exposure regimes and specific endpoint issues.

WE376

Pulsed exposure of fish at sensitive life stages: The 'worst case' challenge.

M. Teigeler, E. Eilebrecht, Fraunhofer IME / Ecotoxicology; A.J. Jones, DuPont Crop Protection / Institute of Environmental Toxicology

Refined exposure tests have become part of the regulation framework for plant protection products in the EU (EFSA Aquatic Guidance Document 2013). A pulse dose test can be used to address areas of risk that cannot be satisfied with the standard suite of aquatic toxicity tests. A pulse dose considers situations where the expected exposure events in the field are significantly shorter than in the standard laboratory tests. However, the challenge is often to cover exposure profiles from multiple scenarios within one test. Therefore, the maximum exposure (peak) concentration, the number of peaks, the duration of the peaks, and the interval between peaks are considered to simulate a realistic profile covering a large number of scenarios. In this study, three different life stages of rainbow trout (*Oncorhynchus mykiss*) were exposed to nine pulses of the test chemical. To set these pulses as sharp as possible, the fishes were transferred from treatment vessels to untreated vessels at each time of pulse application. All vessels, including controls, were kept under flow through conditions. The concentrations of the test chemical were measured at start and end of each pulse event. Fertilised eggs, newly hatched fry and juveniles, already swimming up, were exposed. Glass aquaria with a total volume of 30 L were used. The evaluation of biological effects was based on mean measured concentrations measured for the test substance pulses and could be compared with the predicted environmental concentrations based on FOCUS modeling simulations. In contrast to a continuous exposure, the procedure of several pulse applications may have an impact and possible impairment of the sensitive stages. However, it was demonstrated that the performance of the life stages exposed was acceptable and conforms to quality criteria set by the test guidelines (OECD, USEPA). The test design was shown to provide a suitable approach to address a very complex exposure regime to cover the 'worst case' when a typical laboratory exposure is unrealistic.

WE377

TIER2+: Developing the Tools for Future Risk Assessment - New Chronic Invertebrate Test Systems and the Application of Realistic Exposure Scenarios

A. Dabrunz, F. Kümmich, C. Lang, Eurofins Agroscience Services Ecotox GmbH / Aquatic Ecotoxicology

According to recent and proposed guidance of the European Food Safety Authority (EFSA) current aquatic toxicity test systems should be critically reviewed, adjusted in regards of realistic (e.g. pulsed) exposure and complemented by establishing new test systems. To account for these challenges we will present data on a twofold strategy including A) test systems and B) exposure conditions:\n \n Experimental data of newly established (sub)chronic test systems, which are suitable meeting Tier 2 regulatory requirements (e.g. SSD approaches) will be given. Test systems evaluated include, for example, ostracods, cyclopids, nematodes, oligochaetes and amphipods, with a focus upon experimental conditions, suitability of standard toxicity testing endpoints and experimental validity. Subsequently, data on selected test systems under flow through test conditions simulating pulsed dose exposure scenarios will be given.

WE378

Optimisation of a chronic toxicity flow-through set up to investigate the adverse effects of chemicals to Daphnia magna

C. Beyer, IES Ltd; A. Peither, Innovative Environmental Services IES Ltd; S. Höger, Innovative Environmental Services (IES) Ltd / Environmental Toxicology; A. Zenker, University of Appl. Sc. Northwestern Switzerland / Institute of Ecopreneurship; P. Corvini, University of Applied Sciences Northwestern Switzerland

Daphnids are playing an important role as representative or indicator species for aquatic invertebrates in the environmental risk assessment for plant protection products, chemicals and pharmaceuticals. To investigate chronic toxicity, semi-static *Daphnia magna* reproduction tests following Test Guideline OECD 211 have to be performed. So far this test design is also used for highly degrading substances, despite the fact that by using semi-static test design the exposure concentration of the parent test compound is decreasing and metabolic products are accumulating durimg the renewal intervals of 2-3 days. To ensure a steady exposure level and to avoid the accumulation of degradation products, the studies have to be performed in a flow-through system. We are presenting a new flow-through system for reproduction testing with *Daphnia magna*. The flow-through system may pose additional stress for the daphnids, since interval dosing of test medium is causing turbulences within the test vessels. To preclude that stress effects are given in the new flow-through system, a reproduction test with a hydrolytically stable test substance was carried out. This comparative test allowed studying stress parameters by comparing the effect level concentrations and the EC_x values. Furthermore, a reproduction test with a fast degrading substance was tested to prove the ability of the new flow-through system to maintain a parent compound concentration above 80% throughout the test. In the comparative test both test systems showed the same dose-response-curves and gave the same No Observed Effect Concentration and Lowest Observed Effect Concentration for cumulative offspring per survivor after 21 days. Thus it can be assumed that the new flow-through system does not cause additional stress on *Daphnia magna* in a 21-day reproduction test. In the reproduction test with a non-degrading test item, the new flow-through system could dose the test item concentrations very reliable and precisely (dosed concentrations between 98 and 105% of nominal). In this study, it was proven that the new flow-through system does not cause unacceptable additional stress for the daphnids and can be utilized for reproduction testing with *Daphnia magna*. All tests conducted in the flow-through system were valid in accordance with OECD Guideline 211. The dosing system is very precise and reliable and is capable to maintain a parent compound concentration above 80% for a test item with half-life of 5 hours.

WE379

Eggs and larval fish test, an alternative method to marine fish exposure: Sensitivity and interest of early life stage.

r. lanchec, C. DUPUY, A. Jourdan, Groupe SGS France; j. bertin, SGS Multilab / Ecotoxicology

Multi-trophic level bioassays are usually carried out to determine toxicity of effluents, chemicals, cosmetic ingredients, etc... Toxicity to species is different according to the compound type considered, therefore tests on plants, invertebrates and vertebrates should be conducted. On the other hand, vertebrate organism tests should be avoided when possible, including test on juvenile fish. Alternative methods need to be developed for juvenile fish testing, with equivalent predictability and sensitivity. Several alternative methods exist for freshwater studies but there is no standardized method available for seawater fish. This study is a first step in the evaluation of a marine fish embryo and larvae test as alternative to adult marine fish test OSPAR HOCNF guideline, for example. The embryo and larvae sensitivity of turbot (*Scophthalmus maximus*) and sea bass (*Dicentrarchus labrax*) to reference substances were evaluated. To this purpose, within 72 hours post-fertilisation, the blastula stage eggs were exposed to reference substances separately. The effects of the toxicants on embryos and larvae were observed daily on a 10-days exposure period. For each reference substance and species, EC50 and mortality rate were calculated. To avoid vertebrate testing, results of this study were compared to published data. Several reference substances had a significant impact on survival of eggs and/or embryonic development. The sensitivity of the organisms is significantly different according to reference substances. Moreover, this test was used to evaluate toxicity of waste seawater samples in multi-trophic level bioassays (i.e. combination with single-species on alga, copepod and oyster larvae tests). Based on these results, marine fish embryo test appears as a credible alternative to juvenile fish testing. Therefore additional experiments will be conducted to validate this model.

WE380

Lack of Relevance of Normalized Hindlimb Length Measurement in Assessment of Thyroid Disruption in the Amphibian Metamorphosis Assay

S. Pawlowski, M. Dammann, S. Champ, BASF SE; M. Mathis, D. Fort, Fort Environmental Labs, Inc.

The OECD Test No. 231 and USEPA Test Guidelines OPPTS 890.1100 (amphibian metamorphosis assay -AMA) represents a Tier 1 ecotoxicity test designed to evaluate thyroid disruption. The test exposes Nieuwkoop and Faber (NF) stage 51 *Xenopus laevis* larvae to different concentrations of the test substance for 21-days and the following endpoints are measures: mortality, hind limb length (HLL), body length (snout to vent -SVL), body weight, developmental stage, asynchronous development, and thyroid histopathology. Of these endpoints, SVL and body weight are measures of growth, whereas developmental stage, asynchronous development, HLL, and thyroid histopathology are in the assessment of thyroid axis disruption. Recently, the relevance of suitability of hindlimb length normalized to SVL as a marker of thyroid disruption has been questioned based on its relationship to the growth endpoints (SVL, weight) and the relationship between limb length and differentiation. To evaluate normalized HLL, the correlation between HLL and either SVL or body weight was evaluated in the controls from 10 independently performed AMA studies at study day (SD) 21. Eight of the 10 AMA studies did not have significant late stage development per OECD Test No. 231 and USEPA Test Guidelines OPPTS 890.1100. For the 2 studies, data were censored to separate ≤ NF stage 60 from the >NF stage 60. Negative or no correlation between hindlimb length and SVL was found in 7 of the 8 studies examined without late stage development (r^2=-0.315-0.275, 0.553). Negative or no correlation between hindlimb length and body was found in 6 of the 8 studies examined without late stage development (r^2=-0.347-0.156, 0.429, 0.564). For the censored studies, correction between HLL and SVL or body weight was found in 1 of the 2 studies (r^2=0.452, 0.511). In each of the 10 studies, no asynchronous development was consistent with the absence histopathological findings in the control. The degree of HLL differentiation relative to other morphological markers of developmental stage determine if asynchronous development occurred. Since hindlimb differentiation is controlled by the thyroid axis during metamorphosis, it represents a more suitable endpoint in assessing potential thyroid disruption. To conclude, hindlimb differentiation, developmental stage and thyroid histopathology should be used in a weight-of-evidence based assessment of thyroid axis disruption. Normalized HLL should not be included in the assessment.

WE381

Acute toxicity test using Mediterranean fish species (Dicentrarchus labrax L., 1758): Intercalibration exercises towards standardized procedure

L. Mariani, CNR-IRSA / IRSA; F. Savorelli, ARPA EMR; B. Di Lorenzo, ISPRA Institute for Environmental Protection and Research; F. De Luca Picione, ENEA; M. Francese, Shoreline, Trieste; E. Di Capua, Regional Agency for Environmental Protection, ARPA Toscana; E. Giacco, university of genoa; s. manzo, ENEA / SSPT-PROTER-BES; P. Masulo, Università degli Studi di Napoli Federico II; A. Mazzola, Regional Agency for Environmental Protection, ARPA Sicilia; D. Palazzi, Regional Agency for Environmental Protection, ARPA Emilia Romagna; L. Pane, University of Genova; G. Sansone, Università Federico II Napoli; V. Bellaria, ISPRA Institute for Environmental Protection and Research; G. Sbrilli, Regional Agency for Environmental Protection, ARPA Toscana
The necessity to develop appropriate methods for the assessment of water quality and effluent toxicity was recognized by environment protection organisations and indicated by legislation. In Europe, the EU Water Framework Directive (2000/60/EC) and the new Regulation on Registration, Evaluation, Authorisation and Restriction of Chemicals (EC 1907/2006), both require data from ecotoxicological tests by using algae, crustaceans, and fishes. Some tests procedure have already standardised by organizations (ISO, OECD, USEPA, ASTM), but the freshwater test organisms were indicated more frequently than marine ones. In particular, the lack of specific acute toxicity methods on Mediterranean fish species have involved the adaptation of procedures available for freshwater fish (OECD, 203, 1992). In order to standardize the acute toxicity method for European sea bass (*Dicentrarchus labrax* L., 1758) larvae (species widespread in Mediterranean sea), two intercalibration exercises were conducted by 7 Italian laboratories, according to ISO/IEC 43-2:1997 and ILAC-G13:2000. To this end, for every exercise, the laboratories have conducted 24h-48h acute tests (2 trials each test) exposing sea bass larvae (50-70 days old) to the toxicant reference (Sodium Dodecyl Sulfate) concentrations: 6.31-3.98-2.51-1.58-1.00 mg/L) and control. The LC_{50} (Trimmed Spearman-Karber method: TSK) mean valueranged from 2.93±0.52 mg/L to 3.98±0.99 mg/L to 24h; and from 2.90±0.50 mg/L to 3.87±1.03 mg/L to 48h, respectively. The intra and inter laboratory variability of the tests were verified and Z-score values (< 2) were computed. Statistical analyses showed no significant differences in the data produced by most of the laboratories. The results indicate the standardization procedure is in advanced stage.

WE382

Introduction of a New Dosing System for Chronic Fish Tests Conducted with Difficult Substances

S. Höger, Innovative Environmental Services (IES) Ltd / Environmental Toxicology; A. Peither, J. Schreitmüller, Innovative Environmental Services IES Ltd
Chronic toxicity tests with fish are required for the risk assessment of plant protection products, pharmaceuticals and chemicals (depending on the tonnage and characteristics of the chemical). As chronic standard toxicity test the fish Early Life Stage (ELS) test following the OECD Guideline (GL) 210 has to be conducted, in certain cases this test system can be extended to an OECD GL 229, 230, 234 or to a full life cycle test. All these tests include the evaluation of sublethal effects on the test fish. This extension is recommended in case an influence of the substance towards the endocrine system (and finally the reproduction) cannot be excluded. The addition of some “endocrine endpoints” avoids additional tests, which may be requested at a later stage from the competent authorities. Typically, for these chronic fish tests very low concentrations have to be tested and in many cases the substance to be tested can be classified as difficult according to the OECD criteria, for instance low water solubility, high toxicity to fish, volatility or degradation during the test. For chronic fish tests preferable a flow through test design is used and difficult test items with specific properties as described above request a highly sophisticated flow through test device to guarantee the success of the test. In cooperation with an external company specialized on providing flow through technique to science and industry, IES developed a new, highly flexible dosing system. This very flexible and computer controlled dosing device is a modular system which provides several new technical features for important steps during the test e.g. dosing of the test substance, preparation of the test media and distribution of the test medium to replicates. In this presentation several examples for the testing of difficult substances are shown and the advantages of this dosing system are explained. The biological and analytical results demonstrate that difficult test items can be successfully handled using all available technical options, which are provided by the presented flexible flow through dosing system. Considering the increasing complexity of ecotoxicological tests and the methodical challenges during the testing of difficult substances, this presentation also intends to underline the importance of a continuous improvement of the technical setup for a successful performance of ecotoxicological test.

WE383

Difficult Substances as Challenge for the Algal Growth Inhibition Test According to OECD Test Guideline 201

S. Höger, Innovative Environmental Services (IES) Ltd / Environmental Toxicology; A. Dupont, J. Schreitmüller, Innovative Environmental Services IES Ltd
Photosynthetically active organisms such as green algae, blue green algae and diatoms are not only part of the risk assessment (RA) for plant protection products, but also standard test organisms for the RA for pharmaceuticals and chemicals (REACH). Especially chemicals as basic element for the synthesis of more complex products show a broad variety of characteristics from well water soluble, stable and non-toxic to hardly water soluble, unstable, volatile and toxic for water organisms. The group of chemicals with one or more of the latter characteristics is a challenge for the toxicity test with aquatic organisms. The OECD guidance document on aquatic toxicity testing of difficult substances and mixtures (23) provides some hints how the “standard testing” with such difficult test items should be conducted, but due to the numberless combinations of characteristics of these difficult substances, some innovation is required to find the best test design for the individual chemicals. We show examples for the toxicity testing of difficult test items starting with the investigation of the characteristics of the test item in the respective test water (water solubility, stability in water, photolysis effect, adsorption, storage conditions), followed by the development of a specific test design to determine the toxicity, the testing itself and finally the choice of the most suitable evaluation method within the various possibilities of calculation and interpretation of the results. In this presentation we focus on the testing of algae, but many aspects can be transferred directly to acute and chronic toxicity testing with daphnids or fish. This is important as in daily business in most cases a package of aquatic studies has to be conducted and results shall be comparable. The presented working procedures demonstrate that every test item - independent from its characteristics - can be tested according to established OECD Test Guidelines, but in some cases extensive biological and chemical background and innovative capacity is required to find the best test design. To make it even more complicated, there are different ways to interpret the analytical data and the most appropriate is chosen to provide the required endpoints. The different possibilities are introduced and discussed as well.

WE384

Activity based Collembola sampling may improve the data of field studies for regulatory purposes

P. Mack, A. Appeltauer, J. Illig, Eurofins Agroscience Services Ecotox GmbH; S. Knaebe, EAS Ecotox GmbH / Ecotox Field
Soil micro-arthropod field studies are carried out as part of the risk assessment of plant protection products. Those studies followed the proposed study design for soil organisms by Römbke *et al* (2009). Soil cores are taken in the field and afterwards soil organisms are extracted from the soil using high gradient extraction. Until now little is known about vertical movements of collembolans. Especially in long periods with high temperatures and low precipitation, a high number of collembolans might mitigate in deeper soil layers as included in the standard sampling scenario of 5 - 10 cm soil cores. Therefore it might be useful to cover also deeper soil layers, which contain potentially more specimens at the sampling time. One advantage of activity-based sampling is also that the time period of samplings can easily be increased in case that low numbers of individuals are caught. This would then increase the possibility of a robust evaluation of treatments. One activity based trapping method for soil microarthropods would be the slide traps which were presented at SETAC 2016 by Dehelean et *al*. 2016. Our poster will discuss possible advantages for the combination of soil core and slide trap sampling and will present first results from the comparison of soil core and slide trap catches.
Römbke, J., Schmelz, R., Knäbe, S., 2009: Field studies for the assessment of pesticides with soil mesofauna, in particular enchytraeids, mites and nematodes: Design and first results. Soil Organisms, 81: 237-264 Stefan-Bogdan Dehelean et al., 2016 Stratification of soil arthropods in topsoil layers, SETAC Europe 26th Annual Meeting, Nantes, France

WE385

New Technology evaluating Acartia tonsa as a biological model

S. Abreu, University of Aveiro / Dep. Biology & CESAM; S.M. Leandro, Polytechnic Institute of Leiria / MARE Marine and Environmental Sciences Center; A.M. Soares, University of Aveiro / department of Biology & CESAM; R.E. Martins, M. Oliveira e Silva, University of Aveiro / DETI IEETA
Copepods play an important ecological role on marine ecosystems and may act as a sentinel of environmental degradation resulting from direct or indirect human impacts. The copepod *Acartia tonsa* is a calanoid species with a worldwide distribution and relatively easy to maintain for several generations under captive conditions. These characteristics allow this species to be a potential biological model to be used on ecotoxicogical studies or live food for larviculture. On of the bottlenecks for its massive utilization relies on the time consumption procedures related with counting and cultures monitoring. To overcome such constrain, the aim of the present study was to evaluate the application feasibility of a new technology based on automatic counting and cromatic characterization in real time of the particles and/or organisms. The technology D Counter constitutes an innovative approach, by the fact of turning the data harvesting process much more efficient and accurate, breaking the traditional, error-prone, human-based counting methodology. The obtained results for *A. tonsa* cultures indicates a high significant correlation between manual and automatic counting, constituting the first step for the use of this biological model on experimental studies.

WE386

Solubility limits of lanthanides in standardized ecotoxicological media

D.A. Vignati, CNRS / LIEC UMR7360; C. Hissler, Luxembourg Institute of Science and Technology; J.E. Groenenberg, Université de Lorraine and CNRS / LIEC UMR7360; C. Cossu-Leguille, Universite de Lorraine; L. Giamberini, Université de Lorraine, CNRS UMR 7360 / LIEC, CNRS
The biogeochemical cycles of several lanthanides (LNs) are being progressively disrupted by their increasing use in industrial sectors such high-tech applications and clean energy generation. Except for a few hotspots located close to (industrial) wastewater discharges, LNs concentrations remain essentially low (i.e., in the µg/L range or lower), but the paucity of available data has fostered research on their possible effects on biological organisms. Getting reliable information on the ecotoxicological potential of LNs is also important in view of the possible (re)opening of mining activities in response to the current monopoly of LN production by the People's Republic of China. In this context, testing LNs ecotoxicity following established standard protocols must consider the peculiar chemistry of these elements if meaningful results are to be obtained and used to establish regulatory limits. After addition to ecotoxicological media, typically in the form of soluble chloride salts, LNs can rapidly form complexes with phosphates or carbonates or undergo hydrolysis. Due to the low solubility of LN-phosphates and LN-carbonates, precipitates may be formed which can markedly lower LNs solubility and reduce to a fraction of the expected value the concentrations to which organism are exposed. The presence of possible LN-containing precipitates can further complicate the interpretation of the corresponding biological responses. In this contribution, we use thermodynamic speciation modeling to examine the equilibrium theoretical speciation of LN in standardized ecotoxicological media for algae, daphnids, fish and other laboratory organisms. Using the range of concentrations reported in published literature studies, attempts are also made to model the fraction of added LN predicted to occur in precipitated forms. Finally, depending on the availability of the appropriate complexation constants, the predictions for laboratory media are compared with those for natural waters where the presence of dissolved organic matter can cause important differences in comparison with laboratory media. While thermodynamic modeling does not account for the kinetics aspects of speciation, it still provides useful indications as to the actual exposure conditions likely to be experienced by organisms in standard ecotoxicity tests.

WE387

Improving ecotoxicity tests for trace elements forming poorly soluble chemical species in test media

D.A. Vignati, CNRS / LIEC UMR7360; F.G. Acanfora, University of Salerno / Department of Chemical and Biology; G. Cornelis, Swedish University of Agricultural Sciences / Soil and environment; M. Carotenuto, University of Salerno / Department of Chemical and Biology; B.J. Ferrari, Centre Ecotox EAWAGEPFL; G. Lofrano, University of Salerno / Department of Chemical and Biology
In order to get an appropriate interpretation of ecotoxicological results the exposure concentrations of test organisms to the contaminant of concern must be kept constant and stable over the test duration. Increasing evidence suggests that this is often not the case when dealing with elements that tend to form chemical species with low solubility (e.g., oxides and oxyhydroxides for Cr(III) and Sn, phosphates in the case of some lanthanides). In such situations, the calculation of meaningful effect concentrations for hazard and risk assessment must consider the temporal decrease of exposure concentrations to avoid erroneous conclusions. We performed ecotoxicity tests using semistatic exposure conditions to assess if periodical medium renewal could compensate for the temporal decrease in element concentrations. We also tried to evaluate the possible contribution of soluble vs. colloidal/particulate elemental species to biological effects by testing the ecotoxicity of solutions aged for different periods. Chromium(III) was chosen as a model contaminant, but the general approach is applicable to all elements forming poorly soluble species and potential colloidal precipitates in ecotoxicological test media. In medium aliquots amended with Cr(III) (range 0.005 to 1,25 mg/L), renewal every 24 h was not sufficient to obtain stable exposure concentrations (i.e. ±20% of the initial value) throughout the duration of the test. The actual exposure range (estimated as time weighted mean concentrations) was between 5 and 275 µg/L and was used to estimate the Cr(III) EC50. On the other hand, concentrations remained stable over time and agreed with expected values in comparative experiments performed with Cr(VI). When accounting for the temporal decrease in Cr(III) level during tests, Cr(III) appeared about 10 times more ecotoxic than Cr(VI); in contrast with the current consensus. Ecotoxicological effects persisted in solution aged for 4h and 72 h which, based on previous research, would be long enough to remove ionic Cr(III) via hydrolysis (4 h) and to form colloidal Cr-bearing particles (72 h and, possibly and to a lesser extent, 4 h). The use of semistatic exposure conditions and the assessment of persistent effects in spiked, aged test media would allow better hazard assessment for several elements (e.g., lanthanides, Sn, Ga, In) whose concentration may strongly fluctuate in standardized ecotoxicological media.

WE388

Long term ecotoxicity testing of limonene for hazard classification: not such a lemon after all

P.C. Thomas, CEHTRA SAS / Ecotoxicology and Risk Assessment; F. Balk, Royal Haskoning DHV; H. van Bergen, Para-Celsus concept; K. Jenner, Givaudan / Global Regulatory Affairs & Product Safety; A. Kamper, DHI; S. Gimeno, Firmenich / Product Safety and Regulatory Affairs
Limonene is a stereoisomeric substance taking its name from lemon, which dominates the composition of the essential oils of citrus fruits. When synthesised as dipentene it is a racemate. However, in botanical sources it is present as the D-or L-enantiomer in Natural Complex Substances (NCSs) obtained at anything from traces in some plant extracts to over ninety percent in orange oil. These oils are commercially used as fragrance and flavour ingredients in a wide range of applications (cosmetic products, food manufacture, fragrance perfumery, botanical insecticide, household cleaning products, etc). Therefore, it is predominantly released back to the environment after use. The racemate and both D- and L-limonene received a harmonised classification under Annex VI of the EU C&L legislation as Aquatic acute category 1: (Very toxic to aquatic life) and Aquatic chronic category 1: (Very toxic to aquatic life with long-lasting effects). Both classifications have a severe impact on storage, handling and transport requirements of limonene and the many (natural) complex substances and fragrance and flavour mixtures that contain even small amounts of it. The existing classification is a result of a limited data set notably for chronic endpoints. The chronic category 1 classification is extrapolated from the acute category 1 toxicity, log Kow >4 and erroneously assumed not-rapid biodegradability of the substance. As limonene is a narcotic substance its Chronic 1 assignment was expected to be conservative. Due to doubts on the chronic classification and the consequences for labelling, storage, handling and transport, long term aquatic ecotoxicity studies were conducted to obtain a solid basis for the environmental classification. Contrary to expectations and despite the existence of a guidance document, considered the ecotoxicologists' Bible for testing difficult substances, the chronic studies were fraught with difficulties. This poster describes the problems encountered by the laboratories when testing a highly volatile, rapidly biodegradable, hydrophobic, non-polar narcotic substance like limonene under chronic conditions, how these were countered by the monitoring team and after a persistent effort, eventually overcome. The poster takes the reader through a long adventure lasting several years with results supporting a chronic 3 classification. The subsequent regulatory procedure to implement the classification in the EU regulations is currently ongoing.

WE389

Is that an effect? The importance of using all relevant data in mesocosm studies

J. Ashford, Cambridge Environmental Assessments; A.C. Brooks, Cambridge Environmental Assessments / Department of Enviromental Science; A. Lawrence, Cambridge Environmental Assessments / Regulatory Ecotoxicology; M. Allen, Cambridge Environmental Assessments
In long-term multispecies studies, such as mesocosms, a complex statistical analysis is required to decipher the data and determine whether a test-item effect has occurred. It is sometimes the case that regulators and applicants have differing opinions as to what the no observed effect concentration (NOEC) or no observed adverse effect concentration (NOAEC) should be, based on the accompanying data analysis. This is sometimes because statistically significant differences are viewed in isolation from the rest of the available data from the study. The aim of this poster is to urge regulators to not just focus on statistically significant differences, but to take into account all available and relevant data to assess the biological relevance of any differences observed. At CEA we use a range of sampling methods to collect data on aquatic macroinvertebrates within our mesocosm studies, including emergent traps, colonisers and sweep nets. Each sampling method targets a particular species; sweep nets are used to capture fast-moving pelagic organisms whereas colonisers are left in-situ to allow benthic organisms to enter the trap. In some cases, the same species can be caught using different sampling methods, such as mayflies, damselflies and caddisflies, and this can be from various stages in their life cycle. Comparing the data for the same taxa from different sampling methods can be helpful in creating and overall picture of how that taxa is responding within the mesocosm study. This can be helpful in deciding whether statistically significant differences are biologically relevant; for example, if a statistically significant difference to the control is only observed in one out of three sampling methods used for a particular taxa, this may indicate this is due to natural variation rather than the influence of the test item. Here, we will review caddisfly, damselfly and mayfly data from past CEA studies where statistically significant differences have been observed and highlight cases where different sampling methods (emergent, colonisers and sweep nets) support or contradict a test-item effect.

WE390

Evaluation of the environmental risk assessment procedure according to Directive 2001/18/EC for Gene Modified Organism used as medicinal products

H. Weigt, Fraunhofer Institute for Toxicology and Experimental Medicine ITEM / Chemical Risk Assessment; E. Weber, Fraunhofer Institute for Toxicology and Experimental Medicine ITEM; M. Batke, Hochschule Emden/Leer; S. Schwonbeck, Fraunhofer Institute Toxicology and Experimental Medicine / Chemical Risk Assessment; A. Bitsch, Fraunhofer ITEM / Chemical Risk Assessment
The deliberate release of genetically modified organisms (GMOs) including GMOs

used as medicinal products, e.g. gene therapies, into the environment is regulated by directive 2001/18/EC of the European Parliament and of the Council of 12 March 2001. An integral part of the directive regulates the provision of information on the GMO and, based on this, the risk management with regard to the environmental effects of such releases. As regulated by this directive, a publicly accessible database is the "GMO Register" of the JOINT RESEARCH CENTER of the EC (http://gmoinfo.jrc.ec.europa.eu/Default.aspx), which contains information about all releases under the guideline. As of 07.11.2016, there were 238 entries of medicinal GMOs" in the "Summary Notification Information Format (SNIF). SNIFs are prepared as a summary document of the confidential environmental risk assessments (ERA) by the respective Sponsors of clinical trials in the EU and evaluated during the clinical trial application by the national competent authorities. They comprise, inter alia, information regarding the GMOs and the parental organism's nature, release, environmental interactions, monitoring, waste treatment and emergency response plans. We strived to assess information concerning the environmental risk, derived measures and the overall standard of SNIFs concerning compliance with the regulatory requirements. To do so, we picked a homogeneous group of GMOs, namely gene modified Adenovirus, the most frequently used vector in gene therapy trials worldwide. Relevant information were entered into a database and categorized, applying unified vocabulary. Different challenges regarding the information available within the SNIFs were identified by analyzing the database: in several cases mandatory information was not available, e.g. monitoring plans, and in other cases the SNIF documents were misinterpreted, e.g. the connection between replication, dissemination and survivability was interpreted heterogeneously. Although this analysis has been performed using only Adenovirus data, information gaps and inconsistencies are transferable to other species as well. Consequently, it is proposed to specify some parts of the SNIFs in order to make more reliable information transparently available.

WE391

PBT evaluation 20 years on: is it time to reconsider the technical progress made in risk assessment methodology?

P. Thomas, C. Durou, CEHTRA SAS / -

In the EU, the ecotoxicological dataset for a chemical is used for the purposes of prospective risk assessment (PRA) and of PBT assessment. While the PRA aims at determining the use conditions and risk management for which the environment is safe, the PBT assessment aims at identifying chemical for which effects on the long-term are considered unpredictable and that environmental exposure is difficult to reverse. The scope of this poster is to discuss, in the light of technical and scientific progress: For which PBT-Like and certain PBT chemical, PRA can now be carried out The justification of the numerical criteria behind the identification of PBT/vPvB property Unpredictable effects are fundamentally linked to uncertainty in: Assessing accurately a chemical's potential to amplify along the food chain, and Whether a chronic value can be established based on its MoA. Significant scientific progress has been made in the field of aquatic toxicity testing of difficult chemicals, evaluation and interpretation of ecotoxicological data since the PBT/vPvB criteria were originally designated. The numerical criteria were established in the late 1990s by OSPAR with the primary aim to protect the marine environment and used by the EU (TC NES) from the early 2000's enlarging the protection goal to any environment. The criteria became applicable to any chemical produced over 10 TPA when REACh regulation came into force, thus, treating chemicals as a homogeneous group. Interestingly, the criteria were originally defined by using data from a set of chemicals known as highly hazardous for the environment. Such compounds (e.g. polychlorinated aromatic compounds) were non-ionic and hydrophobic while the chemical space is much more diverse. In other words, a BCF value of 2000 may be a good cut-off for chemicals which are both highly hydrophobic and slowly metabolised to anticipate amplification along a food chain but may be of limited meaning for other chemical classes. In the US, B is defined as a BCF of >5000 which is the vB criteria in the EU while perhaps the only meaningful way to determine B is to consider bioaccumulation in the food chain which has no legal relationship with the B criterion. Further questions can be posed of the true meaning of the half-life cut-off values for P and vP in terms of environmental persistence and the meaningfulness of using a standard mg/L cut off basis for T blanketing all MoAs.

WE392

UVCB block method for estimating expected mixture toxic pressure of substances of Unknown or Variable composition, Complex reaction products or Biological materials

D. van de Meent, Association of Retired Environmental Scientists ARES / Environmental Science; J. Struijs, Association of Retired Environmental Scientists ARES; D. De Zwart, DdZ Ecotox / Centre for Sustainability Environment and Health; J. Hermens, Utrecht University / Institute for Risk Assessment Sciences; N. van Straalen, Association of Retired Environmental Scientists ARES / Department of Ecological Sciences

We have developed a spreadsheet calculation tool for chemical safety assessment of UVCB substances. The tools adopts the approach of Concawe's Hydrocarbon Block Method for chemical safety assessment of complex petroleum substances. The tool is meant to be used for demonstrating 'safe use' of chemicals, as required for registreation of substances under REACH. The tool makes use of scientifically up to date versions of the exposure models SimpleTreat and SimpleBox, combined with state-of-art expected mixture pressure calculation using the Van Straalen-Aldenberg convolution integral. The tool estimates the addition of toxic pressure to the ambient toxic pressures in local, regional and continental EU environments, expected from the UVCB mixture. It is proposed that 'safe use' is demonstrated sufficiently well by showing that the UVCB under study is expected to contribute negligibly to ambient toxic pressure in the environment. The proposed 'safe use' calculation method has been tested on a selection of relatively well studied UVCBs.\n The poster briefly explains the new UVCB block method and illustrates its potential with the outcomes of test calculations.

WE393

Evaluation of hypopharyngeal glands development in Honeybees (Apis mellifera L.) from toxicity studies in the light of current guidelines (EFSA and OECD)

A. Drzewiecka, M. Napora-Rutkowska, J. Faron, Institute of Industrial Organic Chemistry Branch Pszczyna / Department of Toxicological Studies; E. Kulec-Ploszczyca, P. Parma, M. Grzesica, Institute of Industrial Organic Chemistry Branch Pszczyna / Department of Ecotoxicological Studies; N. Lemańska, Institute of Industrial Organic Chemistry, Branch Pszczyna; A. Daniel-Wójcik, Institute of Industrial Organic Chemistry Branch Pszczyna / Deparment of Toxicological Studies

Honey Bee (*Apis mellifera* L.) is a species that belongs to a group called 'beneficial insects'. All arthropods from this group play the important roles in nature, albeit bees go a few steps forward and they also find application in the food, pharmaceutical and others industries. With the current decline in bees' colony numbers, these arthropods should be handled with extreme caution. Therefore, it is extremely important to assess the risk for non-target organisms for which plant protection products are often more toxic than pests of agricultural crops. This assessment is related not only to the direct influence of chemicals on the number of bee populations, causing morphological mortality or morbidity, but also indirectly - through the impairment of the ability to raise the larvae, for example by disturbing the work of hypopharyngeal glands (HPG)responsible for the production of 'milk' containing proteinic substances to feed larvae and queen. By 2017, the only document regulating the toxicity study of chemicals on bees was the EFSA document (EFSA Journal 2013;11(7):3295), which included continuous access to distilled water and pollen and evaluation of HPG. However, in 2017, the new OECD guideline (no. 245) was introduced, in which the methodology of chronic toxicity testing was changed compared to the EFSA document. Changes occurred in the way of dealing with bees – there is no access to distilled water and pollen, and no evaluation of HPG. It is a significant change, because according to the literature and our studies, hypopharyngeal glands do not develop correctly in these conditions, which exclude the assessment of HPG. Hence OECD guideline probably does not consider it as endpoint in the study. As previous study has shown, chemicals can have influence on development of hypopharyngeal glands, without causing mortality and morbidity. This matter is worthy considering and should be investigated further, in order to introduce the evaluation of hypopharyngeal glands as an endpoint in toxicity testing of chemicals on bees.

WE394

Assessing toxicity to Daphnia magna using movement parameters

T. Đerđ, Department of Biology, University of Osijek / Department of Biology; D. Hackenberger, GeoNatura; B. Hackenberger, Department f Biology, University of Osijek / Department of Biology

Daphnia are among the most common settlers of freshwater habitats. These planktonic invertebrates show high sensitivity to various toxicants, therefore representing a useful model organism in ecotoxicological research – with common endpoints being survival, reproductive success and observable morphological changes. Some of recently conducted scientific investigation involving these organisms focused on examining the effects of various substances on their mobility. The aim of this work was to compare and examine the changes of swimming behaviour of *Daphnia* over time and under the influence of sub-lethal concentrations of $ZnCl_2$, based on 12 chosen movement parameters. Organisms obtained from a natural habitat acclimatized to laboratory conditions were exposed to 5 different sub-lethal concentrations of $ZnCl_2$, in 3 replicates. Five organisms were placed in each transparent plastic Petri dish in prepared solutions of the selected toxicant. The recording started instantly upon exposure of the organisms to the toxicant (t_0), as well as 1 h, 24 h and 48 h of exposure (dt=1 h, dt=24 h, dt=48 h). The recording and analysis of motion were carried out in Python, implementing OpenCV, TrackPy and NumPy packages. Analysis of the obtained data showed that the duration of exposure affects the movement parameters, regardless of the concentration of the toxicant. Although, some of movement parameters showed significant correlation with concentrations of toxicants and hence can be used as an early biomarker of exposure.

WE395

The validation of analytical methods in ecotoxicology

I. Pedall, A. Rastall, A. Sagner, M. Faupel, Rifcon GmbH

The validation of analytical methods (regulated by SANCO/3029/99 rev 4.) used in support of ecotoxicological studies has become an important aspect of the registration process for plant protection products. When Regulation (EC) No. 1107/2009 came into force, adherence to the SANCO/3029/99 rev 4. Guidelines became obligatory. This has resulted in serious implications for the registration process because ecotoxicological studies may well now be rejected on the basis of inadequate analytical methodology or incomplete analytical data although the studies have previously been accepted. Here we give an overview of current requirements and provide a checklist that can be used to evaluate analytical methods in ecotoxicological studies.

WE396

A new pulsed-exposure early life stage test design for rainbow trout on an insecticide. Refining OECD Guideline 210 to meet the needs of EFSA Aquatic Guidance 2013

C.S. Ramsden, AgroChemex Environmental Ltd / School of Biomedical and Biological Sciences; C. Gamblin, AgroChemex Environmental Ltd; W.R. Jenkins, W R Jenkins / Regulatory Affairs Ecotoxicology; S. Norman, RidgewayEco

Constant-exposure in OECD TG 210 Fish Early Life Stage studies is unrealistic for fast-dissipating pesticides compared to edge-of-field water-bodies. EFSA Aquatic Guidance (2013) allows aquatic toxicity studies to be modified so the exposure-profile (peak-height, pulse-duration, number of pulses) is comparable to the worst-case predicted field-exposure. In the present study on a synthetic pyrethroid (SP) insecticide, a novel method was developed to *simultaneously* assess effects on 3 early-life stages of rainbow trout. The TG 210 design was modified to incorporate a worst-case time-variable exposure profile in tanks containing a 10 mm sediment layer and stainless steel mesh barrier to allow water movement but prevent fish disturbing the sediment. Life stages used were newly fertilised ‘eggs’, ‘alevins’ (non-feeding larvae) and free-feeding ‘swim-up’ fry. To ensure physical separation of the 3 life stages within the tank, eggs and alevins were each held in a glass incubation tube with a mesh base. A control group plus 5 concentrations were used. To start, each group had 4 replicate tanks each with 50 eggs, 20 alevins and 20 swim-up fry. There were two 72 hour static exposure phases on Days 0 and 14. The study duration for organisms starting as ‘eggs’, ‘alevins’ and ‘swim-up’ fry was 72, 45 and 31 days respectively. This allowed for the assessment of effects over a period including at least 2 weeks of growth after initiation of free-feeding for each of the 3 life stages. Standard end points were assessed including hatch success, survival, growth and clinical signs (e.g. loss of equilibrium and coordination). To assess the potential neurotoxic action feeding behaviour was categorised as *active*,*passive* and *not feeding*. This refined-exposure study showed that 3 critical life stages of fish can be tested *simultaneously*, whilst complying with the fundamental elements of TG 210. Control hatch and survival rates were both > 95%. The design allowed the direct comparison of the sensitivity of each life stage to identical pulsed exposure profiles. Free-feeding swim-up fry was the most sensitive exposed life stage, based on clinical signs, feeding and slightly reduced growth. Swim-up of exposed alevins was delayed at high treatment levels. Exposed eggs were unaffected.

Distribution, transformations and biological effects of incidental nanoparticles and nanoplastics in the environment from a more realistic point of view (P)

WE397

Dissolution of Different Silica Nanoparticles in Aqueous Matrices

M. Maceroni, Adolphe Merkle Institute / BioNanoMaterials; D. Bossert, A. Petri-Fink, B. Rothen-Rutishauser, F. Schwab, Adolphe Merkle Institute / BioNanomaterials Group

Since centuries, silica (SiO_2) is used in large scale industrial applications, such as cement manufacture or glass production. In these applications, SiO_2 is used in his bulk form. Recently, SiO_2 in nanoparticulate form has brodened the range of applications, both in large scale, *e.g.* as anti-cake agent in food industry, or owing to its abrasive effect in cosmetics; and in small scale, for the production of biosensors, biocatalysts, as well as polymer composites [1]. The use of silica nanoparticles (SiO_2-NPs) as a delivery vehicle for molecules in plants is being investigated [2], demonstrating their potential in environmental applications. Many of the current uses increase the likelyness that SiO_2-NPs could accumulate in the environment and in food webs. Therefore, it is crucial to investigate the dissolution of SiO_2-NPs in different environments. Besides information about the degradeability of the nanomaterials, this knowledge will also allow to produce more environmentally friendly products. Here, we present the results of a method development to quantify the dissolution of different SiO_2-NPs in aqueous media in order to determine the dissolution kinetics. This method relies on the use of inductively coupled plasma optical emission spectroscopy (ICP-OES) for the quantification and the parallel characterization of the particles by transmission electron microscopy (TEM) and dynamic light scattering (DLS). A simple setup based on dialysis membranes, and a sampling protocol are in the process of being established. The first results indicate that is possible to detect the dissolved fraction of SiO_2-NPs roughly 60 nm in diameter within about one day. Long-term dissolution experiments will be performed in the coming months to determine the dissolution kinetics more precisely. This setup will serve as a tool to assess the behavior of SiO_2-NPs in environmental media. [1] Barik TK, Sahu B, Swain V. 2008. Nanosilica—from medicine to pest control. Parasitology Research. 103:253. [2] Torney F, Trewyn BG, Lin VSY, Wang K. 2007. Mesoporous silica nanoparticles deliver DNA and chemicals into plants. Nature Nanotechnology. 2:295. *Acknowledgement* - The authors thank the Swiss National Science Foundation (project 168187) and the Adolphe Merkle Foundation for the support and funding of the study, and Laura Rodriguez-Lorenzo for her precious advice and suggestions.

WE398

Occurrence of fullerene aggregates in Mediterranean rivers: Two cases of study

J. Sanchís, IDAEA-CSIC / Water and Soil Quality Research Group; R. Milacic, Jožef Stefan Institute (JSI) / Department of Environmental Sciences; M. Farre, IDAEA-CSIC / Environmental Chemistry; D. Barcelo, IIQAB-CSIC / Department of Environmental Chemistry

Fullerenes are carbon nanomaterials that have many potential applications in nanotechnology and that can be generated in some combustion processes. Since their environmental effects and behaviour are uncertain, fullerenes are considered emerging contaminants and during the last years they have been included in some environmental monitoring studies [1-3]. However, few of these studies have reported the presence of fullerenes in water systems. In order to assess the environmental risk of fullerenes it is important to enlarge the dataset and to characterize the concentrations of these nanomateriales in different rivers. In the present work, C_{60} fullerene, C_{70} fullerenes and five functionalized derivatives were determined in water and sediment samples from two Mediterranean rivers. The first case of study was located in the Sava River (Souteastern Europe), where more than 30 samples were studied in two sampling campaigns. In the second case of study, samples of estuary water, wastewater, river water and coastal water from the Ebro River delta were analysed. In both studies, C_{60} was the most ubiquitous compound and it was detected for the first time in the marine environment, although its concentrations were below the ng/l order in all the cases. The exotic fullerene [60]PCBM was also detected in a punctual sample from the Sava River. The results show that fullerenes are present in the aquatic environment although at concentrations far below than those levels that are expected to cause any toxic effect to aquatic organisms [4]. However, the presence of C_{60} may modulate the toxicity of some co-contaminants, as described elsewhere [5] Acknowledgement: This works has been supported by the European Communities 7th Framework Programme under Grant Agreement No. 603629-ENV-2013-6.2.1-Globaqua and by the Spanish Ministry of Economy and Competitiveness through the project Integra-Coast (CGL-2014-56530-C4-1-R). It has also received funding from the Generalitat de Catalunya (Consolidated Research Groups “2014 SGR 418” Water and Soil Quality Unit). References [1] Astefanei, Alina, et al. Analytica chimica acta 882 (2015): 1-21. [2] Carboni, Andrea, et al. *Environmental Pollution* 219 (2016): 47-55. [3] Zakaria, Susanna, et al. Environmental Science and Pollution Research (2017): 1-10. [4] Freixa, Anna, et al. *The Science of the total environment* 619 (2017): 328. [5] Sanchís, Josep, et al. Environmental science & technology 50.2 (2015): 961-969.

WE399

Occurrence, fate and behaviour of fullerenes in the environment

M. Farre, IDAEA-CSIC / Environmental Chemistry; J. Sanchís, IDAEA-CSIC / Water and Soil Quality Research Group; Y. Aminot, University of Plymouth; E. Abad, IDAEA-CSIC; A.N. Jha, Plymouth University / Biological Sciences; J.W. Readman, University of Plymouth / Biogeochemistry Research Centre; D. Barcelo, IIQAB-CSIC / Department of Environmental Chemistry

The occurrence, fate and behaviour of carbon nanomaterials in the aquatic environment are dominated by their functionalization, association with organic material and aggregation behaviour. In particular, the degradation of fullerene aggregates in the aquatic environment is a primary influence on their mobility, sorption potential and toxicity. In this presentation, a summary of the occurrence of fullerenes in environmental matrices performed in different studies of our group will be presented. The analytical approach to investigate seven fullerenes (C_{60}, C_{70}, *N*-methylfulleropyrrolidine, [6,6]-phenyl C_{61} butyric acid methyl ester, [6,6]-thienyl C_{61} butyric acid methyl ester, C_{60} pyrrolidine tris-acid ethyl ester and [6,6]-phenyl C_{71} butyric acid methyl ester) in waters, soils and sediments combines an ultrasound-assisted solvent extraction (UAE) with toluene followed by liquid chromatography (LC), using a pyrenylpropyl group bonded silica based column, coupled to a high-resolution mass spectrometer (HRMS) using atmospheric pressure photo ionisation (APPI) in negative ion mode. Main results of these studies showed levels of pg/m^3-ng/m^3 in atmospheric aerosols, pg/g-ng/g in soils and pg/l-ng/l in river waters. The composition of different fullerenes including pristine fullerenes (C_{60} and C_{70}) and functionalized ones from the engineered origin will be discussed. In addition, different degradation studies of fullerenes in water suspensions emulating different environmental conditions and during a wastewater treatment will be presented. Degradation studies have been carried out under controlled conditions of salinity, the humic substances content, the pH and the sunlight irradiation. The results of degradation studies will show that up to ten transformation products are produced, including epoxides and dimers. Finally, the kinetics of generation of each transformation product will be as well presented

WE400

The influence of engineered surface coatings on nanomaterial stability in a complex, natural medium

M. Surette, Oregon State University / School of Chemical, Biological, and Environmental Engineering; J.A. Nason, Oregon State University / Chemical Biological and Environmental Engineering

The colloidal stability of engineered nanomaterials (ENMs) within aquatic environments has been shown to control ENM environmental fate. Prior research using simplified, synthetic mediums has demonstrated that ENM stability is strongly influenced by the engineered surface coating enveloping the ENM. However, it is unknown whether this coating will continue to dictate particle stability when the ENM is dispersed within a complex, natural medium. Instead, it has been suggested that the "eco-corona" acquired by the ENM via interactions with the surrounding environment will govern ENM stability. This implies that within these mediums, engineered surface coatings will have a negligible impact upon ENM stability. The aim of this research was to investigate this subject further and determine whether an ENMs' engineered surface coating remains a relevant factor effecting ENM stability in a complex, natural medium. \n Using samples of a local freshwater river to represent a complex, natural medium, a suite of batch experiments were conducted. Each batch was dosed with a single model ENM, which included 12-15 nm gold-core nanoparticles (AuNPs) with different surface coatings (*e.g.*, diverse surface charges, stabilization mechanisms). Aliquots were collected from each batch over time, immediately centrifuged to remove large particles/aggregates, and the supernatant collected for analysis via ICP-OES. From this, the concentration of unaggregated AuNPs remaining in the supernatant over-time was tracked. Using a method from the literature, heteroaggregation attachment efficiency factors (α_{hetero}) were then calculated. As an indicator of ENM stability, α_{hetero} provides a quantitative metric to determine whether the engineered surface coatings influence ENM stability in the representative medium chosen for this work. \n Despite the uniformity in the medium, significant differences in the stability of the model ENMs were observed. As was expected, the ENMs that maintained a positive surface charge after interacting with the surrounding medium were destabilized. More importantly, however, was that both the neutral and negatively-charged ENMs remained stable throughout the duration of the experiment (8 hrs.). This suggests that the surrounding environment did not affect the stability of these ENMs and demonstrates that ENM stability is influenced by the engineered surface coating, even after interacting with a complex, natural medium.

WE401

Engineered Nanoparticles interactions in secondary wastewater treatment: removal kinetic and efficiency during activated sludge stage.

V. Cappadona, University of Strathclyde / Civil and Environmental Engineering; R. Skuce, Scottish Water Horizons Ltd; C. Knapp, V. Phoenix, University of Strathclyde / Civil and Environmental Engineering

The rapid evolution of nanotechnology poses a unique and significant challenge for wastewater treatment plants (WWTPs). Engineered Nanoparticles (ENPs) are already utilized in a diverse array of applications, including cosmetics, optics, medical technology, textiles and catalysts. As the market grows, we will see increasing input of ENPs into WWTPs. Therefore the increased use and potential toxicity of ENPs poses a challenge for WWTPs due to their potential harmful effects towards activated sludge. The extent to which WWTPs can remove ENPs from the sewage must also be explored, to determine not only likely outflow into receiving waters but also accumulation within the activated sludge itself. To this end we focused on the activated sludge treatment, as the majority of ENPs can remain in wastewater stream throughout preliminary and primary stages. We investigated a range of ENP digestion and analysis protocols to determine the most reliable procedure for ENP analysis from activated sludge. From this, we developed an analytical method involving H_2SO_4-HNO_3 microwave assisted digestion coupled with ICP-OES to measure ENP concentrations. Following this, using laboratory microcosms we assessed the kinetics of ENP removal by activated sludge. The kinetic design we adopted provided different ENPs-activated sludge contact times. ENP concentrations were then analysed in both effluent and settled fraction. Similarly to previous reports, high and quick TiO_2 removal rate (>80% and >99% after respectively 5 and 60 min) during activated stage treatment have been found. However, the details of the mechanisms involved in the removal of ENPs from sewage by activated sludge remain not fully understood, but results of laboratory test and site samples indicate ENPs are rapidly captured-associated with activated sludge. Following this, we exposed activated sludge to repeated cycles of ENP exposure indicative of the cycling of activated sludge in a WWTP. During each cycle, ENPs were efficiently removed. As consequence of this exposure, the significant enrichment of activated sludge biomass with metal based ENPs can result in a secondary hazard, as ENPs rich biomass acting as a "sponge" can accumulate high concentrations of ENPs, which may be released when recirculated within the wastewater treatment or when applied to land. A similar approach is now being adopted to investigate the fate of mixture of ENPs and ENPs from real products within the activated sludge treatment.

WE402

Fate factor of engineered TiO2 nanoparticles in aquatic and terrestrial natural environments

A. Schulz, University of Strasbourg (UdS); G. Quaranta, Université de Strasbourg / CNRS / EOST/LHYGES; S. Lawniczak, Universty of Strasbourg / LHYGES

Nanoparticles are defined as nano-objects between 1 and 100 nanometers in size. Engineered TiO_2 nanoparticles are used in several fields such as construction, cosmetic and food which leads to an important production and inevitably to emissions generating environmental impacts. To quantify them, the Life Cycle Assessmentis a powerful method that is able to characterize TiO_2 NPs according to their fate in environmental media and their effects on ecosystems and human health. The main objective of this study is to determine the engineered TiO_2 nanoparticles (TiO_2 ENPs) fate according to two approaches: experimental and by calculation. For this purpose, it requires firstly to detect and quantify TiO_2 NPs in water, soil and sediment near a production site in Vieux-Thann (68) to determine parameters which contribute on TiO_2 NPs fate in soils, water and sediments. Several analysis methods are used in particular inductively coupled plasma with atomic emission spectrometry (ICP-AES), transmission electron microscopy (TEM) and conductivity or potential of hydrogen (pH) measurements. Then, parameters determined in the previous steps are used to calculate a fate factor of TiO_2 ENPs in a natural environment according to the life cycle impact assessment method calculation. During the study, it was found that ionic strength, pH, percentage of organic matter, soil composition (percentage of clay, silt and sand) or size and concentration of TiO_2 NPs are parameters which matter in TiO_2 ENPs fate in soils, water and sediments. Furthermore, the first results obtained show that the sampling point located upstream of the production site has the lowest concentrations of titanium dioxide in soil and sediments. This point is used as a reference and allows us to consider it as the geochemical background. The sampling points located near and downstream of the site production have higher concentrations of titanium dioxide that may be due to transport of TiO_2 ENPs by wind and effluent released into the Thur. Further studies are needed to determine whether the additional titanium dioxide comes from the production site or not and will confirm or invalidate the presence of TiO_2 NPs manufactured in the different environmental media.

WE403

Assessing the fate and transport of engineered TiO2 nanoparticles in sewer pipes through a dynamic multimedia model (SWNano)

K. Kim, Seoul National University; D. Lee, Seoul National University / Environmental Planning Institute Graduate School of Environmental Studies

During the past decades, the nanotechnology and use of engineered nanoparticles (ENPs) have rapidly developed. It is therefore inevitable that they enter the natural environment such as rivers and marine environments. So various models have developed to expect exposure amounts in environment for proper risk assessment of ENPs. Because there is a lack of filed observation data due to deficiency of monitoring techniques for ENPs. However, previous developed models have not considered fate and transport of ENPs in intermediate path before reaching the each environmental compartment in spite of its importance. For example, more than 60% of TiO_2 ENPs does not directly enter into natural surface water after usage, but they first transported into domestic wastewater. This study analyzed spatiotemporal concentration changes and flux of TiO_2 ENPs in a sewer network through newly developed dynamic model, SWNano. Generally, heteroaggregation of ENPs with suspended particulate matters (SPM) is major process determining the fate and transport of ENPs in water. Water quality such as pH, DOM contents, ionic strength and Suspended solids, etc. and characteristics of ENPs such as shape, size and surface treatment strongly affect aggregation rate of ENPs. Therefore, we estimated attachment efficiency through sedimentation experiment of TiO_2 NP in real wastewater and we compared sedimentation rate with the values experimented in other kinds of water from previous studies. As well as experimental attachment efficiency, various input data of SWNano model such as SPM particle size, number concentration of SPM, zeta potential, etc. were also obtained through measurement by dynamic light scattering (DLS) and nano tracking analysis (NTA), etc. It was confirmed that the TiO_2 NPs aggregated relatively quickly with the SPM in sewage considering that the attachment efficiency was estimated to be about 10^{-1} to 10^{-2}. When applying experimental values to input parameters of SWNano model, the range of predicted concentration of SWNano model were almost matched the concentration range from other model results. The model results showed that dispersed TiO_2 NPs decreased with time, while heteroaggregates (TiO_2 NPs+SPM) increased with time. Besides, it was also verified that the degree of decrease of dispersed TiO_2 NPs concentration in sewer with time is significantly different according to the difference of attachment efficiency.

WE404

The importance of cell wall of marine microalgae in preventing the toxicity of nanoparticles

M. Sendra, CSIC - Spanish National Research Council; J. Blasco Moreno, CSIC Spanish National Research Council ICMAN / ECOLOGY AND COASTAL MANAGEMENT; C. Araujo, Consejo Superior de Investigaciones Científicas / Department Ecology and Coastal Management

Metallic and oxide metallic nanoparticles (NPs), such as Ag and CeO_2 NPs, have increased their global production because they have been widely used in new applications and consumer products such as textile, personal care product, biomedicine and catalysis. NPs-containing wastes discharged in aquatic systems have produced undesirable effects in many marine organisms. Marine phytoplankton is vital in marine ecosystems, as microalgae are at the bottom of the food web and, therefore, any change in microalgae population will have an important effect into the rest of food web. The direct mechanism of NPs toxicity is the physical damage in cell membrane through adsorption of NPs onto the cell wall leading to NPs uptake, bioaccumulation and toxicity in different organelles. Therefore, the hypothesis of this work is that microalgae lacking of cell wall will be more vulnerable to toxic effects of NPs than microalgae with a typical cell wall. To test this hypothesis two microalgae species, *Dunaliella salina*, lacking cell wall, and *Chlorella autotrophica*, with a typical cellulosic cell wall were chosen. Species were exposed to ionic ($AgNO_3$ and $Ce(NO_3)_3$) and NPs (Ag NPs and CeO_2 NPs) forms of Ag and Ce over 72 h and the following responses were assessed: cell density, cell viability, cell size, cell complexity, autofluorescence of chlorophyll *a*, active chlorophyll, effective quantum yield of photosystem II and reactive oxygen species (ROS). Metals in both forms (NPs and ionic) caused negative effects in cell division, inherent cell properties and physiological mechanisms of both microalgae. The general trend was a decrease in active chlorophyll, effective quantum yield of PSII, and cell density and an increase in cell complexity and percentage of intracellular ROS. For both marine species, Ag was more toxic than Ce and ionic forms of both metals were more toxic than NPs. Contrarily to our hypothesis, *D. salina*, despite not having a cell wall, showed to be less sensitive to metals than *C. autotrophica*. Therefore, the cell wall of *C. autotrophica* seemed not to suppose higher protection preventing toxicity of NPs. The higher resistance of *D. salina* against the metals and metallic NPs tested might be related to: (i) its ability to metabolically prevent metal exposure and (ii) the high production of exopolymeric substances that isolates this microalgae from the surrounding contaminated environment.

WE405

Environmental screening of structured hybrid nanoporous materials developed for industrial adsorption applications

A. Booth, J. Farkas, SINTEF Ocean / Environmental Technology; R. Blom, SINTEF Materials and Chemistry

Inorganic-organic hybrid nanoporous materials (NPMs), such as amorphous mesoporous aluminosilicates and Metal-Organic Frameworks (MOFs) are designed and developed for numerous application fields including health care, industrial cooling systems, air purification and gas storage. Their usage and production is expected to increase significantly within the next years, with new applications such carbon capture and storage becoming increasingly important. In this study, we investigate the environmental behaviour of 6 NPMs and determine their toxicity towards the freshwater algae *Raphidocelis subcapitata*. The size, surface charge density and dispersion stability of UiO-66-COOH, Al(OH)-fumarate, HKUST, Zn-CPO, FeBTC-JM-AR and CPO-27-Ni are investigated in relevant environmental test media. Furthermore, we study the dissolution of metals and other elements from NPMs in test media, and their contribution to the observed effects on *R. subcapitata*. Particle size measurements showed that the NPMs have a primary particle size between 200 nm and several micrometres. In freshly sonicated stock dispersions, the measured z-averages ranged from 600 nm (CPO-27-Ni) up to 8 µm (HKUST). ZnCPO and CPO-27-Ni had the most negative zeta-potential of -25 and -20 mV respectively, with Al (OH) fumarate and FeBTC-JM-AR featuring a positive surface charge. UiO-66-COOH and HKUST had very weak surface potentials, which was also reflected in their instability in the stock and exposure media. In a first dissolution study, 5 out of 6 materials (100 mg/L) caused an increase in specific dissolved metals or elements in the exposure media, both directly after dispersion preparation and after a 72 h incubation period, reflecting the duration of an *R. subcapitata* standard toxicity test. Most notable releases after 72 h were from Zn-CPO (Zn, 3457 µg/L), CPO-27-Ni (Ni, 235 µg/L) and HKUST (Cu, 143 µg/L). UiO-66-COOH caused a 100 % increase in S in the exposure media, while Al(OH)-fumarate caused an increase of Al from 11 mg/L to around 60 mg/L. FeBTC-JM-AR was the most inert material regarding release of dissolved metals. Due to their adsorption properties, the materials also drastically reduced amount of P in the exposure media, with UiO-66-COOH also decreasing Ca and Mn. Potential mode-of actions, i.e. impact of NPM particles through depletion of nutrient elements, toxicity of dissolved metals, or effects from organic components will be identified in the *R. subcapitata* toxicity tests.

WE406

Tracking Physicochemical Changes of PAHs in the Presence of TiO2 Nanoparticles by Assessment of Biological Responses

L. St Mary, Heriot-Watt University / EGIS; D. Patsiou, Heriot Watt University / School of Life Sciences; M.R. McCoustra, Heriot Watt University / School of Engineering Physical Sciences; T.B. Henry, Heriot-Watt University / The School of Energy, Geoscience, Infrastructure and Society

Polycyclic aromatic hydrocarbons (PAHs) are a class of persistent environmental contaminants with diverse physicochemistry and toxicity. In contaminated aquatic environments, PAHs frequently accumulate in sediments or are sorbed to particulates in the aqueous phase. Some PAHs are photoactive and have photo-induced toxicity, but little is known about interactions between PAH photoactivity, sorption, environmental fate, and toxicity. Engineered nanoparticles (NPs) can behave as particle agglomerates that participate in sorption/desorption reactions in the aqueous phase, and some NPs (e.g., TiO_2-NPs) also have photoactivity. Aqueous-phase interactions between PAHs and TiO_2-NPs are of interest because they are becoming more environmentally relevant (i.e., as NPs are increasingly released into the environment), and because investigations of sorption/desorption processes, in the context of photoactivation, can provide important new information on physicochemistry of both PAHs and NPs. Previous work conducted by our research group has found that sorption of PAHs onto photo-active NPs promotes photo-catalysis of PAHs thus altering PAHs bioavailability and toxicity under UVA radiation. In these experiments, bioavailability (cytochrome P4501A *cyp1A* gene expression in larval zebrafish) is used as an analytical tool to demonstrate sorption of anthracene and benzo(a)pyrene to NPs in water. Our objective is to investigate PAH/TiO_2-NP sorption under UVA and determine formation of PAH decomposition products (e.g., oxidized polycyclic aromatic hydrocarbons (OPAHs) and their bioactivity. Various combinations of PAH/TiO_2-NP preparations will be exposed to UVA, and changes in gene expression of genes involved in Phase I metabolism (cytochrome P450 *cyp1a1* and *cyp1b1*) and Phase II metabolism (*gst*, *ephx*, *gsh*; and epoxide hydrolases *ephx1* and *ephx2*) in early life stages of zebrafish will be assessed. The exploitation of biological responses to investigate changes in PAH and PAH decomposition product bioavailability during sorption processes will provide novel insight into these processes tested directly within the environmentally relevant aqueous phase.

WE407

Toxicity of TiO2 nanoparticles to freshwater chironomids - pointing out the relevant endpoints

D. Savić Zdravković, Faculty of Sciences and Mathematics, University of Niš / Department of Biology and Ecology; B. Jovanovic, Iowa State University / Natural Resource Ecology and Management; A. Đurđević, J. Stanković, Faculty of Science and Mathematics, University of Niš / Department of Biology and Ecology; D. Milošević, Faculty of Sciences and Mathematics, University of Niš / Department of Biology and Ecology

In the environment, nanomaterials are present in a number of chemical forms, exhibiting specific interactions, mobility, biological availability and potential toxicity. Both ecotoxicologists and pathologists have expressed their concerns regarding the potential negative effects of nanomaterials in live systems and the environment. The present study was carried out in order to assess the influence of TiO_2 nanoparticles (in the form of human white food colorant E171) toxicity on the freshwater midge *Chironomus tentans*. The safety and consequences of the intake of this form of E171 TiO_2 for human health have been recently reconsidered. The experimental design was constructed for the sediment dwelling chironomid larvae according to OECD guidelines. Concentrations of 125, 250, 500, 1000, 2000 and 4000 mg of E171 TiO_2 per 1 kg of sediment were used to assess lethal effects (life traits: mortality, emergence ratio, developmental time and rate). Concentrations of 2.5, 25 and 250 mg of E171 TiO_2 per 1 kg of sediment were used to assess sublethal effect (morhometric changes of mentum, mandibles and wings). The mortality and emergence ratio was affected by a higher nanoparticulate TiO_2 concentration in the sediment (>1000 mg/kg). Sublethal effects on *Chironomus tentans* larvae at environmentally relevant concentrations were shown through morphological changes, which were qualified and quantified using the geometric morphometry approach, principal component analysis and canonical variate analysis. This was the first time a geometric morphometric approach was used to assess the deformities in chironomid larvae exposed to nanoparticles. Geometric Morphometrics revealed the tendency of the mentum teeth to narrow and elongate and the mandibles to widen and loose the first inner tooth, with a rise in the TiO_2 concentration. The present study revealed most suitable endpoints in the case of TiO_2 nanoparticle contamination in freshwaters, using *Chironomus tentans* as a bioindicator. The results show that morphological changest of *C. tentans* could be used as an endpoint in nano-TiO_2 monitoring together with geometric morphometry.

WE408

Multigenerational exposure of the nematode C. elegans to Silver Nanoparticles at the expense of oxidative stress defence mechanisms

L. Rossbach, Norwegian University of Life Sciences UMB / IMV; E. Maremonti, Norwegian University of Life Sciences UMB; M. Kleiven, Norwegian University of Life Sciences; D. Oughton, Norwegian Public Roads Administration / Norwegian University of Life Sciences / Centre for Environmental Radioactivity (CERAD CoE); D.A. Brede, Norwegian University of Life Sciences / Centre for Environmental Radioactivity

Adverse effects of Ag are widely known, with effects ranging from oxidative stress, DNA damage and genetic changes, and inhibition of physiological properties, such as growth and reproduction. Most toxicological studies, however, only cover a limited timescale of the organism's life stage, rather than the whole lifespan, or even across generation. Therefore, knowledge on multigenerational effects is lacking. The current study was conducted in order to determine whether the six generational exposure to sub-lethal concentrations of either ionic or nanoparticulate silver (AgNP) could induce alterations in sensitivity to Ag exposure using the nematode *C. elegans* as a model. Further, changes in susceptibility to other metals and the role of ROS as well as metabolic changes were investigated. Exposure to sub-lethal concentrations revealed increased susceptibility to ionic Ag, while

AgNPs tolerance increased. Results show that adaptation development may occur after just a few generations. Subsequent exposure to paraquat, a known ROS inducer, indicated the involvement of ROS defense mechanisms. Therefore, changes in glutathione redox potential and *sod-1* gene expression were measured, employing the genetically encoded fluorescent biosensors *Grx1-roGFP2*, and the reporter strain *Sod-1::gfp*, respectively. Further, effects of the AgNPs on the central metabolism and implications on energy production are investigated by a measurement of the bioluminescence of luciferase expressing nematode PE255. Findings of this study will aid to further improve the understanding of the toxicity of nanoparticles, as well as contribute to our knowledge about the behavior of *C. elegans* in response to toxicants. **Acknowledgements**: Karl Andreas Jensen and Solfrid Lohne. This work was supported by the Norwegian Research Council funded NanoCharm (221391/E40) and NorNanoReg (239199) projects, and the EU NANoREG project grant agreement n° 310584.

WE409

Effect of silver nanoparticles layer on soil surface to terrestrial species

J. Kwak, S. Nam, Konkuk University; Y. An, Konkuk University / Department of Environmental Heath Science

With developing nanotechnology, uses and release of engineered nanomaterials are increasing. Landfill of biosolid after wastewater treatment is considered as one of indirect exposure sources of nanomaterials. This study focused on the simulation of exposure scenarios of nanomaterials landfills, and set the aim to investigate different toxic effects derived from different scenarios. Silver nanoparticles (AgNPs) was selected as test nanomaterials and 4 different exposure scenarios were considered; 1) control, 2) layer of AgNPs with low concentration (Low-Layer), 3) layer of AgNPs with high concentration (Low-Layer), and 4) mixture of AgNPs and soil with low concentration (Low-Mix). Plant microcosm experiment was conducted in the greenhouse for 9 weeks. Soybean plant was most inhibited in Low-Mix exposure group which mimicked whole mixture of nanomaterials and soil because root surface area exposed to AgNPs were most larger than Low-Layer and High-Layer. In case of soil enzymes, activities were depended on exposure concentration. This study concluded that exposure concentration of nanomaterias as well as depth of nanomaterials layer should be considered in the soil ecotoxicity research area. *This research was supported by Basic Science Research Program through the National Research Foundation of Korea (NRF) funded by the Ministry of Science, ICT and future planning (2016R1A2B3010445).*

WE410

Fragmentation of nano- and microplatics from expanded polystyrene exposed to sunlight

Y. Song, Korea Institute of Ocean Science and Technology; W. Shim, S. Hong, Korea Institute of Ocean Science and Technology / Oil and POPs research group; S. Eo, Korea Institutue of Ocean Science and Technology

Production of nano- and micro-sized plastics through weathering of plastic surface by photo-oxidation was regarded as one of major input sources of secondary nano- and microplastics. Its fragmentation process according to exposure duration and size distribution of fragmented particle down to nanometer scale, however, was not revealed yet. Expanded polystyrene (EPS), one of common marine plastic debris, was known to weather more rapidly than polyethylene and polypropylene in our previous study. Fragmentation of nano- and micro-sized particles was qualitatively and quantitatively determined from the expanded polystyrene (EPS) exposed to sunlight for 9 months. The exposed EPS cubes (3x3 cm surface area) were sampled in duplicate at 2 (2M), 5 (5M) and 9 month (9M) after sunlight exposure. The surface colour was changed from white to dark yellow during exposure. The fragmented particles at the top surface of each cube directly exposed to sunlight were collected in 2 ml solution consisting of HPLC grade pure water with 0.1% tween 80 by sonication for 1 min. The collected particles in solution were sequentially filtered with 10 µm and 0.8 µm pore-size filter paper. The mass of >10 µm EPS particles produced per EPS cube surface area (g/m^2) significantly (p< 0.05) increased according to exposure time; 0.1±0.1 g/m^2 for control, 2.6±0.3 g/m^2 for 2M, 3.9±0.4 g/m^2 for 5M and 7.2±0.2 g/m^2 for 9M. The mean and median size of >10 µm EPS particles measured by laser diffraction was 26-29 µm and 18-20 µm, respectively. The hydrodynamic diameter of the EPS particles in the filtrates of < 0.8 µm pore filter-paper was 532 nm for 2M, 530 nm for 5M and 752 nm for 9M by dynamic laser scattering. Their particle abundances measured by nanoparticle tracking analysis were $1.8x10^9$ particles/ml for 2M and $3.2x10^9$ particles/ml for 2M and 9M. Two month exposure of EPS to sunlight was enough to produce a large number of micro- as well as nano-sized plastics by surface weathering.

WE411

Effects of nano-plastics on natural marine aggregates and their associated microbial communities

S. Summers, SCELSE Nanyang Technological University / SCELSCE; T.B. Henry, Heriot-Watt University / The School of Energy, Geoscience, Infrastructure and Society; T. Gutierrez, Heriot Watt University / School of Life Sciences

Plastic debris in the marine environment is of particular interest, as the issue is one on a truly global scale. The ubiquitous presence of micron sized plastic particles and the knowledge that these will break down into ever smaller, nanometre sized particles, has resulted in a surge of recent research into nano-plastics. However, the difficulty in detection of nano-plastics has made it difficult to predict the specific risks involved in their presence within a marine environment. We recently showed that nano-plastics are readily incorporated into marine snow (MS) particles as mediated by 'sticky' polymeric substances and other particulates. MS particles are described as a heterogeneous matrix composed of polymeric substances, such as EPS, faecal pellets, invertebrate casts and microorganisms. It is therefore expected that the association of nano-plastics with MS would include plastics to the total pool of suspended particulate matter in the ocean. Studies to assess the fate and impacts of this pool of MS-associated nano-plastics, however, are lacking, including the microorganisms found colonizing these particles. Since microorganisms, in particular bacteria, are major colonizers of MS, we hypothesized this would also be the case for MS-associated nano-plastic particles. To assess this, we generated MS-associated nano-plastic particles using natural seawater collected from a subarctic northeast Atlantic region and nanometre-sized polystyrene spheres. Analysis of the nano-plastic-MS particulates by barcoded 16S rRNA gene MiSeq sequencing revealed that the addition of nano-plastics introduced some minor variability within treatments, with respect to microbial composition. The presence of the nano-plastics marginally increased the *a*-diversity of the community associated with the particles, compared to the community associated with MS in the absence of nano-plastics. Statistical analysis, however, did not provide substantive evidence to suggest that these differences and variabilities were significant. Therefore, while nano-plastics may have some minor effect in terms of the diversity of bacteria that colonize MS, it was less than the natural variability observed for the microbial communities that colonized non-exposed (no nano-plastics) MS particles. These results suggest that pristine polystyrene nano-plastics do not exert a major influence in altering the bacterial communities associated with MS particles.

WE412

Tracking nanoplastics in marine bivalves at environmentally realistic concentrations

M. AL SID CHEIKH, University of Plymouth / Marine sciences and engineering; S.J. Rowland, University of Plymouth / School of Geography Earth and Environmental; K. Stevenson, Charles River; C. Roleau, Peches et Oceans Canada; T.B. Henry, Heriot-Watt University / The School of Energy, Geoscience, Infrastructure and Society; R.C. Thompson, Plymouth University / School of Marine Science and Engineering

Awareness campaigns on plastic pollution in oceans are backed by governments worldwide with recent initiatives to ban plastic products such as micro-beads from cosmetics or single-use plastic bags. The fragmentation, the persistence and the production of plastic particles (micro < 1mm, MPs, to nano-size < 1µm, NPs) are among the most prominent environmental issues faced by government environmental agencies. A relevant aspect to assess the risk of a pollutant such as plastic particles is their tissue distribution if ingested by an organism. Therefore, the success of environmental plastic monitoring programs will ultimately depend on the reliability of extraction and detection of plastic particles in tissues of diverse organisms. However, most exposure experiments performed with plastic particles are carried out with unlikely high doses of particles, typically above 1 mg/L while the environmental concentration is expected to be about a part per billion. Detection and tracking of such small particles at these low concentrations are indeed major analytical challenges for environmental and laboratory studies. Our study focuses on the tissue distributions of nanoplastic in marine bivalve in a single pulse exposure of 6 hours and depuration of 48 days in open clean seawater. Here we present, preliminary results where we track nanoplastics at environmentally realistic concentrations in marine bivalves.

WE413

Plastics: does size matter? Impact of environmentally relevant nanoplastics identified in the Nordic environment

T. Gomes, Norwegian Institute for Water Research (NIVA) / Ecotoxicology and Risk Assessment; A. Lusher, NIVA Norwegian Institute of Water Research / Marine and Freshwater Research Centre; I. Nerland Bråte, Norwegian Institute for Water Research NIVA; S. Brooks, NIVA / Ecotoxicology and Risk Assessment; A. Macken, NIVA / marine pollution; D. Eidsvoll, Norwegian Institute for Water Research NIVA / Ecotoxicology and Risk Assessment; M. Reid, Norwegian Institute for Water Research NIVA / Environmental Chemistry Section; A. Georgantzopoulou, Norwegian Institute for Water Research NIVA

Plastic pollution is a widespread concern worldwide. Substantial amounts of plastics are produced and discharged into the environment every year, which will potentially impact aquatic ecosystems and consequently aquatic organisms. Plastic in the aquatic environment can undergo mechanical, chemical and biological degradation that can give rise to the formation of smaller plastic particles, which can be denominated as micro- (< 1 mm) or nano-plastics (< 100 nm) depending on size range. Microplastics are ingested by a range of aquatic organisms and this ingestion might cause adverse biological effects, however less research has been conducted on their smaller counterparts, nanoplastics (NPLs). Similarly to other nanomaterials, NPLs possess size specific properties which could increase their toxic potential towards aquatic organisms depending on surface characteristics and interactions with the surrounding environment. Nonetheless, their presence in the environment and any toxic mechanisms are, to a large extent, unknown. In this study, the impact of environmentally relevant plastics identified in Norwegian environmental samples will be evaluated at the nanoscale in three key marine species, the cryptophyte algae *Rhodomonas sp*, the harpacticoid copepod *Tisbe battagliai* and the blue mussel *Mytilus edulis* and compared to its microscale counterpart. The uptake, accumulation and elimination kinetics of NPLs in the three species will be evaluated under ecologically relevant conditions, as well as their potential transfer along the aquatic food chain. Furthermore, the acute and sublethal ecotoxicological effects of both plastic sizes will be investigated at individual, cellular and molecular levels using different biological endpoints. With the results obtained in this study we aim to discuss the differences in uptake, accumulation and biological responses between different sized plastics identified in the Nordic environment, and consequently bridge the current knowledge gap on the assessment of their potential hazardous effects in marine biota.

WE414

Ecotoxicity of engineered nanomaterials in relation to ecosystem complexity and functioning

W. Peijnenburg, RIVM / Center for Safety of Substances and Products; Y. Zhai, CML Leiden University; M.G. Vijver, CML Leiden University / Conservation Biology

Engineered nanomaterials (ENMs) are widely applied, and the release and accumulation of ENMs through waste effluent and deposition are threatening the aquatic and terrestrial ecosystem. While important knowledge has been gained about the uptake of ENMs on cultured invertebrate and microbial species based on simplified microcosm and mesocosm model systems, extrapolation of results of these studies to events actually occurring at field conditions can be hampered by the plethora of environmental variables that affect the fate and toxicity of ENMs over various spatial and temporal scales. In this context, an overview is given on recent achievements in assessing nano-specific effects on systems varying in physico-chemical and biological complexity. Amongst others, it will be illustrated how ENM toxicity can be affected by the intrinsic physico-chemical characteristics of ENMs (e.g. shape, size, surface charge, coating) and extrinsic environmental characteristics (NOM, pH, electrolytes) and how ENMs interact with various components of food webs. ENMs in the environment may directly or indirectly affect a diverse array of organisms and microorganisms, which likely cascades towards distorted ecosystem processes. We further identify challenging yet promising research areas in this emerging field that are essential in pursuing a realistic risk assessment that accounts for ecosystem complexity and functioning. The take home message is that there is a need of studies assessing not only impacts of ENMs on single species but also a need of a comprehensive framework of nano-specific toxicity in complex ecosystems. Considering the abiotic complexity of the transport of ENMs in the natural environment, studies performed with laboratory-cultured species need to include proper characterization and quantification of the environmental factors that impact fate and effects of ENMs. In addition, the biotic complexity in the ecosystem especially in the aquatic and terrestrial environment indicates that risk assessment of ENMs should be conducted in an integral multi-dimensional perspective. For instance, relatively simple studies on uptake and accumulation of ENMs by invertebrate species need to further investigate the perturbation caused by interactions between the intestinal microbiome and the host. Also, the link between microorganisms and invertebrates in a detrital food web should be included for a systematically evaluation of ENMs toxicity.

WE415

Development of rapid reacting automatic mobile lab responding chemical accident of aquatic environment in Korea

H. PARK, Hanyang Univ. / Regulatory Chemical Analysis & Risk assessment Center; S. ok, Kitech / Regulatory Chemical Analysis & Risk assessment Center; M. Song, J. Ra, Korea Institute of Industrial Technology

Most of available mobile lab developed and operated in Korea are air quality monitory system and there is no rapid reacting mobile lab responding chemical accident of aquatic environment in Korea. We designed rapid reacting mobile lab with two major factors, 24hr operating and rapid starting within 1hr after arrival. We also considered system stability during transportation and accessibility to target river or stream, where we collected vibration information of a vehicle by exposure to off-road and raised spot of a road reducing speed. Vibration vulnerability assessment were conducted with vibration testing shaker. We also collected topographical information related to diverse accessibility characteristic to river by conducting sampling at the selecting sampling point. Lastly, we adopted special air-conditioning system to control the system contamination from exposure to vaporized chemicals at the accident location. We adopted dual power supplying system with 5.5kWh generation capacity for 24hr operation equipped with 2hr supplementary battery system of 10kWh assisting and initiating the system on arrival and in between generator exchange. Vibration testing shakers are established with vibration information collected. Activated carbon proved to be most effective to control our target chemicals, which was composited onto COMBI type filter. These findings will be modulated and structured to maximize system stability. [keyword] chemical accident, mobile lab, rapid monitoring system

WE416

Trophic Interactions in the Bioaccumulation and Depuration of Silver in Fish from a Lake Dosed with Nanosilver

C.D. Metcalfe, Trent University / Water Quality Centre; V.V. Yargeau, McGill University / Chemical Enineering; K. Newman, J.D. Martin, Trent University / Water Quality Centre

Bench-scale tests have shown that silver (Ag) accumulates in the tissues of fish exposed to silver nanoparticles (AgNPs). However, these experiments cannot replicate the complex biogeochemical processes and trophic interactions in natural aquatic ecosystems. This study was conducted as part of whole lake addition project in which a total of 15 kg of AgNPs was added over two ice-free field seasons to a small lake (i.e. Lake 222) in the Experimental Lakes Area in Canada. Both yellow perch (*Perca flavescens*) and northern pike (*Esox lucius*) accumulated Ag in their tissues. The greatest bioaccumulation was observed in the liver tissues of pike, and a single pike sampled in the second year of additions had the highest concentration observed in liver of 5.1 µg/g wet weight. In perch, the highest concentrations of Ag were observed in gill tissue. Monitoring in the lake using passive sampling devices and single particle ICP-MS confirmed that Ag nanoparticles were present in the water column and that Ag was distributed throughout the lake at estimated concentrations in the range of 1-11 µg/L. These data indicate that the primary mode of Ag bioaccumulation in perch was probably through uptake into the gill, whereas pike probably accumulated Ag from the diet. The transfer of Ag from forage fish to piscivorous fish can occur in natural lake ecosystems, leading to concentrations in some tissues that are 3 orders of magnitude greater than the concentrations in water.

WE417

Hepatotoxicity of iron oxide (maghemite) nanoparticles in the guppy Poecilia reticulata

G. Qualhato, Federal University of Goias / Department of Morphology; T.L. Rocha, University of Algarve / CIMA; S.M. Sabóia-Morais, Federal University of Goias / Department of Morphology

Although there are many applications of iron oxide nanoparticles (IONPs) in the nanomedicine and nanoremediation, its ecotoxic effects to aquatic organism remains unclear. In this study, the hepatoxic effects induced by citrate-functionalized IONPs at environmentally relevant iron concentration (0.3 mg L^{-1}) was investigated in female guppy *Poecilia reticulata* by histopathological approach based on qualitative analysis and histopathological index after acute and long-term exposure. The animals were collected at the beginning of the experiment and after 3, 7, 14 and 21 days of exposure. TEM results demonstrate crystalline and rounded IONP with an average size of 3.97 ± 0.85 nm, and DLS and ELS analysis showed that the IONPs has low hydrodynamic diameter and high surface charge in ultrapure water (14.11 ± 0.2 nm; -51.1 ± 7 mV) compared to reconstituted water (21.4 ± 0.39 nm; -19.5 ± 6.5 mV). The histopathological results showed an increase in the frequency of histopathological changes in fish after the 7 days of exposure to IONPs, such micro- and macro-steatosis, melanomacrophage aggregates, exudate and hemorrhagic foci. The acute (3 and 7 days) and long-term (14 and 21 days) exposure of *P. reticulata* to IONPs induced high histopathological indexes associated with circulatory disorders and inflammatory responses with high foci of melanomacrophages indicating an increase of hepatotoxicity according the exposure time. Furthermore, guppies exposed to IONPs showed increasing in the number of MMC when compared to the unexposed ones. This is a first study about hepatoxicity of IONPs in guppies. The results indicated that the hepatotoxicity estimated by qualitative parameters and histopathological index are important biomarkers to indicate the animal health and the environmental impact of IONPs. The present study confirming that the guppy *P. reticulata* is a suitable model to test the hepatotoxicity of IONPs. *Keywords*: Nanomaterials; biomarkers; nanoecotoxicity; guppy. **Session: Ecotoxicology and human toxicology: from molecules to organisms, from omics to in vivo (Fish model species in human and environmental toxicology) Presentation preference: Poster presentation**

(Eco)toxicity tests for hazard evaluation of recycling materials and waste (P)

WE418

Biotests for Hazardous Waste Classification (HP14): benchmarking Limits for Tolerable Ecotoxicity.

R. Weltens, VITO / ABS; e. rossi, OVAM; g. vanermen, VITO; K. Tirez, Flemish Institute for Technological Research VITO

The current HP14 classification is based on the chemical composition of the waste: i.e. the sum of the concentrations of individual chemicals with ecotoxic properties (substances labeled H400, H410, H411, H420) are compared to defined limit values (CLP). In the case of complex waste materials however the analytical data are not suitable for classification and a stepwise approach is proposed using additional to the chemical evaluation as step 1 biotests in steps 2 (acute aquatic tests on eluate fractions) and step 3 (acute terrestrial tests on solid waste). Limit values are needed for tolerable ecotoxicity (TE) that are in line with the chemical limit values (step 1). In the study presented here we benchmarked biotest results against waste materials that were proven to be toxic in step 1, and it was conclusion that LID4 as TE was a suitable option for our data set. The main conclusions were: The proposed set of biotests is essential for proper HP14 evaluation. Poor mass balance in the chemical data should be the trigger for the additional use of biotests. LID 4 is proposed as TE for steps 2 and 3 (to be further evaluated for a larger data set). Both aquatic and terrestrial tests are needed for complete HP14 evaluation. *This study was funded by OVAM, the Flemish Waste Agency! The kind help of the technicians Guy Geukens, Cis Boonen, Wilfried Dumortier is highly appreciated.*

WE419

What is the future for the waste wood in terms of ecotoxicological testing?

S. Legay, FCBA / Chimistry Ecotoxicology Lab; C. Martin, FCBA / Gironde

In Europe, the classification of waste is carried out by an assessment of the hazardousness of the waste using data of known waste composition according to the properties of danger. This classification can be based only on the waste composition if the available data are sufficient and relevant. This method is based on the sum of compounds classified in accordance with the CLP (Classification, Labelling, Packaging) regulation [European regulation [EC] 1272/2008]. In the majority of cases under complex mixtures, or of unknown nature (e.g: exterior and interior joinery, furniture, panels, wooden paneling, wood flooring, construction waste and demolition,..) including wood preservative, paints, glues, the characterisation of their wastes is considered to be difficult. Eco-toxicological testing seems to be the most relevant because the effects of all contaminants (synergistic effects, additives and antagonists) are integrated. It is a major advantage in the characterisation of waste. In this case, the waste has to be then subjected to a battery of bio tests (aquatic and terrestrial) in order to evaluate one of the 15 existing properties: Eco-toxicity for the environment (HP14). Test strategies will allow wood wastes to be recovered or recycled.\n

WE420

QUALITY STANDARDS FOR URBAN WASTE FERTILIZERS: PUTTING ECOTOXICOLOGY IN THE PICTURE

S. Chelinho, CFE   Centre for Functional Ecology / Department of Life Sciences of University of Coimbra; C. Venâncio, Department of Biology / Biology; I. Lopes, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro; J. Sousa, University of Coimbra / Department of Life Sciences

The recently adopted EU Circular Economy Package intends to boost the production and EU movement of fertilizing products, such as those obtained from urban wastes (UWF) and the harmonization of quality standards (certification) for such products, to avoid market rejection. The Portuguese and EU regulation on UWF production/commercialization relies only on physico-chemical/biochemical analysis, which do not give any insight on the fraction of contaminant/mixture of contaminants bioavailable for organisms, nor the existence of potential antagonistic and/or synergistic effects. The main objective of this study is to develop an environmental quality certification system for the use of UWF in agricultural systems. In the present contribution, it was intended to characterize the ecotoxicological potential of the selected UWF, by evaluating both soil habitat and retention functions using lower-tier laboratory tests. Five UWF, two with origin in source separated organic wastes (group I; theoretically with higher quality, ex. lower metal content) and three originated from the organic fraction of mixed municipal solid waste (group II) were selected and tested using a battery of standardized ecotoxicological assays with plants, soil invertebrates and freshwater species. Five soil-UWF dilutions (0.7; 2.1; 6.3; 18.9; 56.7%) and eluates of pure UWF were used as test-medium. The results show that the highest and lowest toxicity were observed in the two UWF from group I. Among soil organisms the range of sensitivities was: *E. andrei* > *F. candida* > *E. crypticus* > *L. sativa* > *T. aestivum* while for aquatic organisms was: *H. viridissima* > *R. subcapitata* > *C. vulgaris*> *H. incongruens* > *B. calyciflorus*. The observed toxicity was probably related with UWF high salinity rather than with metal contents. The obtained data also reinforce the need to include information from biological susceptibility of the receptors potentially at risk on the available regulation to obtain a more realistic view of the potential risks and to adapt the UWF application practices. Ultimately, a sustainable economic growth based on the efficient use of resources/ waste valorization can be promoted.

WE421

Chemical and Ecotoxicological Assessment of Reclaimed Asphalt for their Subsequent Use

V. Jandova, M. Buckova, R. Licbinsky, J. Hegrova, J. Huzlik, K. Effenberger, Transport Research Centre

Reconstruction and repair of the road infrastructure are a source of the reclaimed asphalt, which is suitable to continue to use. It is also necessary to deal with the environmental impact of these materials within their ongoing life cycle, except testing their mechanical properties. Currently, the environmental impact tests of reclaimed asphalts are carried out in crushed condition, according to the leachability test of granular materials with grain size Scenedesmus subspicatus, *Sinapis alba*, *Daphnia magna*) were carried out in aqueous extracts. The results were compared with the legislation and were evaluated in terms of the content of the monitored substances and the type of test material.

WE422

Leaching tests - a useful tool for the environmental impact assessment of construction products

N. Bandow, Federal Institute for Materials Research and Testing / Contaminant Transfer and Environmental Technologies; F. Jürgens, BAM Federal Institute Materials Research and Testing; U. Schoknecht, BAM Federal Institute for Materials Research and Testing

Construction products and waste materials used for construction can be in contact with the environment and may release potential harmful compounds. Information on the total content of these substances in the product is not sufficient to assess its environmental impact since it does not consider realistic exposure conditions. Concerning the pathway to soil and groundwater by contact with rain or seepage water leaching tests are available. The aim of this presentation is to show exemplary results of existing leaching methods and underline the strength and weaknesses of this kind of test with selected examples of our work: Concrete roofing tiles with terbutryn were leached according to CEN/TS 16637-2 with permanent immersion into the water and according to EN 16105 with nine immersion cycles each consisting of immersion and dry stages. The eluates were divided into subsamples for different paramters as pH, conductivity, total organic carbon, anions, cations and terbutryn. Concerning the assessment of this leaching data it is important to notice that the eluate concentration do not represent necessarily environmental concentrations. Thus, the concentration in the leaching test cannot simply be compared to limit values as e.g. environmental qualtity standards. Further considerations are necessary inlcuding exposure scenarios and environmental pathways before leaching tests can be used in risk assessment.

Advances in monitoring and evaluating remedy effectiveness for in situ amendments in soils and sediments (P)

WE423

Assessment and management of stormwater on sediment recontamination: you don't need to measure everything, just the right things

I. Drygiannaki, Texas Tech University / Department of Civil Environmental and Construction Engineering; B. Rao, Texas Tech University / Deparment of Civil Environmental and Construction Engineering; M. Bejar, Texas Tech University; M. Rakowska, Texas Tech University / Civil and Environmental Engineering; D. Athanasiou, Texas Tech University / Civil, Environmental, and Construction Engineering; D.D. Reible, Texas Tech University / Civil and Environmental Engineering; G. Burton, University of Michigan / School for Environment and Sustainability; G. Rosen, SPAWAR Systems Center Pacific; B. Chadwick, Us Navy Spawar Systems Center; M. Colvin, SPAWAR Systems Center Pacific; R. Pitt, The University of Alabama; E. Strecker, B. Steets, M. Otto, Geosyntec Consultants

Stormwater sources are difficult to understand because of the poor characterization of the irregular, event-driven inputs and the difficulty of managing diffuse sources of large volumes of runoff. The study objective is to develop methodologies to quantify the consequences of ongoing low-level sources on chemical concentrations, contaminant availability, and effects on biological receptors in surficial sediments. The study area was in Paleta Creek near Naval Base San Diego (NBSD), in California. Two storm-events were captured for particle size characterization and chemical analysis. Receiving and outfall waters collected using auto-samplers, which were triggered at each location during two different seasons. The samples were analyzed for a variety of metals, PAHs, and PCBs, as a function of particle size. Sediment traps and sediment cores were also collected from the Creek and subjected to bulk chemical analysis. The fractionated water and sediment samples were processed for metal extraction using the modified EPA method 3005A and 3050B, respectively, and were analyzed using ICP-MS and MERX-T. Persistent organic pollutants in water samples were Liquid-Liquid Extracted (LLE) using the modified EPA Method 3510C, while sediment was extracted by Pressurized Fluid Extraction (PFE with ASE 350) using the modified EPA Method 3545A. PAH analysis was performed on HPLC and PCBs on GC/MS. Results showed that storm-events were dominated by coarse particles initially most likely to lead to sediment recontamination in the near field of the receiving water (e.g., PAH, and Cd). Cu was associated to the dissolved and clay fraction, however the depositing loads were more influenced by resuspension and redistribution of sediment than stormwater. Data suggested that PAHs and PCBs, due to low bioavailability as determined with passive sampling and bioaccumulation testing, are not a strong contributor to sediment toxicity which appeared to be better correlated to the presence of pyrethroids in the traps. The study indicated that the size-segregated contaminant loads and simultaneous receiving water measurements were very helpful in relating the stormwater discharges to sediment recontamination. The particle associations in stormwater along with spatial distribution particularly in sediment traps, and less in the sediment cores, can identify contributing locations, effective remedial approaches, and help to propose best practices for stormwater and sediment management.

WE424

Development of active capping materials for oil spill contaminated sediment remediation

L. Silvani, Norwegian Geotechnical Institute; P. di palma, IRSACNR; C. Riccardi, INAIL; E. Eek, s.e. hale, Norwegian Geotechnical Institute; p. viotti, Università La Sapienza; M.P. Papini, Università La Sapienza / Chemistry

Petroleum is extensively used for making oil-based chemical and energy; its daily consume is on average 20 million tons and this is not predicted to decrease. The increasing use of the petroleum is inevitably connected to an increase in oil spills. Oil spills can occur for many reasons including human error or equipment failure and whenever an oil spill occurs it can represent a worldwide environmental problem. Effective remediation strategies are required. The aim of this study is to develop different sorbent materials for the active capping of oil spill contaminated sea-sediment. The experimental investigation was performed on an oil spill contaminated sea sediment. Different sorbent materials were tested for the active capping: a commercial Activated Carbon (AC) Carbonitalia, an organophilic clays (OC) CETCO Inc. and a biochar (BC). The sorption properties of the materials were first investigated in aqueous solution by performing equilibrium tests (isotherm) using a mixture of polycyclic aromatic hydrocarbons (PAHs) as target contaminants. The bioavailable concentration was assessed by using polyethylene (PE) (26 µm) passive samplers. Capping experiments were performed in glass cylinders, where a layer of contaminated sediment (7,5 cm) was capped with a layer of 3 cm of sand mixed with the different tested materials (AC, OC and BC) and synthetic sea water (4 cm) on top. The capping efficiency was assessed by using polydimehylsyloxane (PDMS) fibers as passive samplers. The PAH profiles in the different capping systems were compared in order to assess the capping efficiency. Biochar showed a greater capping efficiency than activated carbon and organoclay. For example, anthracene porewater concentrations in the first centimetre of capping material, were reduced by 69%, 56% and 99% respectively for activated carbon, organoclay and biochar after 1 month. The porewater concentrations were also used to model the long term ($\geq$12 months) behaviour of various cap configurations with a numerical simulations. This study demonstrates that biochar can be considered a cost-effective alternative to the more widely used sorbent materials for capping oil spill contaminated sediments.

WE425

PCB Tissue Concentrations and Benthic Community Impacts at a Carbon Amendment Pilot Study in the Intertidal and Subtidal Zones of San Francisco Bay

C.J. McCarthy, CH2M / Environmental Services; C.A. Irvine, RBI / Ecosystem Services; T. Himmer, CH2M; s. clark, Pacific EcoRisk; R. Zajac, J. Eby, CH2M

Historical site activities at the Hunters Point Naval Shipyard (HPNS) in South San Francisco Bay resulted in the release of chemicals, including polychlorinated biphenyls (PCBs), to offshore sediments. To inform remedy selection at this urban site, activated carbon (AC) amendments alternatives were evaluated in a pilot treatability study. Two 0.4 acre plots extending from the intertidal to the subtidal zone were treated with either AquaGate + PAC™ or SediMite™ were assessed for their potential to reduce ecological risks associated with PCB-contaminated sediment. Previous treatability studies indicated that AC may be effective at reducing the bioavailability of PCBs to the bent-nose clams (*Macoma nasuta*) in shallow intertidal sediments when aided by mechanical mixing. This study assessed the effectiveness of AC placements without mechanical mixing in deeper water that is more representative of conditions where full-scale remediation is expected. Tissue bioaccumulation, benthic invertebrate community composition, and chemical analyses were measured as indicators of remedy effectiveness. Comparisons were made between baseline, reference, and post-amendment conditions (8 and 14 months post-placement). PCB tissue concentrations in *Macoma* sp. were measured *in situ* (field) and *ex situ* (bench-top) after 28-day exposures. Developing field exposure chambers that allowed sediments to infiltrate the chambers and expose clams upon deployment and then retrieve the sediment and exposed organisms for chemical analyses was a challenge. Modifying a chamber design used in previous studies by Luthy et al. (2009) proved successful. Test organisms were another challenge. Tissue bioaccumulation was planned to be conducted with *M. nasuta* but instead, initial measurements were made with *M. secta* (white sand clam) collected at a nearby reference location where *M. nasuta* had been previously found. The species have a similar appearance and life histories but *M. secta* had low survival in the field (< 20%), lab exposures (< 60%), and lab controls (10%). Additional field pilot testing led to the use *M. nasuta* from a supplier for post-amendment monitoring. PCB tissue concentrations were reduced by up to 85% in both pilot amendment areas after 14 months with clam survival greater than 90%. Benthic invertebrate communities in test plots were not significantly different from baseline conditions or among treatments 14 months after AC deployment.

WE426

Remediation of mine wastes with biochar: effect on metal bioavailability to earthworms

M. Almira-Casellas, Leitat Technological Center / HEHS; V. Gonzalez, M. Diez-Ortiz, Leitat Technological Center

The impact of two biochar-based amendments (one from pruning trees and other from sewage sludge) in metal toxicity and bioavailability was assessed in an acid (A) and an alkaline (B) mine waste before and after incubation under different simulated field conditions (irrigation versus drying flooding periods). Metal $CaCl_2$-extractable fraction, survival and metal tissue concentrations in the earthworm *Eisenia fetida* exposed to bulk (mine wastes) were measured. Survival of *E. fetida* was recorded after 21 days of exposure to six serial dilutions of mine wastes mixed with uncontaminated Lufa 2.2 natural soil containing waste concentration of 100, 50, 25, 12.5, 6.25 and 3.13% (w/w), and internal tissue metal concentration in surviving earthworms were measured . No signs of toxicity and no significant effects on survival of the organisms were observed in alkaline mine waste B. In contrast, exposure at time 0 to untreated acid mine waste (A) caused a 71 % of mortality. The addition of biochars decreased toxicity in mine waste A and Cd internal concentrations in surviving organisms, indicating a lower metal bioavailability. Over time, survival in the untreated acid mine waste increased and total internal body metal concentrations in organisms decreased, with no significant differences observed among treatments and incubation conditions. A strong decrease in Cd, Zn and Pb $CaCl_2$-extractable fractions was observed in all the mine waste dilution in both biochar treatments comparing to untreated mine waste, with no significant differences among treatments or flooding conditions. Addition of biochar also leads to an increase of the pH, which might explain the reduction in metal bioavailable fraction and the consequent decrease in organisms' body metal bioaccumulation.

WE427

Remediation of mine wastes with biochar: effect on metal bioavailability to Enchytraeus crypticus.

M. Almira-Casellas, Leitat Technological Center / HEHS; V. Gonzalez, M. Diez-Ortiz, Leitat Technological Center

The overall effect of two biochar-based amendments (one from pruning trees and other from sewage sludge) and their efficacy for metal immobilization in an acid (A) and an alkaline (B) mine wastes were assessed. Two different simulated field conditions, irrigation periods versus alternating flooding-drying periods, were evaluated before, immediately after and after 10 months of incubation. Besides physicochemical characterization, ecotoxicological assays with *Enchytraeus crypticus* exposed to both: i) pore water solutions extracted from mine wastes and ii) bulk mine wastes were conducted to provide a more accurate estimation of metal bioavailable fraction and risk of exposure. Survival of *E. crypticus* exposed to mine waste pore water solution in an inert quartz sand matrix was evaluated after 10 days and internal tissue concentration was measured in surviving organisms. Treated and untreated mine wastes were mixed with uncontaminated Lufa 2.2 natural soil to obtain waste concentration of 100, 50, 25, 12.5, 6.25 and 3.13% (w/w), and effects on survival (LC50) and reproduction (EC50) were recorded after 21 days in all treatments at each dilution concentration. Results showed no significant effects of the alkaline mine waste B and its pore water solution on survival and reproduction of *E. crypticus.* However, exposure to untreated and treated acid mine waste (A) and its pore water caused high mortality in organisms at time 0. Over time, the survival increased and the internal concentrations were lower in the amended mine wastes than in the untreated mine waste A, indicating a lower metal bioavailability. Addition of biochar lead to an increase in the pH and a decrease in Pb, Zn and Cd $Cacl_2$-extractable concentrations in the acid mine waste, suggesting a main role of the pH determining the bioavailable fraction of metals in the soil solution. No significant differences between different type of biochars were found. Our results showed that biochar treatments decreased the bioavailable fraction of Pb, Zn, and Cd in the soil solution, reducing the toxicity of the acid mine waste to enchytraeids.

WE428

Bioavailability-based Methods to Assess Remediation Effectiveness

J. Gan, University of California, Riverside / Department of Environmental Sciences; J. Wang, University of California Riverside; A.R. Taylor, University of California Riverside / Environmental Sciences; D. Schlenk, University of California-Riverside / Department of Environmental Sciences

Compared to the total chemical concentration, bioavailability is a better measurement of risks of hydrophobic organic contaminants (HOCs) to biota in contaminated soil or sediment. Many different bioavailability estimation methods have been introduced to assess the effectiveness of remediation treatments. However, to date the different methods have rarely been evaluated against each other, leading to confusions in method selection. In this study, four different bioavailability estimation methods, including solid phase microextraction (SPME) and polyethylene passive sampling (PE) aiming to detect free chemical concentration (C_{free}), and Tenax desorption and isotope dilution method (IDM) aiming to measure chemical accessibility, were used in parallel to estimate bioavailability of DDT residues (DDXs) in a historically contaminated soil after addition of different black carbon sorbents. Bioaccumulation into earthworm (*Eisenia fetida*) was measured concurrently for validation. Activated carbon or biochar amendment at 0.2-2% decreased earthworm bioaccumulation of DDXs by 83.9-99.4%, while multi-walled carbon nanotubes had a limited effect (4.3-20.7%). While all methods correctly predicted changes in DDX bioavailability after black carbon amendment, passive samplers offered more accurate predictions. Predicted levels of DDXs in earthworm lipid using the estimated bioavailability and empirical BCFs matched closely with the experimentally derived tissue concentrations. However, Tenax and IDM underestimated bioavailability when the available DDX levels were low. Our findings suggested that both passive samplers and bioaccessibility methods may be used in assessing remediation efficiency, presenting flexibility in method selection. While accessibility-oriented methods offer better sensitivity and shorter sampling time, passive samplers may be more advantageous because of their better performance and compatibility for *in situ* deployment.

WE429
Identification, Quantification, and Risk Assessment of Polycyclic Aromatic Hydrocarbons and their Polar Derivatives in Soil After Steam Enhanced Extraction
L.S. Trine, Oregon State University / Chemistry / Environmental and Molecular Toxicology; E. Davis, United States Environmental Protection Agency / Ground Water & Ecosystems Restoration Division; S.L. Massey Simonich, Oregon State University / Department of Environmental and Molecular Toxicology
Polycyclic aromatic hydrocarbons (PAHs) are environmental contaminants produced from the incomplete combustion and pyrolysis of organic matter, and are among the major contaminants in soils. Steam enhanced extraction (SEE) is an *in-situ* thermal remediation technique that uses the addition of steam to soils subsurface to increase the removal efficiency and recovery of volatile and semi-volatile contaminants, like PAHs. However, there is limited research on formation of PAH derivatives during SEE of PAHs. Polar PAH derivatives are more mobile in the environment than PAHs, and some are more toxic than corresponding PAHs. There is an urgent need for analytical methods that can accurately quantify PAH derivatives in complex matrices, to better understand the chemistry occurring during SEE, and how it interferes with the remediation of soils. In this study creosote-contaminated soil from the Wyckoff/Eagle Harbor Superfund Site in Washington, USA was thermally treated with laboratory-scale SEE. Soil pre- and post-SEE, effluent collected during SEE, and pre- and post-SEE leachate samples (mimicking rain runoff and groundwater) were collected and analyzed with gas chromatography/mass spectrometry (GC/MS) for PAHs, polar PAHs, and MW302-PAHs (n=97). Most of analyzed PAHs were quantified in all pre-, post- and effluent samples. PAHs decreased significantly, while polar PAHs increased in mean concentration post-SEE soil. Mass balances were estimated for different PAHs, and some were above 100%, indicating the potential formation of PAH derivatives during SEE. These findings suggest that SEE pilot and treatability studies should include PAH derivatives to risk assessments to assess the full effectiveness of SEE and prevent underestimation of potential risks. A quantitative risk assessment will be performed by calculating $B[a]P_{eq}$ concentrations and estimated lifetime cancer risk (ELCR) ingestion estimates. Developmental toxicity testing will be conducted with dechorionated zebrafish (*Danio rerio*) embryos (n=32/treatment) placed into 96-well plates containing pre- and post-SEE soil, effluent, and pre- and post-leachate extract samples at 6 hours post fertilization. Developmental toxicity will be assessed by evaluating morphological changes, embryonic/larval photomotor behavior, and mortality at 24 and 120 hours post fertilization. This study quantified PAH derivatives after SEE, and it will identify implications for risk assessment and developmental toxicity outcomes.

WE430
Enhanced total petroleum hydrocarbon removal without soil disturbance by serial surfactant foam spraying
R. Bajagain, Y. Park, Kunsan National University; S. Jeong, Kunsan National University / Department of Environmental Engineering
Fuel oils are a complex mixture of hydrocarbons. Low-molecular hydrocarbons are readily biodegraded in the natural soil system. Diesel includes recalcitrant hydrocarbons to the natural degradation and may act as potential and actual sources of harmful human and ecological effects. The purpose of the study was to evaluate serial surface foam spraying technology, which avoids disturbing the soil, to deliver chemical oxidant and oil-degrading microbes to unsaturated soil. Hydrogen peroxide was used for pretreatment of Diesel prior to bioaugmentation. All oxidants were applied to the surface soil by surfactant foam spraying. Surfactant foam would be a good media to spread remediation agents to the surface of contaminated sites with less labor or energy. Surfactant foam was sprayed once onto diesel contaminated soil for oxidation of soil total petroleum hydrocarbon (TPH). Periodic bioaugmentation foam was sprayed every three days for biodegradation of soil TPH. Foam spraying employing oxidation-bioaugmentation serial application significantly reduced soil TPH concentrations to 550 mg/kg from an initial 7470 mg/kg. Application of hydrogen peroxide by foam spraying increased the infiltration of hydrogen peroxide into the unsaturated soil. The easy and even infiltration of remediation reagents increased the contact with contaminants, resulting in enhanced oxidation and biodegradation. Fractional analysis of TPH showed C18-C22 present in diesel as biodegradation recalcitrant hydrocarbons. Recalcitrant hydrocarbons were reduced by 92% using oxidation-biodegradation serial foam, while biodegradation alone only reduced the recalcitrant fraction by 25%. (This work was supported by National Research Foundation of Korea (NRF-2015R1D1A1A01059664)).

WE431
Factors affecting sorption of halogenated phenols to polymer/biomass-derived biochar: Effect of pH, hydrophobicity, and deprotonation
S. Oh, University of Ulsan / Department of Civil and Environmental Engineering; Y. Seo, University of Ulsan / Civil and Environmental Engineering; T. Seo, University of Ulsan / Department of Civil and Environmental Engineering
High performance biochar synthesized via co-pyrolysis of polymer and rice straw (RS) was evaluated as a sorbent for ionizable halogenated phenols. Compared with RS-derived biochar, the sorption of 2,4-dichlorophenol (DCP), 2,4-dibromophenol (DBP), and 2,4-difluorophenol (DFP) to polymer/RS-derived biochar was significantly enhanced by changing properties of biochar due to polymer residues, probably via hydrophobic sorption and electron donor-acceptor interactions. Removal of polymer residues and increasing aromaticity of polymer/RS-derived biochar at elevated pyrolysis temperatures affected the sorption capacity of halogenated phenols. Surface charge of biochar and deprotonation of the halogenated phenols according to solution pH were other factors to be responsible for the sorption to polymer/RS-derived biochar. Competition with other halogenated phenols and dissolved cations implied that similar sorption mechanisms were existed and that surface complexation and electron donor-acceptor interactions were involved in the sorption to polymer/RS-derived biochar. Our results suggest that co-disposal of biomass and thermoplastic wastes through pyrolysis may be an effective option to produce a high-performance upgraded biochar as a sorbent for various types of contaminants.

WE432
Biochar for soil management: interactions with legacy contaminants and current-use pesticides
L. Bielská, RECETOX, Faculty of Science, Masaryk University / Faculty of Science, RECETOX; K. Kročová, Masaryk University / Faculty of Science,RECETOX; L. Skulcova, Masaryk University / Research Centre for Toxic Compounds in the Environment (RECETOX)
Biochar (BC) is a product of thermal decomposition of biomass under oxygen-limited conditions. BC has received extensive attention because of its multi-functionality for agricultural and environmental applications. Its excellent sorption properties make BC a valuable sorbent in the treatment of solids contaminated with hydrophobic organic compounds (HOC) and wastewater. Due to its assumed stability, BC is considered an environmentally compatible approach for carbon sequestration and thus, climate change mitigation. Its application to agricultural soils has been shown to increase soil fertility, mainly due to improved nutrient and carbon availability, protection of microorganisms, and increased water holding capacity. Despite these benefits, very little BC is currently utilized as soil amendment, mainly because the mechanisms improving soil health are poorly quantified and understood. Besides, there are several concerns and unknowns for BC agricultural application: i) the presence of pollutants (HOC, heavy metals) originating in pyrolysis or feedstock; ii) lower specific density resulting in transport of BC and BC-associated pollutants into surface water bodies; iii) substantial reduction of efficacy of agrochemicals; iv) effect of BC stability during weathering on its application potential, particularly long-term fate of agrochemicals. In this study, the role of BC in the management of contaminated soils and in the management of pesticide-treated agricultural soils is addressed. A systematic appreciation is devoted to the positive effects of reducing bioavailability of toxic contaminants and the ambiguous effects of reducing bioavailability of intentionally applied current-use pesticides. For that purpose, two different biochars were applied at increasing doses to soils amended with DDE and with epoxiconazole and tebuconazole as representative of legacy contaminants and broadly used fungicides, respectively. Bioavailability was assessed by means of solid-phase microextraction as well as by measuring the uptake of chemicals by earthworm *Eisenia andrei* as a model ecotoxicologically relevant organism. The influence of biochar properties and biochar dose was considered. At the same time, the well-being of earthworms in biochar-amended control soils was detected. This study aimed at balancing the dosing of biochar in soil management to ensure both an efficient and sustainable control of diffusive contamination and of pests.

WE433
PREPARATION AND CHARACTERIZATION OF COMPOSITES OF TYPE CLAY / POLYMERS AND THEIR USE IN THE REMOVAL OF CONTAMINANTS ORGANICS OF AQUATIC ENVIRONMENTS.
M.S. Rodrigues, Instituto Federal do Maranhão; L. Aguiar Vieira, Universidad Federal do Maranhão; A. Costa Filho, S.G. RIBEIRO, Universidade Federal do Maranhão
Clays have been used by mankind to many years due to its easy to obtain, its high technical viability rate and low economic cost. Among countless uses for the clay, research related to its various properties and also the adsorption capacity of this material has gained prominence in various scientific sectors. With the modification and / or activation of these properties can be improved, resulting in a direct increase in the adsorption rate. The changes made in the samples of clays of this work were made from 2,6-bromoioneno polymer, the process of cation exchange, which granted organophilic characteristics for samples, allowing comoadsorventes of organic compounds were used. Serious damage to fauna and flora are caused by the use of organophosphate pesticides, this leads to search for new methods aimed at removing it mainly aquatic environment. Given that the application of this work aims at the preparation and characterization of organophilic clays, as well as to evaluate its adsorption rate of these compounds orgânicos.Para it was used a natural clay from the Carolina-MA region. infrared techniques, X-ray diffraction, scanning electron microscopy (SEM), mass determination tests adsorbent and determining the concentration of contaminants were used to characterize the samples. At the end of the process it was noticed a structural modification of the clay shown in SEM, but also the vibrational result given by the infrared showing major carbon chains present in the composite nano derived polymer, it caused a significant increase of

adsorption capacity the material was 40% in natura, reaching a value of 78.4% after modification, demonstrating the feasibility of the process and material.

WE434

Field sampling and ex-situ bioassays for assessing the ecotoxicological risk of trace elements in different rehabilitated bauxite residues

E. Di Carlo, R. Courtney, University of Limerick / Department of Biological Sciences & The Bernal Institute; A. Boullemant, RioTinto; L. Poizat, Alteo-Alumina

Bauxite residues are the by-product of the aluminium industry, resulting from the extraction of alumina from bauxite ore through the Bayer process. They could pose an environmental risk mainly because of their alkaline nature and the presence of trace elements (TE). For their rehabilitation, phytostabilization seems to be a promising, cost-effective and non-resource intensive option. However, there are still few studies evaluating the long-term success of the rehabilitation programmes and most of them do not consider ecotoxicology. The present study aims at assessing the success of rehabilitation strategies for bauxite residues considering the ecotoxicological risk of TE to organisms that live in the rehabilitated areas. To this end, two approaches were adopted: field sampling and ex-situ bioassays. On the one hand, samples of bauxite residues, plants and soil macroinvertebrates were collected from a range of field sites, over different seasons, and the environmental concentrations of TE were measured through ICP. On the other hand, exposure tests with earthworms (following OECD guidelines) and RHIZOtest (ISO 16198) with three herbaceous species were carried out, under controlled laboratory conditions, in order to investigate the bioavailability of TE. Data from the field represent the first step to understand whether there is a possible ecotoxicological issue for wildlife. Comparing the TE concentrations measured in the environment with the literature thresholds, some problematic elements (such as V) and sites (such as the one with compost capping) are identified. Regarding the laboratory bioassays, data show that the bioavailable fraction (taken up and accumulated by both plants and earthworms) does not always correlate with the fractions measured in the chemical extractions of the bauxite residues, indicating the inadequacy of chemical methods when comes to evaluate the ecotoxicological risk. In conclusion, the outcomes of this research are likely to shed light on the bioavailability of TE in a complex matrix, such as bauxite residues, and to provide a more realistic risk assessment for the organisms living there. Our data clearly show that the chemical total concentrations measured in the bauxite residues do not predict the bioavailable (potentially toxic) fraction of the TE, therefore bioassays should be taken into account when fixing the rehabilitation goals or assessing the rehabilitation success of a contaminated area.

Ecotoxicology of micro and nanoplastics: Mechanistic approaches to understand their risk for the environment and human health (P)

TH001

Synthetic textile fibers end up in agricultural soils - Can these microplastics pose a threat on soil organisms?

S. Selonen, University of Helsinki / Department of Ecological Science; C. van Gestel, Vrije Universiteit Amsterdam / Ecological Science

An important route of microplastics (MPs) to the environment is the release of synthetic textile fibers to waste water due to laundry. The major part of the fibers is retained in the sewage sludge during waste water treatment and ends up in green spaces and agricultural fields. In this study, we explored the accumulation and effects of polyester (PES) fibers in soil invertebrates, using exposure concentrations of 0.02 %, 0.06 %, 0.17 %, 0.5 % and 1.5 % of PES fibers in dry Lufa 2.2 soil. The fibers were mixed in with the soil to achieve as homogenous mixture as possible. Polyester fibers in soil affected the survival and reproduction of the enchytraeid worm *Enchytraeus crypticus*, but the effects did not show a consistent dose-related pattern. Compared to the control soil, the survival was decreased at fiber concentrations of 0.17 % and 0.5 %, whilst the reproduction was decreased in all other treatments except for the 0.06 % concentration. Polyester fibers did not affect the survival and reproduction of the springtail *Folsomia candida* and the oribatid mite *Oppia nitens*, the survival and feeding activity of the woodlice *Porcellio scaber*, nor the survival of the earthworm *Eisenia andrei.* Some of the measured endpoints seemed to show a negative response to the fibers, but the variation within the treatments was high, possibly also because it was difficult to achieve a completely homogenous distribution of the fibers in the test soil. The results of this study indicate that synthetic textile fibers can have slight negative effects on soil-dwelling enchytraeid worms, whilst arthropods were not markedly affected by the fibers. However, not only enchytraeid worms (*E. crypticus*), but also isopods (*P. scaber*) ingested the fibers in when exposed in soil, but the rate of accumulation was not related with the fiber concentration in the soil. As the accumulation of microplastics in soils is evident, the effects of different types of microplastics and their possible role as carriers of xenobiotics to soil organisms need to be further investigated. This study is part of the project IMPASSE – Impacts of Microplastics in Agro Systems and Stream Environment.

TH002

Effects of microplastic particles of polyhydroxybutyrate towards photosynthetic aquatic organisms

M.G. Pleiter, Universidad Autónoma de Madrid; G. AMARIEI, Universidad de Alcala; M. Tamayo-Belda, Universidad Autónoma de Madrid; F. Leganes, Universidad Autónoma de Madrid / Biology; R. Rosal, University of Alcala; F. Fernandez-Piñas, Universidad Autónoma de Madrid / Biology

Nowadays, the ecological impact of microplastics and nanoplastics in freshwaters are not well understood [1]. Here, we have investigated the effects of microplastic particles (nominal size 5 mm) of polyhydroxybutyrate (PHB) in two organisms representative of freshwaters, the filamentous cyanobacterium *Anabaena* sp. PCC7120 and the green alga *Chlamydomonas reinhardtii*. Firstly, we have performed the physicochemical characterization of microplastic particles of PHB and their potential degradation products in MilliQ water by nanoparticle tracking analysis (NTA), dynamic light scattering (DLS) and infrared spectroscopy (IR). Then, we have evaluated the biological effects of PHB on cellular growth, pigment content and several physiological parameters (metabolic activity, formation of intracellular reactive oxygen species and cytoplasmic membrane) in both photosynthetic organisms by flow cytometry using several fluorochromes. The results indicate that PHB released nanoparticles. NTA allowed to analyze the abiotic depolymerisation of PHB after 72 h in MilliQ finding a wide range (75 - 300 nm) of PHB nanoparticles. PHB induced a decrease in cellular growth and chlorophyll content in both photosynthetic organisms. Furthermore, PHB induced an increase in the level of intracellular reactive oxygen species and induced changes in membrane potential. In conclusion, microplastic particles of PHB exhibited toxicity towards photosynthetic aquatic organisms probably due to the release of a wide range of nanoparticles as a consequence of its own abiotic depolymerisation.

[1] Koelmans AA, Besseling E, Shim WJ. 2015. Nanoplastics in the aquatic environment. Critical review. In Marine anthropogenic litter (pp. 325-340). Springer International Publishing. *Acknowledgement* - This research was supported by CTM2016-74927-C2-2-R grant from MINECO/FEDER EU.

TH003

Differential responses of biomarkers in tissues of the blue mussel Mytilus edulis exposed to microplastics at environmentally relevant concentrations

M. Revel, Catholic University of the West / UBL, Mer Molécules Santé; f. lagarde, Institute of molecules and materials of Le Mans / Institute of Materials and Molecules of Le Mans IMMM UMR CNRS; H. Perrein-Ettajani, Catholic University of the West / UBL, Mer Molécules Santé; M. Bruneau, Catholic University of the West / UBL MMS Angers; F. Akcha, R. Sussarellu, J. Rouxel, IFREMER / Laboratoire décotoxicologie; P. Decottignies, B. Cognie, Universite of Nantes / UBL MMS Nantes; A. Chatel, Catholic University of the West / UBL, Mer Molécules Santé; C. Mouneyrac, Université Catholique de lOuest / UBL, Mer Molécules Santé

Since the early 1970s, the occurence of floating plastic has been reported in marine waters with great accumulation in gyres. In recent years, the presence of plastic debris < 5mm called microplastics (MPs) which result mainly from macroplastic's fragmentation has also been reported in aquatic ecosystems even in remote areas. Several studies have reported the presence of MPs and their effects in marine organisms. However, it appears necessary to investigate their potential toxicity especially at environmentally relevant concentrations. The aim of our study was to evaluate the bioaccumulation and toxicity of polypropylene (PP) and polyethylene (PE) fragments towards the blue mussel *Mytilus edulis.* These polymers were selected according to a previous study conducted *in situ* in the Region Pays de la Loire. Mussels were exposed in laboratory during 10 days at two environmentally relevant concentrations of 0.008 and 10 µg/L (9 and 11,250 particles/L respectively) (Desforges et al., 2014), and to a higher concentration of 100 µg/L (112 500 particles/L) of each MPs type. The exposure was followed by 10 days of depuration in clean seawater (without MPs). MPs fragments were prepared in the laboratory from commercially available products by milling; characterized in terms of size, shapes and they were counted. Following exposure, tissues and biodeposits (faeces and pseudofaeces) were chemically digested and analyzed for MPs recovery using infrared micro-spectroscopy. Regarding potential toxic effects, detoxification and oxidative stress mechanisms through measurement of enzymatic activities of Glutathione-S-transferase (GST), Catalase (CAT) and superoxide dismutase (SOD) were evaluated as well as markers of the immune system and DNA damage. Results showed the presence of PE and PP particles in digestive glands of mussels exposed to the highest concentration tested (100 µg/L) of MPs, and in biodeposits where MPs were observed at all tested concentrations. Significant increases in SOD and CAT activities were observed in the digestive glands of mussel's exposed to 0.008 and 10 µg/L and in gills from mussels exposed to 100 µg/L of MPs that could be indicative of an oxidative stress. This study brings new results on the potential sublethal effects of MPs at environmentally relevant concentrations of MPs.

TH004

Effects of zebrafish exposure to high-density polyethylene and polystyrene microplastics at molecular and histological levels.

G. Limonta, University of Siena / Department of Physical Sciences, Earth and Environment; A. Mancia, L. Abelli, University of Ferrara / Department of Life Sciences and Biotechnology; M. Fossi, C. Panti, University of Siena / Department of Physical Sciences, Earth and Environment

Due to the constant increase of plastic use and production, microplastics (MPs) have become a contaminant of serious concern for the marine environment. However, detailed information about biological pathways affected by the exposure to different MP polymers is still lacking, in particular at transcriptome level. The present study focused on the identification of the molecular pathways affected by a chronic exposure of zebrafish (*Danio rerio*) to different concentrations of a combination of two environmentally relevant MPs for 20 days. Adult zebrafish were fed daily with dry fish food (control group, N=12) and food supplemented with a mix of pristine high-density polyethylene and polystyrene microplastics (two experimental groups: 0.1 and 1 mg/L, N=12 each). The microplastics dimension ranged from below 25 µm to 90 µm for both polymers. At the end of the exposure period, the liver was dissected and its whole transcriptome analyzed by next-generation RNA-sequencing technologies on an Illumina platform. In addition, the gastrointestinal tract and the gills were dissected and fixed for histology and immunohistochemistry. The exposure to polyethylene and polystyrene microplastics affected the liver transcriptome in a dose-dependent way, inducing the differential regulation of specific suites of genes. Histological analyses evidenced changes in the inflammatory response occurring at the two mucosal tissues selected for observation. The correlation of histological alterations with differential gene expression will be addressed and discussed. This study provides a comprehensive transcriptomic dataset useful for ecotoxicological studies on other fish species.

TH005

Uptake and Effects of Synthetic and Natural Microparticles in the Shrimp Palaemon varians

M. Weidung, University DuisburgEssen; R. Saborowski, L. Gutow, Alfred Wegener Institute for Polar and Marine Research

Microplastics (< 5 mm) have become ubiquitous in waters. The smaller they are the easier they can be taken up by aquatic organisms. Once ingested they can cause various harmful effects. This study investigates the effects of size of artificial and natural particles on the induction of cellular stress in the Atlantic ditch shrimp (*Palaemon varians*). The study includes feeding experiments with different sizes of fluorescent microplastic particles, nanosized titanium dioxide particles and silica powder of diatoms as a reference for natural particles. The uptake and distribution of particles in the digestive organs was observed by fluorescence microscopy. As marker for oxidative stress we measured the activities of the antioxidant enzymes catalase and superoxide dismutase (SOD) in extracts of the midgut glands of animals which were fed with particles from 2 to 48 hours. The larger particles (2 µm and 10 µm) remained in the stomach and in the lumen of the gut. The smaller particles (0.1 µm) were translocated into the surrounding tissues and entered the cells of the midgut gland. Decapods have a stomach with fine-meshed filter structures which prevent the uptake of particles > 170 nm into the digestive gland. Superoxide dismutase activity was rapidly induced when the animals were exposed to 0.1 µm plastic particles. The activity increased within 2 hours after microplastic ingestion and remained high after 48 hours. Slight difference appeared between natural and synthetic particles. The diatom powder also induced SOD activity which, however, continuously decreased with time. Due to the enzyme cascade where SOD reacts first, the activity of catalase was clearly lower. It can be assumed that any particles < 170 nm enter the cells of the midgut gland and induce oxidative stress. Following optical detection of reactive oxygen species (ROS) via confocal laser scanning microscopy will help to identify cellular reactions after exposure to microparticles.

TH006

Microplastics in the sub-surface layers of the South Atlantic Ocean

V. van der Schyff, North-West University / Unit for Environmental Sciences and Management; J. Karstensen, GEOMAR - Helmholtz Centre for Ocean Research Kiel; R. Morard, Bremen University / MARUM - Centre for Marine Environmental Sciences; S. Speich, Ecole Normale Supérieure de Paris; H. Bouwman, North-West University / Unit for Environmental Science and Management

Microplastic particles in the ocean is a major environmental concern. Most studies tend to concentrate on the ocean surface when examining microplastic pollution. However, it is known that, for various reasons, microplastics can lose buoyancy and sink. During this study, the stratification of microplastic particles in the Southern Atlantic Ocean (SAO) was determined. The study was conducted from the *RV METEOR*, a German research vessel. The cruise was from Cape Town, South Africa, to Rio de Janeiro, Brazil, from the 29th February 2016 to 18th March 2016. A multinet with a mesh size of 25 µm was deployed at fourteen stations across the SAO, and sampled at at increments of 20 m (0-20 m, 20-40 m, 40-60 m, 60-80 m, and 80-100 m). The contents of the multinet samples were filtered through a 1 mm^2 sieve. The remainder of the samples was pressure filtered through black filter paper (to ensure optimal visibility of the microplastic particles), and air-dried. The dried samples were examined under a dissection microscope, and the microplastic particles counted visually. The highest density of microplastic particles were found in the top layer (0-20m), at 52%. Seventeen percent of the particles were found at 20-40m, 14% in 40-60m, 9% in 60-80m, and 8% in 80-100m. There was a high microplastic count near the South African coast (10°E-0°). After crossing the Walvis Ridge and sailing into the high pressure system over the SAO, the plastic count decreased dramatically. A fairly homogenous stratification was observed in the high pressure system. Near Brazil, the microplastic concentrations increased again. In the eye of a cyclonic eddy, microplastics were slightly less stratified. This study was intended as a pioneer study to determine whether microplastics are stratified in the water column. This was found to be the case.

TH007

Effects of dietary microplastic exposure on fish intestinal physiology

G. Asmonaite, H. Sundh, N. Asker, University of Gothenburg / Department of Biological and Environmental Sciences; B. Carney Almroth, University of Gothenburg Sweden / Department of Biology and Environmental Sciences

The accumulation of plastics in digestive tracts of aquatic biota has been extensively documented and ingestion has been proposed as a prominent exposure route for plastic debris for a variety of aquatic animals, including fish. Large plastic items are known to physically block the intestinal passage, exert physical damage, impair food uptake and/or cause false satiation. Whereas, the biological effects resulting from ingestion of smaller micro-sized particles or microplastics (MPs) are considerably less documented and potential (negative) consequences on the alimentary tract are largely unknown. To address this, we designed a study aimed at assessing if/ how ingestion of MPs can affect physiological function of the intestine in fish. We hypothesized that ingestion of MPs cause inflammatory responses and disturb intestinal barrier and transporting functions. Juvenile rainbow trout (*Oncorhynchus mykiss)* were exposed via diet to polystyrene (PS) particles (50-250 µm, 10mg of PS MPs/fish/day) for a period of 4 weeks. Fish were fed four types of diets: control (no PS MPs) and diets containing untreated PS particles (PS-virgin) or particles exposed to sewage (PS-sewage) and industrial harbor (PS-harbor) effluent. To assess the functional adversity of dietary PS MPs exposure, integrity and transport function of the proximal and distal intestine was investigated. Metabolically active intestinal epithelia was mounted in modified Ussing chambers. Epithelial integrity was monitored as the transepithelial electrical resistance (TER;Ω*cm^2) and the diffusion rate of $^{14}C^{-}$mannitol. Active transport was monitored as potential difference (TEP;mV) and short-circuit current (SCC;µA) together with uptake rate of ^{3}H-lysine. Overall morphology was observed using histology. Gene expression analysis of immune related genes (TGFβ, TNFα, IL-8, IL-10, IL-17, IL4/13A) and tight junction proteins (Occludin, ZO-1, Tricellulin) was performed to examine if PS particles and chemical contaminants induced inflammation in intestinal tissue. The innate immune response (lysozyme stability and complement system) in blood plasma was evaluated to assess the presence of systemic inflammation. The findings of the study indicated no or minor functional effects on fish intestinal tissue inflicted by particle exposure. Signs of inflammation were detected and were accompanied by upregulation of tight junction proteins, suggesting activation of intestinal homeostasis in response to PS MPs exposure.

TH008

Biochemical responses and histological effects resulting from foodborne exposure to post-consumer microplastics in juvenile Solea senegalensis.

M. Martins, Faculty of Sciences andTechnology, Universidade Nova de Lisboa / Dep. Science and Environmental Engineering; J. Neves, Faculty of Sciences and Technology Universidade Nova de Lisboa / Department of Environmental Sciences and Engineering; C. Gonçalves, MARE - Faculty of Sciences and Technology, Universidade Nova de Lisboa / Department of Environmental Sciences and Engineering; P. Sobral, MARE - Faculty of Sciences and Technology, Universidade Nova de Lisboa / DCEA; M. Costa, MARE - Faculty of Sciences and Technology Universidade NOVA de Lisboa / Department of Environmental Sciences and Engeneering

Microplastics (MPs) have dramatically increased in marine environments, being recognized as ubiquitous environmental pollutants. Two types of microplastics are usually detected in environmental samples: the primary MPs originally and intentionally manufactured MPs and the secondary MPs which result from the fragmentation of dumped plastic items. The latter are normally referred as post-consumer microplastics and usually present different shapes, colors, composition ad irregular surface. However, few toxicological studies have addressed exposure of marine organisms to post-consumer microplastics. The goal of the present research is to assess the effects of post-consumer microplastics in juvenile *Solea senagalensis*, using diet as the microplastic vehicle. For this purpose, four distinct diets were prepared using commercial fish pellets incorporated with two MPs sizes (< 200 µm and 300-500 µm) and two concentrations of each (562 and 56 MPs per day) and other without MPs (control), making a total of five treatments. Sixty-day laboratory assays were conducted, in duplicate, and the test pellets were provided to fish once a day. After 14, 30 and 60 days, fish were collected from each treatment and excised. The liver, gills and brain of each fish were excised and stored at -80ºC for biochemical analysis. The digestive tract was also sampled and immersed in Davidson´s fixative for histopathological analysis. Biomarkers related with oxidative stress were analysed, namely the lipid peroxides, glutathione and the activity of catalase, superoxide dismutase and acetylcholinesterase. Histopathological analyses were also performed in the digestive tract to assess the presence of MPs and possible histopathological effects.

TH009

Nanoplastic impacts on physical, biochemical, and nutritional characteristics

of pacific whiteleg shrimp

Y. Chae, Konkuk University; D. Kim, Konkuk University / Department of Environmental Health Science; Y. An, Konkuk University / Department of Environmental Heath Science

Because of enormous amounts of plastic wastes in marine environment, the concerns about marine pollution and ecological damages on marine organisms have increased. Especially, among these plastic wastes, small sized plastic particles like microplastics (< 5 mm) and nanoplastics (< 100 nm) are getting a lot of attentions and the researches about their impacts and effects in environments are under way. In this study, we assess various physical, biochemical and nutritional changes in the bodies of pacific whiteleg shrimps (*Litopenaeus vannamei*) exposed to nanoplastics. For 21 days, shrimps were fed mussels (*Mytilus edulis*) contaminated with nanoplastics (44 nm diameter) and their physical (length, weight, water contents, body mass index), biochemical (catalase, CAT; glutathione s-transferase, GST; superoxide dismutase, SOD; fecal microbiota viability), and nutritional (crude lipid, crude protein, amino acids, and fatty acids) changes were assessed after exposure. In results, nanoplastics attached on the filter and ingested to mussels entered the bodies of shrimps and affected the health and physicochemical properties of shrimps. Especially, biochemical changes were significantly induced in the bodies of shrimps. These results can be the evidence of the impacts of small sized plastics on marine organisms. *This research was supported by Basic Science Research Program through the National Research Foundation of Korea (NRF) funded by the Ministry of Science, ICT, and future planning (2016R1A2B3010445).*

TH010

Brood Pouch-mediated Polystyrene Nanoparticle Accumulation During Daphnia magna Embryogenesis

M.G. Vijver, CML Leiden University / Conservation Biology; N.R. Brun, CML Leiden University / Institute for Environmental Sciences

Nanoplastic debris is ubiquitously distributed in aquatic environments and are considered an emerging environmental issue affecting organisms across trophic levels. While ingestion of particles receives most attention, other routes of uptake and cellular uptake remain unexplored. Here, the planktonic filter feeder *Daphnia magna* was used to track routes of uptake and target tissue of polystyrene nanoparticles (PSNP). A sub-lethal concentration of 5 mg L^{-1} fluorescent PSNPs (25 nm) was used to monitor accumulation in adult animals as well as their embryos in the open brood pouch. A time series throughout embryonic development within the brood pouch revealed accumulation of PSNP in lipophilic cells in the early stages of embryonic development while the embryo is still surrounded by a chorion and before beginning of organogenesis. In contrast, PSNP particles were neither detected in the gut epithelium nor in lipid droplets in adults. An *ex vivo*exposure of embryos to PSNP demonstrated a similar accumulation of PSNP in lipophilic cells, illustrating the likelihood of brood pouch-mediated PSNP uptake by embryos. Whether the observed brood pouch-mediated PSNP uptake ultimately translates to long-term effects under chronic exposure to environmentally relevant concentrations remains a challenging area for further research. By demonstrating embryo PSNP uptake via the brood pouch, data presented here give novel insights in bioaccumulation of nanoparticles and likely other lipophilic contaminants. Since this uptake route can occur within a diverse array of aquatic organisms, this study warrants consideration of brood pouch-mediated accumulation in efforts studying the hazards and risks of nanoparticle contamination.

TH011

Micro- and nanoplastic ingestion in blue mussel larvae

S. Rist, DTU (Technical University of Denmark) / Department of Environmental Engineering; A. Baun, Technical University of Denmark / DTU Environment; N.B. Hartmann, Technical University of Denmark (DTU) / DTU Environment

A large number of aquatic species have been found to ingest microplastics in the field and in laboratory studies. Benthic invertebrates seem to be especially exposed to this form of pollution and the blue mussel *Mytilus edulis* is one of the species that has been investigated most in this respect. Studies have not only shown that the mussels ingest microplastics but have also reported diverse adverse effects on a cellular to a physiological level. However, the work has so far only focused on adult mussels and it is unclear how blue mussel larvae interact with and are affected by plastic particles in the micrometre and nanometre size range. Therefore, this research aimed at studying microplastic ingestion and potential physiological effects in blue mussel larvae. The first experiment aimed at quantifying the amount of ingested and egested particles. Ten day old larvae were exposed to two different sizes of fluorescent polystyrene microbeads (2 µm and 100 nm) and body burdens of particles were quantified after 4h. Subsequently, larvae were transferred to clean water to analyse the amount of egested particles after 24h and 72h. The second experiment investigated potential effects of plastic particles on growth and development of the larvae. They were exposed to 3 different concentrations of the 2 µm and 100 nm beads, representing low (0.45 µg/L), medium (28.7 µg/L) and high (287 µg/L) exposure levels, for 2 weeks. Every 2-3 days larvae from the different treatments were sampled, fixed and photographed to analyse larval size and morphology. Results showed that the larvae readily ingested both particle sizes although ingestion appeared to be more efficient for the 2 µm beads. Egestion of micro- and nanoplastic particles did take place but was not complete within 72h, with 43% of the 2 µm and 61% of the 100 nm particles remaining in the animals. Potential effects on larval growth and development remain to be analysed. By taking other life stages into account and using a quantitative approach for analysing particle ingestion and egestion, this study contributes to enhancing the mechanistic understanding of microplastic – blue mussel interaction.

TH012

The sub-lethal impact of polystyrene microplastics and nanoplastics on the Mediterranean mussel M. galloprovincialis

M. Capolupo, erasmus mudus PhD in Marine and coastal management (MACOMA) - University of Cadiz / Inter-Departmental Research Centre for Environmental Science (CIRSA); P. Valbonesi, University of Bologna / Departmento of Biological, Geological and Environmental Science (BiGeA); S. Franzellitti, University of Bologna / Department of Biological, Geological and Environmental Science (BiGeA); E. Fabbri, University of Bologna / Bigea Department via Selmi Bologna

The contamination of marine environments by microscopic plastic debris is a current threat to the fitness of the exposed biota, and even higher concerns are risen on its potential fragmentation to the nanoscale. In the framework of the JPI OCEANS project 'PLASTOX', we investigated the chronic effects induced by a 21-day exposure to 1.5, 15 and 150 ng/L of polystyrene microplastics (MP, 3µm) and nanoplastics (NP, 50 nm) on the fitness of the marine mussel *Mytilus galloprovincialis*. To do so, we employed a multibiomarker approach encompassing immunological responses (lysozyme and phagocytosys), lysosomal endpoints (lysosomal membrane stability and neutral lipids), oxidative stress (catalase activity, malondialdehyde and lipofuscin content) and detoxification (glutathione S-transferase) parameters and neurotoxic effects (acetylcholinesterase activity). The lysosomal membrane stability, whose impairment is a known general stress symptom, was generally reduced in mussel hemocytes after exposure to both MP and NP; however, only in MP-treated mussels this effect was accompanied by a decreased phagocytic activity. Lysozyme activity in hemolymph was affected by either MP or NP treatments, suggesting a generalized alteration of the immune system efficiency. All the performed treatments led to an accumulation of neutral lipids in the mussel digestive gland. Moreover, an accumulation of malondialdehyde and lipofuscin was observed at 150 ng/L NP and at 1.5 and 150 ng/L MP. In gill, catalase was up-regulated following either MP (1.5 and 15 ng/L) or NP (1.5 ng/L) treatments, while a decreased acetylcholinesterase activity was noted only at 15 ng/L NP. Biomarker data were integrated in the Mussel Expert System (MES), which estimates the stress level induced on mussels by calculating a A-E scaled health status index (HSI). The MES did not identify health alterations in control and at 1.5 ng/L MP (HSI = A), while the onset of a low stress level (HSI = B) was detected at 15 and 150 ng/L MP. Differently, the stress level associated to NP treatments was moderate (HSI = C) at 1.5 and 15 ng/L, and low at 150 ng/L. Overall, results show that both polystyrene MP and NP induce a chronic stress syndrome in mussels by affecting lysosomal integrity and generating pro-oxidant conditions. However, the two particle types can differentially alter immunological and neurological processes, with the exposure to NP resulting in a higher impact on the overall mussel fitness compared to MP.

TH013

Effect of cationic amino (PS-NH2) polystyrene nanoparticles in brine shrimp Artemia franciscana nauplii: biochemical and molecular responses

I. Varo, CSIC Spanish National Research Council / Biology, culture and pathology of marine species; A. Perini, CSIC Spanish National Research Council; E. Bergami, University of Siena / Physical, Earth and Environmental Sciences; G. Liberatori, University of Siena / Department of Physical, Earth and Environmental Sciences; I. Corsi, University of Siena / Physical, Earth and Environmental Sciences

The accumulation of plastic litter on beaches and open oceans has been identified as one of the major threats in marine ecosystems worldwide. Laboratory experiments have proved that the formation of nano-sized plastics during the polymer degradation may reach marine ecosystem, considered as the most in danger. In present study, the effect of 50 nm cationic amino polystyrene (PS-NH_2) was investigated in nauplii of *Artemia franciscana*, which is commonly used as aquatic model organism in toxicity tests. Acute toxicity tests were performed on nauplii exposed to sub-lethal suspensions of PS-NH_2 (0.1, 1 and 10 µg/mL) in natural sea water (NSW) for 48 hours. The toxicity was evaluated by measuring growth and several biomarkers as carboxylesterase (CbE), glutathione-S-transferase (GST), cholinesterase (ChE), heat shock protein (HSP70), lipid peroxidation (LP) and catalase (CAT), involved in important physiological processes, such as biotransformation of xenobiotics, neuronal transmission and oxidative stress. The effect of PS-NH_2 (at 0.1 and 1 µg/mL) on the expression of genes related to metabolism, biosynthesis and embryogenesis during the development of brine shrimp was also investigated.Genes included *HSP26*, *HSP70*, mitochondrial uncoupling protein 2 (*UCP2*), chaperonin-containing TCP (*TCP*) and late embryogenesis abundant (*LEA*). Acute exposure to sub-lethal suspensions PS-NH_2 caused a significant decrease in growth in *A. franciscana* nauplii, as well as significant changes in all biomarkers studied, except for LP. A significant up-regulation of *HSP26* and *HSP70* was observed in nauplii exposed to 1 µg/mL of PS-NH_2 as well as the modulation of *TCP*, the latter not significant. This supports the results obtained from biomarkers, suggesting a stress response and potential apoptotic pathway following PS-NH_2 exposure. On the contrary, no significant effect on gene expression related to the brine shrimp's metabolism (*UCP2*) was observed, and *LEA* was significantly modulated only at the lowest concentration tested. These findings indicate that stress-related responses are taking place in exposed nauplii after acute exposure to sub-lethal suspensions of PS-NH_2, and confirm the general concern about PS-$NH_2$and their ability to represent an ecological treat for marine organisms. Given the increasing levels of plastic pollution in the oceans, additional studies should be done considering long-term exposure to analyze the potential risk of nano-sized plastics in marine environments.

TH014

The impact of nanoplastics on Antarctic krill Euphausia superba

E. Bergami, G. Liberatori, University of Siena / Department of Physical, Earth and Environmental Sciences; C. Manno, C.M. Waluda, British Antarctic Survey; S. Cappello, CNR IAMC; I. Corsi, University of Siena / Physical, Earth and Environmental Sciences

Under current climate change scenarios, Antarctic krill (*Euphausia superba*) is facing multiple stressors, which could affect its abundance and distribution. Microplastics have been recently reported in Antarctic waters, representing an additional potential impact on krill population. In this study we investigated the effects of model nanoplastics (< 1 µm) on krill juveniles through short-term exposure (48 h) of polystyrene nanoparticles (PS NPs) with different surface charge. The behaviour of anionic (60 nm PS-COOH) and cationic (50 nm PS-NH_2) NPs in Antarctic natural seawater (NSW, 34‰, 2°C) was also investigated by Dynamic Light Scattering. PS-COOH formed nanoscale aggregates (average size of 862 nm) in Antarctic NSW, while PS-NH_2 maintained their nominal size. No mortality was observed upon exposure to 2.5 µg/ml PS NPs after 48 h. However, krill exposed to PS-NH_2 showed lower motility than individuals exposed to PS-COOH and were characterised by significant up-regulation of *cb6* gene involved in new cuticle formation. Similar findings reported for other microcrustaceans have been associated with mortality over long-term exposure. Both PS NPs also accumulated in faecal pellets (FPs), which were characterised by lower density and sinking rate compared to control. Our findings demonstrate that PS NPs are able to affect swimming behaviour, cuticle formation and FPs properties of Antarctic krill, with potential serious consequences on Southern Ocean food web and biogeochemical cycle.

TH015

Exposure to nanoplastics as a potential stressor on Mytilus galloprovincialis

I. Brandts, M. Teles, Universidad Autonoma de Barcelona; A.P. Gonçalves, . Barreto, University of Aveiro / Biology Department & CESAM; L. Franco-Martinez, A. Tvarijonaviciute, Universidad de Murcia; M.A. Martins, University of Aveiro / Chemistry Department & CICECO; A.M. Soares, University of Aveiro / department of Biology & CESAM; L. Tort, Universitat Autonoma Barcelona / Department of Cell Biology Physiology and Immunology; M. Oliveira, University of Aveiro

Plastic pollution in the marine environment is a worldwide problem, especially since plastic materials degrade into nano-size particles, becoming more bioavailable and constituting a source of entry of other contaminants into marine organisms. The present study aimed to assess the effects of polystyrene nanoparticles (PSNP) and clarify their modulation of short-term carbamazepine (cbz) toxicity on bivalve *Mytilus galloprovincialis*. Mussels were exposed for 96h to a concentration range of PSNP, to cbz alone and to the mixture of PSNP and cbz. Molecular and biochemical biomarkers were evaluated in digestive gland, gills and hemolymph. Abundance of mRNA in digestive gland and gills revealed significant alterations in expression of genes associated with biotransformation, DNA repair, cell stress-response and innate immunity. Combined exposure induced significant down regulation in gene expression when compared to individual exposure. Total oxidant status values suggest oxidative stress after exposure to 0.5 mg/LPSNP, whereas increased total antioxidant capacity and esterase activity suggest activation of antioxidant defenses after exposure to 50 mg/L PSNP. Exposure to 0.05 and 0.5 mg/L PSNP induced effects on neurotransmission in hemolymph. In gills, almost all experimental exposures induced inhibition of AST and ALT values. Genotoxicity was found in hemocytes after exposure to PSNP, cbz and their mixture.

TH016

The role of microplastic size and type on PAH sorption and bioavailability to copepods

L. Sørensen, SINTEF Ocean / Environmental Technology; E. Rogers, Norwegian University of Science and Technology; M.U. Rønsberg, SINTEF Ocean / Environmental Technology; D. Altin, BioTrix; A. Booth, SINTEF Ocean / Environmental Technology

It has been proposed that microplastic (MP) may act as a vector for a wide range of chemical pollutants already present in the environment. Polycyclic aromatic hydrocarbons (PAHs) are ubiquitous pollutants in the environment, known to cause adverse effects to a variety of marine organisms. Most PAHs have relatively high polymer-water partition coefficients, meaning their potential for sorption to, and transport by, MPs is high. In both field and laboratory studies, a broad range of marine species have been shown to ingest significant quantities of MP, with extended periods of retention observed in some cases. If PAHs are adsorbed to the MPs, this could present an alternative exposure route to PAHs for such species. However, the effect of MP sorption on PAH bioavailability in the marine environment remains poorly understood. Although several studies have attempted to study bioavailability of MP-adsorbed PAHs to aquatic organisms, most studies employ approaches that are unable to accurately determine if PAH bioavailability results from adsorbed compounds or from compounds that have dissolved from the MPs into the exposure media. Here, we investigate the sorption kinetics and present adsorption isotherms for three model PAHs (fluoranthene, phenanthrene and 1,3-dimethylnaphtalene) to a range of different MP's in natural seawater. The selected PAHs exhibit different sizes and hydrophobicities, thus having varying seawater solubility (two orders of magnitude). In the case of the least soluble compound, fluoranthene, MP sorption could prove an important route of uptake in pelagic organisms. To account for the natural variability of MPs present in the marine environment, test materials with different sizes, shape (particles, fibres) and polymer compositions (polyethylene and polystyrene microbeads, polyester microfibres) were used. Using a novel approach, the influence of MP sorption on PAH bioavailability to two marine copepod species (*Acartia tonsa* and *Calanus finmarchicus*) was investigated using polyethylene particles with size ranges above and below the ingestion limit for the two species. The range of MP diameters used in the experiments was ~10-300 µm. Chemical body burden was measured after exposure to determine bioavailability. \n

TH017

Limited influence of microplastics on the effects of an endocrine disruptor on the African clawed frog (Xenopus laevis)

S. Rehse, Leibniz-Institute of Freshwater Ecology and Inland Fisheries / Ecophysiology and Aquaculture; A. Zikova, W. Kleiner, W. Kloas, Leibniz-Institute of Freshwater Ecology and Inland Fisheries; C. Zarfl, University of Tuebingen / Center for Applied Geoscience

Microplastics are ubiquitously distributed in freshwater ecosystems. Besides physical effects of the mere microplastic material itself, chemical effects by associated contaminants are discussed as potential threat to aquatic organisms, in particular after ingestion. Anthropogenic hormonal active substances, so called endocrine disruptors (ED), can affect juvenile sex determination and development of vertebrates like fish and amphibians but also humans. Tadpoles are not only vulnerable to ED, but potentially also to particulate contaminants like microplastics because they filter the surrounding water unspecifically during the first phase of their development. Until now, it is not clear if microplastics have negative impacts on amphibians and if they alter toxic effects of chemical contaminants like ED. This study focused on potential effects of microplastics on tadpoles of the amphibian model species *Xenopus laevis*. The aim of this study was to analyse (1) if microplastics alone can have negative impacts and (2) if the presence of microplastics increases the effects of an endocrine disruptor. The oral contraceptive 17α-ethinylestradiol (EE2) was used as model ED; polyamide (PA) particles in the size range of 15-20 µm (mean diameter) with an irregular shape were used as model MP. Tadpoles were exposed in batches with chronic exposure for 21 days to one concentration of EE2 (10^{-8} M) and a low and a high concentration of PA-particles (1 and 100 mg L^{-1}) separetely and in combination with each other. Stress hormones and larval development as well as sexual differentiation were assessed by gross-morphology and histology. Biomarkers, e.g. vitellogenin, were analysed as EE2 specific endpoints. The concentration of EE2 in water was assessed analytically for treatments including EE2. Physical effects of the microplastic particles themselves on larval development and sexual differentiation were not observed. Only increased levels of the hepatic biomarker vitellogenin showed higher exposure of EE2 in treatments including PA particles in comparison to treatments without microplastics. All other EE2 specific endpoints were not influenced by PA particles. These results indicate that microplastics only play a minor role for the effects of a hormonal active chemical in amphibians and thus provide insights for an indepth risk assessment of MP in the environment.

TH018

Kinetics of POPs sorption and plastic additives release to a variety of polymers under Arctic conditions

D. Herzke, NILU - Norwegian Institute for Air Research / FRAM Centre Tromso; K. Sakaguchi-Soeder, Technische Universitaet Darmstadt / Institute IWAR Chair of Material Flow Management and Resource Economy Germany; R. Sempere, V. Fauvelle, Mediterranean Institute of Oceanography, Marseille; A. Booth, SINTEF Ocean / Environmental Technology

The PLASTOX project investigates the ingestion, food-web transfer, and ecotoxicological impact of microplastics (MPs), together with the persistent organic pollutants (POPs), metals and plastic additive chemicals associated with them, on key European marine species and ecosystems. PLASTOX combines field-based observations, laboratory tests and manipulative field experiments to study the ecological effects of MPs. The use of common microplastic reference materials, including a marine litter-derived MP produced from an environmentally weathered fish box, allows a meaningful comparison of data generated by different partners and across the different activities of PLASTOX. As part of a long-term field experiment conducted at marine locations across Europe (Mediterranean to

Arctic), a range of different virgin polymer pellets(LDPE, PP, PS and PET), as well as marine litter-derived microplastic particles from the fish box, were deployed underwater in the small boat harbor at Tromsø, Northern Norway for up to 12 months. The deployment device consisted of an empty stainless steel SPMD canister, with the various plastic types placed in reusable, empty teabags made of PP, placed separately in nylon netting. Sampling was conducted 1 week, 1 month, 3 months, 6 months and 12 months after deployment. Hydrophobic persistent organic pollutants such as PAHs, PCBs, DDTs, PBDEs and pesticides were measured to establish the adsorption kinetics in seawater under Arctic conditions. Samples were extracted using ultrasound and nonpolar solvents, followed by GPC and SPE clean up. Chemical analyses using GC/MS/MS and GC/qMS was done in the laboratories of the TU Darmstadt and NILU, Tromsø. In addition, release kinetics of common plastic additives including phthalates, organophosphate esters, bisphenols and perfluorinated chemicals were estimated from other four post-industrial virgin pellets (LDPE, PS, PVC, PET) according to the same sampling protocol. Chemical analysis was performed using either GC/MS or LC-QTOF.

TH019

Characterization of microplastics present in personal care products and the study of its toxicity mixed with chlorpyrifos on juveniles of Solea senegalensis.

G. Albendín, Universidad de Cádiz (Spain) / Toxicology Area; I. Cabrera-Pozo, University of Cadiz / Toxicology Area; D. Coello, R. Rodríguez-Barroso, J. Quiroga, University of Cadiz / Environmental Tecnology; J. Arellano, University of Cadiz / Toxicology Area

In the last decades, different components from personal care products have arrived at aquatic ecosystems because these products are not biodegraded or removed in wasterwater treatment plants. Some of the personal care products contain plastic microbead such as exfoliating shower gel, toothpaste and make-up. Creams commonly used and available in supermarkets of our area were used by these assays. The microspheres available in these samples were separated and characterized. The particles were identified by Fourier transform-infrared (FT-IR) spectroscopy using a PerkinElmer Spectrum 100. The spectra are recorded in reflection mode in the spectral range 4000-650 cm^{-1} by co-adding 128 scans at a resolution of 4 cm^{-1}, the particles were identified by comparing FT-IR absorbance spectra of the microplastics to those in a polymer reference library. The microplastics were used in toxicity test. In aquatic ecosystems and their organisms are exposed to complex mixtures of environmental contaminants as pesticides and microplastics. Thus, the effects of microplastics interaction with chlorpyrifos, an organophosphate pesticide, have been studied in this work. The toxicity studies were carried out during 96 hours with continuous ventilation and water renewal every 24 hours, at a temperature of 19-20ºC and under 12h light/12h dark exposure. The juveniles of *Solea senegalensis* (weigth 3,07 ±0,49 g) were exposed to five nominal concentrations of chlorpyrifos (5—80 µg/l), three concentrations on this compound mixed with microplastics (chlorpyrifos: 5-10-20 µg/l; microplastics: 0.150 mg/l), microplastics alone (0.150 mg/l), plus an untreated control and a solvent control (acetone). In these assays not mortality was observed on juveniles with both compounds and their mixtures. Cholinesterases (ChE) have been used as specific biomarkers to diagnose exposure of natural populations to organophosphates pesticides. In general, there are two type of ChE presented in fish, acetylcholinesterase (AChE) and butyrylcholinesterase (BChE). The AChE was analyzed on the head homogenate of juveniles, after this crude was inhibited with iso-OMPA, which is a specific inhibitor of BChE. The results showed that there was inhibition activity in the head of *Solea senegalensis* in presence of chlorpyrifos. However, it was not observed significant differences between the same concentrations of chlorpyrifos and its mixture with microplastics.

TH020

Are microplastics inhibitory to Daphnia magna and are they significant vectors for hydrophobic organic pollutants?

C.K. Frydkjær, Aalborg University / Biology and Environmental Science; N. Iversen, Aalborg University / Department of Chemistry and Bioscience; P. Roslev, Aalborg University / Biology and Environmental Science

The presence of microplastics in aquatic ecosystems is of increasing global concern. Ingestion of microplastics may result in adverse effects in aquatic organisms, and microplastics may increase exposure to harmful chemicals by serving as vectors for hydrophobic pollutants. This study investigated: i. the amount of regular and irregular shaped microplastics ingested and egested by the filter feeder *Daphnia magna* during exposure and gut clearance; ii. the adverse effect of microplastic with and without sorbed phenanthrene; and iii. the significance of phenanthrene sorption by microplastic compared to sorption by naturally occurring plankton organisms (bacteria, yeast and algae). *Daphnia magna* rapidly ingested regular shaped microplastic beads (10-106 µm) and irregular shaped microplastic fragments (10-75 µm) with uptake rates between 0.7 and 50 plastic particles/animal/day. Microplastic exposure concentrations ranged between 0.0001 and 10 g/L. Gut clearance was slower and apparent gut residence time was longer for irregular shaped microplastic fragments compared to regular shaped microplastic beads. The acute inhibitory effect of irregular shaped microplastic fragments was also more pronounced with an EC50 (48 h) value of 0.065 g/L whereas regular shaped microplastic beads were much less inhibitory. Microplastic morphology is therefore a factor that should be considered when conducting experiments with filter feeders because most environmental microplastics are likely irregular in shape. The potential of microplastic to act as vectors for hydrophobic organic pollutants was examined using [^{14}C]phenanthrene as tracer. Radioactivity measurements showed that polyethylene microplastic particles sorbed less [^{14}C]phenanthrene compared to natural plankton organisms (bacteria, yeast and algae). The abundance of phytoplankton and bacterioplankton is often much greater in aquatic ecosystems than the present concentrations of microplastic particles. Hence, live and dead plankton organisms are likely more critical carriers of hydrophobic pollutants than microplastics. This suggests a more limited role of microplastics as significant aquatic vectors for hydrophobic pollutants under current environmental conditions.

TH021

Microplastics as vector for hydrophobic organic chemicals in fish: a comparison of two polymers and silica particles, using three different model compounds

M. Tivefälth, University of Gothenburg Sweden; G. Asmonaite, University of Gothenburg / Department of Biological and Environmental Sciences; E. Westberg, IVL Sweden Environmental Research Institute; B. Carney Almroth, University of Gothenburg Sweden / Department of Biology and Environmental Sciences

Plastic pollution is a recognized global issue and the ubiquity of microplastics (MPs) in aquatic environment is a cause for concern. Potential effects on organisms are still not fully understood, and mechanistic understanding, required to fully reveal consequences of exposure, especially in connection to chemical mixtures, is still lacking. Studies have shown that MPs have capacity to sorb and concentrate hydrophobic organic chemicals (HOCs) in the aquatic environment. There is an ongoing debate about MPs as vectors for chemical contaminants and their relative importance compared to other naturally occurring particulates. The goal of present study was to quantify particle-mediated chemical transfer and using biomarker approach examine associated biological effects in three spined stickleback (Gasterosteus aculeatus). An experimental feeding study with chemically spiked particles (250µm) was conducted. Two types of synthetic polymeric particles (PE and PS) and non-plastic polymer particles (silica), which were selected as reference material, were used. Selected particles were loaded with three model compounds (benzo(α)pyrene, ethinylestradiol and chlorpyrifos) having distinct toxicological modes of action and different hydrophobicity (log Kow) values. Eight different experimental diets: control diets (negative control), diets with clean particles (PE, PS, silica), diets containing, particles spiked with a chemical mixture (PE-mix, PS-mix, silica-mix) and, finally, diets loaded with only chemical mixture (chemical control) were developed. During the experiment, fish were fed daily (6 % of body weight and 5 % particles) for a period of two weeks. Gene expression of well-established biomarkers (CYP1a, ERα, VTG and AChE) were then quantified at mRNA level in the liver and gut. Acetylcholineesterase (AChE) activity was measured in brain. Results showed that all treatments containing chemical mixtures caused changes in gene expression and altered enzymatic AChE activity. Differences could also be seen between particle types, where PS contaminated particles showed similar pattern with the non-plastic particles, while PE contaminated particles showed lower expression levels, indicating a smaller transfer of chemicals. The chemical control was for some treatments lower than the particle groups indicating a particle-mediated chemical transfer in fish.

TH022

Dietary exposure to polystyrene microplastics contaminated with environmental pollutants induce hepatic biomarker responses in fish

G. Asmonaite, University of Gothenburg / Department of Biological and Environmental Sciences; B. Carney Almroth, University of Gothenburg Sweden / Department of Biology and Environmental Sciences

In the field of microplastics (MPs) research, polystyrene (PS) particles have become reference material not only for investigating the uptake of the particles, but also for assessing biological effects. There is a growing body of (eco)toxicological information, suggesting that PS MPs, in a range of nano- to micro- meters (< 50µm), have a potential to impact aquatic organisms. On the other hand, there is an evident lack of toxicological information in regards to bigger size ranges of these MPs (>50µm), at sizes, detectable in the environmental matrixes. We conducted an experimental study aimed at elucidating effects resulting from dietary exposures to PS particles ranging in size 50-250 µm. In this study, juvenile rainbow trout (*Oncorhynchus mykiss*) were exposed diets, enriched with PS particles (10mg of PS MPs/fish/day) for 28 days. We used environmentally contaminated PS particles from *in situ* exposures from two environmental matrixes (undiluted sewage effluent and industrial harbor runoff). As PS MPs largely exceeded sizes relevant for biological uptake, it provided us an opportunity to study particles' role as vector for environmental pollutants. Three different experimental diets, containing virgin (PS), sewage (PS-sewage) and harbor (PS-harbor) exposed particles, were developed. Thereafter, a suite of oxidative stress biomarkers was investigated in the hepatic tissue of fish exposed to PS MPs via gene expression analysis (NRF2, GR, GST, GS, GPx, CAT, GCLmod, GCLcat, SOD) and enzymatic assays (GR, GST, GS, CAT). Additionally, mRNA levels of established biomarkers (CYP1a, ERα and β, AR, MT, VTG) were quantified to provide additional insights into xenobiotic-related hepatic responses to dietary PS MPs exposures. The findings of this study revealed an indication of NRF2-mediated oxidative stress regulation.

Fish from PS-sewage and PS-harbor treatments had altered expression levels of multiple antioxidant enzymes in liver. Dietary exposure to PS MPs resulted in low-level activation of hepatic oxidative stress, which may not necessarily exert harmful effects on hepatic physiology, but may rather indicate adaptive homeostatic regulation. Differential responses to different PS MPs treatments (PS-sewage and PS-harbor) potentially could be explained by different chemicals associated with particles during *in situ* exposures.

TH023

Effects of Nanopolystyrene and the Co-Contaminant Tributyltin on the Nematode Community Structure in Sandy Sediments

A. Catarino, A. Homer, Heriot Watt University / ILES; L. Duran Suja, Heriot Watt University / EPS; M. AL SID CHEIKH, University of Plymouth / Marine sciences and engineering; A. dos Santos, Faculdade de Ciências Farmacêuticas - USP / Departamento de Análises Clínicas e Toxicológicas; M. Al-Sid-Cheikh, ISMERUQAR; A.J. Sweetman, Lancaster University / Lancaster Environment Centre; T.B. Henry, Heriot-Watt University / The School of Energy, Geoscience, Infrastructure and Society

Nanoplastics (NPs, ≤ 1, µm) may result from larger plastic debris released in the environment and can pose a risk to marine organisms and ecosystems. The risk of NPs can be exacerbated because toxicants sorbed to NPs may be transported to and become more bioavailable in organisms. It is likely that NPs are the most abundant plastic particles present in marine environments, and as in the case of microplastics, they are expected to accumulate in benthic ecosystems. However, there is no information on the impact of NPs on benthic meiofauna assemblages. It is critical to understand impacts of NPs on sediments of NPs because meiofauna communities play key roles on ecosystem functions such as food production and nutrient cycling. Nematodes are well established as pollution indicators and structural shifts in their communities reflect environmental changes. The goal of our work was to assess the effects of nanopolystyrene (nPS) and nPS with the sorbed co-contaminant Tributyltin (TBT) on free living nematodes on sandy sediment within a mesocosm experiment. Sediment was collected (up to 5 cm depth) at the Eden Estuary, St Andrews, Scotland, UK. The mesocosms (12ºC) consisted of glass beakers (1 L) and the exposure took place for up to 2 months. Core samples of sediments were taken each week from the following treatments: 1) Control sediment, 2) Sediment with spiked TBT (0-100 mg/kg), 3) Sediment with nPS (0-12 mg/kg) and 4) Sediment with nPS spiked with TBT (0-10 mg/kg nPS). Oxygen penetration depth (OPD) was determined by measurement of the oxygen saturation in the sediments using a microprofiler equipped with oxygen microsensors. Changes in the nematode community structure were measured by assessment of changes nematode diversity (nematodes identified to genus) and dose responses analysed according to nPS and/or TBT concentrations in the sediments. We anticipate that our results (ongoing data analysis) will contribute to a better understanding of the environmental risk of NPs and their co-contaminants within a relevant scenario.

TH024

Nanopolystyrene Induces a Decrease in the Oxygen Uptake of Zebrafish Larvae and Enables Sorbed Benzo(a)Pyrene Bioavailability

A. Catarino, Heriot-Watt University / ILES; M. Clement, Polytech Nice Sophia; M. Tait, Heriot Watt University; D. Boyle, Plymouth University; M. AL SID CHEIKH, University of Plymouth / Marine sciences and engineering; T.B. Henry, Heriot-Watt University / The School of Energy, Geoscience, Infrastructure and Society

Microplastics (MPs, 5 mm–1 µm) and nanoplastics (NPs, ≤ 1 µm) can result from larger plastic debris released in the environment and can pose a risk to aquatic organisms. Potential effects of MPs include disruption of gut physiology after ingestion, release of substances (co-contaminants) sorbed to MPs into organisms, and occlusion of tissue surfaces by accumulation of MPs. Although not yet effectively measured in aquatic environments, NPs may be the most abundant plastic particles present, but little is known about their effects in organisms. Because the relative surface area is greater for NP than MPs, there is greater potential for co-contaminant sorption to NPs and subsequent co-contaminant release into organisms upon ingestion. We evaluated the bioavailability of the co-contaminant Benzo(a)Pyrene [B(a)P] sorbed to nanopolystyrene (nPS, 500nm) by assessing the expression of cytochrome P450 1A (cyp1A) in zebrafish (*Dania reno*) larvae (96h postfertilization, hpf). The effects of nPS and nPS with sorbed B(a)P on larval (96 hpf) metabolic rate were assessed over a 24h exposure. The concentrations tested for nPS and nPS with sorbed B(a)P were 0-50 µg/ml of particles, and 0-40 µg/L for B(a)P. Proof of particle ingestion by larvae was observed using fluorescent nPS (500 nm, µg/ml). Whole-organism metabolic rate (MR) was assessed by measuring oxygen uptake (MO_2), using respirometry chambers (24 ml) and Pre-Sens optodes. The expression of hypoxia related molecular biomarkers [cytochrome c oxidase subunit IV isoform 1 (cox4i1), hypoxia inducible factor 1, alpha b (HIF-αb)] was assessed in the same larvae. Gene expression was measured by quantitative reverse transcription PCR (RT-qPCR) after RNA extraction from whole larvae. Sorption of B(a)P to nPS was confirmed using analytical chemistry techniques [gas chromatography–mass spectrometry (GC-MS)]. Preliminary dose-response analysis showed that nPS, B(a)P and nPS with sorbed B(a)P induced a decrease on MO_2 by zebrafish larvae, indicating a higher energetic cost of physiological functions maintenance. The expression of cyp1A was up to 9 fold change in the highest concentration of nPS (45 µg/ml) with sorbed B(a)P, whereas this gene did not expression when larvae were exposed just to nPS, indicating desorption of B(a)P. We anticipate that our results (ongoing data analysis) will contribute to a better understanding of the effects and risk of NPs and their co-contaminants within a more environmental relevant scenario.

TH025

Impacts of exposure to microplastics alone and with adsorbed benzo(a)pyrene on biomarkers and scope for growth in marine mussels M. galloprovincialis*

J. Hatfield, N. González-Soto, University of the Basque country UPVEHU; A. Katsumiti, University of the Basque Country / CBET Research Group, Dept. Zoology and Animal Cell Biology Faculty of Science and Technology and Research Centre for Experimental Marine Biology and Biotechnology PIE; N. Duroudier, Univeristy of the Basque Country (UPV/EHU); J. Lacave, University of the Basque country UPVEHU; A. Orbea, University of the Basque country UPVEHU / CBET Research Group, Dept. Zoology and Animal Cell Biology Faculty of Science and Technology and Research Centre for Experimental Marine Biology and Biotechnology PIE; E. Navarro, University of the Basque country UPVEHU; M.P. Cajaraville, University of the Basque Country / CBET Research Group, Dept. Zoology and Animal Cell Biology Faculty of Science and Technology and Research Centre for Experimental Marine Biology and Biotechnology PIE

Due to their hydrophobicity and relatively large surface area, microplastics (MPs) can act as carriers of hydrophobic pollutants in the ocean and may facilitate their transfer to organisms (so-called “Trojan-horse effect”). This study examined the effects of exposure to polystyrene MPs of 0.5 and 4.5 µm alone and with sorbed benzo[a]pyrene (BaP) on mussels *Mytilus galloprovincialis* in order to elucidate the effects of MP size and the presence of adsorbed contaminants on the organism. MPs were provided daily, mixed with algae, at 1000 particles/ml/day, during 7 and 26 days. Effects were determined on early cellular biomarkers (catalase activity [CAT], neutral red uptake [NR] and DNA damage in hemocytes; histopathology in digestive gland) and on whole organism responses (scope for growth [SFG] and condition index). Chemical analysis showed that BaP concentrations in mussels increased with time (up to 150 times greater than background levels) and that smaller MPs pose an increased hazard in terms of the transfer of adsorbed BaP. In histology, large MPs were abundant in the lumen of stomach, mixed with stomach contents, and in the lumen of digestive tubules (DTs), associated to cell debris. Occasionally they appeared within epithelial cells of the stomach, ducts and DTs and in the connective tissue. Small MPs were also abundant in the lumen of stomach. In all samples, DT appearance indicated a high digestive activity, confirmed by hexosaminidase histochemistry. Overall, effects in all treatments increased with exposure time. Increased effects of MPs+BaP compared to MPs alone were seen in NR and CAT but not in DNA damage despite the genotoxicity of BaP. An apparent increased effect of smaller MPs on DNA damage was also found. A general hormetic effect was demonstrated on SFG across MP treatments. This may be due to a compensatory effect whereby mussels increased their absorption efficiency in order to increase energy intake to make up for energy used dealing with stress observed in biomarker responses. This evidenced a link between MP effects at different levels of biological organisation. Further work is required to understand the effects of a variety of plastic type, size, shape combinations together with a wide variety of pollutants.*Funded by Spanish MINECO (NACE project CTM2016-81130-R), Basque Government (consolidated group IT810-13) and UPV/EHU (UFI 11/37, VRI grant PLASTOX). Work carried out within EU project PLASTOX (JPI Oceans 005/2015).

TH026

Characterization of the adsorption/desorption of benzo(a)pyrene to/from polystyrene micro- and nanoplastics for further toxicity assessment

I. Martinez, University of Basque Country; K. Le Menach, UMR CNRS EPOC Universite Bordeaux / EPOC UMR 5805; M. Dévier, University of Bordeaux / EPOC / LPTC UMR 5805 CNRS; M.P. Cajaraville, University of the Basque Country UPV/EHU / Dept. Zoology and Animal Cell Biology Faculty of Science and Technology and Research Centre for Experimental Marine Biology and Biotechnology PIE; A. Orbea, University of the Basque Country / Dept. Zoology and Animal Cell Biology and Research Centre for Experimental Marine Biology and Biotechnology PIE; H. Budzinski, University of Bordeaux

Degradation processes that large plastic items undergo in the sea have led to the appearance of small plastic pieces known as micro- (MPs) or nanoplastics (NPs), depending on their size. MPs and NPs can also be specifically manufactured for industrial and domestic applications, which results in an additional source of pollution. MPs and NPs become available to biota and enter into the food web. Moreover, due to their physico-chemical properties, such as large surface to volume ratio and hydrophobicity, plastics can adsorb/absorb other pollutants present in the water column acting as Trojan Horses. In order to further investigate the ecotoxicological aspects of this phenomenon, the characterization of the adsorption of benzo(a)pyrene (BaP), as a model polyaromatic hydrocarbon, to polystyrene MPs and NPs (4.5, 0.5 and 0.05 µm), was undertaken. 50 mg.l^{-1} of plastics of the three sizes were incubated for 24 h in an orbital shaker at 300 rpm (21°C) in three BaP solutions (100, 10 and 1 µg.l^{-1} containing 0.01% DMSO) in MiliQ water. After the adsorption period, centrifugation was performed in order to settle the plastic and allow the removal of non adsorbed BaP (NA-B(a)P). NA-BaP was quantified in the

supernatant by SPME/GC/MS. To measure BaP ad/absorbed to the plastics (A-BaP), plastic solutions were freeze-dried and subjected to microwave-assisted extraction before GC/MS analysis. The ad/absorption capacity of the plastics was calculated in mass of ad/absorbed BaP per gram of plastic ($\mu g.g^{-1}$)for the different sizes of plastic in order to determine the capacity of ad/absorption of polystyrene microbeads and whether this process was directly dependent on plastic size. Results indicated that 0.5 µm MPs showed a higher capacity of ad/absorption of B(a)P than 4.5 µm MPs. The percentages of ad/absorbed B(a)P from the total B(a)P solution were 90.88% and 37.18% with a Q_{max} of 217.39 $\mu g.g^{-1}$ and 18.83 $\mu g.g^{-1}$ (Langmuir model; R^2: 0.9862 ;0.9477) for 0.5 µm and 4.5 µm MPs, respectively. In both cases the applied methodology was successful to characterise the ad/absorption process of B(a)P to MPs and is currently being applied to NPs. * Funded by French ANR (No.–10–IDEX-03-02 and Cluster of Excellence COTE (ANR-10-LABX 45), Spanish MINECO (NACE project — CTM2016-81130-R), Basque Government (consolidated research group IT810-13) and UPV/EHU (UFI 11/37 and grant to IMA).

TH027

Occurrence of microplastics in epibenthic and sediment-dwelling species in a Norwegian fjord

a. bour, ECOLAB UMR5245 CNRS UPS INPT; C.G. Avio, L. Pittura, S. Gorbi, Department of Life and Environmental Sciences, Polytechnic University of Marche, Ancona, Italy; F. Regoli, Università Politecnica delle Marche; K. Hylland, Department of Biosciences, University of Oslo, Oslo, Norway / Department of Biosciences

The exponential production and use of plastics has generated millions of tons of plastic waste over the past decades, and the presence of microplastics has been reported throughout the world's oceans. The ingestion of microplastics *in situ* has been shown in various species, but important knowledge gaps remain, as most studies focus on pelagic fish species or bivalves used for human consumption. Here, we report the presence of microplastics in ten sediment-dwelling and epibenthic species representative of different feeding modes and trophic levels. The species analyzed include fish, bivalves, echinoderms, crustaceans and polychaetes. Organisms were sampled in the inner Oslofjord (Oslo, Norway), which is a fjord subject to strong anthropogenic pressures. High occurrence of plastic contamination was observed, with microplastic particles found in all species and in half of the individuals on average, and present in 75% of the individuals for some species. The extracted microplastics had various shapes (fibers, fragments, flakes), colors and sizes. Micro-FT-IR analysis revealed the presence of various plastic polymers: polyethylene, polypropylene and polyamide were the most commonly found, with 37%, 25% and 15% respectively. We hypothesize that maritime and fishing activities are the main source of release for these compounds. Indeed, ropes and fishing lines are usually made of these polymers. Six other types of microplastics were also found, less frequently: PET, PBT, EVA, polyester, polyacrylic, and copolymers. These results underline the potential risk posed by microplastics in sediments and the importance of assessing microplastic occurrence and impacts in benthic environments.

TH028

Development of an optimal analytical protocol for the extraction of persistent organic pollutants adsorbed on plastic debris in the environment

K. Sakaguchi-Soeder, Technische Universitaet Darmstadt / Institute IWAR Chair of Material Flow Management and Resource Economy Germany; T. Neuveglise, Technische Universitaet Darmstadt / Department of Civil and Environmental Engineering; A. Goharnia, Technische Universitaet Darmstadt / Institue IWAR Chair of Material Flow Management and Resource Economy; M. Gottschling, Technische Universitaet Darmstadt / Institute IWAR Chair of Material Flow Management and Resource Economy Germany; A. van Oyen, Plastic Partner GmbH

Accumulation of persistent organic pollutants (POPs)-loaded microplastics (MPs) in the aquatic milieu is an emerging issue of international concern. Qualitative/quantitative determination of POPs on MPs is essential for the estimation of the impact of POPs-loaded MPs on a range of marine organisms. In general, hydrophobic pollutants like POPs are first extracted from the matrix, in this case plastic debris, using non-polar solvents. Then, the extract will be cleaned-up and analysed in e.g. gas-chromatography/mass-spectrometry (GC/MS). Some non-polar solvents applied for POP extraction, however, may dissolve plastic debris partially or completely, which disturb subsequence analyses. A number of methods have been reported for the extraction of POPs from MPs. Yet, the validity of these methods have not been fully discussed and the influence of polymers in extraction solvent on subsequent POP analysis has not been thoroughly investigated. The goal of the current study is the development of an optimal analytical protocol to extract POPs from different MPs. Known amounts of POPs were artificially charged on the surface of selected polymer particles, including preproduction resin pellets from different polymer type (polyethylene, PE; polystyrene, PS; polyethylene terephthalate PET, polypropylene, PP; poly vinyl chloride, PVC) in the laboratory. The POPs on plastic particles were extracted in selected solvents using soaking and sonication methods under different conditions. Solvents used in this study include n-hexane (nHex), isopropanol (iPrOH) and dichloromethane (DCM). Extraction methods and conditions were evaluated for a high extraction recovery, a high reproducibility, as well as for a minimal damage of polymer particles, i.e. carriers of POPs. The recovery rate and analytical reproducibility of POP was determined using gas chromatography-mass spectrometry (GC/MS). The loss of plastic weight was measured for the evaluation of the stability of plastic particles under given extraction conditions. Further, we investigated the influence of polymers dissolved in solvent on quantitative analysis for POPs. TU Darmstadt and CARAT are participants of an EU project "PLASTOX", a consortium of a JPI Oceans' Joint Action. TU Darmstadt is funded by BMBF.

TH029

Comparison of spiking and dialysis tubing methods for the determination of sorption capacity and plastic-water partition coefficient of three different polycyclic hydrocarbons on microplastics

M. Gottschling, Y. Zhou, I. Schwabe, Technische Universitaet Darmstadt / Institute IWAR Chair of Material Flow Management and Resource Economy Germany; A. van Oyen, Plastic Partner GmbH; L. Schebek, Technische Universitaet Darmstadt / Institute IWAR Material Flow Management and Resource Economy Germany; K. Sakaguchi-Soeder, Technische Universitaet Darmstadt / Institute IWAR Chair of Material Flow Management and Resource Economy Germany

Determination of sorption capacity of persistent organic pollutants (POPs) on microplastics (MPs) is essential to study ecological effect of POPs-loaded MPs in the aqueous environment. However, due to high octanol-water partition coefficients (K_{ow}) as well as low water solubility, determination of sorption capacity of POPs on MPs in the laboratory is challenging. Here we present two methods to determine plastic-water partition coefficient of three polycyclic aromatic hydrocarbons (PAHs) on low-density polyethylene (LDPE): conventional spiking method and cellulose dialysis tubing method in batch test in the laboratory. PAHs selected for this test were naphthalene (log K_{ow} = 3.3), phenanthrene (log K_{ow} = 4.46) and fluoranthene (log K_{ow} = 5.16). The plastic samples tested here are LDPE pellets with a low amount of additives. LDPE pellets were previously characterised by CARAT GmbH (Bocholt, Germany). For the spiking method, batch reactors containing given amount of LDPE and MilliQ water were prepared. A high concentration of single PAHs in ethanol solution was injected into each batch reactor until the PAH concentration became stable. PAH concentration in the batch was controlled using high performance liquid chromatography (HPLC). For the dialysis tubing method, on the other hand, a closed dialysis tubing (permeability of 12,000-14,000 Dalton) containing a given amount of LDPE and MilliQ water was placed in each batch reactor filled with MilliQ water with single PAHs far above solubility. The water concentration outside of the tubing was expected to stay constant (= water solubility) during the entire experiment. The PAH concentration in the dialysis tubing was controlled using HPLC until the PAH concentration became stable. All batch reactors were placed on a horizontal shaker. When the adsorption of PAHs is completed, PAHs on LDPE are to be extracted and quantified using gas chromatography-mass spectrometry (GC/MS). Sorption capacity of each PAH was derived from the experiments and methods were compared. PE-water partition coefficient of these PAHs were derived based on the sorption capacity using adsorption models. TU Darmstadt and CARAT are participants of an EU project „PLASTOX", a consortium of a JPI Ocean's Joint Action.

TH030

Microplastics in food and beverages - a distorted perspective on risk

S. Rist, DTU (Technical University of Denmark) / Department of Environmental Engineering; B.C. Almroth, University of Gothenburg / Department of Biological and Environmental Sciences; N.B. Hartmann, Technical University of Denmark (DTU) / DTU Environment; T.M. Karlsson, University of Gothenburg

Microplastics are ubiquitous in aquatic environments and they are ingested by a wide range of animals, including species for human consumption, i.e. bivalves and fish. Additionally, plastic particles have been reported in other food products and beverages, like honey, salt, beer and drinking water. This has triggered a discussion on the human health implications of this contamination – an aspect that has gained increasing attention in the scientific and public debate in recent years. The focus and extent of this debate, however, stands in contrast with scientific findings, which merely show the presence of microplastics in certain products but no actual effects on humans. It is without question that plastics can constitute a human health risk due to toxicity of associated chemicals and to particle toxicity. However, the degree to which microplastic exposure via food products and beverages contributes to this health risk is likely insignificant at present time. When considering the magnitude of plastic usage and consequential exposure to plastic materials in our everyday lives, the relatively few microplastic particles that have been reported in food products and beverages will likely only constitute a minor exposure pathway for both microplastics and associated chemicals. But as this is rarely put into perspective, the current debate creates a distorted picture of plastic exposure and risks to humans, resulting in a misdirected outrage when people find out about plastic particles in fish, while they at the same time not reflect on the plastic container in which the fish is packaged for transport to stores and homes. In this way, the focus is taken away from the root of the problem, namely our use, consumption and disposal of plastic materials. We therefore want to encourage a more balanced and careful discussion on human health implications of plastics that takes these aspects into account.

TH031

Is the Arctic threatened by plastics? Identifying sources and determining the distribution of microplastics around Svalbard

L. Winberg von Friesen, University of Gothenburg, Sweden / Marine Sciences; M. Hassellöv, University of Gothenburg / Department of Marine Sciences; G.W. Gabrielsen, H. Hop, Norwegian Polar Institute; T. Brown, Scottish Association for Marine Science; M.E. Granberg, IVL-Swedish Environmental Research Institute / Research

Where is all the plastic, and what concentration of microplastics are ecosystems being exposed to? These are puzzling questions to the scientific community when comparing estimated values on annual plastic pollution with the actual measurements in the world's ocean habitats. Recent investigations find plastic far away from any potential source, such as in deep ocean sediments and buried within polar sea ice. The Arctic Ocean has, despite its remoteness, been suggested as a sink for plastic. Fed by the thermohaline circulation driven deep-water formation, the Arctic Ocean's bottom might be a dead end for plastics. However, very few studies have quantified the actual environmental concentrations of plastics in this remote area, thus the exposure to organisms living in this environment and potential effects are unknown. In this study, we investigated the occurrence, potential sources and distribution of microplastics in the Norwegian Arctic. We quantified and characterized anthropogenic particles >10µm in different environmental compartments (sediment, water, benthic invertebrates and sea ice) around the western and northern coasts of Svalbard. Samples were collected close to a sewage outlet and far from human activities close to the sea ice front in the Arctic Ocean. By sampling at several depths throughout the water column, microplastics associated with different water masses (Atlantic, Arctic and sewage water as well as sea ice) could be quantified. Simultaneous measurements of organic matter tracers for sea ice microalgae (IP25), pelagic microalgae (C25:3) and sewage (coprostanol) enabled correlations to be made on potential sources, pathways and fate of microplastics in the Arctic. Additional analyses of the presence of plastic specific contaminants in sediment and biota facilitated a discussion on potential exposure independent of particle accumulation in the gut. One of the primary objectives of the investigation was to determine the relative importance of local and remote sources for plastic contamination in the Arctic, and preliminary results indicate a clear signal from local sources and sea ice. In order to evaluate the risk posed by microplastics in the Arctic, a system already burdened by multiple stressors, knowledge about sources, fate and concentration of microplastics in different environmental compartments is crucial.

TH032

Microplastics - an ecotoxicological issue? How to balance facts and perception without marginalizing an environmental problem

C. Völker, ISOE - Institute for Social-Ecological Research / Water infrastructure and risk analyses; J. Kramm, ISOE - Institute for Social-Ecological Research

While plastic has been known as a factor for environmental pollution – symbolized by the plastic bag – for a long time, recent scientific evidence on the massive accumulation in the oceans and the environmental risks associated with microplastics and chemical additives have led to an upswing of the debate. In the 2000s, small plastic particles that had already been detected in seawater in the 1970s were labeled "microplastics" for the first time. Since then, the number of studies on the occurrence and effects of microplastics has risen exponentially. 13 years after the publication of the article by Thompson et al. the question of whether microplastics actually pose a risk to the environment could not yet be answered conclusively. In laboratory studies, biological effects have so far only been detected at high particle concentrations with limited environmental relevance. Independently of this, the impacts of microplastics are perceived by the general public as a serious threat to the environment and human health. The societal perception and the great mobilization potential proved to be important drivers for risk management: In 2015, the Microbead-Free Waters Act was passed banning microbeads from rinse-off cosmetics in the USA. But how did this happen without an environmental risk being scientifically proven? In the public, the presence of plastic waste is usually equated with negative biological effects, without taking into account effective thresholds and environmentally relevant exposure concentrations. From a scientific perspective, this representation leads to a dilemma: It is crucial to stay scientifically correct, to adequately categorize the effects of microplastics and even communicate null effect studies. But does this presentation affect public perception? Can we maintain the public interest in this environmental issue without propagating effects that are not there? After all, we agree on one thing: plastics do not belong into the environment. For the presentation, we draw on results of our interdisciplinary research group on plastics in the environment ("PlastX"). Our team comprises researchers from ecotoxicology, chemistry, geography and sociology analyzing plastics from different environmental as well as societal perspectives.

BiER is good for you: How biotransformation and elimination rate information can improve chemical assessments (P)

TH033

Assessing biotransformation and bioconcentration factors (BCF) of fragrance materials using in vitro approaches utilizing rainbow trout liver S9 sub-cellular fractions and cryopreserved hepatocytes

A. Lapczynski, RIFM / Environmental Science; K.M. Johanning, KJ Scientific LLC / dba of Pura Vida Connections LLC; A. Jenkins, EAG Laboratories

Bioaccumulation potential measured as the bioconcentration factor (BCF) is one of the components for the PBT (persistent, bioaccumulative, toxic) criteria and risk assessment of chemicals by regulatory agencies in some regions of the world (e.g. REACH and ECHA). Biotransformation of chemicals is the largest source of uncertainty in bioaccumulation assessment. Currently, *in vitro* methodologies utilizing the rainbow trout metabolic assay not only are gaining interest, but are being used increasingly by several sectors as a crucial component in model-based estimates of BCFs and as part of a line of weight of evidence presented to regulators. The rainbow trout metabolic assay utilizing liver S9 fractions and cryopreserved hepatocytes to test chemical biotransformation has gone through a Ring Trial for an OECD validation process (OECD Project 3.13 coordinated by ILSI HESI) and the Test Guidelines and Guidance are being reviewed by the OECD assigned panel. In the present study four fragrance materials (Cyclabute, Melafleur, Trimofix and Verdox) with known measured BCF values obtained using OECD 305 method were tested for biotransformation utilizing both rainbow trout liver S9 fractions and cryopreserved hepatocytes. The results indicate that all four fragrance materials were metabolized in both biological systems at different rates, but in all cases the BCFs determined were comparable to the measured *in vivo* BCF values. The *in vitro* metabolic assay is a powerful tool that can be used to determine BCF of test chemicals and provide data to build the database information on fragrance materials for fish metabolism and modeling.

TH034

Addressing species diversity in biotransformation: variability in expressed transcripts of hepatic biotransformation enzymes among fishes.

K.A. Fay, CSRA, Inc.; J.A. Doering, US EPA / Mid Continent Ecology Division; S. Poole, US EPA Mid-Continent Ecology Div / National Health and Environmental Effects Research Laboratory; C. LaLone, U.S. EPA / Mid Continent Ecology Division; D. Feifarek, SCJohnson; D.L. Villeneuve, U.S. EPA / National Health and Environmental Effects Research Laboratory; J.E. Cavallin, U.S. EPA / US EPA MidContinent Ecology Division; G.T. Ankley, U.S. EPA / National Health and Environmental Effects Research Laboratory

There is increasing evidence that diverse xenobiotic metabolizing enzymes exist among fishes, potentially resulting in different chemical sensitivities and accumulation, but this has never been systematically evaluated. One concern is that model test species such as rainbow trout, zebrafish and fathead minnows may not adequately represent the xenobiotic metabolizing capacity of other fish species. Our current study mined available fish liver transcriptome data and performed full-transcript, isoform sequencing on liver samples from two dozen phylogenetically diverse fish species. This novel RNAseq approach eliminated the need for transcriptome reconstruction resulting in reference genomes of the highest precision, allowing for detection of enzyme isoform orthologs among the species, as well as the nuclear receptors that control expression of the enzymes. Species were selected for broad phylogenetic coverage, as well as economic, research, and conservation importance, and included: sea lamprey (*Petromyzon marinus*), lake sturgeon (*Acipenser fluvenscens*), American eel *(Anguilla rostrate)*, alligator gar *(Atractosteus spatula)*, paddlefish *(Polyodon spathula)*, rainbow trout (*Oncorhynchus mykiss*), rainbow smelt *(Osmerus mordax)*, fathead minnow *(Pimephales promelas)*, Antarctic icefish (*Trematomus loennbergii*), common carp (*Cyprinus carpio*), and channel catfish (*Ictalurus punctatus*). In addition to comparing information across fish species, the resolved isoforms were compared to human xenobiotic metabolizing enzymes. This comparison aids in evaluating the utility of human-based biotransformation tools such as ToxCast chemical screening assays or metabolism prediction software for potential relevance in fish. *The content of this presentation neither constitute nor necessarily reflect US EPA policy.*

TH035

Metabolism of Organophosphate Flame Retardants (OPFRs) in Freshwater Fish: Field and Laboratory Studies

Y. XU, R. HOU, Research Center for EcoEnvironmental Sciences, Chinese Academy of Sciences / Key Laboratory of Drinking Water Science and Technology; Z. WANG, Research Center for Eco-Environmental Sciences, Chinese Academy of Sciences / State Key Laboratory of Environmental Aquatic Chemistry

Organophosphate flame retardants (PFRs), as widely used alternatives of brominated flame retardants (BFRs), are commonly detected in environmental matrices. Considering the adverse effects of PFRs, many researchers have paid their attention on the absorption, bioaccumulation, metabolism and internal exposure processes of PFRs in wildlife and human. PFRs can be rapidly metabolized in the body. The general metabolic pathway of PFRs revealed by certain in vitro studies includes dealkylation, hydroxylation, carboxylation, oxidative dehalogenation and phase II conjugation, resulting in a wide array of metabolites. Di-alkyl phosphates (DAPs) from the dealkylation metabolism were recently deemed important biomarkers in human biomonitoring studies. As very limited information is available on DAP metabolites in environmental biotic samples, we first investigate the accumulation and tissue distribution of eight common OP.FRs and their four DAP metabolites in three freshwater fish species from locations around Beijing, China. Accumulation of DAPs were relatively lower but comparable to those of PFRs in freshwater fish. DAPs had low limited affinity to lipid content in tissues, similarly like their parent compounds PFRs. Liver was identified to have a higher accumulation of PFRs and DAPs than the other tissues of fish. It suggested the exposure indicator role of DAPs in wild animal studies. In the consequent laboratory control study, we screened the metabolites of alkyl-PFRs by in vivo exposure of *Gobiocypris rarus*. Metabolites of alkyl-OPFRs in fish liver after 30-day exposure were analyzed with UPLC-(QTOF)MS in MSE mode. The qualitative results verified the metabolic pathway of dealkylation, hydroxylation, dihydroxylation, desaturation, and phase II glucuronide conjugation for all the tested three alkyl-PFRs. We identified and accurately quantified the metabolites 3-OH-TBOEP, BBOEHEP, and 3-OH-TNBP formed in fish liver microsomes. Liver rather than intestine, plays the primary role in PFR clearance in fish. The significance of these metabolites is in good agreement with human urine monitoring and in vivo rat exposure studies. Overall, the results emphasized the importance of hydroxylated metabolites as biomarkers for alkyl-PFRs exposure.

TH036

Bioaccumulation and biotransformation of prochloraz in the aquatic invertebrate Hyalella azteca

D. Fedrizzi, Eawag - Swiss federal Institute of Aquatic Science and Technology / Environmental Chemistry; Q. Fu, Eawag, Swiss Federal Institute of Aquatic Science and Technology / Department of Environmental Chemistry; A. Rösch, Eawag / Environmental Chemistry; N. Cedergreen, University of Copenhagen / Department of Plant and Environmental Sciences; J. Hollender, Eawag / Environmental Chemistry

Prochloraz is a widely applied fungicide for pest management purposes. Due to spray drift and surface runoff, prochloraz enters the aquatic environment where it can pose a risk for non-target organisms. Among others, *Hyalella azteca* has been recognized as a model organism to test toxicity of organic chemicals due to its rapid life cycle, the feasibility of cultivation and its sensitivity to xenobiotics. Biotransformation is a primary detoxification process through which organisms defend themselves from xenobiotics. Biotransformation can reduce the internal concentration of parent compounds hence, influencing their bioaccumulation. The aim of this study was to assess the toxicokinetics of prochloraz and its biotransformation products (BTPs) in *Hyalella azteca.* Adults of *Hyalella azteca* were exposed to prochloraz at the concentration of 100 μgL^{-1} during a 24-hour uptake phase and a subsequent 120-hour depuration phase. Organisms were sampled over time and after extraction, the internal concentration of prochloraz and its BTPs were quantified using reverse phase liquid chromatography coupled to high resolution mass spectrometry with electrospray ionization. Prochloraz and its 30 BTPs were detected, quantified, and respective toxicokinetic profiles were obtained. In every profile, an increase in the internal concentration was seen during the uptake phase followed by a decrease during the depuration phase. The bioaccumulation factor was calculated to be 110 Lkg^{-1}. Finally, the data will be modeled using a toxicokinetic model and thus uptake, elimination and transformation rate constants will enable determining the role of biotransformation in the detoxification of prochloraz in *Hyalella azteca*.

TH037

Toxicokinetics and metabolite identification of two emerging pollutants, Acesulfame-K and 4-MBC, in the manila clam Ruditapes philippinarum.

N.C. Ruiz, INMAR - University of Cadiz / Physical Chemistry; F. Tonini, Alma Mater Studiorum University of Bologna; P. Lara-Martin, University of Cadiz / Physical Chemistry; J. Blasco, Inst. Ciencias Marinas de Andalucia / ECOLOGY AND COASTAL MANAGEMENT; M. Hampel, INMAR University of Cadiz

Marine ecosystems have been historically sinks for many pollutants and chemicals whose effects awoke social concern, triggering the implementation of legislations. Nowadays, new compounds are developed at increasing rates and eventually discharged into marine ecosystems in unknown quantities and with no regulation. Due to the improvement of new analytical techniques, many of these chemicals, the so called “emerging pollutants” (EPs), are being currently identified and their occurrence is being proved in the environment. However, very little is known about the possible adverse effects of these emerging pollutants in exposed non-target organisms. In this context, the present work evaluates the **toxicokinetics** (TK) of two EPs (the UV filter **4-Methylbenzylidene-camphor** (4-MBC) and the artificial sweetener **acesulfame K** (ACE-K)) in the Manila clam *Ruditapes philippinarum,* focusing on determining the **bioconcentration factors** (BCF) and identifying **metabolites** and **transformation products** (TPs) of the studied pollutants. After 7 days of exposure and 3 days of depuration, target compounds were extracted from both water phase and organisms and their concentrations were measured by liquid and gas chromatography coupled to tandem mass spectrometry (UPLC/GC-MS/MS). Additionally high resolution mass spectrometry (HRMS) and automated data analysis software (Metabolynx™) were used to identify possible TPs in the tissue of the Manila clam at different nominal concentrations (from 1 to 100 $\mu g\ L^{-1}$). For the UV filter, the estimated BCFs were between 61 553 and 539 143 $L\ Kg^{-1}$, and several metabolites were identified, such as the reduction or hydroxylation of the compound. On the other hand, the artificial sweetener BCF was consistently lower, around 7 $L\ Kg^{-1}$ and no metabolites were identified. These results suggest that 4-MBC was highly bioaccumulated and metabolised to facilitate its excretion and they are directly related to the physicochemical properties of the target EPs, since ACE-K is highly soluble in water (log k_{ow} -1.33) and excreted unchanged in comparison to the very hydrophobic UV filter (log k_{ow} 5.92).Additionally, the present study provides important information about the toxicokinetics of 4-MBC and ACE-K, which will be helpful for fully understanding the mechanism of action of these compounds. Furthermore, this work demonstrates the potential of the UPLC-GC/HRMS approach using Metabolynx™ software for fast and accurate identification of metabolites of EPs.

TH038

Organophosphate Esters, Including Alkyl-Substituted Triphenyl Phosphates, in East Greenland Polar Bears and Ringed Seals: Adipose Tissue Concentrations and In Vitro Depletion and Metabolite Formation

A. Strobel, Carleton University; W.G. Willmore, Carleton University / Biology department; C. Sonne, R. Dietz, Aarhus University / Department of Biosciences, Arctic Research Centre; R.J. Letcher, Environment and Climate Change Canada / Ecotoxicology and Wildlife Health Division

East Greenland is a contamination “hot spot” for long-range transported anthropogenic chemicals, including organophosphate esters (OPEs). High concentrations of OPEs have been reported in Arctic air (particles) while very little is known for wildlife although recent reports for Hudson Bay polar bears (*Ursus maritimus*) indicate that OPE residue levels in fat tissues are very low or non-detectable and appear to be strongly influenced by biotransformation. In the present study, the hepatic *in vitro* metabolism of six environmentally relevant organophosphate (OP) triesters and corresponding OP diester formation were investigated in East Greenland (Scoresby Sound region) polar bears (PBs) and ringed seals (RSs; *Pusa hispida*). The *in vitro* OP triester metabolism assay results were compared to fat (adipose) levels of selected OP triesters in field samples from the same individual animals. *In vitro* OP triester metabolism was generally rapid and structure-dependent, where PBs metabolized OPEs more rapidly than RSs. Exceptions were the lack of triethyl phosphate metabolism and slow metabolism of tris (2-ethylhexyl) phosphate in both species. OP diester metabolites were also formed with the exception of triphenyl phosphate (TPHP) which was not metabolized at all in the RS assay. Tris(1,3-dichloro-2-propyl) phosphate was completely converted to its corresponding diester. However, the mass balances showed that OP diester formation corresponding to tris (2-ethylhexyl) phosphate, tri (n-butyl) phosphate, and tris (2-butyoxyethyl) phosphate did not account for 100 % of the OP triester depletion, which indicated alternate pathways of OP triester metabolism. TPHP was completely converted to its OP diester metabolite in PBs but not in RSs suggesting species-specific differences. Alkyl-substituted TPHP analogues also showed that the number and position of the phenyl ring substitution heavily influenced the rate of metabolism. The results demonstrated that OP triester bioaccumulation and fate in PBs versus their RS prey is substantially influenced by biotransformation.

TH039

Proteomics of a metabolic simulation system - a look inside rat S9

A. Schiwy, EWOMIS; B. Thalmann, RWTH Aachen University, Institute for Environmental Research / Bio5 - ESA; P. Huesgen, Forschungszentrum Jülich GmbH / Central Institute for Engineering, Electronics and Analytics (ZEA); S. Schiwy, Institute of Environmental Research-RWTH Aachen / Department of Ecosystem Analysis; H. Hollert, RWTH Aachen University / Institute for Environmental Research

The liver is the key organ in metabolism and detoxification of xenobiotics. Simulation of this organ in various bioassays is achieved via the application of either single cytochrome P450 enzymes produced via biotechnological processes or complex enzyme mixtures obtained from animals. Especially, this second process is regarded under various animal welfare regulations as an animal experiment. The animals have to be maintained to a specific age and following their livers have to be induced via various chemicals. Furthermore, this treatment may cause pain to the animals. Finally, the animals have to be killed to harvest the organ (predominantly liver) for further downstream processing. The most common procedure is a mincing of the organ and subsequent centrifugation to separate the different components of the cells. One of these animal-derived products is called rat liver homogenate (S9) and includes the cytosolic and the microsomal fractions. It consists of enzymes for phase I and phase II biotransformation reactions. This product is very prominent in various OECD and ISO test guidelines for bioassays that are not by themselves capable of a metabolic transformation. The most prominent example is the Ames bacterial reverse mutation assay according to the ISO 11350 or the OECD 471. However, the application for S9 is much more diverse and spans from various not guideline-based bioassay towards the ad-hoc production of metabolites for chemical analysis. It is also applied for stability testing of pharmaceuticals and the observation of the potential in bioaccumulation of chemicals and their metabolites. In this study, we look at the proteomics of multiple rat S9 products and compare them with an animal component free biotechnological alternative.

TH040

A critically evaluated database of in vitro and in vivo toxicokinetic data for

mammals and fish

J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology; A. Looky, ARC Arnot Research and Consulting Inc.; K.L. Foster, ARC Arnot Research and Consulting Inc. / Adjunct Professor, Trent University, Applications of Modelling & Quantitative Methods (AMOD); J.M. Armitage, University of Toronto - Scarborough / Physical and Environmental Sciences; M. Halder, European Commission Joint Research Centre / DG Joint Research Centre IHCP EURL ECVAM; A. Lostia, European Commission Joint Research Centre; A. KIENZLER, JRC-EC / F3-Chemical Safety and Alternative Methods Unit-EURL ECVAM

Toxicokinetics (TK) plays an important role in ecological and human health assessments. Relatively few measured TK data exist compared to the number of chemicals subject to regulatory assessment requirements. It is not feasible to measure TK data in all organisms (e.g., humans, rodents, fish) and there is a recognized need to reduce animal testing. Reliable (evaluated), high-quality existing in vitro and in vivo TK data could help evaluate in vitro-in vivo extrapolation models (IVIVE), parameterize TK and bioaccumulation models, and develop and validate quantitative structure-activity relationships (QSARs) for predicting TK parameters from chemical structure. Biotransformation and elimination rate data can be used in diverse contexts for chemical assessment. For example, biotransformation rate constants (k_B) are key determinants and sources of uncertainty in bioaccumulation assessment. k_B can be determined in vivo with whole animal models or from in vitro assays using intact cells or subcellular fractions from the liver or other tissues (e.g., gastrointestinal tract, kidney). We have developed a new database (funded by the JRC CCR.F.C931336.X0) containing TK data (i.e., biotransformation rates) for fish and mammal species (i.e., rat, mice) derived from in vivo and in vitro (S9 fraction, hepatocytes, microsomes) methods. The database entries are scored based on a data quality evaluation. The data quality assessment methods and criteria have been developed from standardized testing guidance when such guidance exists and from professional judgement in the absence of standardized guidance. In total the new database includes approximately 9000 entries for organic chemicals. There are approximately 4,000 and 400 chemicals from in vitro and in vivo studies respectively from rodent species. There are approximately 120 and 700 chemicals from in vitro and in vivo studies respectively from fish species. The database can be used as a source of information for chemical assessments and can help identify future research needs (i.e., chemicals that require chemical evaluation and for which reliable quality data are not available). We believe the database will also be a valuable source information for model developers (e.g. for in vitro-in vivo extrapolation models, kinetic models, models to predict exposure and internal concentration in an organism) and chemical evaluators. The database will be publicly available at the Joint Research Centre website.

TH041

A tiered testing strategy for rapid estimation of bioaccumulation by a combined modelling - in vitro testing approach: derivation of kinetic rate constants in different in vitro models

K. Schirmer, Eawag / Environmental Toxicology; J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology; N. Bramaz, Eawag Swiss Federal Institute of Aquatic Science and Technology / Environmental Toxicology; N. Bury, University of Suffolk / Division of Diabetes and Nutritional Sciences; M.R. Embry, ILSI Health and Environmental Sciences Institute (HESI); J. Fitzgerald, University of Exeter / Biosciences College of Life and Environmental Sciences; C. Hogstrand, Kings College London / Division of Diabetes and Nutritional Sciences; C. Kropf, University of Bern / Centre for Fish an Wildlife Health; H. Segner, University of Bern / Centre for Fish and Wildlife Health; R. Schoenenberger, Eawag Swiss Federal Institute of Aquatic Science and Technology / Environmental Toxicology; J. Stadnicka-Michalak, EPFL - Swiss Federal Institute of Technology

Our research seeks to improve alternative methods to estimate bioaccumulation of organic chemicals in fish. We follow a tiered strategy that integrates toxicokinetic (TK) models, quantitative structure-activity relationships (QSARs), in vitro experimental data from fish liver, gill, and intestinal tissues, and in vitro-to-in vivo extrapolation methods. In a first step, we derived at a list of candidate chemicals for in vitro testing based on model discrepancies, availability of reliable in vivo BCF and BMF data, and availability of in vitro biotransformation rates. The resulting chemicals were divided into three K_{ow} categories based on predominant exposure route(s) to guide in vitro testing: 1) log K_{ow} < 4 (aqueous exposure dominates – to be tested in gill and liver models); 2) log K_{ow} 4 - 5.5 (mixed exposure routes – to be tested in gill, liver and intestine models); and 3) log K_{ow}>5.5 (predominantly dietary exposure dominates – to be tested in liver and intestinal models). In vitro testing is now on-going. Specifically, primary gill cell cultures grown on permeable support are used to determine the combined biotransformation/permeation rate through this epithelial model. Primary suspension preparations from hepatocytes and intestine are explored for biotransformation rates (i.e. loss of parent compound). Permanent cell lines of gills, liver and intestine, exposed in monolayer, complement the set of in vitro methods applied, yielding parent compound loss rates as well. In vitro models are each applied under their respective optimal conditions, taking e.g., temperature and media composition into account. Chemical starting concentrations are set uniformly for all models based on non-toxic concentrations and analytical method sensitivity. Thus far, permeation/biotransformation was observed for all chemicals applied. The resulting rate constants are subject to comparison between the different in vitro models and are input into the TK and QSAR models for model development and hypothesis testing. This poster will describe the overall in vitro testing strategy, the different in vitro models and the results of the chemical testing with regard to in vitro-derived rate constants.

TH042

Update on development of OECD Test Guidelines and Guidance Document on determination of fish in vitro hepatic clearance

M.R. Embry, ILSI Health and Environmental Sciences Institute (HESI); K.A. Fay, CSRA, Inc.; I. Bischof, Fraunhofer Institute for Molecular Biology and Applied Ecology IME; J.W. Davis, Dow Chemical Company / Toxicology and Environmental Research and Consulting; J. Domoradzki, Dow Corning Corporation / Health and Environmental Sciences; M. Halder, European Commission Joint Research Centre / DG Joint Research Centre IHCP EURL ECVAM; J. Hu, The Dow Chemical Company / Toxicology and Environmental Research and Consulting; K.M. Johanning, KJ Scientific LLC / dba of Pura Vida Connections LLC; H. Laue, Givaudan Schweiz AG / Fragrances S & T; D. Nabb, DuPont; J.W. Nichols, U.S. EPA / ORD NHEERL Mid Continent Ecology Division; C. Schlechtriem, Fraunhofer IME / Department Bioaccumulation and Animal Metabolism; H. Segner, University of Bern / Centre for Fish and Wildlife Health; J. Weeks, Weeks Entox LLC

Chemical biotransformation represents the largest source of uncertainty in chemical bioaccumulation assessments, and model-based estimates of chemical bioconcentration in fish may be greatly improved by including biotransformation rates, as measured in vitro. Substrate depletion assays, using rainbow trout hepatocytes (RT-HEP) or liver subcellular fractions (RT-S9), have been successfully developed to provide estimates of fish biotransformation. A multi-laboratory ring trial, coordinated by the ILSI Health and Environmental Sciences Institute (HESI), was recently completed which demonstrates assay reliability within and across laboratories and similar performance of substrate depletion assays using the two biological systems. Based on the successful results of this ring-trial, two OECD test guidelines (TG) (*"Determination of in vitro intrinsic clearance using cryopreserved rainbow trout hepatocytes"* and *"Determination of in vitro intrinsic clearance using rainbow trout liver S9 sub-cellular fractions")* have been drafted and are accompanied by a Guidance Document (GD). The OECD GD provides detailed information on how to conduct the tests as well as how to apply the measured in vitro biotransformation rates to predict bioconcentration factors (BCFs). In addition, guidance on selection of the assay system (e.g., primary hepatocytes vs. liver S9 fractions), specific considerations for testing chemicals, use of negative and positive controls, BCF extrapolation models, and application of the two test methods beyond BCF prediction are also covered. Draft TGs, the GD, and the ring trial report underwent two OECD public commenting rounds during 2017 and submission to OECD WNT final approval is planned for 2018

TH043

The Bioaccumulation Assessment Tool (BAT): A quantitative weight of evidence approach for bioaccumulation assessment

L. Toose, ARC Arnot Research & Consulting; J.M. Armitage, University of Toronto - Scarborough / Physical and Environmental Sciences; K.L. Foster, ARC Arnot Research and Consulting Inc. / Adjunct Professor, Trent University, Applications of Modelling & Quantitative Methods (AMOD); M. Embry, ILSI; J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology

Chemicals are being assessed for bioaccumulation (B) potential in regulatory programs using various methods, metrics and criteria. B data can be obtained from various data streams including laboratory studies, field studies and model predictions using mass balance models and quantitative structure-activity relationships (QSARs). Examples of bioaccumulation metrics include: the bioconcentration factor (BCF), bioaccumulation factor (BAF), biomagnification factor (BMF) and trophic magnification factor (TMF). We have developed the Bioaccumulation Assessment Tool (BAT) to collect, evaluate and integrate various lines of evidence (LOE) associated with these B-metrics and related B classification criteria to aid decision-making. The BAT provides a transparent and consistent framework for evaluating neutral and ionizable organic chemicals in aquatic and terrestrial organisms. It uses a quantitative a weight of evidence (QWOE) approach which includes evaluations for the relevance, reliability (confidence) and outcome of each B-metric. Each substantive LOE (e.g., BCF, BMF, biotransformation rate) is subject to data quality evaluation resulting in a data confidence score. The Data Evaluation Templates (DETs) have been derived from standard test protocols and expert judgment when standard protocols are not yet developed. Physical-chemical properties can be used or the user is allowed to enter biologically relevant partition coefficients in place of default assumptions that assume octanol as surrogate for biological components (i.e., lipid). Estimates for biotransformation rates can be included from in vitro assays (i.e., S9, hepatocyte, microsomal) and from in silico (QSAR) predictions. Empirical data such as lab BCFs and BMFs and field data as well as in silico data (e.g., BCF-QSARs) can be included in the QWOE. This presentation provides an overview of the BAT and demonstrates its application with two case studies. The first example is a typical “data poor” scenario in which only chemical structure information is available. From chemical structure relevant physical-chemical property and biotransformation rate data are obtained from QSARs and entered into the system. The second case study is for a relatively data rich chemical for which various LOE exist (e.g., 3 lab BCFs, various BCF-QSARs, biotransformation rate QSARs, in vitro biotransformation rates). Future work for improving the BAT is discussed.

TH044

Towards the use of elimination rates in bioaccumulation assessment –Current challenges and future needs

G. Treu, German Environment Agency / REACH Chemicals; W. Drost, Federal Environment Agency (UBA) / Chemicals; C. Rauert, Umweltbundesamt / International Chemicals Management

The capacity of chemicals to bioaccumulate in biota is recognized as critical property that contributes to a chemicals risk. The bioconcentration factor (BCF) reflecting the uptake of a chemical from water and the biomagnification factor (BMF) following dietary uptake in fish remain the preferred metrics in bioaccumulation assessment. The test systems are expensive, time consuming and are not suitable for screening purposes. Still, terrestrial bioaccumulation is hardly considered. A comprehensive bioaccumulation assessment should consider both, the aquatic and terrestrial organisms. Recently, it has been suggested that BCF and BMF can be derived by only determining the elimination rate constant (k_2) experimentally while the uptake rate (k_1)is estimated. Following this concept the need for animal tests is reduced if the metabolic contribution to k_2 is from *in vitro* experimentswhile the effect of the other pathways (excretion via urine and feces, and ventilation)are estimated with *in vitro* to *vivo* extrapolation models. Biotransformation often reduces the extent to which chemicals accumulate in fish and mammals. Thus, a Tier 1.5 can be introduced between Tier 1 (screening based on physico-chemical data) and Tier 2 (exposure studies with animals) where *in vitro* biotransformation rates (k_m) obtained from *in vitro* tests with fish or mammalian cells are extrapolated to whole organisms and then incorporated into to existing chemical mass-balance models to predict a BCF or BMF. Only if this model indicates an increased bioaccumulation a potential a higher-tier vertebrate test is then needed. In practice, animal tests are mandatory for chemicals exceeding a certain level of hydrophobicity but may turn out as non bioaccumulative due to metabolisation. A k_2 based extrapolation model allowing to estimate BCF and BMF values by incorporating *in vitro* $k_m$of different tissues, e.g. gills, liver and gastro intestinal tract, could be serve as alternative screening criterion under REACH. This would allow to experimentally cover species differences currently ignored in bioaccumulation regulation. However, uncertainties remain related to the validity of this approach, e.g. for ionic substances, and should be addressed in future research by taking into account specific metabolic pathways. This poster aims at demonstrating current limitations and future needs for the k_2 based bioaccumulation assessment under REACH from a regulatory agency’s perspective.

TH045

SETAC Bioaccumulation Science Interest Group

L.P. Burkhard, U.S. EPA / ORD/NHEERL/Mid-Continent Ecology Division

Advances in evaluating and regulating endocrine disruptors (P)

TH046

Progress of the Japanese Program on Endocrine Disrupting Effects of Chemicals: EXTEND2016

K. Yamazaki, Ministry of the Environment / Environmental Health

Ministry of the Environment, Japan published its fourth program on endocrine disrupting effects of chemical substances “EXTEND2016” in June 2016. It is developed upon achievements on development of framework, development and improvement of test protocols and implementation of testing and assessment in the preceding program “EXTEND2010”. While basic concepts and framework was inherited from EXTEND2010, EXTEND2016’s focus has been shifting to implementation of testing and assessment and consideration of appropriate risk management measures. During fiscal years 2016-17 progress has been made in development of test protocols, evaluation of existing knowledge, identification of candidate chemicals for testing, implementation of testing and assessment and communication to the public, as well as in international collaborative projects with the United Kingdom and the United States. One of the most significant achievements should be finalization of the data obtained from the medaka extended one generation reproduction test (MEOGRT) for 4-nonylphenol, which are expected to be referred to in regulatory environmental risk assessment. The reproduction tests are being conducted for additional chemicals within the program. Updated progress in testing and assessment under EXTEND2016 will be presented at the Annual Meeting.

TH047

Effects of endocrine disruptors on reproductive health: A new approach to integrating ecotoxicological and human health data

L. Parent, Télé-université / UER Science et TEchnologie; P. Grigorova, Université TÉLUQ / Département Science et Technologie; M. Nikolaros, Université TÉLUQ / Science et Technologie

Exposure to synthetic and natural chemicals is almost inevitable in our daily lives. Some of them raise concerns with their endocrine disruptive potential and possible interference with endocrine system leading to the variety of adverse health effects. It was initially through clusters of presumptions that the potential effects of endocrine disruptors (EDs) on human health and the environment were highlighted. EDs, as a growing source of concern, now need to better document the complexity of the relationship between exposure and effects, hence the development of new evidence-based approaches to better document decision-making in health policy. Among these approaches, we retained the systematic reviews, based on objective methods, to integrate multiple sources of evidence (epidemiology, wild animals, laboratory animals, in vitro and in silico data) relevant to the evaluation. Our project aims to systematically review the data published the last 10 years linking the exposure to EDs (polybrominated diphenyl ethers (PBDE), alkylphenols, bisphenol A (BPA), parabens, phthalates, perfluorinated compounds) with the effects on the development and reproductive health as changes in sex ratio, congenital malformations, sperm quality disruption, alteration of plasma levels vitellogenin, sex hormone levels as well as anomalies of gonad development. 16 701 articles were screened and 744 met the inclusion criteria for the review. The data was extracted from 155 epidemiological studies, 242 in vivo studies, 377 ecotoxicological studies and the ROB (risk of bias) analysis was performed for the relevant outcomes, confidence in the body of evidence for an effect was rated, and scores are given. In this presentation, we will show what is the strength of the evidence for the association between exposures and (adverse) effect, and we will discuss of the role of ecotoxicological studies in the global analysis: prioritizing EDs, understanding mechanisms of action, establishing standards or impact criteria, identifying sensitive biomarkers and bioindicators for each of the EDs.

TH048

Pros and cons of fish toxicity tests in detecting chemicals with endocrine disrupting activities

A. KIENZLER, JRC-EC / F3-Chemical Safety and Alternative Methods Unit-EURL ECVAM; Z. DANG, RIVM / LIEC CNRS UMR; S. van der LINDEN, JRC-EC

In the last decade, with the increased awareness of potential effect of endocrine disruptors (EDs) on wildlife, fish toxicity test guidelines have been developed by adding biomarker endpoints that indicate chemicals with EAS (estrogen, androgen and steroidogenesis) activities. These tests have been summarized in the OECD guidance documents 150 and 171 and specific guidance on the diagnosis of endocrine-related histopathology in fish gonads is available (GD 123). However, while the relevance of fish toxicity tests is clear in the assessment of endocrine disruptors, comparison of these tests in response to EDs has not yet been made in these documents. In the EU legal frameworks, these fish tests are often required on the basis of the existing information case by case. Due to a difference in sensitivity of species and life stages, many EU discussions have focused on which test should be suitable. However, due to practical considerations, (e.g. regional preference, practical use, and specifically sensitivity.) it is unlikely to meet all of the requirements within one test. But in order to avoid further additional testing, species selection should always consider these factors as much as possible. This work intends to summarise the pros and cons of the available test guidelines and to address some issues e.g. sensitivity in different life stages and in species. Available fish toxicity tests include test guidelines (TG) 229, 230, 234, 240 and guidance document (GD) 148. The number of fish used in each fish test, the covered lifestage, the investigated EDs-related endpoints, their robustness (and to which extend these have been validated) and the species sensitivity in response to chemicals with EAS modes of action will be compared. To this aim, publically available data on different fish species tested according to TGs or TG-like protocols will be collected and analyzed. Analysing these fish toxicity data will help identifying which fish test, which species, which life stage of test are needed for the identification and/or risk assessment of EDs. Based on the overall data analysis, we will propose an environmental testing strategy, which is important for minimizing vertebrate testing and costs.

TH049

Towards developing a list of reference chemicals for endocrine assay validation

C. Prosser, ExxonMobil Biomedical Sciences, Inc.; M.R. Embry, ILSI Health and Environmental Sciences Institute (HESI)

Compared to other areas of human and environmental hazard assessment, evaluating the potential for endogenous compounds to interact with endocrine pathways is relatively nascent. However, recognizing the possibility of a public and environmental hazard, many national governments, international organizations, industry bodies, public interest groups and academic institutions established research programs to address the impacts of endogenous substances on the endocrine system. This has resulted in attempts to develop and validate a battery of tests to screen for endocrine active compounds with multiple publications by both regulatory agencies and academics aimed at identifying appropriate *in vitro* and *in vivo* assays. Thus, there has been considerable effort to establish criteria and interpret results for the identification of potential of endocrine active compounds. However, despite all the attention on test development, little consideration has been given to establishing a list of reference compounds to be used in the validation process. Without establishing a set of criteria it may prove problematic to assess intra-assay variability for the same endocrine mode of action (e.g. estrogenic/androgenic effects). When evaluating the current, validated, assays in OECD Guidance Document 150, there is a great disparity in the reference chemicals selected, and no discussion as to why various chemical were chosen for the validation procedure. Additionally, reference chemical selection is often not consistent within multi-laboratory ring trials during assay validation. This presents challenges to regulators and researchers in selecting assays with needed sensitivity and/or appropriateness of use. Here, we attempt to identify the parameters that should be evaluated when selecting validation chemicals. These range from simple physical/chemical properties, to more complex information related to a known mode of action. Additionally, reference chemicals used during assay validation should span a range of potencies incorporating both positive and negative controls. Some well accepted and commonly used chemicals are provided as a realistic starting point to compile a set list of reference chemicals for the validation of endocrine assays.

TH050

Assessment of endocrine disrupting properties of pesticides and biocides: data processing to support data analysis

A. Lostia, Joint Research Centre - European Commission - Institute for Health and Consumer Protection; S. Munn, European Commission; S. van der Linden, European Commission Joint Research Centre / Directorate F Health, Consumers and Reference Materials; A. KIENZLER, JRC-EC / F3-Chemical Safety and Alternative Methods Unit-EURL ECVAM; E. Grignard, European Commission Joint Research Centre; E. Joossens, European Commission DG Joint Research Centre

In 2016 the Commission completed an impact assessment to estimate possible health, environmental and socio-economic consequences of adopting different options, formulated as scientific criteria, for identifying Endocrine Disruptors (EDs) under the Plant Protection Products Regulation and Biocidal Products Regulation. JRC developed a central element of this assessment, namely, a science-based methodology to screen over 600 chemicals in about 10 months, including all EU-registered biocides and pesticides. The methodology was based solely on already existing data. To achieve the objective of screening hundreds of substances in a limited time, all retrieved data for a substance (toxicological studies, effects observed, NOAEL, etc.) are captured in an excel template (consisting of 40 columns), developed by JRC, in order to systematically organise the information to then facilitate data-analysis. As a result a large and curated database is available summarising relevant existing data collected for the 600 substances screened. A major accomplishment was the development of an innovative way to process and visually represent the data captured in the excel template as a mean to facilitate the data analysis in a systematic manner and in medium-throughput to ensure meeting the objective defined in the Commission Roadmap of screening 600 substances in a limited time and by using high-quality science-based strategy. Briefly the data collected are re-organised and processed into a data-matrix which is built automatically after the template has been filled. The data-matrix, available for each of the screened chemicals, allows getting an overview of all toxicological information available for a certain chemical thus facilitating the data analysis to identify EDs. For instance the data-matrix visually reports if certain toxicological endpoint has been observed across different studies to support evaluation of consistency and reproducibility of toxicity findings. Focusing on all the pesticides and biocides screened (about 400 substances), the data-matrix for all these substances were merged together in order to build a heat-map summarising all the toxicological information collected by endpoint. The heat-map can be used to group chemicals based on the similarity of their toxicological behaviour as a mean to prioritise chemicals for further analysis or to build read-across strategy to fill data-gaps.

TH051

Assessment of endocrine disrupting properties of pesticides and biocides: data requirements, availability and needs

S. van der Linden, European Commission Joint Research Centre / Directorate F Health, Consumers and Reference Materials; A. Lostia, European Commission Joint Research Centre; A. KIENZLER, JRC-EC / F3-Chemical Safety and Alternative Methods Unit-EURL ECVAM; E. Grignard, European Commission Joint Research Centre; S. Munn, European Commission

Before pesticides and biocides are allowed to enter the European market, a minimum set of toxicological data is required to be submitted in order to evaluate their (unintentional) toxicity and approve their use. In case of concern(s), specific conditions may apply to limit their use or approval might not be granted at all. The data that is required to be submitted is (mostly) coming from standardized test guidelines (TGs). While these TGs focus on a diverse range of toxic effects, none of the TG studies currently in the data requirements are specifically developed for the assessment of endocrine disruption (ED). However, ED specific findings can potentially be extracted from these studies and supplemented with data coming from other sources. OECD Guidance Document 150 can help with the ED specific interpretation of data and a guidance document for assessing pesticides and biocides is currently being developed by EFSA, ECHA and JRC. In the context of the recent ED impact assessment, we screened the regulatory dossiers, scientific literature and other available information to assess and categorise all pesticides and biocides currently registered in the EU. This assessment is performed for both human and environmental health. This presentation will provide an overview of the results of this categorisation, combined with indications of the origin of the data driving the categorisation: i.e. data obtained from the regulatory dossiers or other scientifically relevant information. Examples are highlighted where the data obtained from the regulatory dossier would potentially lead to different conclusions compared to when all additional data are taken into account.

TH052

Plausible or Causal: Bioactivity and mechanistic potency as a critical piece in hazard characterization of endocrine active chemicals

E.M. Mihaich, ER2; K. Plotzke, Dow Chemical Company / Toxicology, Environmental Research & Consulting

While methods have been and are being developed and validated, and regulatory programs around the world are moving forward with evaluating chemicals for their potential interaction with the endocrine system of humans and wildlife, the challenge still remains in distinguishing between effects that are specifically endocrine mediated from those that are a result of general stress or some other mode or mechanism of action (MoA). Under certain legislations, understanding the potential MoA is particularly important because regulatory decisions might categorize a substance as an endocrine disruptor (ED) through a hazard characterization process rather than taking into consideration exposure and risk. The WHO/IPCS definition of an endocrine disruptor requires that a substance alter the function of the endocrine system and consequently result in adverse effects in an intact organism. Pathway models are being established that provide plausible links between molecular initiating events, key events and ultimately adverse effects. However, when several potential pathways or MoA converge on the same adverse effect in an intact organism it becomes challenging to identify the biologically plausible causal link between the MoA and the environmental or health effect of regulatory concern. The first part of the WHO/IPCS definition, that of the chemical acting through an endocrine MoA to alter the function of the endocrine system, focuses on the need for the substance to have sufficient affinity for and activity with components of the endocrine system, compared to endogenous hormones, in order to compete with the normal hormonal signaling and feedback mechanisms that maintain homeostasis. Although, certain in silico or in vitro screens may predict endocrine-related bioactivity and the potential for a chemical to interact with the endocrine system, affinity and activity, also known as mechanistic potency, can be used in comparison to that of the endogenous ligands to support or reject the biological plausibility of a causal link between an endocrine MoA and an adverse effect. A hypothesis testing, weight of evidence framework and case study examples will be used to illustrate the use of bioactivity and mechanistic potency data, along with other lines of evidence, in the assessment of endocrine activity.

TH053

Addressing endocrine concerns for the environment in dossier evaluations with an FSDT - possibility to avoid further vertebrate tests

F. Kaßner, Umweltbundesamt / Federal Environment Agency / IV2.3 Chemicals; J. Arning, German Environment Agency UBA; E. Hassold, German Environment Agency UBA / IV 2.3 Chemicals; S. Germer, German Environment Agency UBA

In the context of the REACH regulation, long-term toxicity testing on fish is a standard information requirement for substances manufactured or imported in quantities of 100 or more tonnes per year. Additionally, some substance properties, for example a low water solubility, lead to the necessity to conduct a long-term toxicity test on fish. If a data gap in a registration for long-term toxicity to fish is identified in the process of a dossier evaluation (Dev), the preferred option is to request a Fish early life-stage Test (FELS - OECD 210). However for a substance with hints for endocrine disrupting properties, further tests would be needed to clarify the concern in a substance evaluation. In our opinion it is possible to avoid additional vertebrate tests in a SEV by requesting a Fish Sexual Development Test (FSDT – OECD 234) as a standard long term fish toxicity test under dossier evaluation. . This would make it possible to clarify the endocrine disrupting properties of the substance, if they are revealable in an FSDT, what is the case for most oestrogenic or androgenic acting chemicals. Similar to the FELS the FSDT assesses fish early life-stage effects and covers all relevant standard endpoints. The investigation of the endocrine disruptive potential of substances to environmental organisms is not explicitly part of the standard information requirements under the REACH Regulation. However, the legal text of REACH Regulation does not refer to a specific OECD test guideline, but to a type of the test. ECHA's Guidance on Information Requirements R.7b indicates that the need to conduct further tests is triggered e.g. by information on a specific mode of action and unexpected sensitivity of a group of organisms to the substance under investigation. As the FSDT is a level 4 test, according to the OECD conceptual framework no other fish (vertebrate) test may be necessary to identify the substance as an endocrine disruptor according to the WHO/IPCS definition.

TH054

Structural Alerts for Potential Endocrine Disruptors

R. Kühne, N. Ost, Helmholtz Centre for Environmental Research UFZ / Department of Ecological Chemistry; L.A. Baumann, University of Heidelberg / Aquatic Ecology and Toxicology; H. Segner, University of Bern / Centre for Fish and Wildlife Health; J. Arning, German Environment Agency UBA / Chemicals; G. Schuurmann, Helmholtz centre for environmental research - UFZ / Department of Ecological Chemistry

Endocrine disrupting chemicals interact with the hormone system. They may trigger adverse effects on organisms. Endocrine disrupters are labelled as substances of very high concern (SVHC) and are subject of regulations as REACH. However, there are so far no internationally harmonised criteria for endocrine activity. Furthermore, the endocrine system is rather diverse. Existing tests are rather expensive, and it is still not clear whether they comprise all relevant pathways. Thus, the number of existing data is limited. In silico tools may provide alternatives at least to allow prioritisation of tests by screening compound lists. This study aimed in identifying structural alerts for potential endocrine disruptors of two relevant hormone systems, estrogen/androgen and thyroid hormones. Chemicals binding to the estrogen/androgen receptors may either yield an agonistic effect by mimicking the hormone, or an antagonistic effect by blocking the receptor site and thus preventing the hormones from binding themselves. Thyroid hormones bind to the ligand binding domain (LBD) of the receptor, and secondly binding of a co-activating protein to a part of the LBD (AF-2) triggers gene expression. Chemicals binding to LBD may again yield agonistic or antagonistic effects, for binding to AF-2 only antagonism is known. However, adverse effects to thyroid hormones can also result from other mechanisms as enzyme/protein interaction e.g. with the transporter protein and aryl-hydrocarbon-receptor interaction. Structural alerts to predict chemicals with potential effect on these systems have been developed. For the estrogen/androgen system, an existing approach has been refined. The model identifies 91% of the active chemicals, and false negative results are weakly active only. The models for LBD and AF-2 binding only miss one active compound. For other thyroid hormone effects a screening level model detects ca 95% of the known active compounds, but there is suspect of missing compound classes due to the lack of respective experimental data. Particular remark was given to characterize the applicability domain and reliability of the predictions. All models are implemented as automated tools in the software system ChemProp (UFZ Department of Ecological Chemistry 2017. ChemProp 6.6. http://www.ufz.de/ecochem/chemprop). Acknowledgment: This study was financially supported by the German Federal Environment Agency, FKZ 3714 63 412 0.

TH055

Mixtures of endocrine disrupting chemicals disrupt behaviour and thyroid hormone related gene expression in Zebrafish (Danio rerio) larvae

L. Birgersson, J. Sturve, University of Gothenburg / Department of Biological and Environmental Sciences

Endocrine disrupting chemicals (EDCs) in the aquatic environment can have severe effects on the health of aquatic organisms as well as human health. Numerous anthropogenic EDCs, such as plasticizers, fire retardants and antibacterial agents, enter aquatic ecosystems from wastewater treatment plants and land runoff. Several of these have been shown to have adverse effects on fish, including disruption of reproduction, neurodevelopment and brain formation. Previous studies have mainly focused on single compound exposures or simple mixtures and further evaluation of complex mixtures at low concentrations is needed. Within the framework of the European Horizon2020 project EDC-MixRisk, EDCs linked to adverse effects on neurodevelopment and growth in a pregnancy cohort study have been identified. Mixtures of these chemicals (phthalate metabolites, phenols and PFASs) were synthesized to be tested in a range of *in vivo* and *in vitro* systems. The thyroid hormones (THs) are one of the targets of interest as they are essential for brain development and disruption of this axis may lead to alteration of neurodevelopment. The current study aimed to determine the effects of the EDC-mixtures on larval behaviour and to identify disruption of TH-related gene expression in zebrafish (*Danio rerio*) during early development. Zebrafish embryos were exposed to Mix N and Mix G (mixtures correlated with adverse effects on neurodevelopment or growth in the epidemiological study) for 48h in concentrations equivalent to 0.01x – 100x human levels. Alterations of larval behavior caused by the exposures were studied as an endpoint for neurodevelopment since behavior integrates many biochemical processes and can be a sensitive endpoint for sub-lethal toxicity of endocrine disruptors. Larval locomotion was tracked using the ViewPoint ZebraBox and a protocol of alternating dark/light cycles. Quantitative PCR was used to determine the effects of the EDC mixture on the expression of thyroid related genes. Our results show that acute exposure to the mixtures significantly alter larval locomotion and expression of genes involved in TH signaling, including thyroid hormone receptors *thra* and *thrb* as well as the deiodinases *dio1* and *dio2* at concentrations corresponding to those found in pregnant mothers. These results will be combined with results from other model systems in the EDC-MixRisk project to improve risk assessment of EDC-mixtures.

TH056

Contaminants of emerging concern in the North American Great Lakes: Assessing environmental mixtures in multigenerational exposure studies

N. Cipoletti, St. Cloud State University / Aquatic Toxicity Laboratory; L. Wang, St. Cloud State University; H.L. Schoenfuss, St. Cloud State University / Aquatic Toxicology Laboratory

In aquatic ecosystems such as the North American Great Lakes watershed, organisms are exposed to complex chemical mixtures throughout life, producing effects not anticipated in laboratory settings designed to test acute effects of single chemicals. By exposing fathead minnows through three generations, we aim to capture exposure effects during sensitive life stages. Through two separate multigenerational studies, we analyzed the effects of both urban and agricultural co-occurring contaminants at environmentally relevant concentrations in the Great Lakes watershed. Fathead minnows were housed in a flow-through exposure system and propagated for three generations (approximately one year of continuous exposure). Larval fish were analyzed for predator avoidance performance, feeding efficiency, and growth. Adult fish were analyzed for fecundity, biological indices, and hematological characteristics (VTG, glucose). Both urban and agricultural exposures resulted in growth alterations between treatments most likely due to density-dependent growth. Urban exposure indicated higher fecundity (both first and second generation) at low and environmentally relevant concentrations as compared to control and high treatments, potentially as a therapeutic hazard associated with the estrogenic nature of the mixture. Agricultural exposure indicated higher fecundity at low and environmentally relevant concentrations for first generation only, whereas the second generation demonstrated decreases in environmentally relevant concentrations as compared to low and control treated fish, highlighting the differences between adult only exposed fish (first generation) and lifetime exposure (second generation). Urban exposure demonstrated no changes to male plasma VTG (egg-yolk precursor protein), but agricultural exposure demonstrated increases in high exposed male fish (first and second generation) and environmentally relevant exposed male fish (second generation) due to the estrogenic nature of the mixture and exposure during different developmental stages between generations. Results indicate that mixtures, environmentally relevant in composition and concentrations, have the potential to alter growth, lead to reductions in fecundity, and elevated egg-yolk precursor protein in male fish. Potential, yet unknown, consequences to the population level of exposed aquatic organisms may exist and warrant further study.

TH057

Contaminants of emerging concern in the North American Great Lakes: Assessing species sensitivity using environmental mixtures

S. Kohno, St. Cloud State University / Aquatic Toxicology Laboratory; N. Cipoletti, St. Cloud State University / Aquatic Toxicity Laboratory; L. Wang, St. Cloud State University; U. Hasbay, H.L. Schoenfuss, St. Cloud State University / Aquatic Toxicology Laboratory

Contaminants of emerging concerns (CECs) have been detected ubiquitously in aquatic environments, and their endocrine-disrupting effects concern us. We identified typical CECs in rivers associated with agricultural and urban land use in the North American Great Lakes watershed. A mixture of agricultural CECs (AG) was assembled to mimic the environmentally occurring compounds and consisted of 8 chemicals, while an urban CEC mixture (UB) contained 11 chemicals including the known estrogenic compounds, bisphenol-A, estrone and nonylphenol in both mixtures. In addition to estrogenicities of CECs, Minnesota Pollution Control Agency reported that an exposure to CECs in the river water activated a peroxisome proliferator-activated receptor (PPAR)/retinoid X receptor (RXR) pathway in the transcriptome analysis of fathead minnow. Our laboratory found that an exposure to CECs induced a higher incidence of hepatic vacuolization in fathead minnow, which would be an obesogenic effect of CECs via a PPAR/RXR signaling pathway. Two isoforms of estrogen receptor (ESR) of fathead minnow, bluegill sunfish, American alligator or human was examined in the human embryonic kidney 293T cells by quantifying their transcriptional activities using estrogen-response elements and luciferase reporter gene in an exposure to agricultural or urban CECs. Utilizing the same method *in vitro*, alligator PPAR-gamma and RXR-alpha were also examined. Both AG and UB mixtures were estrogenic, however, their estrogenicities varied depending on isoforms of ESRs and 4 species. Human ESR1 was the most sensitive to AG based on their estrogenicities, while minnow ESR1 was the least sensitive to AG. Bluegill ESR1 was the most sensitive to UB based on their estrogenicities, whereas BG ESR2 was the least sensitive UB in receptors we tested. Both AG and UB CEC mixtures did not activate neither alligator PPAR-gamma nor RXR-alpha. Although further investigations of PPAR/RXR signal are required in fathead minnow, PRARG/RXRA signals might not be involved in a CECs-exposure inducing a hepatic vacuolization in fathead minnow. These results indicate that efficacious receptors and species differ between CECs mixture, and further endocrine studies of CECs are required for a better understanding and prediction of CEC effects by utilizing a variety of receptors cloned from diverse threatened and endangered species.

TH058

Contaminants of Emerging Concern in the North American Great Lakes: Effects from simple exposures to complex mixtures
U. Hasbay, H.L. Schoenfuss, St. Cloud State University / Aquatic Toxicology Laboratory
Contaminants of emerging concern (CEC), including pharmaceuticals, personal care products and industrial agents may impact aquatic life. Previous studies have documented reduced predator escape performance in fathead minnow larvae exposed to diverse CECs. However, these studies did not consider the complex mixtures of CEC present in the environment. The purpose of the current study was to understand the change on apical endpoints as the complexity of CEC mixtures increases. We tested the hypothesis that as the complexity of CECs in exposures increase, the apical endpoints observed will differentiate from simple exposure endpoints. We assessed the potential of 21 commonly detected CECs on three life stages fathead minnows: embryo (developmental abnormalities, transcriptomics), juvenile (survival, escape performance, feeding efficiency, qPCR) and adult (survival, secondary sex characteristics, nest defense, courtship, boldness, qPCR) after 96-hour flow-through exposures. In addition, we began the process of building a series of complex mixtures to study the CEC effects using neural network methodology. Individual compound concentrations and mixture composition were based on an analysis of nearly 500 water samples collected as part of the Great Lakes Restoration Initiative. Our current findings show that the larval survival was significantly reduced ($p< 0.05$, ANOVA) by diverse CECs including estrone, desvenlafaxine, and tris(2-butoxyethyl) phosphate exposures. Interestingly, exposure to ibuprofen showed a potential therapeutic effect at the medium concentration (environmental concentration) on adult fathead minnow. We also observed indication of endocrine disruption on our mixture exposure which includes all studied chemicals. We expect that these evaluations will lead us to improve adverse outcome pathway concepts by testing same chemical effects at different life stages of fathead minnows, and forming a linkage between behavioral responses and adverse outcomes.

TH059
Contaminants of emerging concern in the North American Great Lakes: Load reduction and biological recovery after wastewater treatment upgrades
H.L. Schoenfuss, St. Cloud State University / Aquatic Toxicology Laboratory; D. Martinovic-Weigelt, University of St. Thomas / Biology; P. Edmiston, The College of Wooster / Chemistry; T. Minarik, Metropolitan Water Reclamation District of Greater Chicago
Many urban aquatic ecosystems are becoming effluent dominated, resulting in the presence of contaminants of emerging concern and subsequent adverse effects on aquatic wildlife. Despite these dramatic alterations, effluent dominated urban systems support many ecosystem services and are used by the nearby human population for recreation. The Metropolitan Water Reclamation District of Greater Chicago upgraded two wastewater treatment plants (one million cubic meters/day each) to disinfection (UV; chlorination/de-chlorination). The receiving aquatic ecosystem adjacent to these two wastewater treatment plants has been the focus of intense biological and chemical study for the past seven years and provides a unique opportunity to assess two divergent treatment technologies (UV disinfection vs. chlorination/de-chlorination) and to examine how adverse biological effects in exposed fish may be mitigated through effluent disinfection. We exposed male fathead minnows (*Pimephales promelas*) in on-site flow-through exposure systems four times prior to the treatment upgrades and four times since to examine these questions. In addition, we conducted extensive analytical chemistry on effluent samples and employed in-vitro assays to examine overall biological activity of effluents prior and following disinfection treatment. Both disinfection methods transformed many CECs, in some instances reversibly (UV disinfection). UV disinfection resulted in enhanced maturity of male fathead minnows when compared to fish exposed to non-disinfected effluent or a control treatment ($P< 0.05$, ANOVA). Disinfection by chlorination/dechlorinating reduced testis maturity and changes liver hepatocyte appearance (both $P< 0.05$, ANOVA). Expression of vitellogenin, an indicator of estrogenic exposure, was not altered by treatment at either facility ($P>0.05$, ANOVA). These results indicate that biological activity of effluent is altered by both disinfection treatments but with more advantageous outcomes using UV disinfection.

TH060
Contaminants of emerging concern in the North American Great Lakes: Validation of effects through field-based exposures
V. Korn, St. Cloud State University / Aquatic Toxicology Laboratory; N. Cipoletti, St. Cloud State University / Aquatic Toxicity Laboratory; C. King, H.L. Schoenfuss, St. Cloud State University / Aquatic Toxicology Laboratory
Agricultural and urban pollutants are an environmental health concern as their presence in aquatic ecosystems often results in increased stress in aquatic organisms. The effects of agricultural and urban mixtures, each having different chemical signatures, have been studied rather infrequently. The objective of these field-based studies was to assess the impacts of agricultural and urban pollutants on the physiology, reproduction, and population health in fish. These studies, both part of the Great Lakes Restoration Initiative, utilized two distinct watersheds; the Maumee River watershed (Toledo, Ohio) was used to study agricultural pollutants, while the Milwaukee river system watershed (Wisconsin) was used to study urban pollutants. Laboratory cultured adult and larval fathead minnows were exposed to water samples from different sites in the two watersheds for 21 days. Adult minnows were analyzed for alterations in hematological characteristics (glucose, VTG, E_2, 11-KT) and reproduction, while larval minnows were analyzed for feeding efficiency and predator avoidance performance. The Maumee River indicated reduced reproductive capability, as measured by fecundity, at specific sites along the channel with large seasonal differences between the two sampling years potentially due to altered contaminant loads during heavier periods of precipitation. The Maumee River demonstrated no changes to larval predator avoidance behavior or other apical endpoints (feeding, growth) indicating that agricultural contaminants pose no/little perceived threat to larval development. Conversely, the Milwaukee River indicated increased reproductive capability, as fecundity increased among field sites in comparison to controls. Additionally, urban samples resulted in an increase in larval growth following 21 days of exposure, but did not impact the predator avoidance behavior. The results indicate that agricultural and urban pollutants entering aquatic environments impact fish physiology and reproduction. Further research is underway to determine whether these observed effects have an impact at the population level.

TH061
Towards a multiparallel detection of biological effects caused by anthropogenic micro-pollutants
C.E. Riegraf, German Federal Institute of Hydrology; L. Moscovici, The Hebrew University of Jerusalem / Institute of Life Sciences, Department of Plant and Environmental Science; G. Reifferscheid, German Federal Institute of Hydrology; S. Belkin, D. Shakibay, The Hebrew University of Jerusalem; S. Buchinger, Federal Institute of Hydrology / Department G Biochemistry Ecotoxicology
Organic micro-pollutants (MPs) can enter the aquatic environment via diverse pathways and sources e.g. waste water treatment effluents, agricultural activities or the disposal of various consumer goods. Tracking the occurrence, distribution and fate of MPs in the subsurface can be achieved by using an effect directed analysis, which directly links chemical analysis of contaminants to their potential adverse biological effects. The project "TREES"[1] (**TR**acking **E**ffects of **E**nvironmental organic micro-pollutants in the **S**ubsurface) aims to develop an innovative technological platform for monitoring MPs based on the assessment of their biological effects. The proposed setup will be composed of the following steps: (a) Extraction and pre-concentration of MPs and their possible transformation products in soil or water samples by solid phase extraction. (b) Separation of the extracts using high performance thin layer chromatography (HPTLC). (c) Biological effect measurement of the individual separated constituents by using genetically engineered yeast (*Saccharomyces cerevisiae*) or bacterial (*Escherichia coli*) bioreporters. A main goal of our study is to develop tools and methods for a multiparallel effect detection covering a range of potentially adverse biological effects. This can be achieved by the construction of yeast strains using specific fluorescence reporters for the various endpoints to be detected. By coupling these strains with HPTLC and mass spectrometry, a wide variety of compounds with biological activity could be screened simultaneously. The first step of coupling HPTLC with different bioreporters for the detection of a series of biological endpoints (androgenic, thyroidogenic, genotoxic, dioxin-like effects, effects on the vitamin D and the retinoic acid receptor) was successfully performed by using various reference compounds. Furthermore, mixing yeast strains with different endpoints yielded to the detection of different adverse effects at the same time. Next steps will include (i) the analysis of real samples, (ii) the further development of advanced sensor-strains suitable for the simultaneous detection of different (eco)toxicological effects and (iii) method development for the detection of compounds by chemical analysis after separation by HPTLC. <br clear="all" /> [1] A German-Israeli research and development project in the field of water technology within the framework of the BMBF-MOST cooperation, FKZ: 02WIL1387.

TH062
Endocrine disruptors used in polymers in the offshore oil and gas industry
C. Phillips, Cefas Lowestoft Laboratory / Science Directorate - advice and assessment; R. Suehring, University of Toronto; A. Smith, Cefas / Ecotoxicology and Molecular Ecology
Concerns were raised by regulatory assessors that a number of polymer substances found in products registered for use and discharged offshore as a result of the activities of the oil and gas industry are based on monomers that are known and suspected endocrine disruptors. These polymers were described by the registration data as being moderately or readily biodegradable substances and might therefore have the potential to biodegrade into the endocrine disruptors on which they were based, or leach endocrine disruptors upon weathering or as a result of well bore conditions. To determine the potential of these polymers to release endocrine disruptors, four substances with known endocrine disrupting monomer groups were extracted using high pressure and temperature as well as acidification and/ or biodegradation and were tested using a Yeast-based estrogen screen (YES) and yeast-based androgen screen (YAS). The results from the presented study show that at least one of the analysed products has a high potential for releasing EDs and highlights the importance of well-informed environmental protection to prevent endocrine disruptors from impacting the marine environment.

TH063

Thyroid disruption screening using zebrafish as vertebrate model

I. Iturria, O. Jaka, C. Martí, A. Alzualde, BioBide; A. Muriana, BBD BioPhenix S.L. / RD

Endocrine disrupting compounds are frequently found in the environment and have a profound impact on the development and physiology of vertebrate organisms. Thyroid Disrupting Compounds (TDC) specifically affects the function of thyroid hormones, interfering with their synthesis, transport and/or binding; altering important physiological processes. Several environmental contaminants such as polybrominated diphenyl ethers or halogenated organophosphates, used as plasticizer and flame retardant, are suspected to produce a thyroid-disrupting effect. Given so, chemical manufacturing entities could benefit from cost-effective methodologies for the screening of TDC in order to deselect candidates during the early phase of the development. In this work, we present an assay for the screening of potential TDC using zebrafish embryo. This vertebrate model is extensively used as a biosensor for the evaluation of acute and developmental toxicity, and several assays in zebrafish are described by the OECD guidelines for the testing of chemicals. Besides, the embryo's small size and transparency allow to carry out fluorescence-based screening assays with medium throughput. In this work, the thyroid hormone disruption potential of several environmentally relevant contaminant was assessed. For this end, an initial acute toxicity assay was performed in order to estimate the EC50 and NOEC of the tested compounds, and subsequently select concentrations with no systemic toxicity. Afterward, change in the thyroglobulin (TG) synthesis was assessed using a zebrafish transgenic line expressing mCherry fluorescent protein under the control of the zebrafish TG gene promoter, by analysis of the fluorescence microscopy images. Finally, a gene expression assay, using rt-qPCR, was performed over known markers of thyroid disruption to further characterize the involved pathways of endocrine disrupting effect. The zebrafish assay showed to be a sensitive and cost-effective assay to evaluate the potential thyroid disruptor activity of chemicals.

TH064

Development of stably transfected cell lines with zebra fish thyroid hormone receptors alfa and beta for assessing endocrine disruption in environmental samples

V. García Herranz, INIA   National Institute for Agricultural and Food Research and Technology; E. Sánchez Martínez, Institute of Aquaculture Torre de la Sal-Spanish National Research Council; M. Fernandez-Cruz, J. Navas, INIA - National Institute for Agricultural and Food Research and Technology / Environment; J. Cerdá Reverter, Institute of Aquaculture Torre de la Sal-Spanish National Research Council

Endocrine-disrupting chemicals (EDCs) are ubiquitous in our environment and can be found in many products including food/feed, containers as plastic bottles or metal food cans, cosmetics, pesticides, flame retardants, detergents… Accordingly, they suppose a threat to animal and human health through different exposure routes. *In vitro* bioassays are valuable tools for detecting and studying EDCs action and provide a sensitive and rapid system to evaluate their potential effects. In addition, the application of this kind of assays is in line with the 3R principles in relation to animal use in research. In recent years, thyroidal endocrine disruption has aroused great interest due to the critical roles played by thyroid hormones in animals. The aim of this work was to develop two cell lines stably expressing zebrafish (*Danio rerio*) thyroid receptors (TR) α and β that could be used for the assessment of disruption of the hypothalamus-hypophysis-thyroidal axis. To do this, the commercial HEK-293 cells was transfected with the zebrafish TRα- or TRβ-pcDNA3 constructs, together with luciferase gene (DR4-TK-Luc reporter construct) under the control of thyroid hormone response elements (TRE). Forty-eight hours post-transfection, cells were harvested and seeded in 96-well plates in Dulbecco's Modified Eagle's Medium (DMEM) supplemented with 0.4 mg/mL G-418, 10% fetal bovine serum and 1% penicillin/streptomycin (selection medium). To test the stable integration as well as the expression level of the receptor positive clones were plated in 96-well plates and exposed to a range (from 1 µM to 1 fM) of triiodothyronine (T3) concentrations. Finally, the clones showing better EC_{50} values were selected to determine the presence of thyroidal activity in livestock residues including agricultural amendments. These transactivation systems allowed distinguishing the contribution of each TR to the residue-induced thyroidal activity. Acknowledgements - Supported by RTA2012-00053-00-00, RTA2015-00041-00-00 and AGL2016-74857-C3-3-R.

TH065

Screening endocrine disrupting potentials of alternative plasticizers using three cell line assays

G. Lee, H. Kang, Seoul National University Graduate School of Public Health; K. Choi, Seoul National University / Environmental Health Sciences

Phthalates have been used as plasticizer in polyvinyl chloride (PVC), food containers, medical devices, building materials, and personal care products. Because of reproductive toxicity of several phthalates including bis(2-ethylhexyl) phthalate (DEHP) and diethyl phthalate (DEP), use of major phthalates are regulated in many products in several countries. Accordingly, many alternative plasticizers have been developed and increasingly used worldwide, but their possible adverse endocrine disruption effects are not well-known. The aim of this study is to screen endocrine disrupting potentials of several widely used alternative plasticizer, cyclohexane dicarboxylic acids (DINCH), acetyl tributyl citrate (ATBC), dioctyl terephthalate (DOTP), trioctyl trimellitate (TOTM), bis(2-ethylhexyl) adipate, and diethylhexyl adipate (DEHA). A series of *in vitro* assays employing a human breast (MVLN), a human adrenal (H295R), and a rat pituitary (GH3) cells, were employed. The test doses for each plasticizers applied were determined based on preliminary cytotoxicity assays for each cell line. While none of alternative plasticizers showed significant estrogen receptor binding affinity in MVLN cells. DINCH and DEHA exhibited significant increase in estradiol (E2) to testosterone (T) ratio in H295R cells. These results suggest that these plasticizers DINCH and DEHA cause increased estrogenicity through altering steroidogenic pathway, similar to DEHP. In GH3 cell line, *tshβ* gene was significantly downregulated by exposure to TOTM, suggesting its thyroid disrupting potential through altering signaling pathway to thyroid gland. Our observation shows that DINCH, DEHA and TOTM may disrupt balance of important hormones. Further investigations using *in vivo* models are warranted.

TH066

Development of reporter gene system for assessing cherry shrimp ecdysone receptor agonists using mammalian cells

K. Chan, The Chinese University of Hong Kong / Life Sciences; Y. Chan, K. Chu, The Chinese University of Hong Kong / School of Life Science

Ecdysteroid is a key steroid hormone that regulates growth, development and molting in animals under the phylum of Arthropod, which includes the insects and crustaceans. The hormone targets the receptor complex which comprises the ecdysone receptor (EcR) and retinoid X receptor (RxR). The activated complex anchoring on the ecdysone responsive element (EcRE) stated on the promoter subsequently initiates transcription of the responsive gene(s). Chemicals act as receptor agonists do not necessarily adopt the structure of the native hormone, as in the case of estrogenic endocrine disruptors. Recently, for insect pest control, synthetic diacylhydrazine (DAH) and bisacylhydrazine (BAH) compounds were developed to disrupt ecdysone/receptor signalling. They work as the ecdysone receptor agonists, which cause premature launching of the molting process and subsequently death. Crustaceans, as a subphylum closely related to insects phylogenetically, also adopt this ecdysone signalling system, as they share the hormone, hormone synthetics enzymes and the receptors. Thus, these endocrine disrupting insecticides, together with other untested potential endocrine disruptors, may post a thread on the crustaceans. Here we report the development of an in vitro reporter assay for the screening of ecdysone receptor agonist in cherry shrimp. The assay is done by transiently transfecting mammalian cells with plasmid vectors expressing cherry shrimp EcR and RxR, together with a vector carrying a luciferase reporter gene fused to a minimal promoter linked to five copies of EcRE. The results show that the system responses well to the native ecdysone hormones in a dosage-dependent manner. The adaptation of mammalian cells in *in vitro* assay for heterogenous receptor is satisfactory. Three DAH/BAH insecticides were also tested and gave minimal to moderate signals. The results suggest that these DAH insecticides aimed for insect pest control can be potential hazards to crustaceans. More studies on different mammalian cells and competition study with mixtures of chemicals are being carried out to validate this reporter gene system.

TH067

Micro-injection as an alternative for aquatic exposure? A case study in zebrafish embryos with 17α-ethinylestradiol.

E.D. Michiels, University of Antwerp / Zebrafishlab Dept Veterinary Sciences; F. Lai, University of Antwerp / Toxicological Centre Dep of Pharmaceutical Sciences; L. Vergauwen, University of Antwerp / Zebrafishlab Dept Veterinary Sciences SPHERE; A. Covaci, University of Antwerp, Toxicological Center / Toxicological Centre Dep of Pharmaceutical Sciences; A.L. van Nuijs, University of Antwerp / Toxicological Centre Dep of Pharmaceutical Sciences; S.J. Van Cruchten, University of Antwerp / Applied Veterinary Morphology, Dept Veterinary Sciences; D. Knapen, University of Antwerp / Zebrafishlab Dept Veterinary Sciences

Pharmaceutical companies have to perform an environmental risk assessment for every drug that is launched to the market. The mandatory tests for potential endocrine disrupting (ED) compounds are mainly based on aquatic toxicity tests. However, it is often difficult to expose fish to poorly water soluble ED pharmaceuticals via water. Micro-injection in the yolk is therefore proposed as an alternative and ecologically relevant exposure route because the yolk of zebrafish embryos contains many lipids, and this route mimics maternal transfer. To be used as an exposure method, micro-injection needs to be characterized and compared to the traditional exposure route via water. In this study, 17α-ethinylestradiol (EE2, an estrogen receptor (ER) agonist) was chosen as a model compound to compare both exposure routes. Zebrafish embryos were exposed either via water or via injection within the first two hours post fertilization (hpf) until 120 hpf. Different endpoints at different levels of biological organization were assessed. Morphological (i.e., different types of abnormalities) and physiological (e.g., heart rate and swimming performance) endpoints were scored, as well as ER binding and qPCR analysis of 14 genes. An LC-MS/MS method was optimized for measuring EE2 levels in medium of the aquatic exposure experiment and the internal dose in embryos after aquatic exposure or injection. The pattern of brain aromatase mRNA expression was different between both exposure routes, while vitellogenin (*vtg1*) and estrogen receptor 1 mRNA levels were similar between both routes after EE2 exposure. At the morphological and physiological level we observed differences as well. However, the degree of ER-binding was similar between both routes from day 1 until day 5. Despite daily refreshment, the EE2 concentration in the medium decreased regardless of the exposure concentration. The internal doses were the highest at the beginning of the exposure for both exposure routes and decreased afterwards. The order of magnitude of the internal dose was also similar between the injection and an aquatic exposure in the µg/L range, which was also seen e.g. for the mRNA expression of *vtg1*. Based on the dose measurements we can conclude that even if the embryos were dosed with EE2 within the same order of magnitude that there were still different outcomes for some endpoints. Therefore micro-injection is rather a complementary method and not an alternative route for aquatic exposure.

TH068

Vitellogenin expression, ovarian growth and hormone levels are affected by atrazine in the crayfish Procambarus clarkii

G.R. Silveyra, University of Buenos Aires / Dept. of Biodiversity and Experimental Biology. Institute of Biodiversity, Experimental and Applied Biology (IBBEA), CONICET-UBA; P. Silveyra, Penn State College of Medicine / Dept. of Pediatrics; I. Vatnick, Widener University; D.A. Medesani, University of Buenos Aires / Dept. of Biodiversity and Experimental Biology. Institute of Biodiversity, Experimental and Applied Biology (IBBEA), CONICET-UBA; E.M. Rodriguez, University of Buenos Aires / Biodiversity and Experimental Biology

Atrazine, a widely used herbicide, has been categorized as a suspected endocrine disruptor for many years. Although several studies have investigated the effects of atrazine exposure on reproductive function, its safety remains controversial and the mechanisms of its toxicity remain unclear. In this study, we tested the hypothesis that atrazine can affect reproduction in crayfish through dysregulation of vitellogenin expression and hormone synthesis. Adult female crayfish (*Procambarus clarkii*) were exposed during one month to atrazine at concentrations of 1 or 5 mg/L. At the end of the exposure, ovaries, hepatopancreas and hemolymph samples were harvested for analysis of vitellogenin expression and steroid hormone levels. Ovarian tissue was also sampled for both biochemical and histological analyses. Atrazine-exposed crayfish had a lower expression of vitellogenin in the ovary and hepatopancreas, as well as smaller oocytes and reduced vitellogenin content in the ovary. Despite these effects, circulating levels of estradiol increased in females exposed to 5 mg/l of atrazine, showing that the inhibiting effect of atrazine on vitellogenin production was not related to a lower secretion of sexual steroids; instead, some early stimulating effects of estradiol on vitellogenesis could have occurred, particularly in the hepatopancreas. Together, our data showed that atrazine exposure was able to inhibit vitellogenin production in the crayfish *P. clarkii*, altering on the other hand the normal balance of sex steroids.

TH069

Identification of molt-inhibiting hormone and ecdysteroid receptor sequences in Gammarus pulex and consequences of endocrine disruptor exposures

E. Gismondi, University of Liege

Endocrine disruptors (EDCs) are well known to disrupt the development and the reproduction of exposed organisms. Although this point has been studied in vertebrate models, the limited knowledge of the endocrine system of invertebrates makes the evaluation of EDCs effects difficult. However, invertebrates represent the major part of aquatic ecosystems, such as amphipods *Gammaridae*, which are crucial for their functioning (e.g. litter degradation, food resource). Moreover, gammarids are hosts of hidden parasites such as vertically-transmitted microsporidia (microsporidia VT), which could be confounding factors in assessment of EDC effects, since microsporidia VT could feminize juvenile males in some *Gammarus* sp. Consequently, currently, no biomarkers (assessment tools) are available to assess the endocrine disruption in gammarids. The present work focused on EDC effects on the molt process of *Gammarus pulex*, by researching the DNA sequences of two main proteins in the endocrine system of amphipods: the molt-inhibiting hormone (MIH) and the ecdysteroid receptor (EcR). Next, the expression variations of these two genes have been measured by RT-qPCR after an exposure of four EDCs: ethinylestradiol (EE_2), 4-hydroxitamoxifen (4HT), 17α-methyltestosterone (17MT) and the cyproterone acetate (CPA), all commonly studied in vertebrates. Sequence research allowed to obtain a 204 bp length and 255 bp length amplimers for EcR and MIH, respectively. The EcR sequence encodes for 68 amino acid fragment while the MIH sequence encodes for an 85 amino acid fragment. Exposure of *G. pulex* males at each EDC highlighted an increased of the MIH expression, whatever the parasitic status. However, a tend to increase was observed for the EcR expression only in uninfected gammarids. This work allowed to identify two main proteins involved in the endocrine system of amphipods. Exposure to each ECD highlighted EDCs affecting vertebrates could also impact invertebrates species. In addition, the presence of microsporidia VT appeared to be a confounding factor which could lead to misinterpretation the endocrine risk assessment. Finally, results the results are promising in the development of PE biomarkers in invertebrates, since this is a tool that is currently missing. However, further studies will be needed to study the variations of these genes and understand their regularization, before to use them as biomarkers.

TH070

Use of in vivo and in vitro assays to investigate the effects and bioavailability of endocrine disrupting compounds in sediment on the benthic invertebrate Chironomus riparius

S. Karnatz, RWTH Aachen University / Institute for Environmental Research Department of Environmental Analysis; S.E. Crawford, RWTH Aachen University / Institute for Environmental Research, Dept. of Environmental Analysis; V. Esser, RWTH Aachen University / Physical Geography and Geoecology; A. Müller, RWTH Aachen University / Institute for Environmental Research; A. Shuliakevich, Institute for Environmental Research (RWTH Aachen University) / Institute for Environmental Research; H. Hollert, RWTH Aachen University / Institute for Environmental Research

Sediments act as a sink and source of chemicals in the environment and, therefore, it is of great importance to know how sediment-bound chemicals affect aquatic organisms. The synthetic hormone 17α-Ethinylestradiol (EE2), a component of oral contraceptives, is ubiquitous in the environment and is a known potent endocrine disrupting compound (EDC) that adversely effects aquatic vertebrates (e.g. reproductive development of fishes). Due to the octanol-water partition coefficient ($logK_{ow}$) of EE2, it is expected to adsorb to organic matter and accumulate in sediment. However, little is known about the effects of sediment-bound EE2 on sediment-dwelling organisms. In the present study, the effects of EE2 were investigated on the freshwater benthic invertebrate, *Chironomus riparius*, in a long-term sediment-water toxicity test using formulated EE2-spiked sediment and field sediments. Field sediments were collected from the Rivers Luppe (silt loam, 8% organic carbon; eastern Germany), Wurm and Inde (sand, 2% organic carbon; western Germany) as these rivers are heavily influenced by anthropogenic activities (i.e., downstream of wastewater treatment plants) and are suspected or known to contain high concentrations of EDCs. Two types of sediment were formulated with silica sand, kaolin clay and peat moss to match the sediment types of the Luppe and Wurm/Inde. Each formulated sediment type was used in the negative control, solvent control and spiked (10 µg EE2/g d.w.) treatments. The survival and growth of the *C. riparius* larvae were measured after 10-d in half of the replicates ($n = 6$), while the number of adults emerged, time to emergence and the sex ratio were evaluated at the end of the 28-d test ($n = 6$). Extracts of the whole-organism were analyzed through a yeast estrogen receptor (YES) assay, a common *in vitro* assay used to estimate estrogenic potential. Additionally, EE2 tissue extracts were quantified through LC/MS-MS with deuterated internal standards which were used to account for any losses during the extraction process. The bioavailability of EE2, inferred through the YES assay and LC/MS-MS analysis, provides insight into the bioaccumulation of EE2 in *C. riparius* larvae. Knowledge about the bioavailability, bioaccumulation, and estrogenic potential of sediment-bound EE2 on benthic organisms is important for understanding the potential effects on vertebrate predators and subsequent upper trophic levels as a secondary source of contamination.

TH071

Assessing acute toxicity of Bisphenol A on Daphnia magna by passive dosing approach

H. Kwon, Y. Jeong, H. Jeon, S. Kim, KIST Europe / Environmental Safety Group

Bisphenol A (BPA) is a raw material for widely used polycarbonate plastics, but it is known to have negative effects on human health and the environment. It was classified as an endocrine disrupting chemical (EDC), which requires an accurate and reliable toxicity data for robust risk assessment. However, currently available eco-toxicity data (48h-EC_{50}) on *Daphnia magna* showed a significant discrepancy of 3.9-16.0 mg/L. Therefore, our aim is to determine the reliable toxicity of daphnia to BPA. Passive dosing as well as the existing toxicity testing protocol (i.e., spiking with co-solvent) were used to administer BPA to daphnia. Conventional spiking method often fails to control the exposure concentration of (semi-)hydrophobic organic compounds due to the loss of target compounds from sorption to the test vessel and volatilization. Here, passive dosing technique compensates for the concentration loss by using a biocompatible polymer as a reservoir. Moreover, the adverse effect of the only target compound can be considered in this format as passive dosing does not require a co-solvent to dissolve and deliver the target compound. A silicone O-ring was chosen as a reservoir for dosing BPA to *Daphnia magna*. The uptake and release kinetics of BPA on the O-ring were investigated until equilibrium. After the concentration of BPA in the test vessel reached equilibrium, we put *Daphnia magna* (< 24h) in it and checked the sub-lethal effect in 48h by following the OECD guideline 202. At the same time, the acute toxicity test by spiking BPA dissolved in methanol (0.01%) was conducted with the same range of concentrations (0-40 mg/L). Through the passive dosing method, we were able to determine the silicone-water partition coefficient of BPA and control stable concentration over the test period. The uniform concentration of BPA induced the half maximal effective concentration of daphnids at the lower concentration. We expect that the application of this method in a chronic toxicity test will provide more reliable environmental hazard and risk assessment of BPA. Furthermore, this result suggests that passive dosing could be adjusted to less hydrophobic compounds like BPA (log K_{OW} of 3.64).

TH072

Toxic effects of juvenile hormone analogue insecticides, methoprene and fenoxycarb, on cherry shrimp (Neocaridina davidi)

X. HU, The Chinese University of Hong Kong; K. Chan, The Chinese University of Hong Kong / Life Sciences; K. Chu, The Chinese University of Hong Kong / School of Life Sciences

Crustaceans are a large group of arthropod, and they are the major constituents to aquatic ecosystems that provide a variety of ecological and economic services. Nowadays, the increasing quantities of insecticides leached into water bodies severely affect the health of aquatic environment globally and heighten the adverse impact on the crustaceans. Among these insecticides, juvenile hormone analogue (JHA) insecticides are a kind of endocrine disrupters known to interfere with the natural hormone action in insects by mimicking the juvenile hormones. However, the structure and functions of the methyl farnesoate (MF) in crustaceans are similar to the juvenile hormone (JH) in insects. Therefore, the exogenous JHA insecticides would cause adverse effects on the development and reproduction in crustaceans as in insects. The aim of our study is to examine the toxic effects of JHA insecticides - methoprene and fenoxycarb on a freshwater shrimp *Neocaridina davidi* which is successfully cultured and maintained in our laboratory as a new crustacean model. These insecticides are growing in use in agriculture both locally and globally, and their impact to the aquatic ecosystem is needed to be further clarified. In the present study, the acute and chronic toxicity effects of two juvenile hormone analogue insecticides, fenoxycarb and methoprene, on newborn *N. davidi* were investigated. The 24h and 48h median lethal concentrations (LC_{50}) of fenoxycarb and methoprene were 1.40, 0.97 mg/L (4.64, 3.20 mmol/L) and 1.96, 1.26 mg/L (6.32, 4.06 mmol/L), respectively. The results of chronic experiments suggest that these two chemicals have adverse effects on the development of juvenile *N. davidi*. After six weeks chronic exposure to 100 μg/L (0.33 μmol/L) fenoxycarb and 200 μg/L (0.64 μmol/L) methoprene, the body length decreased by 20.5% and 11.8% as compared to control, respectively. And the total number of molts of 20 shrimp over this period reduced by 29.2% and 17.7%. Differential expressions of JH signaling pathway genes were observed in this study. The genes hr3 (hormone receptor 3) and e75 in *N. davidi* were up-regulated, while Chd64 (calponinlike protein), CHH (crustacean hyperglycemic hormone), e74, JHE (JH esterase), JHEH (JH epoxide hydrolase) and JHAMT (JH acid methyltransferase) were down-regulated in shrimp treated with fenoxycarb and methoprene. The results indicate the interference of these two JHA insecticides on the juvenile hormone system.

TH073

Development of Multimedia Fate Model for Human Risk Assessment of EDCs in the Asan Lake Watershed, Korea

M. Choi, J. Kim, Greenecos Inc; Y. Kim, Greenecos Inc. / CEO

Multimedia fate model, HURAME (multimedia fate model for HUman Risk Assessment in Multimedia Environment) has been developed considering topographic features and environmental and been meteorological data of watershed region of Asan Lake in Korea. Human risk assessment system using this multimedia fate model was established and integrated to assess spatial exposure and risk about human living in this area. For this purpose, first, GIS database system consisting of environmental and meteorological data and measured data of hazardous chemicals was developed. Second, Environmental concentrations of various chemicals were predicted, applying different fate processes according to different chemical properties. Third, advection and dispersion by wind in air grids, runoff in watershed, flows of water in water segments are considered in the watershed-based multimedia fate model, which was linked to a risk assessment system that predicts environmental multimedia concentrations of EDCs (Endocrine Disrupting Chemicals) and assesses human risk in this area. HURAME is valuable tools for predicting the fate of chemicals in evaluative and real environments with areas consisting of many watersheds. These models are an integral component of exposure assessment and risk assessment strategies, and are used in detailed assessments of contaminant fate. The aim of these models is to describe quantitatively contaminant fate and migration in a defined watershed region, with water segments and air grids inside of the region of interest treated more complicatedly. As a result, regional levels of environmental contamination are controlled by environmental parameters and processes and meaningful evaluation requires assessment of contaminant fate in neighboring regions. A linked set of regional models thus has the potential to describe quantitatively the impact of chemical emissions over a wider geographic scale with significant spatial differences in environmental characteristics and chemical use patterns.

TH074

Comparative toxicity and endocrine disruption potential of urban and rural atmospheric organic PM1 in JEG-3 human placental cells.

B.L. van Drooge, IDAEA-CSIC / Department of Environmental Chemistry; A. Marqueno Bassols, Institute of Environmental Assessment and Water Research IDAEA CSIC; B. Pina, C. Porte, IDAEA-CSIC / Department of Environmental Chemistry; J. Grimalt, Institute of Environmental Assessment and Water Research IDAEA CSIC / Department of Environmental Chemistry

Outdoor ambient air particulate matter and air pollution are related to adverse effects on human health. The present study assesses the cytotoxicity and ability to disrupt aromatase activity of organic PM_1 extracts from rural and urban areas at equivalent air volumes from 2 to 30 m^3, in human placental JEG-3 cells. Samples were chemically analysed for particle bounded organic compounds with endocrine disrupting potential, i.e. PAH, O-PAH, phthalate esters, but also for organic molecular tracer compounds for the emission source identification. Rural samples collected in winter were cytotoxic at the highest concentration tested and strongly inhibited aromatase activity in JEG-3 cells. No cytotoxicity was detected in summer samples from the rural site and the urban samples, while aromatase activity was moderately inhibited in these samples. In the urban area, the street site samples, collected close to intensive traffic, showed stronger inhibition of aromatase activity than the samples simultaneously collected at a roof site, 50 m above ground level. The cytotoxicity and endocrine disruption potential of the samples were linked to combustion products, i.e. PAH and O-PAH, especially from biomass burning in the rural site in winter. Exposure of zebrafish embryos to the same organic PM extracts resulted in high mortality in those exposed to rural samples collected in winter and an induction of genes implicated in basic cellular functions, such as cell proliferation. Moreover, the embryo transcript analysis showed strong correlations between the Aryl Hydrocarbon Receptor signalling pathway and PAH concentrations. On the other hand, in the zebrafish embryos exposure experiment, the urban extracts showed an induction of oxidative stress related genes, which suggest different potential adverse outcomes for exposure to air pollution from specific sources.

TH075

Dietary and non-dietary prenatal exposure to endocrine disruptors (BPA and DEHP). Spanish case study.

M. MARTÍNEZ RODRÍGUEZ, J. Rovira, Universitat Rovira i Virgili; R.P. Sharma, Universitat Rovira i Virgili / Departament d Enginyeria Química; M. Nadal, Universitat Rovira i Virgili / School of Medicine, IISPV; V. Kumar, Universitat Rovira i Virgili / Departament d Enginyeria Química; M. Schuhmacher, Rovira i Virgili University / Departament d Enginyeria Química

Endocrine disruptors (EDs) are chemicals compounds that send confusing messages causing various dysfunctions by mimicking or altering the effect of hormones in the human body. Several EDs such as Bisphenol A (BPA) and di (2-ethylhexyl) phthalate (DEHP), are involved in obesity and diabetes diseases in children. Recent studies have shown evidences that these chemicals can cross the placental barrier making fetal exposure closely related to maternal exposure. The aim of this research is to establish fetus exposure to EDs (BPA and DEHP). To address this issue, recently, pregnant women recruitment has begun. In present work, dietary and non-dietary (dermal, non-dietary ingestion and inhalation) exposure of these women was considered in order to predict the dose of EDs at which the child has been exposed in the early stage of the development. The data obtained from this cohort (such as, physiological data, dietary habits and lifestyle, among others) was implemented into a physiological based pharmacokinetic (PBPK) model, which assesses the absorption, distribution, metabolism and excretion of a chemical compound into human body, as well as the internal exposure to target organs. To estimate the early exposure of the child, the model was implemented with fetus compartment for these chemicals. Results indicates that for both, BPA and DEHP, diet is the main contributor to the total exposure. However, indoor environment and dermal exposure also contribute significantly to the total DEHP exposure. Levels of both EDs were modelled in maternal blood and in fetus blood as well as in other body compartment. These results will be validated with the results of biological monitoring in the current cohort (n=72). The integration of the data obtained from current on-going human biomonitoring campaign and the physiological based pharmacokinetic model, here implemented; predict the early exposure of the child/fetus to EDs. This work is included in the frame of HEALS project (FP7-603946).

TH076

Sensitive Biomarker Assay using LC-MS/MS: Determination of Thyroid Hormones (T3 and T4) in Fetus, Pup and Adult Rat Serum - Sampling Considerations

S. Diaram, Envigo / Bioanalysis (LC-MS/MS)

The regulatory need for the analysis of circulating levels of thyroid hormones in rats (fetus, pup and adult) on reproductive toxicology studies has prompted the need for a sensitive assay for T3 and T4, which could not be fulfilled when using the traditional immunoassay technology platform. A method with a lower limit of quantification (LLOQ) of 5 pg/mL for T3 (final range 5 to 1500 pg/mL), and a final range of 70 to 70000 pg/mL for T4 was developed and validated using LC-MS/MS (liquid chromatography coupled to tandem mass spectrometry detector). The method validated utilzes a 50 µL sample volume of serum to determine both T3 and T4 from the same sample aliquot. Across several studies from various Toxicology facilities it was observed that mainly two different collection tubes were used for the clotting process to generate the serum sample ((a) plain plastic tubes and (b) tubes containing clot activator). A trend was observed in samples obtained using plain tubes for the clotting process resulting in suppressed analytical instrument responses. Hence an appropriately labelled internal standard is imperative, however this does not safeguard data points where low pg/mL concentration levels are present in the samples, which is particularly prevalent in fetus and Day 4 of age pups. Considering that samples are collected from animals of fetus and Day 4 of age pups, which may be triggered for analysis subsequent to Day 13 and adult male samples being analyzed, the emphasis of the integrity of the sample is paramount to

ensure meaningful data can be collected. An experiment was performed to evaluate if tubes containing clot activator could produce 'cleaner' serum samples to avoid loss of data points from analytical instrument signal suppression, yet provide true and accurate data without significant loss of T3 and T4 arising from potential absorption or non-specific binding to the clot activator tube. The CV (precision) and RE (accuracy) for both T3 and T4, across quality control samples (generated from collection tube types (a) and (b)) were within acceptance criteria of ≤20% (25% for the LLOQ) demonstrating that tubes containing clot activator can be used for T3 and T4 sampling.

TH077

Steroid estrogens and estrogenic activity are ubiquitous in dairy farm watersheds regardless of effluent management practices

L.A. Tremblay, Cawthron Institute; J.B. Gadd, NIWA / Department of Chemistry; G. Northcott, Northcott Research Consultants Limited

Steroid estrogens contamination has been linked to adverse effects on exposed aquatic biota. Steroid estrogens are excreted by all mammals and are therefore found in most agricultural wastes including dairy manure and dairy shed effluent (DSE). Some previous studies have demonstrated elevated levels of free and conjugated estrogenic steroids in DSE and this source has increased as New Zealand has experienced rapid expansion and intensification of dairy farming. This research used an approach incorporating analytical chemistry and bioassays to evaluate the levels of estrogenic activity in environmental samples from representative dairy watersheds with differing DSE management practices: either land-applied or discharged to water. The results demonstrated that estrogenic activity and steroid estrogens were prevalent in the waterways within all the studied dairy watersheds. Estrone was the predominant steroid measured in watershed waters because of its presence in dairy cow wastes and as a degradate of the main dairy cow estrogen, 17α-estradiol. Measurable estrogenic activity (17β-estradiol equivalents, EEq) was found at low levels in 83% of the stream samples (highest 1.44 ng L^{-1} EEq) and 75% of the groundwater samples (≤0.15 ng L^{-1} EEq). While estrogenic activity was generally -1, one (of 10) stream with measurable estrone, 17α- and 17ß-estradiol had activity of 1.4 ng L^{-1}, a level potentially harmful to aquatic biota. Comparable steroid estrogen concentrations and estrogenic activity were found whether DSE was spray irrigated on farm paddocks or directly discharged into waterways. This suggests that direct access of cattle to streams, the direct input of DSE into waterways and runoff from land application all require more intervention and effective management.

TH078

Toxic receipt: Why You Should Avoid it?

J. Milic, Institute of Chemistry, Technology & Metallurgy / Department of Chemistry; V. Mart, J. Randjelovic, L. Šojić, ALHem - Safer Chemicals Alternative

Bisphenol A (BPA) is at the moment one of most commercial chemicals at global scale and is used in production of thermal papers as a color developer. BPA is not chemically bound to paper, so in contact with skin, it migrates into the skin, and is absorbed. This chemical is proved to be toxic for fertility, disruptive for endocrine system, it can cause allergic reactions on skin and respiratory irritation, and it can lead to serious eye damage. In December 2016, the European Commission made a decision to ban, i.e. restrict use of bisphenol A in thermal paper, if concentration of the chemical equals or exceeds 0.02 mass %. This decision shall be applied as of 2 January 2020. During 2017, ALHem carried out the campaign **"Toxic receipts"** in order to check the presence of BPA in thermal paper from public and private sectors in Serbia, as well as on paper and plastic packaging for food, comprising 33 samples, out of which: 20 thermal papers (mostly cash receipts), 6 plastic boxes and 7 paper packages for food. The results indicated that all samples of imported thermal paper (rolls) tested in laboratories were positive on the content of BPA. In addition, 87.5% of thermal paper from private sector and 88.9% from public sector contains this chemical. BPA was present in samples in the range of 0.63 and 0.91% (w/w). In this campaign, food packaging was also tested, primarily the one used for packing of fatty food. BAP is soluble in fats so it easily migrates from the packaging into food, which is corroborated by the data that greatest intake of this toxic chemical by humans is by peroral route, i.e. by food. The obtained results indicate that tested plastic boxes for food from supermarkets did not contain BPA. When it comes to paper packaging, French fries bags did not contain BPA, while tested cardboard boxes contained BPA in traces. These cardboard packages are made of recycled paper, so it can be assumed that thermal paper enters the recycling together with other paper waste, thus contaminating final products made of such recycled paper. Taking into account that on European market there are suppliers of thermal paper free of this hazardous substance, ALHem hereby calls upon institutions from state and public sectors, as well as upon companies from private sector, especially upon trade chains, to replace this product with safer alternative free of bisphenol A, so as to contribute to health protection of their workers, especially cashiers, but also of all citizens.

TH079

SETAC Endocrine Disruptor Testing and Risk Assessment Interest Group

H.L. Schoenfuss, St. Cloud State University / Aquatic Toxicology Laboratory

Risk assessment of Nanomaterials: innovative approaches and application of recent research developments to regulatory science (P)

TH080

Evaluate the ecological risk during product development: safe by design case study - Met@Link project

R. Weltens, VITO / ABS; l. geerts, VITO NV / Health; s. verstraelen, VITO / ABS

Safe-by-design requires risk evaluation at critical points during the development of a product to enable a well documented choice for the lowest risk option. In the Met@link project new metal-based inks - enriched with Ag-nanoparticles (Ag-NP) - for printing conductive tags are developed. The environmental risk is assessed to support decissions between different environmental risk options (or risk management options). Environmental risk assessment (ERA) analyses the potential effects on the one hand (concentration effect relations for the target organisms) and evaluates the potential exposure of target organisms on the other hand (i.e. to define the environmental compartments and organisms of concern). Risk management either reduces the potential effects (f.i. redesigning the product) or prevents the predicted exposure (f.i. redesign the production process) to minimize the potential risk. Case study: ERA Ag-NP metal based ink. Potential effects of conceren? Literature data learn that Ag-NP particles are indeed highly toxic to aquatic ecosystems, mainly due to the leaching of Ag+ ions. Potential exposure? Looking at the production process the aquatic ecosystem is of concern because of the waste water generated during production of the inks. Two prototype inks with suitable technical properties were formulated. Both were tested for their leaching potential and for their ecotoxicity to aquatic organisms. Ag-concentrations (ICP-AES) and Ag-NP (SP-ICP-MS) were measured, and the ecotoxic effects on algae (OECD 201) and *Daphnia* (OECD 202) were measured. Results showed that proper coating material helped to prevent leaching of Ag+ and substantially decreased the ecotoxicity of the leaching fraction.

TH081

REACH Substance Evaluation of silver - justification of read-across from ionic silver to nanosilver

K. Arijs, ARCHE; J. Mertens, Precious Metals and Rhenium Consortium c/o EPMF

As part of the REACH Substance Evaluation for silver, new data was required to be generated to further justify the read-across from ionic silver to silver nanoforms: Information on aquatic and soil ecotoxicity of the smallest silver nanoform with the highest specific surface area registered under REACH as compared to ionic silver. The tested nanoform shall be sufficiently characterised; Information on the fate of nanosilver in soil only in case any of the ecotoxicity tests show higher toxicity for nanosilver as compared to ionic silver; Information on the uses for each individual nanoform registered under REACH. An ecotoxicity testing programme was therefore undertaken comparing the effects of nanosilver with silver nitrate using 3 internationally standardised and accepted aquatic ecotoxicity tests: Toxicity to the alga *Pseudokirchneriella subcapitata* (OECD Test Guideline No. 201): nanosilver was less toxic than silver nitrate. Long-term toxicity to *Daphnia magna* (OECD Test Guideline No. 211): nanosilver was less toxic than silver nitrate. Toxicity to soil microorganisms (OECD Test Guideline No. 216) in 3 soils representative for the EU: nanosilver was equally or less toxic than silver nitrate. The silver nanoform was fully characterised (aqueous suspension containing approximately 37% nanoparticles with spheroidal-like shape, mean primary particle size 9.4 nm). The dissolution behaviour of the tested silver nanoform was determined in the test media used in the ecotoxicity tests. The nanosilver dissolution behaviour qualitatively explained the observed toxicity. Since the ecotoxicity testing demonstrated that nanosilver was equally or less toxic than ionic silver, further fate testing in soil was not required. The data collection on the uses of the silver nanoforms covered by the REACH registration dossier showed limited tonnage and use of nanosilver. Furthermore, since nanosilver is transformed to 'bulk' silver during its use, there is limited release to the environment. The generated data show that the read-across of toxicity values from ionic silver to nanosilver as a 'worst case' approach is justified and scientifically defensible.

TH082

Revising REACH technical guidance on information requirements and chemical safety assessment for engineered nanomaterials for aquatic ecotoxicity endpoints - recommendations from the EnvNano project

S.F. Hansen, Technical University of Denmark / DTU Environment; S.N. Sørensen, DTU Environment / DTU Environment; L. Skjolding, DTU / DTU Environment; N.B. Hartmann, Technical University of Denmark (DTU) / DTU Environment; A. Baun, Technical University of Denmark / DTU Environment

The European Chemical Agency (ECHA) is in the process of revising its guidance documents on how to address the challenges of ecotoxicological testing of nanomaterials. In these revisions, outset is taken in the hypothesis that ecotoxicological test methods, developed for soluble chemicals, can be made applicable to nanomaterials. European Research Council project EnvNano - Environmental Effects and Risk Evaluation of Engineered, which ran from 2011-2016, took another outset by assuming that: "The behaviour of nanoparticles in suspension is fundamentally different from that of chemicals in solution. The aim of this paper is to present the findings of the EnvNano project and through these provide the scientific background for specific recommendations on how ECHA guidance could be further improved. Key EnvNano findings such as the need to characterize dispersion and dissolution rates in stock and test media have partially been addressed in the updated guidance. However, it has to be made clear that multiple characterization methods have to be applied to describe state of dispersion and dissolution over time and for various test concentration. More detailed information is called for on the specific characterization methods and techniques available and their pros and cons. Based on findings in EnvNano, we recommend that existing algal tests are supplemented with tests where suspensions of nanomaterials are aged for 1-3 days for nanomaterials that dissolve in testing media. Likewise, for daphnia tests we suggest to supplement with tests where a) exposure is shortened to a 3h pulse exposure in daphnia toxicity tests with environmentally hazardous metal and metal oxide nanomaterials prone to dissolution; and b) food abundance is three to five times higher than normal, respectively. We further suggest that the importance of considering the impact of shading in algal tests is made more detailed in the guidance and that it is specified that determination of uptake, depuration and trophic transfer of nanomaterials for each commercialized functionalization of the nanomaterials is required. Finally, as an outcome of the project a method for assessing the regulatory adequacy of ecotoxicological studies of nanomaterials is proposed.

TH083

Identifying criteria for environmental risk assessment models at different stage-gates of nano-material/product innovation considering requirements of various stakeholders

S.N. Sørensen, DTU Environment / DTU Environment; S.F. Hansen, A. Baun, Technical University of Denmark / DTU Environment; D. Spurgeon, Centre for Ecology & Hydrology; M. Matzke, NERC Centre for Ecology and Hydrology; K. Schirmer, Eawag / Environmental Toxicology; M. Burkard, Eawag Aquatic Water Science / Southern Ocean Persistent Organoic Pollution Program; M. Dal Maso, M. Poikkimaki, Tampere University of Technology / Aerosol Physics; A. Verschoor, RIVM / Centre for Safety of Substances and Products; J.T. Quik, RIVM / DMG; W. Peijnenburg, RIVM / Center for Safety of Substances and Products; H. Wigger, Empa   Swiss Federal Laboratories for Materials Science and Technology; B. Nowack, Empa   Swiss Federal Laboratories for Materials Science and Technology / Technology and Society Lab

The EU H2020 project calibrate aims to establish a state-of-the-art versatile risk governance framework for assessment and management of human and environmental risks of manufactured nanoparticles (MN) and MN-enabled products applicable throughout the innovation process (stage-gates) for these materials and products. Initial efforts have focused on identifying criteria for environmental risk assessment (ERA) models and tools for such governance framework. It was recognized that some criteria are applicable to both environmental and human risk assessment (HRA), and these so-called “overall” criteria were identified through joint efforts of the ERA and HRA working group experts in caLIBRAte. The identified “overall” criteria relate to RA model features and resources needed to use the tools, whereas the criteria specific to ERA models relate to model outcome on hazard, exposure and risks. The identified criteria were listed against the Cooper stage-gates®, thus forming a table in which the importance or relevance of each criterion could be assessed for each of the stage-gates. This was formed into questionnaires with defined response options for each criterion and stage-gate combination, such as picklists and ratings of importance of the criterion. These questionnaires were sent to stakeholders representing regulators, consultants, researchers and industries, who provided their feedback, either by filling the questionnaires or by listing general input on their current RA approaches or needs. Efforts to obtain input from NGOs and insurers remain ongoing. The feedback clearly illustrated different requirements between stakeholder groups. For example, not all use the (same) stage-gate approach or have the same level of expertise for RA. Other criteria were similar or similarly important to most stakeholders. For example, the middle stage-gates are reported as essential for RA issues with regulatory compliance being the main driver. Criteria suggested useful for users by caLIBRAte partners included the use of modeling/estimations and safety-by-design considerations as low cost options to identify “red flags” for hazard and/or exposure at early stage-gates of MN innovation. The criteria and stakeholder feedback generated will be applied to evaluate existing models/tools against, but also to enable the creation of a “System of systems” for RA along stage-gates when developing MN and MN-enabled products, incorporating the needs of different specific user groups.

TH084

Considerations of nanomaterial's environmental fate to support grouping and environmental risk prediction

M. Herrchen, Fraunhofer IME; C. Nickel, Institute of Energy and Envionmental Technology e.V. - IUTA / Air Quality & Sustainable Nanotechnology; K. Hund-Rinke, Fraunhofer IME / Department of Ecotoxicology; D. Kuehnel, UFZ - Helmholtz Centre for Environmental Research; K. Schwirn, German Federal Environment Agency UBA; T. Kuhlbusch, BAuA; C. Asbach, Institute of Energy and Envionmental Technology eV   IUTA

Engineered nanomaterials (ENM) are used in different products with the consequence that they can be released into the environment during their life cycle. Given the large varieties of ENM, the effort for an individual investigation and assessment would be enormous. Therefore grouping of ENM and read across between different materials is a major target for future risk assessment. In this poster we present practicable approaches that can support the discussion on grouping of ENM regarding their environmental fate for a subsequent risk prediction. In our project we focused on the behaviour of the pristine ENM in aquatic and terrestrial compartments. The transformation (chemical transformation and dissolution) and the transport (mobility and agglomeration) of an ENM in the environment was studied. To predict the exposure potential for the environmental compartments both pieces of information were combined to result in a number code (from 1 for low to 3 for high) for a so called “fate bond” which will be included in a matrix of ENM grouped regarding their potential environmental risk. For example, if the transformation via dissolution and chemical transformation is low in the environmental compartment, the transformation potential of the ENM is low. If the mobility is low and the agglomeration potential is high, the transport is also low. Low transformation and low transport means high ENM exposure potential in the considered compartment and leads to a number value of “3” in the fate bond. For simplification, in this project water phase and sediment phase are considered as one compartment (water compartment) and therefore transport and mobility effected by e.g. agglomeration and sedimentation are not needed to be consideredin the presented approach. In contrast, for soil systems the mobility was analysed in detail, as important factor for the exposure concentration. For an environmental risk prediction the fate information (fate bond) is combined with the (ecotoxicological) hazard properties (ecotox bond; presentat at an additional poster) of an ENM. In this poster, the concept to support discussion on grouping and risk prediction will be presented and discussed by using various ENMs as examples. Keywords: transformation, transport, fate grouping *Acknowledgement* - The results are generated in the framework of the project nanoGRAVUR which is funded by the German Federal Ministery for Education and Research (BMBF) under grant no.: 03XP0002

TH085

Matrix to predict possible environmental risk of nanomaterials during use phase

M. Herrchen, Fraunhofer IME; K. Hund-Rinke, Fraunhofer IME / Department of Ecotoxicology; C. Nickel, Institute of Energy and Envionmental Technology e.V. - IUTA / Air Quality & Sustainable Nanotechnology; D. Kuehnel, UFZ - Helmholtz Centre for Environmental Research; K. Schwirn, German Federal Environment Agency UBA; T. Kuhlbusch, BAuA

Grouping of engineered nanomaterials (ENMs) is a strategy in environmental risk assessment that should allow an adequate hazard assessment while reducing the testing effort rneeded for a material-by-material fate and effects testing. We present a practicable matrix that allows to group of ENMs regarding their potential risk to the aquatic and terrestrial environment. This matrix are based on the combination of assumptions regarding release and fate as well as ecotoxicological effect. The assumptions on release are based on a decision tree which combines information on the production volume of the ENM, that portion which is relevant for the considered use, use in closed / open systems, and slow / fast release into the environment. The resulting so called “release bond” classes ENMs to ENMs with low release (release bond =1) to high release (release bond = 3). The release grouping is followed by a detailed description of release scenarios for the considered use. These scenarios allow the identification of the initial environmental compartment into which the ENM is emitted. Furthermore, possible sinks become obvious for which the fate and ecotoxicological effect grouping need to be performed. Basically, surface water, sediment and soil are possible sinks in the environment. To simplify the fate grouping approach, surface water plus sediment are here supposed to be a monophase system. Thus, chemical transformation and dissolution are considered as relevant processes for the aquatic fate grouping. Transformation (chemical and dissolution) and transport (agglomeration and movement) are considered relevant processes for the terrestrial fate grouping. The so called “fate bond” classes ENMs to ENMs with low exposure potential (fate bond = 1) to high exposure potential (fate bond = 3). In a next step, release bond and fate bond are combined to a so called exposure bond (5 groups in total) and subsequently combined with the so called ecotox bond. The latter one is based on information about ecotoxicity of the bulk material, morphology of ENM, and the ion release potential. The combination results in a 5 x 5 risk matrix with 25 possible combinations of exposure and ecotox bonds. These are summarized to three risk groups low, medium and high. The applicability of the approach will be demonstrated by risk grouping of nano-ZnO and nano-TiO_2 used in sunscreen products. Key words: release, fate, exotox bond

TH086

Concepts for nanomaterial categories regarding environmental hazard and for prediction of their environmental risk as well as proof of principle

K. Hund-Rinke, Fraunhofer IME / Department of Ecotoxicology; D. Kuehnel, UFZ - Helmholtz Centre for Environmental Research; C. Nickel, Institute of Energy and Envionmental Technology e.V. - IUTA / Air Quality & Sustainable Nanotechnology; K. Schwirn, German Federal Environment Agency UBA; D. Volker, German Environment Agency; E. van der Zalm, German Federal

Environment Agency UBA
The grouping of engineered nanomaterials (ENMs) is being intensively discussed in order to develop approaches that allow an adequate hazard assessment of ENMs while reducing the testing effort or to rank them regarding their environmental hazard. Two approaches differing in their focus have been developed and evaluated with a set of 25 ENMs. Based on systematic testing using aquatic test designs used in regulatory testing, the physico-chemical (PC) properties, ion release, morphology and reactivity as well as ecotoxicity of the chemical were identified as relevant parameters. The zeta-potential was considered to be less important. Regarding the parameters ecotoxicity of the bulk chemical, solubility and reactivity we decided upon a pragmatic approach with questions which have to be answered "yes" or "no". Approach I (ecotox flow-chart) is characterized by maximum 24 groups where the property morphology is defined by three categories i.e. fibers, small spherical ENMs, others. The ecotoxicity of the ENMs of a specific group is attributed to similar PC-properties thus support discussion on grouping with the final objective of read across. Approach II (ecotox-bond) was developed for risk assessment by using an approach similar to control banding. For risk assessment the hazard information has to be combined with properties influencing environmental fate. For the parameter "morphology" only fibers and small spherical ENMs are considered. In the ecotox-bond every "yes" for an answer gives one point. The points are added together resulting in five groups in a range or band of 1 to 5 which is used for further assessment. The same number of points can be achieved by different properties resulting in groups of ENMs which can differ significantly in their PC-properties. This procedure is considered suitable for the initial prediction of risk. It is more important that the outcome can be combined with environmental fate. Both approaches are considered to be a suitable starting point for further discussions and developments. Besides the definition of threshold values for solubility, fiber morphology and size of small spherical ENMs, further parameters (e.g. attachment of ENMs to algae) have to be explored to improve the consistency of the groups. Regarding the terrestrial ecotoxicity, soil properties seem to reduce the impact of the toxic properties of the ENMs. Currently the prediction of terrestrial toxicity is not satisfactory.

TH087
Forms of released engineered nanomaterials: A systematic assessment in material flow analysis
V. Adam, EMPA Technology & Society Lab / Technology and Society Lab; A. Caballero-Guzman, EMPA / Technology and Society Lab; B. Nowack, Empa Swiss Federal Laboratories for Materials Science and Technology / Technology and Society Lab
The forms in which engineered nanomaterials (ENMs) are released to the environment affect both their fate and toxicity, two parameters essential to risk assessment. Yet, most of current models assessing ENM releases to the environment do not fully account for the transformations that they undergo before release to the environment. This work consists in the development of a method based on current literature, expert elicitation and probabilistic material flow analysis (PMFA) for modelling the proportions of nano-Ag and nano-TiO_2 flowing in dissolved, transformed (particles that have been subjet to chemical transformations), matrix-embedded (ENM released while embedded in a solid matrix), nanoparticulate (non-transformed, not embedded ENM including free, aggregated and agglomerated ENMs) and product-embedded (ENM contained within a whole product, going to solid waste treatment) forms to the environment. Transformations of ENMs in the environment are excluded from the scope of this work. The modelling system includes 10 technical and 3 environmental compartments. The ENMs flow from production, manufacturing and use to wastewater treatment (sewer system, wastewater treatment plant and sludge) and solid waste treatment (landfilling, incineration and recycling) before reaching air, soil or surface water. Each mass flow was described with a probability distribution. The variability of the data obtained in the literature was used to assess the width of these distributions. Nano-Ag is released to surface water and soil mainly in transformed forms (61% and 77%, respectively), while nanoparticulate forms dominate the releases to air (60%). Most transformations occur in water. Nano-TiO_2 presents contrasting results, as most of the releases to air, soil and water are in nanoparticulate forms (80%, 94% and 99%, respectively). The only transformation identified occurs during waste incineration. The quantification of the forms in which ENMs are released constitutes an essential piece of information for the input data to environmental fate modelling. For the first time, a method was developed for a systematic assessment of these released ENM forms. Results show that, especially for nano-Ag, the actual nanoparticulate form represents only a small fraction of the total ENM mass released to the environment, thereby calling for a revision of current exposure levels commonly used.

TH088
Using the SimpleBox4nano tool for predicting the environmental concentration of nanomaterials
J.T. Quik, RIVM / DMG; J.A. Meesters, E.A. Bleeker, J. Slootweg, RIVM / VSP; S. Lofts, NERC Centre for Ecology & Hydrology / Shore Section; W. Peijnenburg, RIVM / Center for Safety of Substances and Products
In environmental risk assessment the risk quotient, predicted environmental concentration (PEC) relative to the predicted no effect concentration (PNEC), is a useful indicator for risk of chemicals. The SimpleBox[1] modelling approach has long been applied in the regulatory framework REACH, as part of EUSES, to calculate PECs. The SimpleBox model was recently extended for use with nanomaterials (SimpleBox4.0-nano), by updating particle specific transport process algorithms and including nanomaterial specific transformation processes, such as agglomeration and dissolution.[2] In this study we show the sensitivity of SimpleBox4.0-nano to the newly added process parameters. This shows that in addition to the dissolution rate and attachment efficiency, also the concentration of natural particles and their size play a role. In order to use SimpleBox4.0-nano we provide guidance on measuring or calculating the relevant input parameters. Furthermore, we indicate the relevance of the different fractions of PECs as calculated by SimpleBox4nano for estimating the risk quotient. 1: www.rivm.nl/simplebox 2: Meesters, J.A.J., et al., *Multimedia Modeling of Engineered Nanoparticles with SimpleBox4nano: Model Definition and Evaluation.* Environmental Science & Technology, 2014. **48**(10): p. 5726-5736.

TH089
Directions of in silico method development to complement the predictive models used in risk assessment of nanomaterials
J.T. Quik, RIVM / DMG; M. Bakker, RIVM / VSP; D. van de Meent, Association of Retired Environmental Scientists ARES / Environmental Science; M. Poikkimaki, M. Dal Maso, Tampere University of Technology / Aerosol Physics; W. Peijnenburg, RIVM / Center for Safety of Substances and Products
There is an increasing need for predictive risk assessment of nanomaterials (NMs) using methods that are rapid, accurate and resource efficient. To fulfill this need, the development and use of more *in silico* methods for estimating the hazard of NMs and NM-related parameters in exposure modelling seems eminent. In order to find the relevant application of new *in silico* methods, we analyze a selection of currently available human and environmental risk assessment models for NMs. This analysis was done by identifying all the NM-related properties used in these models related to three categories of data: 1) Measured hazard or exposure data, 2) Extrinsic NM Properties, e.g. related to the interaction of the NM with the surrounding matrix, and 3) Intrinsic characteristics specific to the NM, matrix or experimental conditions. This analysis is combined with the current state of Quantitative Structure Property Relationships (QSPRs) development, as a specific set of *in silico* methods, to recommend further development of *in silico* methods for predictive risk assessment of NMs. In particular, the use of descriptors related to the interaction between a NM and its surroundings, e.g. the attachment efficiency, is proposed. QSPRs as well as other *in silico* methods are well suited to fill this gap in predictive risk assessment, under the condition that enough data of reliable quality becomes available.

TH090
NanoScreen - Minimizing the risk associated with nanomaterials used in sunscreen at all lifecycle stages
R. Catalano, Aix-Marseille Université; J. Labille, CNRS; D. Slomberg, Aix-Marseille Université; O. Radakovitch, IRSN; M. Zerrad, Institut Fresnel - Aix Marseille Université; S. Robert, CNRS ESPACE
Among cosmetics and personal care products, sunscreen products are of emerging concern regarding both environmental and human healths. While some organic UV blockers have been evidenced to undergo rapid photodegradation, to induce allergene skin reactions due to dermal penetration, or to cause deleterious effects on marine system, the fate of mineral UV blockers is still under consideration from a regulatory perspective. This is largely related to the potential impact of nanotechnology-based products on both environment and human health. The nano-TiO_2 UV-blockers typically used in sunscreen usually consist of rutile nanoparticles coated with a first mineral layer of silica or alumina aimed at blocking the photocatalytic character, and thus passivating the nanomaterial. In addition, the grafting of a second layer of organic coating is aimed at favoring the nanomaterial dispersion in the cream formulation. Once drained from the skin either through bathing activity or everyday usage and cleaning, the nanomaterials contained in the susncreen can be released to the sea shore. Their behavior in this system is largely determined by this industrial coating and by their initial dispersion in the formulation. This project aims to develop the Eco-design of sunscreens through the minimization of risks associated with nanomaterials incorporated into the formulation. The fabrication and end of life steps are mainly considered and studied using the two following approaches. In order to estimate the release of nanomaterials from sunscreen in marine environment and the subsequent bioaccessibility to the living organism, we carried a field campaign on three beaches on the french coast. The titanium concentration was measured in the sea water as a fuction of the number of swimmers. It is fair to say that the higher nanoparticles concentration in the sunscreen, the higher the release factor. In order to decrease the nanoparticles concentration in the sunscream without decreasing the sun protection factor, the filter selection and coating property is a key step. The filter coating determines its dispersion in the cream formulation, and thus the UV ray protection on the skin. In a laboratory approach we aimed to formulate the best filter-cream dispersion, in order to maximise the sun protection factor while maintaining low dose of nanoparticles.

TH091

OECD Test Guidelines and Guidance Documents for Environmental Safety Assessment of Nanomaterials

J. Ahtiainen, Drumsö Ecotox Consultancy; F. von der Kammer, University of Vienna / Department of Environmental Geosciences; M. Gonzalez, Organization for Economic Cooperation and Development; K. Schwirn, German Federal Environment Agency UBA; D. Volker, German Environment Agency

The OECD test guidelines (TGs) for testing chemicals have been widely used for regulatory purposes all over the world since the establishment of the Mutual Acceptance of Data (MAD) principle in 1984. This MAD principle ensures that, if a chemical is tested under the Good Laboratory Practice (GLP) conditions accordingly to an OECD TG, the data should be accepted in all OECD countries. The TGs have been developed, harmonized, internationally validated (round-robin-tests) and adopted by OECD countries to be used for the phys-chem characterisation, fate estimation, and hazard identification for risk assessment of various chemicals. In addition to the TGs, OECD Guidance Documents (GDs) usually guide how to use TGs and how to interpret the results. These GDs do not have to be fully experimentally validated, and hence they are not under MAD, but they are based on the latest published scientific research. But are the existing TGs and the related GDs applicable and adequate for the regulatory testing of nanomaterials? In general, it is accepted that most of the "endpoints" or more precisely measurement variables are applicable also for nanomaterials. However, for some endpoints new TGs are needed. In addition, GDs are needed to give more precise advice on the test performance, e.g. including sample preparation and dosage of the test material, the characterization of the exposure and understanding the results in order to gain regulatory relevant data on nanomaterials. The poster will present the status quo on recent TGs and GDs development for nanomaterials at OECD level with relevance for an adequate environmental safety assessment of nanomaterials. Selected activities on TG/GD development will be presented in detail regarding their objectives, challenges and status. Emphasis will be given to the OECD TG on dispersion stability in simulated environmental media, which was published by OECD in October 2017 and the draft GD on dispersion stability and dissolution rate of nanomaterials, which will support interpretation and utilization of data coming from this TG and a draft TG on dissolution rate which is in preparation. In order to illustrate the effort of TG/GD development the way from the idea for a new TG and new GD to an accepted OECD TG/GD guideline will be presented.

TH092

Applicability of OECD fish bioaccumulation test guideline 305 to nanomaterials

J. Navas, A. Bermejo-Nogales, INIA - National Institute for Agricultural and Food Research and Technology / Environment; D. Hernandez-Moreno, INIA / Environment; F. Torrent, Universidad Politecnica de Madrid / Escuela Superior de Ingenieros de Montes; A. Valdehita, INIA National Institute for Agricultural and Food Research and Technology / Department of Environment; M. Fernandez, E. Conde, I. Rucandio, CIEMAT; M. Fernandez-Cruz, INIA - National Institute for Agricultural and Food Research and Technology / Environment

OECD test guideline (TG) 305 deals with the assessment of chemical compounds bioaccumulation in fish. The last version (October 2012) considers several possibilities for the exposure of fish to chemicals. The first approach, suitable for soluble chemicals, is based on aquatic exposure with two phases: exposure (uptake, usually 28 days) and post-exposure (depuration). A bioconcentration factor (BCF) is calculated both as the ratio of the chemical concentration in/on fish and in the water at steady state (BCF_{ss}), and as a kinetic bioconcentration factor (BCF_k) estimated as the ratio of the rate constants of uptake and depuration assuming first order kinetics. TG 305 also proposes the use of a “Minimised Aqueous Exposure Fish Test” as a simplified approach with the objective of confirming a BCF estimated on the basis of K_{ow} and QSARs. A third approach corresponds to a “Dietary Exposure Bioaccumulation Fish Test” that should be used, for instance, for very low soluble substances that do not allow reaching stable water concentrations. In the uptake phase (normally 7-14 days), fish are fed with feed spiked with the test substance. In the depuration phase (up to 28 - 42 days), fish fed untreated fish feed. This method allows the calculation of a kinetic biomagnification factor (BMF_k). If it is estimated that steady-state was reached in the uptake phase a BMF_{ss} could be calculated. For the study of bioaccumulation of manufactured nanomaterials (MNs) in fish these approaches should cover all possibilities. In a number of cases MNs are highly insoluble and a dietary exposure would be preferred. In this case it is necessary to implement methods ensuring the inclusion of MNs in feed and the total ingestion of the calculated quantities by fish. Similarly, experience should be gained on the applicability of the mathematical approaches to calculate BCF and BMF in the case of MNs. In a series of experiments we have shown that metal MNs (TiO_2, CeO_2, ZnO) can be easily added to fish feed and that they do not pass to water for a period of time long enough for guaranteeing the total ingestion by fish. In addition, bioaccumulation experiments using the dietary exposure approach with these type of metal MNs indicated that the equations used in TG305 for the calculation of BMF are applicable to MNs.
Acknowledgements: National projects AT2011-001 and EG13-074 and EU FP7 project 604387 GUIDEnano.

TH093

A new test method to determine the bioaccumulation of manufactured nanomaterials in filtering organisms (Bivalvia) using the freshwater mussel Corbicula fluminea

S. Kühr, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Bioaccumulation and Animal Metabolism; B. Knopf, Fraunhofer Institute for Molecular Biology and Applied Ecology IME / Environmental Specimen Bank and Elemental Analysis; B. Meisterjahn, Fraunhofer Institute for Molecular Biology and Applied Ecology IME / Ecological chemistry; K. Hund-Rinke, Fraunhofer IME / Department of Ecotoxicology; K. Schwirn, German Federal Environment Agency UBA; D. Volker, German Environment Agency; C. Schlechtriem, Fraunhofer IME / Department Bioaccumulation and Animal Metabolism

The identification and scientific assessment of compounds that bioaccumulate in organisms and biomagnify in food webs play a key role within the PBT-assessment. The bioaccumulation potential of compounds is commonly expressed in form of bioconcentration factors (BCF) determined in flow-through studies with fish according to OECD 305. Comparable studies with manufactured nanomaterials (MNMs) are difficult to carry out due to the lack of suitable test systems that allow a permanent and constant exposure of the compounds. MNMs tend to sediment in water and are supposed to be primarily taken up by benthic species in aquatic ecosystems. Different studies have shown that mussels are able to ingest and to incorporate MNMs suspended in water. However, existing standardised test methods to investigate the bioaccumulation of substances in mussels have been developed and optimized for soluble, non-particulate substances. Therefore, an alternative test concept was developed allowing to investigate the bioaccumulation of MNMs in mussels under flow-through conditions. First studies were carried out with the freshwater mussel *Corbicula fluminea*. By using silver MNMs (NM300K) and silver nitrate we were able to compare the accumulation and elimination of ionic and nanoparticulate silver. Mussels were exposed for a period of 4 - 6 days. In both cases steady state concentrations of total silver in the mussel tissue were reached within 24 hours. The quantification of the total content of silver in water and tissue samples was carried out by ICP-MS or ICP-OES. The determined tissue and water concentrations were used to determine bioaccumulation factors for both test items. In a further study the bioaccumulation of a titanium dioxide nanomaterial (NM 105) was tested. The studies have shown that the new test system is suitable to investigate the bioaccumulation of MNMs.

TH094

Genotoxicity of ZnO nanoparticles. A comparison of methods, tools and mechanisms of action in test experimental models used for human and ecological risk assessment

s. manzo, ENEA / SSPT-PROTER-BES; s. schiavo, ENEA CR; M. Oliviero, University Parthenope; F. Pacchierotti, ENEA; c. arcangeli, ENEA CR; E. Cordelli, ENEA / SSPT-TECS-BIORISC Via Anguillarese, 301, 00123, Rome, Italy; g. leter, ENEA CR

ZnO nanoparticles are considered among the most toxic ones mainly for their capability to dissolve toxic ions. They are largely employed in many productive sectors and primarily in personal care product formulations and then represent a real threat both for humans and for biota. The assessment of the environmental and human health threat posed by engineered nanomaterials (ENM) need a dialogue between toxicologists and ecotoxicologists in order to get a comprehensive understanding of the adverse outcome pathways and to reach a consensus on safe limits for both humans and the environment. Generally, toxicity testing imply the evaluation of multiple endpoints and, especially in ecotoxicological studies, of several test organisms with different biological complexity and representative of the diverse trophic levels. In the case of human health risk assessment, multiple potential target organs are considered, also as a function of exposure routes. DNA damage and consequential genetic alterations are an essential component of toxicity assessment and often showed a very sensitive response to ENM exposure. Moreover, genetic alterations are transmissible to the next generation and can have an impact at the population level. Due to ENM reactive nature and therefore to their peculiar behaviour in the experimental media it is more and more evident the need to investigate the early molecular initiating events triggered by ENM that lead to subsequent DNA damage responses, e.g. oxidative stress and in common events across organisms and cell types. Therefore in the view of a possible unique (eco)toxicological assessment of ENM hazard, here we review computational and experimental methods and tools applied to the evaluation of ZnO nanoparticle (as a prototype of metal oxide ENM) genotoxicity in ecotoxicological test organisms and test systems aimed at human hazard identification. We also compare dose-effect relationships and adverse outcome pathways for possibly drawing a unifying model and for establishing common safe limits.

From detection to action: advancements in assessing and managing highly fluorinated compounds (P)

TH095

Assessment of persulfate oxidation liquid chromatography tandem mass spectrometry for the analysis of perfluoroalkyl and polyfluoroalkyl substances in water

G. Munoz, Universite de Montreal / Chemistry; S. Mejia, McGill University / Civil

Engineering and Applied Mechanics; Y. Yao, K. Volchek, Environment Climate Change Canada / Emergencies Science Technology; S. Vo Duy, S. Sauvé, Universite de Montreal / Chemistry; J. Liu, McGill University / Department of Civil Engineering

The global distribution of certain perfluoroalkyl and polyfluoroalkyl substances (PFASs) in the environment is of concern given their environmental persistence and possible adverse effects. The characterization of the wide variety of PFASs at aqueous film forming foam (AFFF) impacted sites may be compounded by the relatively limited number of certified standards to ensure a rigorous quantification. A possible solution is the implementation of a surrogate approach such as the total oxidizable precursor (TOP) assay, relying on the oxidative conversion of potential perfluoroalkyl acid precursors (Pre-PFAAs) into readily measurable perfluorocarboxylates. However, in order to ensure a fully legitimate comparison between conventional (i.e. before oxidation) and after TOP analyses, a number of critical knowledge gaps remain to be bridged. The two types of water samples (i.e. before TOP *versus* after TOP) might reveal differential instrument matrix effects or necessitate different clean-up strategies, which could *de facto* impact the method reporting limits and preclude a consistent comparison between the two approaches. The present work aimed at assessing the applicability of the TOP assay to various water matrixes through stringent validation. The performance of a workflow involving persulfate oxidation followed by ultrahigh performance liquid chromatography tandem mass spectrometry (TOP-UHPLC-MS/MS) analysis was therefore evaluated using various environmental waters. The validation endpoints ascertained included, notably, the evaluation of oxidation yields in the various matrixes assayed, as well as a careful assessment of the matrix effect that may occur at the instrumental analysis stage. The method was applied to a limited survey of surface water and groundwater samples. It was observed that even though fluorotelomer sulfonates (ΣFTSAs) were the target pre-PFAAs predominantly reported before oxidation in most instances, they could only partially account for the observed ΔPFAA (molar concentration increases upon oxidation). The unexplained ΔPFAA portion likely results from the oxidation of untargeted pre-PFAAs for which oxidation yields are yet to be determined.

TH096

Use of biochars for the sorption of poly- and perfluorinated alkyl substances (PFAS) and heavy metals from contaminated soils

L. Silvani, Norwegian Geotechnical Institute; Y. Zhang, NMBU; G. Okkenhaug, A. Botnen Smebye, g. cornelissen, s.e. hale, Norwegian Geotechnical Institute

The contamination of soil with a mixture of comopunds represents a worlwide environmental issue. Contaminats in soil can leach to groundwater or be transfered to the food chain by crop uptake and affect safety and quality of food resources. Of particular concern is industrially contaminated soils, where high concentration of both organic and inorganic pollutants can be found. In the present study four soils with two different types of pollution (poly- and perfluorinated alkyl substances (PFASs) and heavy metals) and two different total organic carbon contents (high and low), were used. PFASs are a class of compound characterised by hydrophobic, alkylated, fluorine-saturated carbon-chain with a hydrophilic head attached at a terminal end. PFAS have been extensively used by industry and consumers as they are heat, water and oil resistant. In addition, their resistance to chemical, physical and biological degradation renders them persistent. Heavy metal contamination is often a problem in soils from shooting ranges and metals pose a hazard and risk for human health and the ecosystems. The soils used in this work are contaminated with As, Ba, Cd, Cr, Cu, Hg, Mo, Ni, Pb, Sb, Se and Zn. Among the remediation techniques available, adsorption is the most commonly used for both organic and inorganic contaminants removal from soil. Biochar (BC) has a high adsorption potential for organic and inorganic contaminants and can be made at a low cost. Biochar is thus a promising and economic alternative to other carbonaceous materials, such as activated carbon, for this environmental application. In this study, three BCs were used as sorbents: a wood BC (wBC) made from wood chip waste (used for all the treated soils), an iron enriched BC (Fe-BC) (used for the metal contaminated soils) and an activated biochar (aBC) (used for the PFAS contaminated soils). Isotherm batch tests have been carried out using a water and soil mixture (L/S=10) to which BC was added at increasing doses (from 0 to 20%). The aim of this work is to investigate i) whether biochar can be used as a sorbent material for the treatment of industrial contaminated soil, ii) whether BC can sorb PFAS in soils with both high and low TOC contents, iii) if iron enriched BC increases the sorption of metal as compared to non-enriched BC and iv) whether there is a correlation of BC properties (surface area, pores, surface propertie, etc) with sorption.

TH097

Sorption of 14 PFASs to organic soil constituents - the effect of H+, Na+, Ca2+ and Al3+ ions

H.F. de Campos Pereira, Swedish University of Agricultural Science / Department of Soil and Environment; M. Ullberg, Swedish University of Agricultural Sciences / Department of Soil and Environment; D. Berggren Kleja, Swedish University of Agricultural Sciences; J. Gustafsson, Swedish University of Agricultural Sciences / Department of Soil and Environment; L. Ahrens, Swedish University of Agricultural Sciences SLU / Dept. of Aquatic Sciences and Assessment

Environmental risk assessment of perfluoroalkyl substances (PFASs) requires accurate prediction of their sorption in soils. The aim of this study was to investigate sorption of 14 PFASs, including perfluorocarboxylates (PFCAs), perfluorosulfonates (PFSAs) and perfluorooctanesulfonamide (FOSA), to an organic soil horizon and the effect of solution pH and simulated soil organic matter (SOM) net charge as a function of pH and added concentrations of Al^{3+}, Ca^{2+} and Na^+. Generally, the organic C-normalized partitioning coefficients (K_{OC}) were negatively correlated with both pH (–0.32 ± 0.11 log units per unit pH) and the SOM bulk net negative charge (–1.41 ± 0.40 log units per log unit $mol_c\ g^{-1}$). The sorption increased with increasing perfluorocarbon chain length (hydrophobicity) for both PFCAs and PFSAs with 0.60 and 0.83 log units per CF_2 moiety, respectively. Comparing the effect of the PFAS functional head group on sorption, sorption affinity followed the order PFCAs < PFSAs < FOSA. Effects from cation additions on sorption were evident for the C_3 and C_5–C_8 PFCAs and perfluorohexane sulfonate (PFHxS), and for these substances, the SOM bulk net charge was the better sorption predictor as compared to the pH value alone. However, for sorption of the most long-chained substances (i.e. the C_9–C_{11} and C_{13} PFCAs, PFOS and FOSA), cation effects were small and instead sorption was more strongly related to the pH value. This suggests that the most long-chained PFASs have a binding preference towards the highly condensed parts of the humin fraction of SOM, in similarity to other hydrophobic organic compounds, whereas shorter PFASs to a higher degree are bound to humic and fulvic acid where co-sorption of cations gives significant effects. A conceptual model which explains the observed difference in sorption behaviour between shorter and longer PFASs is presented. Progresses made on PFAS binding to organic soil fractions will contribute to more accurate prediction of PFAS sorption in soils and thereby aid in the environmental risk assessment of these chemicals.

TH098

Environmental degradation rates for new PFAS via decarboxylation potential in water, in a MS collision cell and in silico

V. Nikiforov, NILU - Norwegian Institute for Air Research

Straight-chain perfluoroaliphatic carboxylic acids, like PFOA, are extremely stable chemical compounds. In contrast, severel other perfluorinated carboxylic acids are less stable and undergo decarboxylation - spontaneous degradation with loss of carbon dioxide. For instance, perfluorobenzoic acid decomposes slowly in aqueous solution, while perfluoropivalic acid loses CO_2 so fast at room temperature that its spontaneous decomposition is a symthetic method for nonafluoroisobutane. There are indications that novel oxygen-containing analogs of PFOA are lees stable towards decarboxylation. A typical detection method for PFCAs is based on the same decarboxylation process: SRM transition from [M-1] to [M-45]. A collision energy, required for such transition is a measure of intrinsic stabilty of a corrsponding perfluorocarboxylate anion. Relative energies for this transformation can be satisfactorily predicted by DFT calculations at standard RB3LYP/6-31+G(d,p) level. Decarboxylation rates in water for perfluorinated and structurally similar carboxylic acids also correlate well with MS and DFT-derived energies. Thus mass-spectral information and results of simple quantum-chemical modeling can be used as a measure of abiotic degradation potential for per- or polyfluorinated acids in aquatic environment.

TH099

Perfluoroalkylated acids (PFAAs) in soil and invertebrates (Isopoda) near a fluorochemical plant in Flanders, Belgium.

T. Groffen, Systemic Physiological and Ecotoxicological Research (SPHERE), University of Antwerp / Biology; M. Eens, University of Antwerp / Department of Biology; L. Bervoets, University of Antwerp / Department of Biology (SPHERE Research Group)

Perfluoroalkylated acids (PFAAs) have been produced for over five decades. Due to their hydrophobic and lipophobic character they are suitable for a wide range of applications. However, PFAAs may enter the environment, accumulate in wildlife and may cause detrimental effects. The widespread use of PFAAs has resulted in a global presence. Therefore the major global manufacturer, 3M, phased out the production of perfluorooctane sulfonate (PFOS) and perfluorooctanoic acid (PFOA) in 2002. Nevertheless, these compounds are still detected in high concentrations in the environment and biota. Especially the fluorochemical plant has been characterized as a PFAAs hotspot for environmental contamination. In the present study we measured the concentration of 12 PFAAs (8 perfluoroalkyl carboxylic acids (PFCAs) and 4 perfluoroalkyl sulfonic acids (PFSAs) in soil and isopods collected at a fluorochemical plant in Antwerp, Belgium. In addition, samples from four other areas were collected, representing a gradient in distance from the pollution source. We tested for both correlations between soil properties (e.g., total organic carbon (TOC) and PFAAs concentrations in soil, as well as correlations between PFAAs concentrations in soil and invertebrates. In the soil, PFBA, PFOS and PFOA were the only compounds that were detected at all sites. Soil concentrations of all other compounds, with exception of PFDoA and PFBS, were < LOQ in all sites except for the plant site. Median concentrations of 606 ng/g ww for PFOS and 8 ng/g ww for PFOA were detected in soil at the plant site, which are high compared to what has been reported in previous studies conducted in the area. Furthermore, these concentrations decreased significantly with distance from the plant. However, concentrations did not differ between the three locations that were situated farthest away from the plant. No significant differences in TOC were observed among the studied sites, but TOC was positively correlated with multiple PFAAs, including PFOS and PFOA. At this moment (November 2017), isopods have not been tested for PFAAs concentrations yet, but based on the soil concentrations and concentrations detected in previous studies near the fluorochemical plant in Antwerp, we expect high concentrations of multiple PFAAs. The outcome of the present study will be used in further monitoring studies on the effects of soil type on PFAAs bioavailability to invertebrates, as well as effects of PFAAs on multiple biomarkers.

TH100

Occurrence and distribution of legacy per- and polyfluoroalkyl substances (PFASs) and fluorinated alternatives in coastal waters of the German North and Baltic Seas

H. Joerss, Helmholtz-Zentrum Geesthacht / Environmental Chemistry; R. Ebinghaus, Helmholtz-Zentrum Geesthacht / Department for Environmental Chemistry

Long-chain per- and polyfluoroalkyl substances (PFASs) are recognized as global contaminants of high concern as they have been shown to be persistent, bioaccumulative, toxic, and ubiquitously present in the environment. This has led to a number of actions by industry and regulatory authorities aiming at restricting the production, use, and release of long-chain PFASs. Consequently, an industrial shift has been taking place, moving away from long-chain PFASs toward alternative substances, such as per- and polyfluoroether carboxylic and sulfonic acids (PFECAs and PFESAs). Due to structural similarities, the question arises whether the alternatives represent a substantial improvement on their predecessors. Public data on their properties and environmental exposure is still limited. This study aims at investigating occurrence and distribution of legacy PFASs and fluorinated alternatives in surface water samples from coastal areas of the German North and Baltic Seas. In summer 2017, two sampling campaigns were realized using the research vessel *Ludwig Prandtl*, during which 94 water samples were taken along the German coastlines. The analytical method included 26 legacy PFASs and 5 fluorinated alternatives, among them the PFECAs GenX and ADONA. Filtered 1 L water samples were spiked with mass-labelled internal standards (50 µL, 60 pg/µL) and loaded onto preconditioned solid phase extraction cartridges (Waters Oasis WAX; 6cc, 500 mg, 60 µm). After a washing step, the target compounds were eluted using methanol and 0.1 % ammonium hydroxide in methanol. The eluates were reduced to 150 µL under nitrogen and 13C8-PFOA was added as injection standard (10 µL, 100 pg/µL). Instrumental analysis was performed by HPLC-MS/MS, using an Agilent HP 1100 LC system coupled to an AB Sciex API 4000 triple quadrupole mass spectrometer. First results show that the fluorinated alternative GenX can not only be detected in all water samples along the German North Sea coast, but is one of the dominating PFASs with average concentrations of 1.4 ± 0.2 ng/L. Based on these and further results, it will be discussed if regulations on long-chain PFASs and the subsequent ongoing shift to fluorinated alternatives lead to changes in the coastal environment.

TH101

Suspect screening for short chain PFAS in environmental water samples, waste water treatment plants, and building materials

I. Matzenbach, Hochschule Fresenius, University of Applied Sciences / Chemistry and Biology; D. Zahn, Hochschule Fresenius / Chemistry and Biology; T.P. Knepper, Hochschule Fresenius, University of Applied Sciences / Chemistry and Biology; T. Frömel, Hochschule Fresenius, University of Applied Sciences

Perfluoroalkyl and polyfluoroalkyl substances (PFASs) are very persistent anthropogenic fluorinated chemicals that have been detected in remote areas and all compartments of the environment. Historically perfluorooctanoic acid (PFOA) and perfluorooctane sulfonate (PFOS) are the two most frequently used and most well studied PFASs. As a consequence of the bioaccumulative and toxic properties of long chain PFASs, their voluntary phase-out started in the year 2000 and led to an increased prevalence of short chain homologues (C_4 to C_6) in the aquatic environment. Short and ultra-short (>C_4) chain PFASs are quickly eliminated from organisms and thus do not bioaccumulate. However, they are more mobile in the water cycle then their long chain homologues, thus exhibiting higher tendencies to reach raw and drinking water, and are expected to accumulate in the edible parts of plants, which may lead to an increased exposure through drinking water and vegetable consumption. While perfuorobutanoic acid (PFBA) and perfluorobutane sulfonate (PFBS) have been extensively studied information about ultra-short chain PFAS is still scarce and, if available, limited to perfluorocarboxylic acids (PFCAs) and perfluorosulfonic acids (PFSAs). Trifluoroacetic acid has been detected in concentrations in excess of 20 µg/L in tap water, while perfluoropropane sulfonate (PFPrS) and perfluoroethane sulfonate (PFEtS) have been detected in a study of tap water samples from China, Japan, India, the United States of America, and Canada. In 2016, the first C_1-homologue of a legacy PFAS class was detected in the form of trifluoromethane sulfonic acid (TFMSA), which was present in various compartments of the water cycle ranging from waste water treatment plant effluents to finished drinking water. Information about ultra-short chain homologues of other PFASs like perfluoroalkyl phosphonic acids (PFPAs), perfluoroalkane sulfonamides (FASAs), perfluoroalkane sulfonamidoethanols (FASEs), and perfluoroalkane sulfonamidoacetic acids (FASAAs) are to the best of our knowledge not available so far. In an attempt to close this gap in knowledge, we performed a suspect screening for (ultra-)short chain PFASs of several substance classes in environmental water samples, waste water treatment plants and building materials.

TH102

Utilization of passive samplers to detect poly- and perfluoroalkyl substances (PFASs) in wastewater treatment plants and estuarine environments

C. Gardiner, Graduate School of Oceanography, University of Rhode Island / Chemical Oceanography; A.R. Robuck, Graduate School of Oceanography, University of Rhode Island, Narragansett, Rhode Island 02882 / Chemical Oceanography; M.G. Cantwell, D.R. Katz, Office of Research and Development, US Environmental Protection Agency, Narragansett, Rhode Island 02882 / Atlantic Ecology Division; J. Bečanová, R. Lohmann, Graduate School of Oceanography, University of Rhode Island, Narragansett, Rhode Island 02882 / Chemical Oceanography

Poly- and perfluoroalkyl substances (PFASs) are of growing concern worldwide, due to the linkage of these compounds to adverse effects in humans and the environment. Surface waters in the northeastern United States in particular have displayed elevated concentrations of PFASs. Here we utilize passive samplers to gain a better understanding of the sources and spread of these contaminants. Thirty-two microporous polyethylene (PE) passive samplers (containing Hydrophilic-Lipophilic-Balanced sorbent) were deployed across nine sites in Narragansett Bay (RI, USA) in the fall of 2017 for a one month duration each. Deployment sites ranged from wastewater treatment plant and industrial outfall, military and fire training bases, and more pristine areas. 25 PFASs (including sulfonates, carboxylic acids, and GenX) were measured across all sites in the passive samplers, as well as water and sediment samples. For a more direct point source evaluation, 10 additional samplers were deployed in two waste water treatment plants of a large urban area. By analyzing the spatial and temporal trends of these fluorinated compounds we plan to assess their longevity in water and sediment of the Bay. Lastly, we aim to understand and predict potential effects on the environment and better advise on regulatory practices.

TH103

Distribution of per and polyfluoroalkyl substances in sediments of the Spanish coast

E. Concha-Graña, Universidade da Coruña / Grupo Química Analítica Aplicada (QANAP); P. López-Mahía, Universidade da Coruña / Analytical Chemistry Department; D. Prada-Rodríguez, Universidade da Coruña / Grupo Química Analítica Aplicada (QANAP); L. Viñas, Instituto Español de Oceanografia / Centro Oceanográfico de Vigo; J.A. Campillo, Instituto Español de Oceanografia / Centro Oceanográfico de Murcia; S. Muniategui, Universidade da Coruña / Analytical Chemistry

Per- and polyfluoroalkyl substances (PFASs) configuration, consisting in an alkylated hydrophobic chain fully or partially fluorinated, hydrophilic group terminated, provides to PFASs simultaneous hydrophobicity and lipophobicity. Their persistence, bioaccumulation and toxicity make them a source of increasing environmental and public health concern. Presence of PFASs in sea environment is caused by discharges of wastewater effluents or river flows, urban runoff, atmospheric deposition of volatile precursors and subsequent transformation, or direct application of fire-fighting foams containing PFASs, among others. Samples were collected in two semiconfined coastal areas, one of them an area with high industrial and port activities (Ría de Vigo) and the other one with high touristic and agricultural activity (Mar Menor). PFOA, PFOS, PFOSA, n-MeFOSA and n-EtFOSA were extracted from sediments by sonication, cleaned up by dispersive solid phase extraction and the analyzed by LC-LTQ-Orbitrap-HRMS in full mode (Concha-Graña E. et al, 2017). This is the first time that these compounds were measured in these areas. N-MeFOSA and N-EtFOSA were not detected in any sample, whereas PFOSA was only detected in two samples, but below the quantitation limit. PFOS was measured in 39 % of samples, most of them from Mar Menor. In Ría de Vigo PFOS was detected in a point close to a ceramic factory. Regarding PFOA, this compound was measured at level higher than quantitation limit in 2 Mar Menor samples. Sum of PFASs in each sampling point was below 0.4 ng/g in Mar Menor and below 0.1 ng/g in Ría de Vigo, being the total concentration of PFOS similar than the detected in similar areas. Some characteristic of the sediments were taking into account in order to find the correlation between these parameters and the obtained data. Moreover, the environmental risk was evaluated. Acknowledgements: Financial support by the Program of Consolidation and Structuring of Units of Competitive Investigation of the University System of Galicia (Xunta de Galicia) (reference: ED431C 2017/28) potentially co-financed by ERDF, and by the Ministry of Economy and Competitiveness (IMPACTA, project reference: CTM2013-48194-C3-1-R/2-R, and ARPA-ACUA, project reference: CTM2016-77945-C3-3-R). References: Concha-Graña E. et al, VIII Reunión de la Sociedad Española de Espectrometría de masas, V Reunión Nacional de Dioxinas, Furanos y Compuestos Orgánicos Persistentes Relacionados (2017)

TH104

Utilization of Polyethylene Passive Samplers to Detect volatile PFAS precursors in water and air

E. Dixon-Anderson, R. Lohmann, University of Rhode Island / Graduate School of

Oceanography

Fluorotelomer alcohols (FTOHs) and other poly- and per-fluorinated alkyl substances (PFASs) are common and ubiquitous by-products of various industrial telomerization processes. They can degrade into various perfluorinated carboxylic acids (PFCAs) including perfluorooctanoic acid (PFOA) and perfluorooctanesulfonic acid (PFOS), which are persistent organic contaminants of concern. This study assessed the use of polyethylene passive samplers as a sampling tool for volatile PFAS precursors coupled to their analysis via gas chromatography-mass spectrometry (GC/MS). Parallel active and passive sampling was performed in ambient air in Providence (RI, USA) in April 2016. Atmospheric concentrations were dominated by FTOHs (average 9.9 – 16 pg/m^3), with traces of other volatile PFASs also present. Polyethylene-air partitioning constants, log K_{PEA}, were derived. A deployment at a Waste Water Treatment Plant (WWTP) was also performed in 2016, and sampling rates derived from the loss of performance reference compounds. Best-fit curves were used to determine polyethylene-water partitioning constants, log K_{PEw}, during the 3-week uptake experiments. Derived log K_{Pew} values for 6:2, 8:2 and 10:2 FTOHs were 3.8, 4.4 and 4.8, respectively. For MeFOSE and EtFOSE, derived log K_{Pew} values were 4.0 and 4.4, respectively. Based on these partitioning constants, aqueous concentrations in the effluents were below 1 ng/L for the FTOHs, MeFOSE and EtFOSE.

TH105

Occurrence and Removal of perfluoroalkyl and polyfluoroalkyl substances (PFASs) in full-scale water and wastewater treatment plants

H. Chen, National University of Singapore; K. Gin, National University of Singapore / Civil & Environmental Engineering

Perfluoroalkyl and polyfluoroalkyl substances (PFASs) comprise a group of compounds that are widely used in the markets for stain repellents, polishes, paints and coatings. In recent years, the occurrence of PFASs in the environment has been recognized as emerging environmental problem due to their persistence, bioaccumulation potential and possible adverse effects on living organisms. Unlike most other persistent and bioaccumulative orgnanic pollutants, PFASs are water soluble. Therefore, removal of PFASs by water treatment processes could be a challenge. The objective of this study was to evaluate the ability of different water treatment techniques to remove PFASs from water. In this study, three full-scale water treatment plants were investigated during a one-year monthly sampling for the removal of 31 PFASs, including 20 perfluoroalkyl acids (PFAAs) and 11 PFAA precursors. The treatment processes include conventional activated sludge system (CAS) and membrane bioreactor (MBR) system in plant 1, sand filtration (SF) and microfiltration (MF) in plant 2, and microfiltration, reverse osmosis (RO), ultraviolet disinfection (UV) in plant 3. Short-chain PFASs (e.g. PFBA, PFPeA and PFHxA) are presented at relatively high concentrations (several hundred ng/L) in the influent. Total PFASs concentrations (∑PFASs) were highest in Plant 1 (227 – 1,279 ng/L), followed by Plant 3 (174 – 215 ng/L) and Plant 2 (61 – 109 ng/L). Total PFASs concentrations in the treated water were 119 – 483 ng/L, 50 – 127 ng/L and 0.8 – 3.1 ng/L in Plant 1, 2 and 3, respectively. Results showed that RO is the only efficient process for removal of PFASs (>98%) for both short-chain and long-chain PFAAs. Both CAS and MBR system have limited removal efficiency (< 50%) for PFAAs. In some cases, the effluent concentrations of PFAAs were even higher than the influent, suggesting potential degradation of PFAA precursors. The biodegradation of PFAA precursors also leads to the higher removal of some PFAA precursors. Considering the low removal of PFAAs in most of the treatment processes, further research is needed to improve the efficacy and efficiency of their removal.

TH106

Perfluorooctane sulfonate (PFOS) and perfluorooctanoic acid (PFOA) occurrence in biota in Czech rivers

V. Kodes, D. Leontovycova, Czech Hydrometeorological Institute / Section of water quality; R. Grabic, University of South Bohemia in Ceske Budejovice / South Bohemian Research Center of Aquaculture and Biodiversity of Hydrocenoses

Objective of the Study The study compares PFOS and PFOA concentrations detected in biota within CHMI bioaccumulation monitoring program from years 2010 – 2016. Material and Methods A bioaccumulation monitoring of selected pollutants in biota is a part of surveillance monitoring of surface waters in the Czech Republic. Monitoring comprises two profile sets containing 21 and 22 monitoring sites. Sampling at those two site sets alternates in the three-year cycles. Sites are located at important parts of main Czech rivers (country borders, before confluences, downstream industrial sites or large cities, etc.). An assessment was made for following matrices: juvenile fish, benthos (*Hydropsyche sp., Erpobdella sp., Gammarus sp.*), mussels (*Dreissena polymorpha*) and fish (*Leuciscus cephalus*). The analyses of fish were conducted for following tissues: muscle, blood and liver. In total, following number of samples of various matrices were analysed using LC-MS/MS and LC-HRMS: fish blood 105, fish liver 15, fish muscle 78, juvenile fish 149, benthic organisms 126, mussels 73. Results PFOS highest values were detected in fish blood (10 - 3030 μg.l^{-1}). Wet weight concentrations in fish liver (0,5 - 317 μg.kg^{-1}), juvenile fish (1,2 - 312 μg.kg^{-1}) and benthic organisms (0,05 do 61 μg.kg^{-1}) significantly exceeded levels of PFOS found in fish muscle (0,4 do 38 μg.kg^{-1}). The lowest PFOS concentrations were found in mussels (0,01 - 2 μg.kg^{-1}). PFOA concentrations compare to PFOS reached significantly lower levels in all monitored matrices. Range of values was between 0,01 - 3,1 μg.kg^{-1} , where minimum represents the smallest concentration found in mussels and the maximum represents concentrations in juvenile fish. PFOA highest values were detected in juvenile fish (0,01 - 3,1 μg.kg^{-1}), followed by benthic organisms (0,02 - 2,5 μg.kg^{-1}) and fish blood (0,06 - 1,8 μg.l^{-1}). Small concentrations were found in mussels (0,01 - 1 μg.kg^{-1}), fish muscle (0,02 - 0,5 μg.kg^{-1}) and fish liver (0,02 - 0,07 μg.kg^{-1}). Conclusion In general the highest PFOS concentrations were found in fish, except fish muscle as expected. PFOS concentrations in fish blood, kidney and liver were higher than concentrations found in muscle tissues due to its binding to proteins in a blood and a liver. All collected fish blood samples and more than 50% of collected samples of fish liver and juvenile fish exceeded EQS for PFOS (9,1 μg.kg^{-1}).

TH107

Analytical strategy to study the distribution of perfluoroalkyl substances in fish tissue of Italian deep subalpine lakes

M. Mazzoni, University of Insubria, DISTA / Water Research Institute; R. Bettinetti, University of Insubria / DiSTA; A. Buffo, F. Cappelli, Water Research Institute - Italian National Research Council IRSA-CNR; B. De Felice, M. Parolini, University of Milan / Department of Environmental Science and Policy; S. Polesello, Water Research Institute- CNR / Water Research Institute; S. Valsecchi, Water Research Institute - Italian National Research Council IRSA-CNR

Perfluoroalkyl substances, such as perfluorinated sulfonic acids (PFSAs) and perfluorinated carboxylic acids (PFCAs), are ubiquitous contaminants in the aquatic environment, including wildlife and humans. Perfluoroalkyl acids bind to proteins, resulting in bioaccumulation behaviour differently from that of lipophilic substances. Therefore, conventional methods (fish fillet analysis, lipid normalization, etc.) to monitor their concentrations in aquatic biota cannot be used as such to assess the bioaccumulation and biomagnification of PFCA and PFSA. In this study, conventional monitoring approaches and new strategies are compared to assess the best methodology to be implemented in biota monitoring plans for these compounds. Several fish specimens from Italian southern alpine deep lakes were seasonally collected for the analysis of 10 perfluorocarboxylates, 7 perfluorosulfonates and 5 perfluorosulphonamides. Individual fish were measured, weighed and dissected in three fractions: whole viscera, the muscle and the rest of the carcass (head, fishbone, skin and fins). The fractions of six fish were analysed separately or pooled in one or two samples for the subsequent analysis. The dry weight, the lipid and the protein content were measured in each fish fraction (muscle, viscera and the rest of carcass). PFAS analysis were carried out with fresh samples but some samples of fillet were also freeze-dried in order to compare the concentrations. Extraction of the animal tissues (2-5 g) was performed by sonication with ACN/H_2O mixture enhanced by salting out and acidification; extracts were purified on HybridSPE-Phospholipid to remove matrix suppression effects by phospholipids. Perfluoroalkyl compounds were determined by liquid chromatography tandem mass spectrometry (HPLC-MS/MS) coupled to an on-line turbulent flow chromatography (TFC) for on-line purification of the extracts. PFAS concentrations in lyophilised samples (expressed on fresh weight basis) are lower than ones determined in fresh samples probably due to evaporation of analytes. The viscera and the rest of the carcass (head, fishbone, skin and fins) showed similar concentrations while the fillet concentrations were from 4 to 8 fold lower than the concentrations of the other two fractions of the fish. Fish fillet analyses proved to be an underestimation of the total body concentrations, which are from 2- to 5-fold higher.

TH108

Potential contribution of targeted and unknown precursors to the apparent biomagnification of perfluoroalkyl acids (PFAA) in the food web of an urban river

C. Simonnet-Laprade, University of Bordeaux UMR EPOC; H. Budzinski, University of Bordeaux; K. Maciejewski, UMR EPOC; K. Le Menach, UMR CNRS EPOC Universite Bordeaux / EPOC UMR 5805; R. Santos, Hepia University of Applied Sciences Western Switzerland; F. Alliot, A. Goutte, EPHE / UMR METIS; P. Labadie, UMR CNRS EPOC Universite Bordeaux / UMR 5805 EPOC

This study assessed the potential contribution of targeted and unknown fluoroalkyl organofluorines to the apparent biomagnification of perfluroalkyl acids (PFAAs) in the trophic web of the urban river Orge (near Paris, France). A total of 16 PFAAs and 10 of their potential precursors (pre-PFAAs) including 4 perfluorooctane sulfonamide derivatives, 4 fluorotelomer sulfonates (FTSAs) and 2 polyfluoroalkyl phosphate (diPAPs) were analyzed in water, sediments and biota samples including invertebrate and fish species. PFASs were ubiquitous in all compartments and 22 compounds were detected in biota. Mean ∑PFASs in biota ranged between 2.0 and 147 ng.g^{-1} wet weight and PFOS, PFDoDA and PFTrDA were the dominant compounds. Pre-PFAAs such as 6:2, 8:2 and 10:2 FTSA, as well as FOSA, N-MeFOSAA, N-EtFOSAA and 6:2 diPAP were also frequently detected (60–100 %) and the sum of these compounds contributed to 1–18 % of ∑PFASs. Trophic magnification factors (TMFs) were estimated using a generalized linear mixed-effect model and were > 1 for C_9-C_{14} PFCAs and C_7-C_{10} PFSAs, as well as several pre-PFAAs such as 8:2 and 10:2 FTSAs, FOSA, N-MeFOSAA, N-EtFOSAA and 6:2 diPAP. However, a significant decrease in pre-PFCAs/PFCAs concentration ratios with trophic level suggested a possible contribution of precursors to the apparent biomagnification of PFCAs, via their biotransformation. In addition, the Total Oxidisable Precursor (TOP) assay was applied to sediments and, for the first time, to biota samples. Results revealed the presence of large proportions of unknown pre-PFAAs in sediments/biofilm/leaf litter samples (64-80 % of total PFAS molar concentration); this proportion was lower in invertebrates (28-54 %) and in fish (15-26 %). These results suggests either the biotransformation of precursors in benthic invertebrates and fish or the limited bioaccessibility of unidentified sediment-bound pre-PFAAs.

TH109

PFAS and their precursors in the Environment. First indications from a large scale environmental monitoring study

M. Kotthoff, Fraunhofer IME / Environmental and Food Analysis; M.W. Bücking, Fraunhofer IME   Institute for Molecular Biology and Applied Ecology / Environmental and Food Analysis; H. Ruedel, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Department of Environmental Monitoring; J. Koschorreck, Umweltbundesamt

Per- and Polyfluorinated Substances (PFAS) have been an ongoing challenge for the environmental sciences for decades. However, the substance versatility, in terms of chemical classes and physico-chemical characteristics yet hinders a full overview of the spectrum. Due to the differential mobility and degradation pathways, the environmental distribution of individual species is complex and requires massive analytical effort. This obscure situation is even stretched by new molecules from international markets, that already travel around the world as industrial substance or as ingredient of commercial applications. Our study set out to apply two large scale multi methods capturing short (e.g. C2 to C6 PFAA), medium and long chain PFAS (e.g. C6 to C14 PFAA and PFSA), and also precursors (e.g. PAPs, diPAPs, FTS, NaDONA) and novel molecules (e.g. F-53B constituents) on samples of the German Environmental Specimen Bank. Samples include rain samples, suspended particulate matter samples, fish liver, mussels, tree leaves and needles, deer liver, earthworm and herring gull eggs. Here, we present first detections of the F-53B constituents in bream liver samples afar from production sites, and provide indications on distribution patterns.

TH110

A physiologically based toxicokinetic (PBTK) model describing the bioaccumulation of two perfluorinated substances in rainbow trout (Onchorynchus mykiss)

A. Vidal, Irstea Lyon; R. Beaudouin, INERIS / Models for Ecotoxicology and Toxicology METO; E. Vulliet, CNRS / TRACES Team; E. Rochard, Irstea Bordeaux / UR EABX; J. Garric, Irstea Lyon / UR RIVERLY   Laboratoire Ecotoxicologie; M.P. Babut, Irstea / Water

Per- and poly-fluorinated substances (PFASs) are ubiquitous in the environment, specifically in aquatic systems. While several PFASs are acknowledged to be bioaccumulated by vertebrate species, including fish, their absorption, distribution, metabolisation and elimination (ADME) remain incompletely understood yet. The aim of this study is to develop a physiologically based toxicokinetic (PBTK) model in order to describe the mechanisms in rainbow trout (*Oncorhynchus mykiss*) exposed through the diet to two selected PFASs, namely perfluorooctane sulfonic acid - PFOS - and perfluorohexane sulfonic acid – PFHxS. Here PFOS is considered as a model compound, as it remains the dominant PFAS in environmental matrices. PFHxS represents an industrial alternative to PFOS, since its addition to Annex B of the Stockholm convention in 2009. Dietary exposure experiments were performed on adult rainbow trouts at two water temperatures (7°C and 11°C). 200 fish were fed daily using pellets spiked with a mixture of PFOS and PFHxS during several weeks. Then, fish were allowed to depurate, in the same tanks, where non contaminated food was supplied daily. During both phases, 5 randomly selected fish were periodically sacrificed. Organs and blood were sampled, weighted, and prepared for PFOS and PFHxS analysis (UPLC-MS/MS). Data from these two dietary experiments, as well as this obtained from the literature, were used for the model calibration. The present PBTK model comprises seven compartments: arterial and venous blood, liver, viscera, brain, kidney, and muscle. Uptake is considered to occur exclusively by diet and no biotransformation of the selected PFASs is considered, since PFOS and PFHxS are final products of precursor degradation. Also, the model took into account growth of individuals and temperature variation, both variables which may influence ADME processes in fish.

TH111

Does water temperature influence the toxicokinetics of perfluorinated substances? Comparison of two dietary experiments in rainbow trout (Oncorhynchus mykiss)

A. Vidal, Irstea Lyon; L. Espinat, INRA; C. Gesset, P. Chèvre, Irstea / UR EABX; F. Lafay, Institut des Sciences Analytiques / TRACES; E. Rochard, Irstea Bordeaux / UR EABX; J. Garric, Irstea Lyon / UR RIVERLY   Laboratoire Ecotoxicologie; M.P. Babut, Irstea / Water

Per- and poly-fluorinated substances (PFASs) are widely found in fresh and marine water environments and accumulate in aquatic organisms. Fish are poïkilotherms and subject to large seasonal changes of temperature, which control physiological processes such as feeding, respiration, fecal egestion, and ultimately growth. Absorption, metabolisation, distribution and elimination (ADME) are concerned as well. For accurate predictions of organic contaminants bioccumulation it is therefore important to take into account temperature variations. Also, to our knowledge, no study on the effect of the temperature on the ADME of PFASs in fish has been carried out yet. The aim of this work is to determine to which extent temperature affects absorption and elimination rates, and distribution within the fish of two perfluorinated acid compounds, namely perfluorooctane sulfonate (PFOS) and perfluorohexane sulfonate (PFHxS). Here PFOS is considered as a model compound, as it remains the dominant PFAS in environmental matrices. PFHxS represents an industrial alternative to PFOS, since its addition to Annex B of the Stockholm convention in 2009. Two dietary exposure experiments were performed on adult rainbow trouts (*O. mykiss*) at two water temperatures (7°C and 11°C). 200 fish were fed daily using pellets spiked with a mixture of PFOS and PFHxS during several weeks. Then, fish were allowed to depurate, in the same tanks, where non contaminated food was supplied daily. During both phases, 5 randomly selected fish were periodically sacrificed for the analysis of the selected PFASs in muscle, liver and blood. Compound-specific tissue distribution, uptake and elimination rate constants in blood were obtained by a simultaneous adjustment to experimental data. Half-lives were estimated for both compounds, in blood, at both conditions. Globally, fish acclimated to the warmer temperature showed faster absorption and elimination rates of PFOS and PFHxS, and their distribution differed between organs, suggesting that temperature represents an important factor in the toxicokinetic profile of PFASs.

TH112

Toxicokinetics of perfluorinated alkyl acids in zebrafish embryo

C. Vogs, Karolinska Institutet; G. Johanson, Karolinska Institutet / Institute of Environmental Medicine; M. Näslund, S. Wulff, Karolinska Institutet / Institute of Environmental Medicine IMM; M. Sjödin, M. Hellstrandh, J. Lindberg, E. Vincent, Swedish Toxicology Sciences Research Center

Perfluorinated alkyl acids (PFAAs) are widely distributed and have been detected e.g. in humans, wildlife and numerous other environmental matrices. These surfactants are highly bioaccumulative as well as persistent and have been associated with several health effects including hepatotoxicity, immunotoxicity and developmental toxicity. The chemical structure of PFAAs mainly differ in two ways: the length of the hydrophobic alkyl chain and the hydrophilic end groups. Little or nothing is known how the structure affects the toxicokinetics (TK) (uptake, distribution, biotransformation, elimination) and, consequently, the toxic effects in different organisms. We therefore studied the TK of four PFAAs; perfluorooctanesulfonic acid (PFOS), perfluorooctanoic acid (PFOA), perfluorohexane sulfonate (PFHxS) and perfluorobutanoic acid (PFBA) in the growing zebrafish (*Danio rerio*) embryo (ZFE). ZFEs are increasingly used as alternative test model for toxicity testing. We exposed ZFEs at three concentrations of each PFAA up to 120 hours post fertilization (hpf). The test concentrations were selected from pilot studies at which the highest would cause developmental effects in less than 20% of the embryos. Exposure medium and ZFEs were sampled separately at nine time points. Water samples from chemical controls without ZFEs present were taken in parallel. Mass-labelled internal standards specific for each PFAA were added prior to further sample treatment and analysis by LC-MS/MS. The exposure concentrations in the TK experiments differed by four orders of magnitude (PFOS< PFHxS< PFOA< PFBA). Chemical control concentrations remained constant until 120 hpf for all PFAAs tested, ruling out unspecific loss due to adsorption to glass. The time courses of the internal concentrations in ZFE indicated biphasic uptake kinetics with slow uptake before hatching compared to a faster uptake after hatching. Apparent steady-state concentrations were reached at 96 hpf for PFOS, PFOA and PFHxS, while PFBA did not reach steady-state within 120 hpf. Moreover, PFOS and PFHxS (sulfonic acid end group) showed a higher bioconcentration than PFOA and PFBA (carboxylic acid end group). In conclusion, these data indicate that the functional group of PFAAs, in addition to the alkyl chain length, may have an important influence on the toxicokinetic processes.

TH113

Role of bioaccumulation in the derivation of environmental risk limits for two perfluorinated substances, PFOA and HFPO-DA

E. Verbruggen, RIVM Expertise Centre for Substance / Centre for Safety of Substances and Products; E. Smit, RIVM / Centre for Safety of Substances and Products; P. Wassenaar, National Institute for Public Health and the Environment (RIVM)

Environmental risk limits (ERLs) were derived in the Netherlands for the substances perfluoro octanoic acid (PFOA) and hexafluoropropylene oxide dimer acid (HFPO-DA; also referred to as GenX, FRD-902 or PFPrOPrA). These ERLs serve as advisory values according to the guidance under the Water Framework Directive to set environmental quality standards (EQS) in Dutch policy. For these two PFAS substances, the assessment of the bioaccumulation potential is a key issue in the derivation of the ERLs. The most critical receptors are humans and wildlife, which are not only exposed directly via drinking water, but also obtain a significant part of the total exposure indirectly through their diets. For this purpose, bioaccumulation through the (aquatic and terrestrial) food chains has to be evaluated. This information is amply available for PFOA, but is very scarce for HFPO-DA. For PFOA, a typical bioaccumulation behaviour has been observed.

The bioaccumulation factors in the aquatic environment appeared to be dependent on the exposure concentration. For both the terrestrial and aquatic food chain, the specific protein-binding behaviour of PFOA requires different methods for normalisation of the concentration values, than those normally applied to hydrophobic substances, i.e. based on lipid and organic carbon. Not only exposure via food, but also the human toxicological threshold value of PFOA itself is dependent on bioaccumulation. Higher safety factors are needed because of the difference in toxicokinetic half-live between humans and laboratory animals, like rats and mice. For HFPO-DA kinetic data are only limited, which hampers the derivation of a human-toxicological threshold. It is further investigated based on the available data for both substances whether these findings for PFOA can be extrapolated to HFPO-DA, taking into account the structural differences between both compounds. Additional experimental bioaccumulation data for HFPO-DA is probably needed to complete the ERL derivation.

TH114

Perfluoroether carboxylic acids - are these substances appropriate PFOA-alternatives regarding their environmental concerns?

C. Staude, German Environment Agency / Chemicals; A. Biegel-Engler, German Environment Agency - UBA / Chemicals; W. Drost, Federal Environment Agency (UBA) / Chemicals; L. Vierke, German Environment Agency / Chemicals

Perfluorooctanoic acid (PFOA) is a persistent, bioaccumulative and toxic substance. To minimize the exposure of humans and environment a restriction according to REACH will come into force in the EU in 2020. For a global regulation, the substance is proposed as a Persistent Organic Pollutant (POP) under the Stockholm Convention. As a result of the regulatory activities as well as voluntary measures, PFOA has been replaced with other fluorinated as well as non-fluorinated alternatives. The use of PFOA as processing aid in fluoropolymer production has been mainly substituted with perfluoroether carboxylic acids (PFECAs). PFECAs are structurally similar to perfluoroalkyl carboxylic acids such as PFOA. The difference is a perfluoroether chain instead of a perfluoroalkyl chain. Due to this structural similarity it could be expected that PFECAs are equally hazardous to the environment. Thus, the German Environment Agency has assessed the environmental hazards in the context of substance evaluations under REACH for certain PFECAs such as ADONA (ammonium 2,2,3 trifluor-3-(1,1,2,2,3,3-hexafluoro-3-trifluormethoxypropoxy), propionate) and GenX (ammonium 2,3,3,3-tetrafluoro-2-(heptafluoropropoxy) propanoate). The poster will present a summary of the substance evaluations. PFECAs are expected to be very persistent under environmental conditions. The substances have a low bioaccumulation potential in aqueous organisms. However, just as PFOA, PFECAs may not fit into the common accumulation pattern. Furthermore, the substances are probably mobile in the aqueous environment and soil and can reach groundwater and consequently drinking water resources. PFECAs have already been detected in surface water, groundwater and drinking water around fluoropolymer production plants [1-4]. In conclusion, further data are necessary, but the available information on PFECAs already demonstrates that these substances are hazardous for the environment and further risk management measures are needed. [1] Gebbink WA, van Asseldonk L, van Leeuwen SPJ. 2017. Environ. Sci. Technol. 51: 11057-11065 [2] Sun M, Arevalo E, Strynar M, Lindstrom A, Richardson M, Kearns B, Pickett A, Smith C, Knappe DRU. 2016. Envrion. Sci. Technol. Lett. 3: 415-419 [3] Schreiber J. 2014. Untersuchung des Transportverhaltens von ADONA in Boden und Grundwasser anhand von Feld- und Laborstudien. Diploma thesis [4] Heydebreck F, Tang J, Xie Z, Ebinghaus R. 2015. Environ. Sci. Technol. 49: 8386-8395/ 49: 14742-14743

TH115

Fluoropolymers: Polymeric PFAS That Satisfy Global Polymer of Low Concern Criteria

B. Henry, W.L. Gore & Associates, Inc.

Fluoropolymers, such as polytetrafluoroethylene (PTFE), constitute a distinct class within the polymeric category of the PFAS group. Fluoropolymers are resistant to chemical, hydrolytic, oxidative, photochemical and biological degradation. They are thermally stable within their intended processing temperatures (e.g., 260°C for PTFE). Fluoropolymers have negligible residual monomer, low molecular weight oligomer, or leachable content. Fluoropolymers have high molecular weights well over 100,000 Da, are practically insoluble in water and are not mobile or subject to long-range transport in the environment. Their very high molecular weight prevents fluoropolymers from crossing the cell membrane and thus they are not bioavailable or bioaccumulative. The nontoxic nature of PTFE is supported by numerous Good Laboratory Practice (GLP) studies including acute and subchronic systemic toxicity, irritation, sensitization, local toxicity on implantation, cytotoxicity, *in vitro* and *in vivo* genotoxicity, hemolysis, complement activation, and thrombogenicity. Clinical studies of patients receiving permanently implanted PTFE-containing medical devices demonstrate no chronic toxicity or carcinogenicity, reproductive, developmental or endocrine toxicity. Fluoropolymer medical devices have been implanted in over 40 million patients for over 40 years. This poster includes fluoropolymer biocompatibility/toxicology, human clinical, and physical-chemical-thermal-biological data to show that fluoropolymers satisfy globally recognized assessment criteria to be considered as “Polymers of Low Concern” and to be recognized as being a low hazard class of PFAS. Fluoropolymers, therefore, are distinctly different from the other polymeric and non-polymeric classes of PFAS and should be separated from all other classes of PFAS for hazard assessment or regulatory actions. Grouping all classes of polymeric and non-polymeric PFAS together for restriction or regulation is not scientifically appropriate. Fluoropolymers, as polymers of low concern, are uniquely benign PFAS.

TH116

Fluoropolymers Are Unique, Low Hazard PFAS Needing Different Analytical and Regulatory Approaches Than Monomeric Fluorinated Substances of High Health and Environmental Hazard

B. Henry, T. Kennedy, W.L. Gore & Associates, Inc.; H. Fiedler, Örebro University, Örebro, Sweden

Fluoropolymers, such as polytetrafluoroethylene (PTFE), differ from other monomeric and polymeric per- or polyfluoroalkyl substance (PFAS) classes, such as perfluoroalkyl acids, or polymeric precursors that degrade to them. Fluoropolymers do not demonstrate the same toxicity or physical/chemical/thermal properties as other PFAS. Fluoropolymers, such as PTFE do not meet the criteria of PBT (Persistent/ Bioaccumulative/ Toxic) or vPvB (very Persistent/ very Bioaccumulative) chemical substances, nor do they meet the Persistent, Mobile and Toxic (PM or PMT) substances criteria proposed by the German Environmental Agency, Umwelt Bundesamt (UBA, 2017). As high molecular weight fluoropolymers (e.g., PTFE) are benignly persistent (i.e., not mobile, bioaccumulative or toxic), all “highly fluorinated” substances do not pose equivalent health or environmental hazards and thus should not be regulated as a single class of chemicals. Chemical analytical techniques useful for differentiating one fluorinated substance from another are readily available, reliable, and reproducible and should be employed to identify and quantify those highly hazardous monomeric per- and poly-fluoroalkyl substances (PFAS) individually, rather than techniques aggregating all fluorine containing substances into one group (e.g, total organic halogen, and total organic fluorine). Therefore, high molecular weight fluoropolymers, as a uniquely benign class of polymeric PFAS, require analytical and regulatory approaches differentiating them from fluorine-containing substances that present high health and environmental hazards.

Advances in Soil Ecotoxicology and Risk Assessment of Terrestrial Ecosystems (P)

TH117

Challenges and Open Questions in Earthworm field testing

T. Vollmer, Eurofins Agroscience Services EcoChem GmbH / Field Ecotoxicology; O. Klein, Eurofins Agroscience Services Ecotox GmbH / Ecotox Field; S. Knaebe, EAS Ecotox GmbH / Ecotox Field

In the risk assessment of plant protection products for in-soil organisms, the earthworm field test following ISO 11268-3 (ISO 2014) is used as the highest tier option. The test protocol is currently under revision and transition to an OECD document under the auspices of UBA (Germany), mainly focusing on improving/testing statistical performance of the test (e.g. effect of replication) and exploring the options to run the test in a dose-response design. In the light of the recently published EFSA opinion on in-soil risk assessment (EFSA, 2017), and with roughly 20 years of experience with the field test under the ISO guideline, other aspects of the test also might require revision, namely: Description of field site requirements (size and composition of initial earthworm population), e.g. minimum requirements, potentially derived from typical MDD values for a given endpoint, Land-use of the field site (arable fields vs. permanent grassland): is there a preferred option, and/or does this depend on characteristics of the substance under test? Site management/maintenance in general (soil cultivation, crop rotation vs. minimum disturbance) and more specifically in the case of testing a substance with herbicidal action (impact on vegetation coverage in test-item treated plots vs. plots of positive and negative control). Plot size and distance between neighbouring plots, plot allocation patterns, and plot separation, especially with a view on potential migration of earthworms between plots and on external re-colonization. Testing of persistent substances (e.g. how to establish a plateau concentration in soil)? Toxic reference (positive control): reduced replication for the toxic reference? Alternatives to the standard reference item Carbendazim? Examples and suggestions will be given and discussed in this contribution and areas for further research will be identified. EFSA [European Food Safety Authority], 2017: Scientific Opinion addressing the state of the science on risk assessment of plant protection products for in-soil organisms. EFSA Panel on Plant Protection Products and their Residues (PPR). EFSA-Q-2011-00978, Parma, Italy. ISO, 2014: ISO Guideline 11268-3: Soil quality – effects of pollutants on earthworms. Guidance on the determination of effects in field situations.

TH118

Regional Differences of the Environmental Risk Assessment of Pesticides in Soil with a special Focus on the European Union

I. Jaabiri Kamoun, National School of Engineering of Sfax, Tunisia / Laboratory of Water, Energy and Environment; J. Sousa, University of Coimbra / Department of Life Sciences; J. Roembke, ECT Oekotoxikologie GmbH

In the European Union (EU) the environmental risk of chemicals is regulated in various ways. Probably the most complex approach in place is the one for pesticides, mainly because these chemicals differ from other chemical groups by three reasons: (1) They are intended to harm organisms, i.e. those which are impacting agriculture. However, many of the pest species affected by pesticides belong to the same taxonomic groups being responsible for many soil functions and services. (2) They are also directly distributed in the environment, usually by spraying, but also in various other ways such as a coating on seed material. (3) Since their effects are supposed to act only against the pests, when being applied regularly the amount of pesticides ending up in the environment is high. Due to the long experience with this very detailed approach the results of the EU ERA for pesticides are often taken over by other countries (e.g. in Africa). But is this procedure reliable? This contribution focuses on the soil compartment and tries to tackle the following questions: (1) Can (and if yes: how) regional differences (e.g. regarding ecological or agricultural factors) influence the performance or the outcome of pesticide ERA? (2) How do ecological and agricultural differences influence the pesticide ERA within the European Union? Our findings show that regional differences in abiotic, biotic and anthropogenic factors can affect the fate of pesticides in soil as well their effects on soil organisms, meaning that these differences should be considered in pesticide ERA. Proposals will be made how to improve the ERA process but keep it at the same time practical (e.g. by using a tiered approach). These ideas will include the selection of representative reference soils and test conditions for Mediterranean regions. In addition, we will discuss whether the range of standard test species used so far is sufficient. In this context comparisons with current developments in tropical soil ecotoxicology might be helpful. Based on the answers to these questions it will be discussed whether (and if yes, how), the ERA of pesticides has to be modified for Mediterranean regions (both inside and outside of the European Union).

TH119

Adaptation of the earthworm field test method: conceptual overview and first results

J. Roembke, ECT Oekotoxikologie GmbH; B. Daniels, RWTH Aachen University / Institute for Environmental Research Institute; B. Förster, S. Jaensch, ECT Oekotoxikologie GmbH; P. Kotschik, Umweltbundesamt / Federal Agency of Environment / Risk assessment for plant protection products; R. Ottermanns, RWTH Aachen University / Institute for Environmental Research; S. Pieper, German Federal Environment Agency (UBA) / Plant Protection Products; M. Ross-Nickoll, RWTH Aachen University, Institute for Environmental Research / Institute for Environmental Research; A. Scheffczyk, ECT Oekotoxicologie GmbH; B. Scholz Starke, RWTH Aachen University / Institute for Environmental Research

In 2016, the German Federal Environment Agency (UBA) launched a project entitled "Necessary adaptations of the standard Earthworm Field Test" to improve the scientific basis when transferring the ISO 11268-3 (1999) guideline to a new OECD standard. As a first step, a literature search was performed to compile available data from earthworm field tests, both from the open literature and from anonymized studies used for regulation. This data set was statistically evaluated to develop a design for a pilot study for the earthworm field test. In February 2017, a workshop was held to discuss the outcome of this statistical evaluation and in particular the proposal for a pilot field study test design together with members of the "OECD-GSIG-Earthworm Field Group". During and after the workshop, the group agreed on a test design including various aspects of statistical robustness, practicability and flexibility. In these discussions various options were checked, all of them with the intention to improve the quality of the output but without increasing the efforts in routine application of the new design. In simulation studies, the number of plot replicates dedicated to either NOEC- or ECx-derivation were varied as well as the number of samples per plot. Additionally, the number of treatments of the chemical to be tested (carbendazim, because it is the reference substance for earthworm field tests for more than 20 years) was also modified in order to cover a broad range. This study can be considered as the biggest earthworm field studies ever conducted. In April 2017, the pilot study was started in a design with 30 plot replicates. After this pre-sampling two further samplings have been performed and the last sampling will be conducted in April 2018. First results of this project indicate a clear concentration-dependent effect of carbendazim on earthworms.

TH120

Soil ecotoxicology research and ecological risk assessment in southern African mining landscapes

M. Maboeta, North-West University / Unit for Environmental Sciences and Management; H. Eijsackers, WUR/NWU

Although there has been a remarkable development in the field of soil ecotoxicology and risk assessment (RA) models, it is debateable if these RA models are representative enough in order to utilise them on larger spatial scales, attuned to specific landscapes and ecosystems worldwide. An example of this is southern Africa where some soil ecotoxicological research has been done. To address this we will present the results of: An inventory of research on the ecotoxicity of metals toward soil life in southern Africa as an example, focussing on southern African soils, soil life and living conditions. The outcomes of an analysis of the geographical surroundings of gold and platinum mine waste deposits in South Africa The field study of open coal mining in and near a national park in Swaziland It is concluded that there is a limited body of information on southern African soil life, and most of these were laboratory based studies done by a small group of researchers. Future research with regards to incorporating the information available into a soil ecosystem assessment procedure is needed and recommended. It is recommended that a starting point to address this might be the development of site-specific guidelines for Ecological Risk Assessment (ERAs) taking into account landscapes, vegetation and faunal characteristics. From our studies in the surroundings of platinum and gold mine waste, we conclude that these wastes still contain considerable amounts of other chemical elements. The extraction methods, moreover, result in very alkaline or acidic conditions. Further the mine waste is very fine grained and therefore susceptible for wind erosion. Consequently these wastes, given the prevailing wind conditions in these areas, will be dispersed over a wide area causing risks for organisms in natural and built areas surrounding these deposit areas. The coal mine study illustrates that mining in or around natural protected area cause risks due to the irradiating impacts of wind and surface and ground water dispersal form the mined area. Therefore ERA should start to assess the impacts on the natural ecosystems present in the area, and compare these with the outcomes of a Potentially Affected Fraction of species PAF analysis. **Key words:** soil ecotoxicology, ecological risk assessments, mining, southern Africa

TH121

Establishment of tiered risk assessment approach of pesticides for soil organisms in China

J. Jiang, Nanjing Insitute of Environmental Sciences, MEP; J. Zhou, Nanjing Institute of Environmental Sciences, MEP

The aim of the risk assessment for soil organisms is to prevent negative acute or long-term effects on soil functions and functionally most important species. The risk assessment approach established in China considers ecologically relevant groups of soil organisms, eartheworm and soil microorganisms, which are involved in a range of soil functions providing essential ecosystem services, e.g. organic matter breakdown and mineralization, water regulation in soil. The tiered approach is a valuable tool to quickly identify those pesticides which do not pose acute or chronic risk (in a certain area of ecotoxicology) on soil organisms – even under worst case assumptions, and to identify those that need more attention and further evaluations. All risk assessments presented are based on Risk Quotients (RQ), calculated by dividing the Predicted Environmental Concentration (PEC) by the Predicted no Effect Concentration (PNEC). This calculation takes into account, that beside the toxicity of a pesticide, the amount of this pesticide in the environment plays a major role when assessing a risk. If RQ > 1, the risk is unacceptable and higher tier risk assessment should be conducted. Exposure analyses employ tiered assessment approach. Tier 1 exposure analysis employs simple model (PECsoil_SFO_China from NIES) to predict exposure to soil organisms. A higher tier exposure analysis can be applied by refining environmental exposure parameters or using semi-field trial test. Currently, the models PRAESS and China-PEARL, which developed by NIES and ICAMA in China, are applicable to predict the exposure concentration at specific depth of soil layer and at specific scenarios in China. Proposed test systems for effect assessment include acute toxicity test or reproduction of earthworm, reproduction test, nitrogen transformation test, litterbag test and earthworm field test. The PNEC can be calculated using the endpoint obtained from ecotoxicological studies and corresponding uncertainty factors (UF). Tier 1 risk assessment mainly focuses on the earthworm acute or chronic (in case pesticide DT_{50}>180 d or RQ_{acute}>1) assessment and N - transformation assessment. High tier risk assessment mainly foucses on the litterbag test assessment and earthworm field assessment. We have used this tiered risk assessment approach to assess the risk of more than 40 common used pesticides in China. **Keywords:** tiered, risk assessment, pesticide, soil organism

TH122

Ecological recovery and terrestrial Non Target Arthropods: abundance, functional roles and networks

M. Hagen-Kissling, Eurofins-Mitox; S. Aldershof, Bioresearch and Evaluation; F.M. Bakker, Eurofins-Mitox

Terrestrial non-target arthropods exhibit a vast array of life history strategies and migration tactics. However, their home range is rather small, and thus, they are a good model group to investigate the factors which influence ecological recovery in risk assessments. Arthropod communities are not stable, but the numbers of species and individuals per species fluctuate over time and space. Part of the variability may be due to the initial disturbance (the application of PPP), part may be seasonal, intrinsic or due to biological interactions, as individuals are embedded in complex food and interaction networks. However, the recovery of a network depends on the fact that all ecological and functional roles within such a complex entity are still realised by a certain number of species (and their individuals). Usually, only abundance criteria are applied in ecological recovery investigations. For example, if numbers of individuals trapped are similar to a control group on two subsequent occasions, recovery is concluded upon. But if the proportion of one functional role (e.g. predators, parasitoids, pollinators or herbivores) in the focal group is far below the proportion of the same functional group in the control group, recovery is not completed, and the stability of the network in focus might be imbalanced. On the other side, abundance might be different to the abundance in the control group, because of a phase shift due to the initial disturbance, but the proportional distribution of functional roles still mirrors the control group. Thus, we feel that pure abundance data are not enough to understand ecological recovery, but suggest to use additional knowledge about the involved species and their interaction network, like the functional roles and their proportional distribution within a community. Investigating the ecological recovery of a community using information from field work and experiments together with additional information about the species and their importance for and embeddedness in the ecological network is of high importance for a better understanding of the ecological recovery of communities.

TH123

Comparing effects of fludioxonil on non-target invertebrates using ecotoxicological methods from single-species bioassays to model ecosystems

A. Hägerbäumer, R. Raschke, Bielefeld University / Animal Ecology; S. Höss, Ecossa / Animal Ecology; W. Traunspurger, Bielefeld University / Animal Ecology

Lower tier toxicity testing used for risk assessments of plant protection products (PPPs) is conducted with single species. Informations from such toxicity tests are, however, limited to direct effects of the respective tested substances. Although the uncertainties regarding the protectiveness of these tests for *in-situ* communities are known, informations on effects of PPPs on community-level of non-target organisms are scarce. Model ecosystems, i.e. microcosms, are suitable to bridge this gap between single-species tests and field studies since they provide controlled experimental conditions and are able to demonstrate direct and indirect effects of the respective substances. In the present study, single-species toxicity tests and soil-spiked microcosms were used to comparatively investigate the toxicity of the non-systemic fungicide fludioxonil (FDO) on non-target soil organisms. Regarding soil invertebrates, nematodes are among the most abundant metazoan organisms and are considered as important components of the soil food web. Since nematodes are suitable for risk assessments via various assay tools ranging from single-species toxicity tests to field studies, potential effect of FDO on these non-target organisms were assessed using standardized toxicity tests with *Caenorhabditis elegans* (ISO 10872) in spiked soil exposure and *in-situ* nematode communities, sampled from microcosms with FDO-spiked soils. In the standardized toxicity tests, FDO inhibited the reproduction of *C. elegans* dose-dependently, with a chronic 96-h EC50 of 363 mg kg^{-1} (dry weight, dw) and a 96-h No Observed Effect Concentration (NOEC) of -1 (dw), which is comparable to the 28-d NOEC of *Chironomus riparius* in sediment (40 mg kg^{-1} dw) and within the range of the 56-d NOEC of *Eisenia fetida* (20 mg kg^{-1} dw) and 28-d NOEC of *Folsomia candida* (125 mg kg^{-1} dw). In the spiked microcosms, distinct effects on *in-situ* nematode communities could be measured, with significantly lowered abundances in spiked soils, (40% and 50% reduction of nematode abundance in soils spiked with 300 and 600 mg FDO kg^{-1} (dw)). Overall, this study provides new insights into the impact of the non-systemic fungicide fludioxonil on non-target soil organisms and demonstrated the general suitability of standardized toxicity testing on *C. elegans* in protecting *in-situ* communities.

TH124

To what extent do soil micro-arthropods facilitate OM breakdown in an arable field soil? - Implications on specific protection goal setting for soil risk assessment of plant protection products

G. Ernst, Bayer Ag / Ecotoxicology; J. Bendall, Dow Agrosciences; T. Carro, FMC; H. Cunningham, Syngenta / Environmental Safety; A. Koutsaftis, ADAMA; S. Loutseti, DuPont De Nemour Hellas S.A.; M. Marx, Bayer AG Crop Science Division / Environmental Safety - Ecotoxicology; B. O'Neill, DuPont Crop Protection; A. Sharples, FMC Agricultural Solutions; F. Staab, BASF SE

Soil functional test systems provide valuable and ecologically relevant information for the risk assessment of plant protection products (PPP). Functional tests directly measure ecosystem functions and services which are provided by soils and soil organisms (e.g. organic matter (OM) degradation and mineralization). Focusing on structural endpoints in the risk assessment for PPP lacks a clear link to the protection goals derived from ecosystem services. Directly measuring soil functions and services can help to better assess the impact of a stressor on the fertility of soils. Furthermore, functional test systems can help to evaluate the ecological relevance of a density change of a soil organism population affected by a certain stressor. To quantify the soil mesofauna and microorganism contribution to the process of OM breakdown, a project on soil functional test systems was initiated by the European Crop Protection Association (ECPA). A field study was set up in 2015 which measures the impact of two insecticides (Methamidophos, Lindane) on organic matter degradation in a minicontainer test. Soil micro-arthropod abundances were monitored in parallel to determine the link between effects on the structure of soil micro-arthropods and their soil functional implications (i.e. OM breakdown). The results indicate that the process of OM degradation is dominated by soil microbes. Soil mesofauna contributed only a minor amount to OM degradation. The minicontainer test did not show a clear effect of insecticides on the mesofauna driven organic matter degradation, although total abundances of Collembola and Acari were heavily reduced by the insecticide applications. In the recently published Soil Scientific Opinion (2017), EFSA proposed Specific Protection Goals for soil micro-arthropods for in-field areas. This foresees that even short-term effects on single species in a magnitude of >65% are considered unacceptable to ensure the provision of Ecosystem Services in agricultural soils. The present study shows that a reduction of the total soil micro-arthropod community by 80% over a period of 6 months has no unacceptable effect on the mesofauna driven OM degradation in a minicontainer test on an arable field. Thus, the relevance of structural endpoints on soil micro-arthropods (i.e. single species population) within an in-field soil risk assessment for PPP, which focus on maintenance of soil fertility (protection of soil functions), is questionable.

TH125

The role of source sink-dynamics in the assessment of risk to non-target arthropods from the use of plant protection products

G. Lewis, JSC International Ltd; S. Braaker, BASF France S.A.S.; C. Mayer, BASF SE / Ecotoxicology

The concept of source-sink dynamics as a potentially important component of metapopulation dynamics was introduced in the 1980's culminating in the paper by Pulliam (1988). Since that time, a considerable body of work has developed to consider its theoretical implications as well as to identify how it may be manifested under field conditions. Most recently, the concept of source-sink dynamics has been considered within the European Food Safety Authority (EFSA) Opinion addressing the state of the science on risk assessment of plant protection products for non-target arthropods (EFSA, 2015). This presentation reviews the available literature that investigates the theoretical implications of source-sink dynamics as well identifying the relevant available evidence from both experimental systems and field observations, primarily in relation to non-target arthropods in an agricultural environment. Consideration of this information clearly shows that metapopulation dynamics are generally more complex than presented by the simple source-sink model as originally proposed and that they are very much species/context dependant. However, this issue does raise important questions about the consideration of spatial and temporal scales when assessing the population dynamics of non-target arthropods in the context of the risk from the use of plant protection products. It is therefore important to consider what the evidence base is for source-sink dynamics in the agricultural environment and what this tells us about whether or how it is manifested in relevant populations. A structured approach can then be adopted in terms of identifying a suitable range of representative surrogatespecies and generating the necessary information for them and at the landscape level to allow the development of suitable population models. These models could then be used in an appropriate way within a risk assessment scheme e.g. at a higher tier level addressing specific issues of concern identified at the lower tiers. They may also have the potential to inform risk managers to improve sustainable landscape management. Practical considerations, in terms of the amount of information needed in relation to the life-cycles of non-target arthropod species and landscape structure, are also identified. Acknowledgements: This work was initiated and funded by the ECPA non-target arthropod group

TH126

Classification of uncertainty in ecological risk assessment of pesticides

A. Hunka, Halmstad University / School of Business, Engineering and Science; M. Meli, ADAMA Agricultural Solutions; S. Pashami, S. Waara, Halmstad University

Uncertainty estimates are inherently built into any prospective risk assessment. Uncertainties need to be correctly recognized, described and presented to provide a basis for decision-making. One important factor to consider is that more data and experimental results often increase the perception of uncertainty instead of reducing it, making this aspect an issue of significant concern. The lack of straightforward presentation of all sources of uncertainty puts an extra burden on risk managers. This issue has been recognized by EFSA recently, but still there is very little research into classifying, visualizing and addressing uncertainty in ERA of pesticides. Currently EFSA recognizes standard and non-standard uncertainties in ERA of pesticides. This classification offers little insight into how those two categories impact ERA conclusions and further risk management decisions. In order to address this gap, we have undertaken an appraisal of a large subset of ERAs (102 up to date) conducted for approval of active substances at the EU level. We have been working on ERA data, conclusions on pesticide peer-reviews and Draft Assessment Reports in order to identify the most commonly acknowledged sources of uncertainty, classify different uncertainties and link them to recognised points of concern, data gaps and risk management decisions. At the moment it is still unclear which sources of uncertainty influence the decision outcome more than others and our preliminary results indicate that it is possible to uncover non-obvious relationships between uncertainty and risk assessment outcomes. It is, for instance, possible to describe how different sources of uncertainty affect the process of ERA (e.g., duration, effectiveness of a Rapporteur Member State in producing a draft assessment report) and compare how uncertainty is addressed in risk assessment for different environmental compartments, especially soil *versus* ERA for aquatic organism which is much more developed. We aim to provide a typology of recognized uncertainties in ERA and discuss how it could help inform the establishment of the surrogate reference tier and the subsequent calibration of lower tiers in the new risk assessment scheme for in-soil organisms, which is currently being developed by EFSA. Ultimately, we aim to link the typology of uncertainties in ERA to risk management techniques, in order to help ERA practitioners to better address and manage uncertainties.

TH127

Derivation of soil threshold concentrations for arsenic: consideration of bioavailability through combination of ecotoxicological and analytical data

J. Roembke, J. Brack, B. Förster, ECT Oekotoxikologie GmbH; E. Giese, Federal Environment Agency; S. Jänsch, R. Minati, ECT Oekotoxikologie GmbH; A. Scheffczyk, ECT Oekotoxicologie GmbH; D. Rückamp, Federal Institute for Geosciences and Natural Resources; K. Oorts, ARCHE

The German Federal Soil Protection Act (1988) defines precautionary values for seven metals which, if exceeded, indicate that concern for a harmful soil change exists. All precautionary values given in the German Soil Protection Ordinance (1999) are based on total concentrations ("aqua regia"). However, a realistic risk assessment of metals should consider their bioavailability in soil. Thus, the aim of this project is to connect bioavailable fractions of arsenic with ecotoxicological effect concentrations, taking into account soil properties (texture, pH, organic matter content etc) and various metal extractions (1M NH_4NO_3,0.01M $CaCl_2$, $Ca(NO_3)_2$ with ionic strength corresponding to soil solution, DTPA/$CaCl_2$, 0.43M HNO_3, plus aqua regia). Arsenic was chosen due to its high relevance as a soil contaminant, its low data availability compared to other metals and is an element of concern included in many soil regulations. Six soils covering a wide range of Central European soil properties were chosen and spiked with sodium arsenate dibasic heptahydrate (Na2HAsO4•7H2O). Chronic toxicity endpoints were tested with microbes, plants and invertebrates, according to ISO standard guidelines, allowing derivation of threshold values via an SSD approach. Test results (given as NOEC, EC_{10} and, preferably, EC_{50} values), based on the six extraction methods, have been determined. The variation in EC_{50} values based on nominal concentrations among the soils tested differed typically by a factor of 2 - 5 for the endpoints tested. The extraction strength of the different methods and soils differ at least by an order of magnitude in the order NH4NO3 < CaCl2 < Ca(NO3)2 < DTPA < HNO3 and aqua regia for most soils. Plants were the organisms reacting most sensitively, partly together with the Bacteria. Both invertebrate species were always less sensitive (i.e. EC50 values (nominal concentration) > 250 mg As/kg soil) than microbes and plants except in one sandy soil (RefeSoil-01A). Currently, chemical and biological results are combined in order to explain the observed variation in toxicity expressed as nominal or total As concentrations in soil. This information will be used to include As bioavailability into the derivation of precautionary values.The representativeness of the different extraction methods regarding bioavailable fractions as well as the properties of the different soils are checked as part of a more realistic risk assessment of metals in soils.

TH128

Activity based in-soil arthropod sampling

S.B. Dehelean, F.M. Bakker, Eurofins-Mitox

Higher tier (field) assessment of effects on soil microarthropods relies strongly on the accuracy of the sampling methodology. Two main classes of trapping methods exist to date, these are either abundance-based or activity-based. Abundance-based trapping usually involves the collection of soil cores followed by heat extraction such as Berlese-Tullgren or McFayden methods. Activity-based sampling implies installing hypogean traps and collecting the catch at pre-determined intervals. Soil core sampling provides an instantaneous assessment of the fauna at the exact moment and at the very location of sampling, whereas hypogean traps provide an assessment of the activity in a wider area and over a longer time span. Clearly, higher abundance may imply higher activity. Soil core sampling is an established and recommended method known to extract springtails, mites and some other small arthropods. Hypogean trapping is a relatively novel approach (cf Dehelean et al. SETAC 2016, Sims et al. 2016, Bakker et al. 2017) and seems to have a certain degree of selectivity. For purposes of method development and evaluation we have performed a comparative study in which soil core sampling and hypogean trapping (mine traps) were performed in the same fields. The study comprised both a hay meadow and an arable field. Soil cores were taken from the top 10 cm of soil, mine traps collected from various depths. With this contribution we will highlight the differences in species spectrum, numbers collected and variability observed with the different methods and discuss the implications for data analysis and interpretation.

TH129

The application of the CPCAT approach reduces shortcomings of effect detection for earthworm field studies

B. Daniels, RWTH Aachen University / Institute for Environmental Research; S. Jänsch, ECT Oekotoxikologie GmbH; P. Kotschik, Umweltbundesamt / Federal Agency of Environment / Risk assessment for plant protection products; R. Ottermanns, RWTH Aachen University / Institute for Environmental Research; S. Pieper, German Federal Environment Agency (UBA) / Plant Protection Products; M. Ross-Nickoll, B. Scholz-Starke, RWTH Aachen University, Institute for Environmental Research / Institute for Environmental Research

Field studies to determine effects of pollutants on earthworm community are generally conducted according to standardized ISO-guidelines (ISO 11268-3). However, statistical test procedures suggested in the guidelines are frequently criticized, mainly for two reasons: test data characteristics do not fulfil test requirements (normal distribution and variance-homogeneity) and the resulting toxicity metrics of multiple testing procedures (NOEC / LOEC) fail to adequately detect the actual level of effects. Lehmann et al. (2016) presented a new approach to overcome these shortcomings by introducing the CPCAT procedure. We applied this statistical method to detect effects in a set of 16 earthworm field studies and provide a comparative analysis with regard to results of well-established multiple testing approaches. This is the first time the performance of CPCAT is assessed within a comprehensive meta-analysis of field study data. Raw data of biomass and abundance on sample level (0.25 square metres) were extracted from original study reports and assessed on sample and plot level. In total, data of 17 different earthworm species, ecological and morphological groups as well as total abundance and biomass for 1-3 treatments and 3 sampling dates after application within test duration of one year were analysed. This led to a total of 4215 comparisons for the detection of differences between control and treatments. We demonstrate that the distribution of both endpoints abundance and biomass can be described by a Poisson model, which is a requirement for the application of CPCAT (variance homogeneity -often not fulfilled in toxicity tests- is not a prerequisite of CPCAT). The number of endpoints showing a significant difference between control and treatment was compared to the outcome of parametric test procedures (pairwise t-test, Dunnett and Williams t-test for multiple testing). The study reveals that the application of standard multiple testing procedures leads to a disguising of possible effects due to relatively high differences to be achieved between control and treatments. This consequently results in uncertainties regarding the actual level of effects at the NOEC. The CPCAT approach offers a more powerful and statistically proper evaluation for these earthworm field studies because data distribution and variance are adequately considered and smaller differences between control and treatments can be detected.

TH130

Relationship between soil microbial biomass methods used in environmental fate laboratory studies

P. Massey, Smithers Viscient; P. Pearson-Davies, B. Earnshaw, Smithers Viscient ESG; S. Swales, Smithers Viscient ESG Ltd

The determination of microbial biomass activity is a pre-requisite for OECD laboratory studies that are designed to investigate the environmental fate of chemicals in soils. By determining soil microbial biomass prior to, during and after a study, the viability and suitability of the experimental soil can be demonstrated. Choosing the appropriate methodology for soil microbial biomass is critical for conducting successful environmental fate studies. One common method, referenced in the OECD guidelines, for determining soil microbial biomass is the fumigation extraction method. This method determines the carbon content of the soil biomass, via fumigation, using ethanol-free chloroform. During fumigation, cells are lysed by the chloroform, which results in a flush of organic carbon into the soil environment. This organic carbon is then extracted and quantified. Another suitable way of estimating soil microbial biomass is by substrate induced respiration. This method uses a suitable labile substrate to promote a respiratory response. The carbon dioxide evolved or the oxygen consumed as a result of this respiratory response is then used to determine microbial biomass activity. Microbial biomass size can then be determined by relating respiration and fumigation extraction data. \n Despite there being multiple recognised ways of determining soil microbial biomass, it is important to recognise that they reflect different aspects of the soil microbial community. One fundamental difference between these methods is that they can potentially distinguish between active and non-active components of the biomass. As noted in OECD 14240-1:1997, substrate induced respiration can be used to estimate the active aerobic biomass, whereas in OECD 14240-2:1997, fumigation can extract carbon from both active and non-active biomass components. With such differences between methods, it is important to consider which method is more appropriate for determining soil suitability for environmental fate laboratory studies.\n Work is currently being undertaken by Smithers Viscient to investigate the relationship between the soil microbial methodologies commonly used for laboratory soil studies. The aim of this work is to better understand how the choice of soil biological methods relates to soil suitability, which will ultimately facilitate and refine our choice criteria when choosing soils for environmental fate studies.\n

TH131

Where are the Springtails? New data on the vertical distribution of Folsomia candida (Collembola) and its population dynamic in artificial soil

L.S. Tzschoppe, RWTH Aachen University / Institute for Environmental Research BioV; V. Roeben, RWTH Aachen University, Institute for Environmental Research / Institute for Environmental Research BioV; M. Roß-Nickoll, RWTH Aachen University / Institute for Environmental Research

Folsomia candida is a non-target arthropod species which is often referred as the „Standard Soil Arthropod" (Fountain and Hopkin, 2005). It is part of the regulatory framework of pesticide risk assessment and in the last years an increasingly important model organism in ecological and effect modelling. However, the knowledge on the population dynamics on a long-term scale and the vertical dispersal within the soil column is still scarce. We will present the results of two experimental studies exploring those unknown topics – one on the population dynamics over time and one on the vertical dispersal in relation to food location.

The population dynamics experiment is a one-year study assessing the dynamics of *Folsomia candida* in artificial OECD soil at constant 20'C. The study started with 25 individuals of different age classes in 100 g OECD soil. Since then the population increase was measured on at least a monthly basis with five replicates per testing day. The food regime is adapted to the increasing population density to make sure that the maximum population level is achieved during the study. We will show a fast growth at the beginning of the experiment and expect to reach an oscillating population size around its maximum capacity at the end of the study. In a second experiment the vertical dispersal of *F. candida* in relation to food location is investigated. Transparent PVC columns were filled with on average 350 g of OECD soil up to a level of 20 cm soil column height and 86 *F. candida* of different age classes. Each column was closed with Parafilm on top and a gauze on the bottom with a small water reservoir beneath it to avoid desiccation. The initial humidity was set to 50 percent of the maximum water holding capacity of the soil. The columns were separated into six compartments at different heights: 1, 2.5, 5, 10, 15 and 20 cm. Septa in the middle (4^{th}) and bottom (6^{th}) compartment allowed for watering and feeding. We varied the location of feeding by four different regimes while all other parameters were kept constant. The columns were provided either with food at the top, in the middle, in the ground or at all three compartments. Our hypothesis is that food is a main trigger for the vertical distribution of *F. candida* in soil. Two of three large examination dates have been processed so far. The data confirms our hypothesis and the results of the study will present new data for the otherwise well investigated Collembola species *F. candida*.

TH132

Why zinc doesn't matter: habitat quality drives invertebrate response to zinc, not concentration

S. Siciliano, University of Saskatchewan / Department of Soil Science; K. Jegede, H. Fajana, University of Saskatchewan   Toxicology Centre

The responses of organisms in soil ecotoxicity tests are often determined by the bioavailable concentrations of contaminants they are exposed to. However, the direct effect of habitat quality on the performance or response of organisms in different contaminated soils is often neglected. Habitat quality is a measure of extent to which habitat promotes individual and population fitness. This study assessed the effect of habitat quality on mite, *Oppia nitens* exposed to different contaminated soils which was corrected for bioavailable metals. Forty-seven (47) soils were ranked into habitat quality by summing up the scores of enchytraeid and collembola survival and reproduction with the plant biomass in each of the soils. From the 47 soils, 18 soils were divided into three habitat quality groups based on high, medium and low habitat quality. The 18 soils were dosed with low to high concentrations of zinc and mites exposed to the soils for 28 days. Mite survival, reproduction, stress biomarkers, and bioavailable zinc were determined after 28 days. Habitat quality did not change zinc bioavailability which remained at 2% across all three habitat indices. Instead, mite fitness improved with increasing habitat quality and mites were able to tolerate higher zinc body burdens in better habitat qualities. Furthermore, the zinc response (measured as the slope of the EC50) was more pronounced in lower habitat qualities. Our data suggest that habitat quality is more important than metal concentration for soil protection. Ecorestoration, rather than remediation, will likely be a more effective means of ameliorating zinc toxicity.

TH134

Effects of atmospheric hydrogen chloride and ammonia on Paronychiurus kimi (Collembola : Onychiuridae)

J. Wee, J. Son, Korea University / Division of Environmental Science and Ecological Engineering; Y. Kim, Y. Lee, J. Hong, M. Lee, Korea University; K. Cho, Korea University / Division of Environmental Science and Ecological Engineering

As the use and distribution of various chemicals increases, there is a possibility of chemical accidents and in Korea, the incidence of chemical accidents is also increasing. Damage caused by chemical accidents is not only widespread but also has a long-term impact, making it difficult to predict damage and respond appropriately however, there are very few studies on chemicals that can cause accidents. Especially for chemicals exposed to gaseous state, little is known about the effect on soil organisms such as Collembola and earthworm. The experiment carried out in PS container filled with 30g of soil according to modified OECD 232 guidelines. Investigating the effects of gaseous hydrogen chloride and ammonia on *Paronychiurus kimi* (Collembola), the test vessels with *P. kimi* were exposed to two different concentration of toxic substances in the enclosed chamber for 20 minutes. After exposure, the test vessels with *P. kimi* were transferred to an incubator (20?, continuous darkness) in a closed state, and the mortality and reproduction rate of *P. kimi* were observed after 1 hours, 2 weeks, and 4 weeks. There were no deaths after 1 hour, but the mortality rate was increased over time from 2 weeks. Also, after 4 weeks, the number of juveniles produced by adults *P. kimi* were decreased as concentration-dependent manner. These results show that the long-term effects of gaseous phase chemicals can occur at concentrations that are not acutely affected.

TH135

Toxicity assessment of methyl ethyl ketone using earthworm and soil algae

R. Cui, Konkuk University / Department of Enviromental Sciences; Y. An, Konkuk University / Department of Environmental Heath Science

Methyl ethyl ketone (MEK) is a kind ofketone-based volatile organic compound and widely used as industrial solvent. There is a high possibility of leakage of this compound into soil environment, but few studies for ecotoxicity of MEK were present. This study evaluated the toxicity of MEK using earthworm *Eisenia andrei* and soil algae *Chlamydomonas reinhardtii* and *Chlorococcum infusionum*. *Eisenia andrei* were exposed with 10 g of control or MEK soils with closed system. After 7 days exposed, mortality and abnormalities including thinning, fragments, swelling, bleeding, and mucous secretion were measured. For soil algae, *Chlamydomonas reinhardtii* and *Chlorococcum infusionum* were exposed with 2.5 g of control or MEK soils in 15 mL glass test tube, and chlorophyll intensity was measured after 6-day exposed. As results, 7d-LOEC and 7d-EC50 of MEK to *Eisenia andrei* were calculated as 1136 mg MEK/kg dry soil and 1910 (1643.00-2221.58) mg MEk/kg dry soil, respectively. For soil algae, *C. infusionum* was more sensitive than *C. reinhardtii* for MEK, 6d-EC50 to *C. reinhardtii* and *C. infusionum* were calculated as 3400.44 (3132.01-3690.94) mg MEK/kg dry soil and 60.97 (51.19-72.62) mg MEK/kg dry soil, respectively. These results can be used for risk assessment of MEK in soil ecosystem. *This work was supported by Korea Environment Industry & Technology Institute(KEITI) through "The Chemical Accident Prevention Technology Development Project", funded by Korea Ministry of Environment(MOE)(No. 2016001970001).*   ***<strong>Key word: metyl ethyl keton, earthworm, soil algae***

TH136

Effects of endocrine disrupt chemicals (EDCs) to soil algae

R. Cui, Konkuk University / Department of Enviromental Sciences; Y. An, Konkuk University / Department of Environmental Heath Science

There were many data for endocrine disrupt chemicals (EDCs) for aquatic organism, but soil toxicity data of them were very limited. This study evaluated the effects of bisphenol A (BPA), bis(2-ethylhexyl)phthalate (DEHP), and nonylphenol using soil algae. Soil algae *Chlamydomonas reinhardtii* and *Chlorococcum infusionum* were exposed at 0.5 g of control or exposed soils in 6-well plate. Algae were extracted for 1 day using algae culture medium after 6-d exposure, and the chlorophyll intensity was measured by fluorescence microplate reader. We observed that the BPA was most toxic following NP and DEHP. The effect of DEHP was insignificant to *Chlamydomonas reinhardtii* and *Chlorococcum infusionum*. The results can be used for risk assessment of BPA, DEHP and NP in soils. *This study was funded by the Korea Ministry of Environment (MOE) as the Environmental Health Action Program (1485014458)*   **Key word: bisphenol A, bis(2-ethylhexyl)phthalate, nonylphenol, soil algae**

TH137

Evaluation of reproduction tests of earthworms and enchytraeids exposed to sugar cane vinasse in natura and after pH adjustment

C.M. Sousa, São Paulo State University   UNESP / Biology; C.P. de Souza, Sao Paulo State University - UNESP / Biology; H. Soares-Lima, Universidade Estadual Paulista Júlio de Mesquita Filho- Unesp- Rio Claro / Departamento De Biologia, Centro de Estudos de Insetos Sociais; A. Marcato, Sao Paulo State University - UNESP / Department of Biology; T. Natal da Luz, University of Coimbra / Department of Life Sciences, University of Coimbra; J. Sousa, University of Coimbra / Department of Life Sciences; C.S. Fontanetti, Sao Paulo State University - UNESP / Biology

The use of sugarcane vinasse as fertilizer in crops has been widely used in order to provide an adequate destination for this residue however, it has properties that can be prejudicial to the animals present in the soil, as already verified in numerous studies. Therefore, the objective of this work was to treat vinasse with lime (CaO) to adjust pH to 7.0 (neutral), in an attempt to reduce its toxicity for later use in the soil. In this context, the development of ecotoxicological tests presents itself as a tool of great assistance in the analysis of residues released to the soil. Thus, reproduction tests were conducted using animal soil biondicators to evaluate the effects of vinasse may have on reproductive behavior of these animals. Earthworms of the species *Eisenia andrei* (Annelida) and enchytraeids of the species *Enchytraeus crypticus* (Annelida); both tests were developed according to the protocols proposed in ISO 11268-2 (ISO, 2011) and ISO 16387 (ISO, 2013), respectively. In the reproduction test with *E. andrei* exposed to vinasse *in natura* in comparison to the animals exposed to the treated vinasse there was an increase in the number of animals, which suggests that the vinasse treatment for pH adjustment was valid for this test. The reproduction test with *E. crypticus* exposed to the same conditions cited above also showed an increase in the number of individuals exposed to treated in bioassays vinasse compared to exposed to vinasse *in natura*. The results allow to infer that the pH adjustment of the vinasse to a neutral level was effective in reducing the toxicity of the residue for the tests of reproduction in both species used, since the environment favored the reproduction of the animals tested.

TH138

Ecotoxicological Characterization of Nitrogen-Based Energetic Soil Contaminants

R.G. Kuperman, Edgewood Chemical Biological Center / Molecular Toxicology Branch; R. Checkai, U.S. Army Edgewood Chemical Biological Ctr / Molecular Toxicology / Environmental; M. Simini, U.S. Army Edgewood CB Center /

Environmental Toxicology
We provide an overview of ecotoxicological effects of nitrogen-based energetic materials (EM) of notable ecological concern, hexahydro-1,3,5-trinitro-1,3,5-triazine (RDX), octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine (HMX), and 2,4,6,8,10,12-hexanitro-2,4,6,8,10,12-hexaazaisowurtzitane (CL-20), 2,4,6-trinitrotoluene (TNT), 1,3,5-trinitrobenzene (TNB), 2,4-dinitrotoluene (2,4-DNT), 2,6-dinitrotoluene (2,6-DNT), 2-amino-4,6-dinitrotoluene (2-ADNT), 4-amino-2,6-dinitrotoluene (4-ADNT), and nitroglycerin (NG). Ecotoxicological effects of these EMs were determined in definitive studies with Sassafras sandy loam (SSL) soil using soil invertebrate, terrestrial plant, and biological activity endpoints. SSL soil was selected because it has physicochemical characteristics (low organic matter and clay contents) that support very high relative bioavailability of organic chemicals. Data for reproduction (soil invertebrates), growth (plants), and critical soil processes (basal and substrate-induced respiration, litter decomposition) were independently analyzed using appropriate regression models to determine the EM concentration producing 20 percent decrease (EC20) in the measurement endpoint compared with carrier (acetone) control. Toxicological benchmarks developed in studies with soil invertebrate and terrestrial plants were used to derive draft Ecological Soil Screening Levels (Eco-SSLs) for use in screening-level ecological risk assessment of EM-contaminated soils. Additionally, we developed species sensitivity distributions (SSDs) for select EMs using toxicity data for all three soil ecological receptor groups (invertebrates, plants, and soil processes). These SSDs were then used for derivation of Soil Clean-up Values (SCVs). Benchmark data plus draft Eco-SSL values developed in these studies will be submitted to the USEPA Eco-SSL Work Group for use in establishing soil invertebrate- or plant-based Eco-SSLs for the individual EMs, and will be made available for use in Ecological Risk Assessment of terrestrial habitats at U.S. Army testing and training sites and other military locations. The SCVs can provide site managers and regulators with a risk assessment tool which allows them to select specific hazardous concentration (HC) values (e.g., HC5 or HC50 protection level) that they wish to use to derive a site-specific SCV protective of plants, soil invertebrates, and critical soil processes.

TH139

Organismal responses of oligochaetes in bacterial inoculum amended copper oxychloride spiked soils

M. Maboeta, North-West University / Unit for Environmental Sciences and Management; O. Oladipo, M. Engelbrecht, North-West University

The excessive release of heavy metals such as copper via anthropogenic sources into the soil environment has raised some global concern. Copper oxychloride, a common agricultural metal-based fungicide applied to vineyards and orchards for fungal control, contains 60% copper. This high copper content may significantly contribute to the soil copper burden and negatively affect the mesofauna. Metal-tolerant bacteria such as *Bacillus cereus* strain have been identified for their bioremediative traits in metal polluted soils. We examined the effect of *Achromobacter spanius - Bacillus cereus* consortium on the ecotoxicity of copper oxychloride ecotoxicity towards *Eisenia andrei* and *Enchytraeus albidus*. In this study, the bacterial strains used (*Achromobacter spanius* and *Bacillus cereus*) were previously isolated from gold and gemstone mining sites and confirmed to tolerate to 200 $mgkg^{-1}$ Cu. Twenty-four hours pure broth cultures of the two bacterial strains were inoculated into fungicide spiked soils. Utilizing standard ISO and OECD protocols, 10 mature *Eisenia andrei* and *Enchytraeus albidus* were exposed separately into bacterial inoculated copper oxychloridespiked soils (200 and 1000 $mgkg^{-1}$). Avoidance behavior, biomass, reproductive success, metal contents in soils and earthworm tissues were determined. Findings revealed that *E. andrei* in inoculated substrates (200 $mgkg^{-1}$) exhibited significantly higher ($p< 0.05$) preference, relative growth rate, survival, cocoon and juvenile counts and soil Cu content (comparable to the control) than non-inoculated soils. Similarly, with the *E. albidus*, significantly higher ($p< 0.05$) preference and reproductive success was recorded. However, at 1000 $mgkg^{-1}$ copper oxychloride soils, no distinct effect was observed on both *E. andrei* and *E. albidus* in bacterial inoculated and non-inoculated substrates. In conclusion, *Achromobacter spanius - Bacillus cereus* bacterial consortium decreased the ecotoxicity of metal-based fungicide towards *Eisenia andrei* and *Enchytraeus albidus* at 200 $mgkg^{-1}$ copper oxychloride concentration. These results further confirm the Cu tolerance potential of these bacterial strains at 200 $mgkg^{-1}$. *Achromobacter spanius* and *Bacillus cereus* are therefore recommended for the bioremediation of soil contamination of copper contaminated environments. **Keywords:** Copper oxychloride fungicide. *Achromobacter spanius - Bacillus cereus* consortium. Ecotoxicity. Oligochaetes

TH140

Development of a terrestrial biotic ligand model (TBLM) for predicting acute toxicity of cadmium and zinc to soil collembolan Paronychiurus kimi

J. Son, K. Cho, Korea University / Division of Environmental Science and Ecological Engineering

Complex interactions between metals and soil properties make it difficult to apply a biotic ligand model widely used in aquatic ecotoxicity studies. In this study, a terrestrial biotic ligand model (TBLM) was developed to predict the acute toxic effects of cadmium and zinc on the survival of soil collembolan *Paronychiurus kimi* in a simplified soil solution system under the assumption that soil pore water is the main route of exposure to metals. After 7 days of exposure, survival and internal metal concentrations in *P. kimi* were determined. The free metal ion activity for each metal was calculated by the Visual MINTEQ using inputs of soil metal concentrations, cation and anion components of the soil solution, and pH of the soil solution. The toxicity of cadmium and zinc was linked to the fraction of biotic ligand sites occupied by free metals (i.e. Cd^{2+} and Zn^{2+}). The results showed that the fraction of the biotic ligand occupied by metal can be used to predict the metal toxicity, indicating the applicability of TBLM to explain metal toxicity to *P. kimi* in a simplified soil solution. Although the approach used in this study may be limited to soil solution, the use of TBLM can be a useful tool for investigating factors affecting bioaccumulation and toxicity of metals.

TH141

Characteristics of metal-tolerant bacterial plasmids from a platinum mine tailings dam

T. Mahlatsi, C. Bezuidenhout, M. Maboeta, North-West University / Unit for Environmental Sciences and Management

The presence of mine tailings may promote the development of both heavy metal and antibiotics resistance among microbes with resistant plasmids. Plasmids provide their hosts with a large array of phynotypes such as heavy metals and antibiotics resistance due to gene transfer. This study describes the characteristics of plasmids isolated from various bacteria that displayed an ability to withstand high metal concentrations. Isolated plasmids were individually transformed into *Escherichia coli* JM109. Transformants were evaluated for metal tolerant capabilities using a microdilution approach where the plasmid DNA concentration ranged between 11.75-118.06 ng/µl after extraction. Incompatibility groups were determined by subjecting plasmids to PCR amplification using IncQ, IncP-9 and IncW specific primers, where only IncW provided positive results. Minimum inhibition concentrations (MICs) were carried out to determine the ability of transformed *E. coli* JM109 to tolerate metals at varying concentrations. Results indicated that transformed *E. coli* JM109 developed ability to grow in the presence of several heavy metals. Some strains were resistant to high concentrations (+10 mM) of Ni^{2+}/Al^{3+}, Pb^{2+} and Ba^{2+} with metal resistance order of Ni/Al=Pb>Ba>Mn>Cr>Cu>Co=Hg. Moreover, protein profiling was used to determine the impact of plasmids on *E. coli* JM109. Proteins were extracted from both transformed and un-transformed *E. coli* JM109 and subjected to one-dimensional (1D) and two-dimensional (2D) SDS- PAGE. One dimension SDS-PAGE illustrated general similarity of the profiles except for two banding positions in the 30 to 35 kDa region where bands were present in the transformants that were grown in the Al/Ni alloy containing media. Two-dimensional electrophoresis PAGE analysis showed that some of the proteins were up-regulated while others were down-regulated. The largest numbers of proteins were from 15 – 75 kDa. Since the plasmids rendered the *E. coli* JM109 tolerant to metals, it can be concluded that the change in the protein profiles was due to the effects of the plasmids. Furthermore, that plasmids isolated from various heavy metal-tolerant bacterial species were successfully transformed into *E. coli* JM109 rendering various new metal-tolerant *E. coli* JM109 strains. Plasmids isolated and characterized have advanced our understanding that these plasmids could be important reservoirs for resistant genes, and may hold significant biotechnology potential.

TH142

Sensitivity of the waterside species, Yuukianura szeptyckii (Collembola: Neanuridae), to cadmium and copper

Y. Lee, Korea University; J. Wee, J. Son, Korea University / Division of Environmental Science and Ecological Engineering; Y. Kim, Korea University; K. Cho, Korea University / Division of Environmental Science and Ecological Engineering

Collembola is the most abundant organism in the soil ecosystem and some species are used as ecotoxicological evaluation species for toxic substances in soil. However, Neanuridae species, which is one of the family of Collembola, are rarely studied as toxicity evaluation species. In this study, the toxicity sensitivity of copper and cadmium of *Yuukianura szeptyckii*, known as the species in which they live water side, and their bioaccumulation amount were examined based on the ISO guideline 11267. After 28 days of exposure to tested metals, LC50 for adult survival and EC50 for reproduction were estimated. These toxicity values of *Y. szeptyckii* were also compared to those of other collembolan species (*F. candida* and *Paronychiurus kimi*) reported in literature to investigate their suitability as a new test species in toxicity test. Not only the adult survival but also the juvenile production of *Y. szeptyckii* was decreased in a concentration dependent manner after 28 days of exposure duration. Although the response of *Y. szeptyckii* to the tested metals was not highly sensitive to the other collembolan species reported in literature, the study of the response of *Y. szeptyckii* to chemicals in the soil is considered to be very important. Because their special habitat can provide an understanding of ecotoxicity against certain environmental conditions.

TH143

Drivers of copper and zinc availability and phytoavailability in agricultural soils receiving long-term organic waste amendments

C. Laurent, CIRAD - Centre de coopération internationale en recherche agronomique pour le développement / PERSYST-UPR Recycling and risk; M. Bravin, CIRAD; C. PELOSI, INRA (Institut National de la Recherche Agronomiqu; O. Crouzet, INRAAgroParisTech; I. Lamy, INRA / UMR ECOSYS
Organic wastes (OW) are used as soil amendments and fertilizers but they are also the major source of copper (Cu) and zinc (Zn) contamination in agricultural soils. The potential ecotoxicological effects of this contamination on soil organisms depend on Cu and Zn availability in soils. The availability of Cu and Zn itself depends on their chemical speciation and consequently to the temporal evolution of soil parameters such as pH and organic carbon content and reactivity. These soil parameters are key parameters both influenced by the application of OW and the activity of soil organisms on the surrounding soil, i.e. the rhizosphere for plants. However, the concomitant influence of OW applications and rhizosphere effect are poorly documented when taking into account long-term impacts. Accordingly, we aimed at studying the relationship between the availability in soil and the phytoavailability of Cu and Zn in four decadal field trials that received different types of OW for more than ten years. Soils in the four field trials exhibited very different pH and organic carbon content. Copper and Zn availability was determined on 102 soil samples from the four field trials by (i) an equilibrium-based method using cupric ion selective electrode and the windermere humic aqueous model (WHAM) to quantify Cu^{2+} and Zn^{2+} activities in soil solutions (pCu^{2+} and pZn^{2+}) and (ii) a kinetic method using the diffusive gradient in thin films (DGT) directly on soil samples. We measured key soil parameters in soil solutions to assess the relationship with pCu^{2+} and pZn^{2+}. Copper and Zn phytoavailability is currently determined using the RHIZOtest which is a standardized biotest that will enable to measure the uptake flux of Cu and Zn in plants and the related availability of Cu and Zn in the rhizosphere that is physically separated from roots. The results already achieved showed no clear relationship between pCu^{2+} and pH or dissolved organic carbon among the four field trials altogether. When studying each trial separately, we observed a pH gradient as a function of the type of fertilizer (mineral or OW) applied, which influence substantially the speciation of Cu in soil. The on-going measurements will enable to test whether Cu and Zn availability in unplanted soils determined Cu and Zn phytoavailability or whether root-induced chemical changes in the rhizosphere additionally determined it.

TH144

Toxic Effects of Cadmium on Chinese Cabbage , Folsomia Candida (collembola) and their Prediction Modes in 18 Soils of China

L. Zheng, Y. Feng, Y. Zhou, Nanjing Insitute of Environmental Sciences

In this paper,we adopted 18 Kinds of typical soils in China,and Chinese cabbage ,folsomia candida(collembola) were used as the research object.The germination and root elongation of cabbage under different concentration of cadmium in soil were measured. The endpoint of the *F. candida* was reproduction.The results show that the soil properties significantly affected the dose effect curve of cadmium, soil pH is the main influencing factor; at the same time, we calculated the toxicity threshold and prediction models. This study has a guiding significance for the plant and invertebrates ecological risk prediction and assessment of heavy metal cadmium.

TH145

Do we use plant protection products correctly? Impact of agrochemicals on non-target beetle, Bembidion lampros (Coleoptera: Carabidae)

J. MOKKAPATI, Institute of Environmental Sciences, Jagiellonian Univeristy / Ecotoxicology and Stress Ecology; G. Sowa, Institute of Environmental Sciences Jagiellonian University; A. Bednarska, Polish Academy of Sciences / Institute of Nature Conservation; R. Laskowski, Jagiellonian University / Ecotoxicology & Stress Ecology Group

Intensification of agriculture and the widespread use of pesticides during the last few decades have led to significant reduction of the abundance of non-target arthropods (NTA), including the ground beetles (Carabidae), which are natural pest enemies in agricultural areas. Due to the growing demand for food, it is not possible at the moment to stop using pesticides. We need, therefore, to make every effort to ensure that they are used in a way that do not jeopardize NTA. In the present study, three commonly used pesticide formulations: Dursban 480 EC, containing the organophosphate insecticide chlorpyriphos (CPF), Sherpa 100 EC, containing the pyrethroid cypermethrin (CYP), and Spekfree 430 SC, containing the fungicide tebuconazol (TEB), were tested for their effects on survival of the ground beetle *Bembidion lampros*. The beetles were collected from agricultural fields either in spring (after overwintering) or autumn (population dominated by newly emerged individuals) and exposed individually to a single pesticide spray applied with the Potter tower. In terms of recommended field dose (RFD), Dursban appeared almost 10 times more toxic than Sherpa: the 96-h LD_{50} for Dursban was 0.057 (CI 0.048-0.071) and 0.054 (CI 0.046-0.066) RFD for spring and autumn beetles respectively, and for Sherpa − 0.556 (CI 0.453-0.704) and 1.003 (CI 0.863-1.214) RFD respectively. However, the toxicity of both insecticides was almost identical in terms of their active ingredients (g a.i. ha^{-1}) – the LD_{50} for CPF was 16.4 for spring and 15.6 for autumn beetles, and for CYP 16.7 and 30.1, respectively. The beetles survival rate decreased significantly with increasing dose of both insecticides, but the spring-collected beetles appeared more sensitive, plausibly explained by their overwintering or ageing. In contrast to insecticides, tebuconazol caused significant increase in survival at higher doses, possibly due to its interference with immune competence of insects or elimination of pathogenic fungi. The results show that at least some insecticide formulations may cause unacceptable effects on NTA when applied according to recommendations, indicating the urgent need for revising current pesticide usage recommendations. The differences in sensitivity between the spring and autumn-collected beetles call for further studies to see whether such seasonal differences can be important for ERA. This study was supported by National Science Centre, Poland (2015/19/B/NZ8/01939)

TH146

The fate and bioavailability of currently used and emerging pesticides in agriculturally used fluvisols - effects of soil and pesticide properties

M. Šudoma, N. Neuwirthová, Masaryk University; M. Svobodová, Masaryk University / Research Centre for Toxic Compounds in the Environment (RECETOX); L. Bielská, RECETOX, Faculty of Science, Masaryk University / Faculty of Science, RECETOX; M. Hvezdova, Z. Simek, L. Skulcova, Masaryk University / Research Centre for Toxic Compounds in the Environment (RECETOX); K. Brandstätter-Scherr, University of Natural Resources and Life Sciences / Institute for Environmental Biotechnology, Department for Agrobiotechnology (IFA-Tulln); J. Hofman, Masaryk University, RECETOX / Faculty of Science, RECETOX

The class of conazole fungicides (CFs), among them epoxiconazole, tebuconazole, flusilazole and prochloraz are currently used pesticides and members of the triazole group, used as broad-spectrum fungicides that inhibit ergosterol biosynthesis and are typically applied as foliar sprays for cereals, sugar beet or oilseed rape. Conazole fungicides are widely used in EU countries and their residues are frequently found in European arable soils which corresponds to their environmental properties. CFs are strongly sorbed to soil ($logK_{OC}$ of 3–4) and have low to moderate water solubility (S_W of 7–150 mg/L).They are very persistent in soils and tend to form long-term residues as their typical DT_{50} values range from 120 days to 1 year. These attributes predetermine them to be highly bioaccumulative and hazardous. However, in real ecosystems, complex interactions occur (between pesticides, soil, microbes, earthworms, plants...) and these are poorly understood. Hence, in this contribution (poster), we would like to present the novel microcosm experiment, where the combined effects of soil properties, microorganisms, plants and earthworms on CF multimedia fate and bioavailability were evaluated. In particular, the CF fate (by means of total, desorbable and freely dissolved concentrations) and bioavailability (by means of uptake to model fauna, flora and passive samplers) is studied in complex microcosm systems consisting of agriculturally used fluvisols under the addition of selected model compounds (epoxiconazole, tebuconazole, flusilazole, prochloraz) at background levels (0.5 mg/kg), seeding plants (*Lactuca sativa*), earthworms (*Eisenia fetida*), SPME passive sampling fibers, Silicon rubber sheets and Chemcatcher® passive samplers. A subset of 10 fluvisols was selected based on the DRIFT mid infrared portion using the Kennard-Stone algorithm. These 10 soils are representative of a large fluvisols range in terms of their qualitative and quantitative SOM properties (TOC, DOC, HA,FA, WHC, pH, texture, etc.).

TH147

A Field Trial to Determine Effects of Thiamethoxam treated Sugar Beet Seed on the Non-Target Arthropod Fauna of Arable Land in The Netherlands

C. Elston, Syngenta Ltd / Product Safety; M. Coulson, Exponent International Limited; F. Bakker, EurofinsMitox; P. Thorbek, Syngenta / Environmetal Safety; M. Finnegan, Syngenta

The aim of this study is to assess the potential effects of thiamethoxam, applied as a seed coating to sugar beet, on the full fauna of naturally occurring non-target arthropods (NTAs) in a commercial arable field in The Netherlands when compared to a non-insecticidal control treatment. This is a three year study which began in March 2017 with the drilling of the sugar beet seed at two different seed treatment rates equivalent to a typical sugar beet seed loading and oil seed rape seed loading using plots of 2 ha each organized in 4 blocks of 8 ha each (32 ha total study area). NTA field studies are important for investigating impacts of pesticides on populations, communities and different life stages of NTAs under realistic exposure conditions, however such studies are challenging to design and conduct. In order to assess the regulatory acceptable risk of a pesticide to NTAs in-field a study should be designed in a way that enables an adverse effect to be detected if present. A toxic reference is used to show the test design is adequate to demonstrate persistent adverse treatment-related effects. Many of the chemicals historically used as toxic reference items with large historical datasets are now not available due to changing regulations, making it difficult to select a suitable toxic reference. Conducting such a study requires multiple sampling methods such as pitfall traps, mine traps, soil cores and sweep nets to account for different life histories of NTA species and a team of qualified taxonomists to identify all organisms. In this study NTA populations will be monitored for a three year period that covers at least two generations to enable the detection of any trans-generational effects that might occur. The current EU risk assessment scheme considers that effects on populations are acceptable for the in-field area above the threshold value of 50% if recovery or potential for recovery is demonstrated within 1 year. This study has been designed to enable the assessment of: (1) the magnitude of treatment effects on non-target arthropod (NTA) populations, (2) the range of NTA taxa affected by the treatment and (3) the duration of treatment effects and the time period until populations recover. The evaluation will be based on (1) time to recovery (population density similar to control) and (2) persistence of effect (population growth similar to the control). Effects will be classified in accordance with DeJong et al. 2010.

TH148

Bioaccumulation kinetics of pesticides chlorpyrifos and tebuconazole in the earthworm Eisenia andrei in two different soils

M. Svobodová, Masaryk University RECETOX; K. Smidova, Masaryk University RECETOX / Research Centre for Toxic Compounds in the Environment (RECETOX); M. Hvezdova, Masaryk University / Research Centre for Toxic Compounds in the Environment (RECETOX); L. Bielská, RECETOX, Faculty of Science, Masaryk University / Faculty of Science, RECETOX; J. Hofman, Masaryk University, RECETOX / Faculty of Science, RECETOX

This study was conducted to investigate the bioaccumulation of two pesticides currently used in large amounts in the agriculture, the insecticide chlorpyrifos and the fungicide tebuconazole. The two compounds have distinct properties suggesting their different fate in the soil and bioaccumulation. Their detailed uptake kinetics in the model earthworm species *Eisenia andrei* were measured to find a sufficient length of exposure to achieve equilibrium in concentration between soil and earthworm and to compare two pesticides in two arable soils differing in the organic carbon (1.02 and 1.93% respectively) and clay content (10.7 and 20.7% respectively). Concentrations of the pesticides in soils and earthworms were determined by LC-MS/MS after QuEChERS extraction which has shown to be rapid, simple and effective approach to determine broad spectrum of pesticides in soil and earthworm samples. According to our results, a steady state was reached after 3 to 5 days for both pesticides and soils. The values of bioaccumulation factors calculated at the steady state ranged from 4.5–6.3 for chlorpyrifos and 2.2–13.1 for tebuconazole. Bioaccumulation factors were also calculated as the ratio of uptake and elimination rate constants with results comparable with steady-state bioaccumulation factors. The results suggested that the degradation and bioaccumulation of tested compounds by earthworms was influenced by more factors than only the organic carbon content in soils. The clay content also probably contributed, namely to degradation of chlorpyrifos by clay-catalysed hydrolysis and to decreased bioavailability of tebuconazole by binding to clay minerals. The lower K_{oc} and hydrophobicity of tebuconazole relative to chlorpyrifos probably led to higher availability of tebuconazole through pore water exposure. On the other hand, higher hydrophobicity of chlorpyrifos probably caused an increase in availability by its additional uptake via ingestion.

TH149

Effects of diuron and imidacloprid on eight nematode species

J.N. Neury-Ormanni, Irstea / EABX-CARMA; C.N. Doose, INRS - Centre Eau Terre Environnement; N. Majdi, Ecolab / UMR 5245 CNRS; J. Vedrenne, S. Morin, Irstea Bordeaux / UR EABX; S. Höss, Ecossa / Animal Ecology; W. Traunspurger, Bielefeld University / Animal Ecology

To assess the lethal effects of diuron (herbicide) and imidacloprid (insecticide) on ubiquist organisms at the basis of food webs, we performed multispecies toxicity tests using nematode species commonly found in soil and freshwater benthic ecosystems. Diuron and imidacloprid belong to the top 15 of the most frequently detected pesticides in French rivers. Both chemicals show an elevated DT50 (time to 50% degradation) in sediments, about 130 days for imidacloprid and more than 30 days for diuron. A standardized toxicity test with *Caenorhabditis elegans*, a representative for the response of nematode species to toxic stress, showed no significant response to imidacloprid at high concentrations up to 119 mg L^{-1} regarding growth or reproduction. Diuron inhibited 82% of reproduction success of *C. elegans*, but showed no significant effect on growth at high concentration (33 mg L^{-1}). Then, we compared the lethal effects of diuron and imidacloprid on eight species of free-living nematodes: *Aphelenchoides sp, Caenorhabditis elegans, Pristionchus pacificus, Diploscapter coronatus, Rhabditoides sp, Plectus velox, Plectus opisthocirculus, Plectus acuminatus*. Nematodes were exposed in water for 48h to two concentrations (35 and 350 mg L^{-1} for imidacloprid and 10 and 100 mg L^{-1} for diuron). Results indicated a low risk of these pesticides to nematodes, as the chemicals did not affect significantly the survivorship at their solubility limit in water for every tested species.

TH150

Multigeneration effects of pentachlorophenol and 2,2',4,4'-tetrabromodiphenyl ether on Folsomia candida

M. Qiao, Q. Zhang, Research Center for Eco-Environmental Sciences, Chinese Academy of Sciences,

The multigeneration effects of pentachlorophenol (PCP) and 2,2',4,4'-tetrabromodiphenyl (BDE47) on the springtail Folsomia candidawere evaluated. Multigenretion tests were performed in accordance to two different test methods. In the first method, the parental generation springtails (F0) were exposed to PCP or BDE47 for 28 days. The first filial generation (F1) springtails were transferred to unpolluted artificial soil for 28 days and reproduced the second filial generation (F2). In the second method, the F0 generation were exposed for 10 days and then transferred to unpolluted artificial soil to generate the F1 generation. The F1 generation were also transferred to unpolluted artificial soil for 28 days and reproduced the F2generation. For PCP, significant effects were observed on F1 and F2 generation in the first method and F1 generation in the second method. This suggests that PCP influences the reproductive capacity of adult springtails and the hatching of eggs or the mortality of juveniles.For BDE47, significant effects were only observed on F1 generation in the first method, which shows that BDE47 affects egg hatching or juvenile survival rather than the reproductive capacity of adults. The affected endpoints of springtails can be inferred by the two methods. PCP and BDE47 do not influence completely the same endpoints.

TH151

Bioaccumulation of lead in earthworms: a comprehensive study to derive a biota-to-soil accumulation factor (BSAF) for risk assessment

K. Oorts, ARCHE; J. Chowdhury, International Lead Association / Senior Scientist -Environment

Secondary poisoning to mammals and birds is a critical pathway for risk assessment of Pb in soil. This risk is generally assessed for the food-chain soil => earthworms => earthworm eating predators. Therefore, a correct evaluation of bioaccumulation of Pb in earthworms is essential for risk assessment of Pb in soils under the REACH Regulation and a literature review of biota-to-soil accumulation factors (BSAF) for Pb in earthworm species was made. To ensure that biota Pb burdens are in equilibrium with soil Pb concentrations, only data from field studies or laboratory studies using soil and biota collected at the same field site were considered. Data from laboratory studies where Pb was added to the soil as a Pb salt were only used as supporting evidence. In total, 248 BSAF values for earthworms were identified that meet the reliability criteria, ranging from 0.01 to 22.05 (dry weight basis, median 0.23). Results are available for several earthworm species, belonging to different ecological groups of earthworms: anecic, endogeic and epigeic earthworms. No distinct differences in BSAF values across these groups could be identified. BSAF values are derived in a wide range of soils and the data available can be considered as representative for soils in Europe. Only the cation exchange capacity (CEC) is significantly correlated with BSAF values. No significant correlation with Pb content, pH, organic carbon content or clay content is observed. The significant negative regression between log BSAF and log CEC was confirmed by laboratory studies and is also consistent with the significant decrease in Pb toxicity observed for *Eisenia fetida* reproduction with increasing CEC of the soil. It was concluded to implement the effect of soil properties on BSAF by using the overall regression between log CEC and log BSAF in the risk assessment of Pb in soil. This yields a generic BSAF of 0.30 on dry weight basis, corresponding to 0.048 on a fresh weight basis, for the median eCEC value of 16 $cmol_c$/kg soil for European arable soils. BSAF values on fresh weight basis vary from 0.089 to 0.028 for soils with an eCEC of 8 and 30 $cmol_c$/kg soil, respectively, corresponding to the 10^{th} and 90^{th} percentile of eCEC in European arable soils. Implementing effect of soil properties on BSAF improves consistency between assessment of secondary poisoning and direct toxicity of metals to soil organisms, where bioavailability corrections for varying soil properties are commonly accepted.

TH152

Hazard assessment of liquid organic hydrogen carriers in terrestrial environment

Y. Zhang, Dresden University of Technology / UFT Centre for Environmental Research and Sustainable Technology Department Sustainable Chemistry; M. Markiewicz, University of Bremen / Centre for Environmental Research and Sustainable Technology; S. Stolte, University of Bremen / UFT Centre for Environmental Research and Sustainable Technology Department Sustainable Chemistry

A proactive environmental hazard assessment of liquid organic hydrogen carriers (LOHCs) – a novel energy system which can efficiently and relatively safely store and transport hydrogen – was conducted by characterisation of potential behaviours and ecotoxicities of these chemicals in soil environment. Adsorption properties of promising LOHC candidates including indoles, quinaldines, carbazole derivatives, benzyltoluenes and dibenzyltoluene in terms of carbon-water partition coefficients (Koc) were investigated via HPLC screening. Further characterisation was performed via adsorption isotherm modeling and soil column leaching with the extrapolation of soil-water partition coefficients (Kd) using the quinaldines (Quin-2Me) as examples. Soil ecotoxicity was estimated for the quinaldines in the soil bacteria *Arthrobacter globiformis* and Collembola *Folsomia candida* in pore-water and soil exposure scenarios. The log Koc values generally increased following indoles < quinaldines < carbazole derivatives < benzyltoluenes < dibenzyltoluene. The mobility of LOHCs was thus classified as highly mobile, moderately mobile or immobile. Adsorption isotherm and column leaching showed the strongest adsorption and retention of the partially hydrogenated form (Quin-2Me-pH) in soils. The H_2-rich form (Quin-2Me-H10) appeared the highest leaching capacity through the soil followed by the H_2-lean form (Quin-2Me) implying the risk of groundwater contamination. Ionic-interaction was considered dominant in the adsorption of Quin-2Me-H10 to soils given its high protonation at the soil pH; while hydrophobicity was the main force in the adsorption of its two analogues. No or only slight toxicity was found for the quinaldines in the *Arthrobacter* at the highest test concentrations (500 mg L^{-1} and 750 mg kg^{-1} dry weight (dw) soil). Higher toxicity was found in the Collembola and malformations of cuticle in the pore-water scenario were observed. Dose-response modeling showed $10 < LC_{50} \leq 100$ mg L^{-1} (liquid-only exposure) and $100 < EC_{50} \leq 1000$ mg kg^{-1} dw soil (calculated soil pore-water based) of the quinaldines assigning these chemicals to category "harmful" to soil organisms. Predicted no-effect concentrations showed 1–3 orders of magnitude higher the effective concentrations than the former suggesting potential risks of the chemicals toward the soil environment and proper monitoring is needed in the application of the LOHCs. Key words: adsorption, bioavailability, hazard assessment

TH153

Combining field measurements and biotest to assess lead and zinc phytoavailability in contaminated urban soils

M. Bravin, C. Chevassus-Rosset, CIRAD; L. Lemal, MetRHIZlab; M. Montes, G. Moussard, E. Simon, M. Tella, CIRAD; M. Valmier, MetRHIZlab; E. Doelsch, CIRAD / UPR Recyclage et risque; F. Feder, CIRAD; S. Legros, CIRAD / LITEN

Along with the French legislation on the recycling of wastes from wastewater treatment plants, a guideline was provided to stakeholders to proof the low mobility and phytoavailability of trace elements exceeding total concentration thresholds in soil. Due to the lack of adequate plant biotest at the time the guideline was published (i.e. in 2005), the guideline suggests to measure trace element phytoavailability in the aerial parts of plants collected in situ in contaminated and uncontaminated soils. The present study aimed at applying the guideline methodology with the combination of a recently developed plant biotest (i.e. the RHIZOtest) and field measurements to lead (Pb) and zinc (Zn) contaminated urban soils on which irrigation with treated wastewater was foreseen. Ten contaminanted and uncontaminanted soil samples (hereafter referred to as soil) were collected in representative sites expected to be irrigated with treated wastewater. The phytoavailability of Pb and Zn was estimated on each soil by measuring Pb and Zn concentration in the aerial parts of field-collected plants and by deploying the RHIZOtest and measuring the uptake flux of Pb and Zn in the whole plants exposed to soils. As expected, field-collected plants exhibited a large range of Pb concentration in leaves, irrespective of total Pb and Zn concentrations and Pb and Zn mobility and phytoavailability measured in soils in the first step. In comparison, RHIZOtest measurements showed that only the contaminated soil 15 exhibited a significantly higher phytoavailbility than other soils and have consequently to be rejected for irrigation with wastewater. This study thus showed how the use of a biotest dedicated to the measurement of trace element phytoavailability in combinaison with field measurements was useful to assess the risk of high phytoavailability in contaminated urban soils.

TH154

Can approaches beyond the traditional ones characterizing the effects on soil microflora provide an added value in the scope of regulation?

K. Hund-Rinke, Fraunhofer IME / Department of Ecotoxicology; A. Huemmler, Fraunhofer IME; K. Schlich, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology; F. Wege, Fraunhofer IME; G. Broll, University of Osnabrueck / Institute of Geography

According to the current regulation, side effects of chemical substances on the soil microflora focus on the determination of the nitrogen transformation (OECD 216). However, according to EFSA a more comprehensive risk assessment is required. We investigated whether a combination of several test approaches addressing various microbial aspects results in a better understanding of the fluctuation of the soil microflora after a threat and a more comprehensive risk assessment taking the new requirement regarding the consideration of ecosystem services and the protection of the biodiversity into account. We used a silver nanomaterial as example and applied three functional approaches to get information on the functional microbial diversity: (i) potential ammonium oxidation activity (PAO) addressing a small, mainly autotrophic bacterial community with comparable low diversity; (ii) respiration activity of the heterotrophic microflora as indicator for the C-transformation with basal respiration activity and activity in the presence of carbon sources (glucose, cellobiose) as well as sulfur or nitrogen containing organic substances (cysteine, alanine); (iii) activity of selected exoenzymes (selection based on the carbon sources used in the second approach). The second and third approach were performed in microwell plates. The three functional approaches (i) PAO, (ii) C-transformation, (iii) exoenzymes seem to be a suitable assessment tool and can provide a benefit in the assessment of chemicals. The combination of results was dependent on the test concentration. The exoenzymes were the most sensitive indicator and seem to be a suitable early warning indicator. An increased concentration of the chemical responsible for the initial effect or a further impact can severely affect the microbial population. Additionally affected nitrifiers indicated a stronger damage. Effects in all three approaches indicated a severe impact. The high sensitivity of the exoenzymes in contrast to the respiration activity of the microbial cells could be due to their location outside the microbial cell and a lower protection level. However, also the small size of the ions as affecting substance has to be considered. In further experiments, the combination of results obtained with the three functional approaches have to be determined with additional chemicals. Due to the use of microwell plates the additional work load seems to be acceptable also for testing in the scope of regulation.

TH155

Re-calibration of the earthworm Tier 1 risk assessment of plant protection products - an update

G. Ernst, Bayer Ag / Ecotoxicology; J. Bendall, Dow Agrosciences; T. Carro, FMC; H. Cunningham, Syngenta / Environmental Safety; A. Koutsaftis, ADAMA; S. Loutseti, DuPont De Nemour Hellas S.A.; M. Marx, Bayer AG Crop Science Division / Environmental Safety - Ecotoxicology; B. O'Neill, DuPont Crop Protection; A. Sharples, FMC Agricultural Solutions; F. Staab, BASF SE

The conservatism in tier 1 earthworm risk assessment for plant protection products (PPP) is expected to increase due to revision of the PEC_{soil} modeling guidance. The new EFSA guidance foresees to use worst case PEC_{soil} values for each European regulatory zone considering a lower soil bulk density, a lower organic carbon content, and a reduced crop interception rate due to consideration of worst case wash-off assumptions. Furthermore, several different soil layers for which PEC_{soil} values could be calculated are under discussion, i.e. 0-1 cm, 0-2.5 cm, 0-5 cm, and 0-20 cm soil depth. Calculated PEC_{soil} values based on the new EFSA guidance are estimated to strongly increase, which might lead to an overly conservative tier 1 risk assessment. In a project published by Christl et al. (2016), results from standard laboratory earthworm reproduction tests are compared with the effect levels in higher tier studies for a representative set of 54 case studies compiled by ECPA companies. In this exercise, the relevant soil layer for PECsoil modeling and assessment factor were adjusted to result in tier 1 protection of unacceptable field study effects. The results of this evaluation give clear evidence that considering a layer of 0-5 cm in combination with the currently used assessment factor of 5 would lead to an appropriate earthworm tier 1 risk assessment (Christl et al. 2016). However, the lower tier earthworm risk assessment also changed in Europe regarding correction of laboratory endpoints for lipophilic compounds (logP >2). A correction of endpoints by a factor of 2 is proposed by EFSA (2015, EFSA Supporting publication 2015:EN-924) for studies containing artificial soil with 5% peat (formerly only endpoints from studies with 10% peat were corrected for high logP). Furthermore, in its Scientific Opinion, EFSA (2017) proposed Specific Protection goals for earthworms which include a maximum acceptable recovery time of 6 months for initial effects in field studies. This deviates from the current procedure of an acceptable recovery time of one year for earthworm populations. The dataset of 54 case studies was re-evaluated considering the new EFSA proposals and the new results will be presented.

TH156

Digging into the soil risk assessment of pesticides: current approach and its uncertainty

M. Arena, EFSA - European Food Safety Authority / Pesticides; D. Auteri, s. barmaz, EFSA - European Food Safety Authority / Pesticides Unit; S. Pieper, German Federal Environment Agency (UBA) / Plant Protection Products

According to the Uniform Principles (Reg No 546/2011) in the context of Regulation (EC) No 1107/2009, all possible sources of uncertainties should be considered when performing a regulatory assessment in the context of pesticides authorization. The current risk assessment for soil organisms, conducted according to SANCO/10329/2002, foresees, at Tier 1, the application of a trigger value of 5 covering uncertainty linked to e.g. intra- and interspecies variability and the extrapolation of toxicity endpoints from lab- to- field. However, the current approach presents additional uncertainties. Test methodology for soil organisms only requires dosing verification after the application of the pesticide to the soil. The determination of the tested concentration at regular intervals is currently not required although it may be very relevant for a proper hazard characterization (e.g. bioavailability), since, for example, during laboratory handling procedures of the spiked soils, possible losses of the pesticide may occur. In case further refinements of the risk are triggered, higher tier tests (semi-field or field studies) under more realistic conditions may be one option. Standardised field protocols are mainly available for earthworms. The available standardised field methods evaluate the effects on abundance and biodiversity of earthworms, taking into consideration the likely level of effects, the species/groups affected, population recovery (within 1 year) as well as information on the application and fate of the pesticide. The magnitude of effects is directly assessed in terms of risk without the application of any assessment factor. However, field studies only give a picture of a particular situation as effect manifestation and recovery are dynamic processes which depend on the local situationand time-scale. Additionally, an assessment at community level which takes into account interspecies interactions and indirect effects is currently not implemented as well as the statistical power of test is not properly evaluated. An approach aimed at defining a Regulatory Acceptable Concentration (RAC) could be useful to address those uncertainties and would allow the harmonisation of the risk assessment of the different taxonomic groups.

TH157

SETAC Soils Interest Group

M.H. Wagelmans, Bioclear earth

Natural toxins and harmful algal blooms (HABs): water and food safety, analysis, toxicity, and risks (P)

TH158

A novel analytical method for simultaneous quantification of Bracken fern produced carcinogenic ptaquiloside-like compounds and their derivatives

V. Kisielius, Metropolitan University College; H. Hansen, University of Copenhagen / Department of Plant and Environmental Sciences; M. Rodamer, Agilent Technologies; D. Lindqvist, L.H. Rasmussen, Metropolitan University College

Significant variety of toxic secondary metabolites produced by plants appear in chemical structures of glycosidic molecules. These compounds are highly water soluble and mobile in soils and sediments. In cases when toxic glycosides are persistent and released in high loads from vegetation, evaluation of the risk to aqueous environments adjacent to drinking water supplies is needed. Nevertheless, the environmental distribution and fate of such natural toxins are largely unknown. This study aims to identify the environmental risk factors that predetermine release of glycosidic natural toxins from non-agricultural lands to aqueous ecosystems. Bracken ferns (*Pteridium aquilinum*) are known to produce up to 6 kg/ha of carcinogen ptaquiloside. Previous studies demonstrate leaching of ptaquiloside from Bracken to soils and upper ground waters. The ptaquiloside-like compounds – ptesculentoside and caudatoside – have recently been studied in Australian Brackens. Except from a few positive samples included in the Australian study, there have been no reports of these compounds in Europe. We hereby report a novel method for quantification of ptaquiloside, caudatoside and ptesculentoside and their respective pterosin-derivatives (6 compounds in total) to be used for the abovementioned study. The novel LC/MS method (Agilent 1260 Infinity HPLC System; Agilent 6130 Single Quadrupole MS; Agilent InfinityLab Poroshell 120 EC-C18 semi-UPLC column (3.0x50 mm, 2.7 μm)), enables simultaneous determination of all 6 compounds with low limits of detection (1 ng/l) using loganin as an internal standard. The total time of analysis is 6 minutes and the system is operated under semi-UPLC conditions with a max. pressure of 400bar. Mobile phase with a low fraction of acetonitrile is applied (10% v/v). These features are favourable for high-throughput analysis and could be practically utilised in, e.g. water supply facilities. The method will be applied for studies of the spatial and temporal variation of the 6 compounds in in plants, soils and surface waters. The project is part of the European Training Network *NaToxAq,* investigating the natural toxins in waters from the perspectives of their physio-chemical properties, spatial and temporal variation, health risks and concepts of water treatment operations for their removal (Horizon 2020 Research and Innovation Programme - Marie Sklodowska-Curie, grant agreement No. 722493).

TH159

A novel method for ptaquiloside and pterosin B preservation in groundwater samples

N. Skrbic, University of Copenhagen / Plant and Environmental Sciences; S.C. Christensen, A. Pedersen, HOFOR A/S, Copenhagen; H. Hansen, University of Copenhagen / Department of Plant and Environmental Sciences; L.H. Rasmussen, Metropolitan University College

Analyzing natural toxins in groundwater is challenging due to their labile and unstable nature. Ensuring sample integrity for analyses is a critical step to facilitate trustful findings, and appropriate preservation methods need to be developed. This research focuses on the development of a preservation technique for ptaquiloside (PTA) and its degradation product pterosin B (PTB) in groundwater. Ptaquiloside is a carcinogenic compound produced by one of the five most common plants on the planet, Bracken fern (*Pteridium aquilinum*). It is highly water-soluble with almost no sorption to soil and sediment, and hence leaches to the aqueous environment. In turn, PTA can potentially contaminate groundwater, which presents a concern for human health if used as a drinking water source. Ptaquiloside is chemically unstable under acidic and alkaline conditions, making it difficult to collect and preserve for analysis. Thus, a controlled and well-designed preservation procedure that can ensure its stability for the subsequent analyses is necessary. In order to develop a technique for preservation of PTA and PTB in groundwater samples, a Plackett-Burnman experimental design is applied. This approach allows assessing the influence of a number of independent factors such as sample bottle type, test time, water type, pH, temperature and transportation conditions by a reduced number of experiments. In each of the experiments, a water sample with known concentration of PTA and PTB was treated with a predefined factor set, and a recovery percentage of the compounds were evaluated by LC-MS system. This led to an optimal combination of factors for the preservation of the compounds of interest. We also performed robustness and range tests to quantify the precision, accuracy and linearity of the method. The optimized technique was further validated by applying it at field sites covering different groundwater types and different spiked toxin concentrations. By developing this method, we facilitate reliable investigation and monitoring of PTA and PTB in groundwater. In that way, we contribute both to the scientific discourse on the topic as well as offer a practical tool for water supply companies. This research project is part of European Training Network - NaToxAq, which is funded by the European Union's Horizon 2020 research and innovation programme under the Marie Sklodowska-Curie grant agreement No 722493**.**

TH160

Harmful algal bloom smart device application: using image analysis and machine learning techniques for classification of harmful algal blooms.

J.M. Lazorchak, U.S. EPA / Office of Research and Development; M. Waters, Northern Kentucky University / Mathematics Statistics; M. Steinitz Kannan, Northern Kentucky University / Biological Sciences Department; H. Mayfield, Foundation for Ohio River Education; J. Allen, U.S. EPA / Office of Research and Development

Northern Kentucky University and the U.S. EPA Office of Research Development in Cincinnati Agency are collaborating to develop a harmful algal bloom detection algorithm that estimates the presence of cyanobacteria in freshwater systems by image analysis. Green and blue-green algae exhibit different Hue-Saturation-Value color histograms in digital photographs. These differences are exploited by machine learning techniques to train a smart device (cellular phone, tablet, or similar) to detect the presence of cyanobacteria in a small surface portion of a freshwater system. The Harmful Algal Bloom Classification Application (HAB APP) has been field tested and verified to classify both green and blue-green algae. Specifically, the APP has been tested on several small streams and ponds, correctly classifying green algal blooms and has been tested on the Ohio River, correctly classifying blue-green algae in the 636-mile cyanobacteria bloom in summer 2015. The application is being tested via fixed camera monitoring stations and optimized at several locations along the Ohio River and in Lake Harsha, a 22,000-acre reservoir which supplies six million gallons per day of drinking water to the Ohio county in which it lies and is a source of many recreational activities, including swimming, boating, and fishing. The presence will be verified by other detection instruments and *in vitro* by agency scientists and hysteresis techniques will be used to monitor the presence of cyanobacteria on a periodic (*e.g.* daily, seasonally) basis at the monitoring stations. Further, the APP is being extended to classify harmful algae microscopically at the genus level using a convolutional neural network approach.

TH161

Matrix-assisted laser desorption/ionization-time of flight mass spectrometry application for rapid screening of microcystins occurrence in northern Taiwan tap-water reservoirs

W. DING, National Central University / Department of Chemistry

Microcystins (MCs) are the most common hepatotoxins and tumour promoters produced by freshwater cyanobacteria. Due to the damaging the liver through inhibition of protein phosphatases 1 and 2A, they pose a serious health threat to humans and animals, and even inducing death. MC-LR and MC-YR are probably the most concern and toxic microcystins. They are also widely distributed and detected in the freshwater system worldwide. In this study, matrix-assisted laser desorption/ionization–time of flight mass spectrometry (MALDI-TOF-MS) technique was developed for the rapid screening of these two MCs in two tap-water reservoirs in northern Taiwan. In addition, solid phase extraction (SPE) was used for sample cleaning-up and target analytes enrichment. The extracted target toxins were separated on a C18 column with washing of 10% methanolic solution, and then eluting with methanol. The limit of quantitation of MC-LR and MC-YR was 0.06 ug/L, which was below the limits recommended by WHO guidelines for drinking water (i.e., 1 ug/L). A preliminary result revealed that trace levels of MC-LR and MC-YR were detected in two reservoirs water samples by using the developed method.

TH162

Smelly HABs: response-surface optimized HS-SPME-GC/MS method for monitoring multi-class HAB odor compounds in water

C. Avagianos, M. Pisania, T. Kaloudis, EYDAP SA / WATER QUALITY CONTROL

HABs are known to produce a wide range of malodorous compounds belonging to various chemical classes such as terpenoids, ionones, amines, aldehydes, ketones and sulfurous compounds. Such compounds have detrimental effects to the aesthetic quality of water, making drinking water unpallatable by consumers and damaging recreational and tourism enterprizes in lakes. Although HAB odorous compounds are generally non-toxic to humans at environmentaly relevant concentrations and they do not inevitably indicate the co-occurrence of cyanotoxins, there is a need for water utilities and water authorities to apply frequent montitoring for early-warning and control of off-odor events. The objective of this study was to develop and optimize an efficient method for monitoring of multi-class HAB odors in freshwaters using automated HS-SPME/GC–MS. The study focused on optimization of the SPME factors, including salting-out effect, pre-incubation and extraction times, extraction temperature and stirring rate. 20 model compounds of various chemical classes were selected as indicative of the wide range of odorous compounds, ranging from volatile, early-eluting (e.g. alkyl sulfides) to late-eluting compounds (e.g. ionones). Factor effects on the responses (extracted ion peak areas) were assessed. Design of experiments (DoE) techniques included preliminary Plackett-Burman factorial designs for screening of significant factors, followed by a 4-factor Box-Behnken design to assess linear and quadratic main effects and factor interactions and to optimize responses. Optimization was based on desirability functions that reflect the objectives for maximum sensitivity screening of the whole range or of certain classes/groups of compounds. Optimization experiments resulted in full-quadratic response models for individual compounds, while desirability functions can be

easily computed for classes of compounds. The most significant factor was extraction temperature, especially for volatile early-eluting compounds where fine-tuning of temperature is essential to achieve the required sensitivity. The optimized automated HS-SPME-GC/MS method is proved to be a valuable tool for high-throughput, efficient and sensitive non-targeted screening of HAB odorous compounds, while sensitivity is further enhanced when certain classes of compounds are targeted, e.g. when sensory pre-evaluation of samples is applied.
Acknowledgement - The authors thank CYANOCOST – COST Action ES 1105 www.cyanocost.net

TH163

Suspected screening of cyanotoxins in freshwater by high performance liquid chromatography coupled to high-resolution mass spectrometry

D. Filatova, IDAEA- CSIC / IDAEA; m. picardo, IDAEA CSIC Barcelona / IDAEA; O. Núñez, Universitat de Barcelona / Department of Chemical Engineering and Analytical Chemistry; M. Farre, IDAEA-CSIC / Environmental Chemistry

Cyanobacteria are one of the components of regular microbial succession in periphyton formation. The community distribution is affected by water quality, flow regime, climate, and geology. During the past decades, there has been a noticeable increase of cyanobacterial blooms, dominating in many freshwater bodies worldwide. Some cyanobacteria species are known to produce toxic secondary metabolites called cyanotoxins, which vary in structure and harmful properties (hepatotoxins, neurotoxins), and being a major concern for drinking water supply and recreational water use. The most widespread cyanotoxins are microcystin (MCs) variants MC-LR, -RR, -YR, with MC-LR being the most toxic one. For this reasons, the World Health Organization appointed a guideline of 1 μg/L in drinking water for total MC-LR. In order to monitor levels of cyanotoxins and prevent both human poisoning and wildlife damage, suitable analytical methods need to be developed. This work presents the development of a sensitive, fast and robust method for the analysis of cyanotoxins in freshwaters based on high-performance liquid chromatography coupled to high-resolution mass spectrometry (HPLC-HRMS). For the sample pre-treatment of cyanotoxins, solid-phase extraction for multiple toxins has been employed, which was recently developed in our research group. The chromatographic separation was achieved using a C18 analytical column (150x2.1 mm, 5μm) using methanol and water as mobile phase. The total chromatographic run was 15 min. The chromatographic separation was coupled to a Q-Exactive Orbitrap instrument (Thermo Fisher Scientific). The interphase used was ESI under positive conditions. The main advantage of high-resolution mass spectrometry will be the target analysis of 10 cyanotoxins, as well as the analysis in scan mass spectrometry to assess the potential presence of transformation products and other non-targeted toxins in the samples. This multi-toxin method has been developed and validated for freshwater cyanotoxins such as microcystins, nodularin, cylindrospermopsin, and anatoxin-a. The developed method was applied for the study and characterization of cyanotoxins concentrations in Catalonia freshwater reservoirs. **Keywords:** cyanotoxins, microcystins, high-resolution mass spectrometry.
Acknowledgement: *This project has received funding from the European Union's Horizon 2020 research and innovation programme under the Marie Sklodowska-Curie grant agreement No. 722493.*

TH164

Oligonucleotide probes for fluorescence in-situ identification of cyanobacterial cells in surface waters

l. dejana, Water Research Institute National Research Council / National Water Research Institute; A. Barra Caracciolo, National Research Council / Water Research Institute; P. Grenni, National Research Council of Italy (CNR) / Water Research Institute; c. fajardo, Facultad de Veterinaria, Complutense University Avenida Puerta de Hierro s/n, 28040 Madrid, Spain; M. Martin-Fernandez, UCM / Biochemistry and Molecular Biology; l. medlin, Marine Biological Association of the UK, The Citadel, Plymouth PL1 2PB, UK; G. Mengs, Natural Biotec SL; m. saccà, Council for Agricultural Research and Economics (CREA), Agriculture and Environment Research Center (AA), Via di Corticella 133, 40128 Bologna, Italy; m. lettieri, European Commission, DG Joint Research Centre, Directorate D Sustainable Resources Unit D.02 Water and Marine Resources TP 121, Via E.Fermi, 2749, 21027 Ispra (VA), Italy

Harmful cyanobacterial blooms have been increasing in freshwater ecosystems in recent decades, mainly because of eutrophication and climate change. In some cases, some cyanobacterial species can produce toxins and this phenomenon can have a negative impact and pose a risk for ecosystem and human health. Of the 150 known cyanobacteria genera, more than 40 species produce toxins, which are natural compounds showing different chemical and toxicological characteristics. Cyanobacterial toxins are responsible for both acute and chronic poisoning in animals and humans. Among the main classes of cyanotoxins, microcystins are among the most frequently found in the environment. These toxins are accumulated mainly in the liver, but also in the intestines and kidneys and can be very dangerous for both animal and human health (Lucentini and Ottaviani, 2011). Fast and sensitive methods to identify unequivocally *Microcystis aeruginosa* and *Planktothrix agardhii* are very useful to discriminate these species with respect to the other non-toxic cyanobacteria. For this purpose, we designed, developed and validated some oligonucleotide probes (GNPlankS02, PkAgD03, MicAerD03) for FISH (Fluorescence *In-Situ* Hybridization) analysis to detect these species in freshwater samples. The FISH probes were designed using the ARB software with the Silva database in the framework of the MicroCoKit project. We tested different fixative methods to minimise the natural autofluorescence from chlorophyll-a (Groben and Medlin, 2005)to visualize *Microcystis aeruginosa* and *Planktothrix agardii* under a laser confocal microscope. Firstly, the FISH probes designed have been tested on pure cultures of *M. aeruginosa* and *P. agardhii* species, then the probes were successfully applied to natural samples collected from surface waters. **Keywords:** *Microcystis aeruginosa*; *Planktothrix agardhii*; FISH probes; algal bloom **References Groben R. and Medlin L., 2005. *In situ hybridization of phytoplankton using fluorescently labeled rRNA probes*.** Methods in enzymology, 395, 299-310. **Lucentini L. and Ottaviani M., 2011. *Cyanobacteria in water for human consumption: Guidelines for risk management.*** "National Group for cyanobacteria risk management in water for human consumption", vol. 2, VIII, p. 67 Rapporti ISTISAN 11/35 Pt. 2

TH165

Adequacy of EPI Suite prediction models to estimate physicochemical properties of natural toxins potentially present in surface water

I. Rodríguez Leal, Stockholm University / Department of Environmental Science and Analytical Chemistry ACES; M. MacLeod, ITM - Stockholm University / Department of Environmental Science and Analytical Chemistry

Natural toxins constitute a potential risk to water supplies in Europe. Only a few systematic risk assessments of individual natural toxins have been conducted in Europe. There is thus a need to conduct new risk assessments, especially to reflect possible effects of climate change on the distribution of agricultural plants throughout the continent and to reflect increasing prevalence of monoculture farming. Furthermore, screening-level assessment of many natural toxins that have been identified but not fully assessed is needed (Bucheli 2014). Persistence and mobility of natural toxins in water might be usefully modelled in screening applications using techniques developed for environmental pollutants of anthropogenic origin, such as EPI Suite™ (US EPA 2017). Environmentally relevant partitioning properties of many natural toxins have not been experimentally determined. To model overall persistence of natural toxins in aquatic environments requires sorption coefficients (e.g., K_{OC}) and estimates of their degradation rate constants in the aquatic environment that have been determined by experimental methods or estimated using quantitative structure-activity relationship (QSAR) and quantitative structure-property relationship (QSPR) models. QSAR predictions should be considered carefully when applied to a set of chemicals that are structurally distinct from those that were used to develop the model. The establishment of an applicability domain of the models provides a range of chemicals where the predictions are expected to be reliable and based on interpolation rather than extrapolation, regarding the structure of the chemicals in the training set (Gramatica 2007). We present here an analysis of the applicability domain of selected EPI Suite™ QSAR models, and interpret the results with reference to natural toxins within these limits that could be included in a database to prioritize natural toxins in water according to their persistence and mobility. **References** Bucheli, Thomas D. 2014. "Phytotoxins: Environmental Micropollutants of Concern?" *Environmental Science & Technology* 48 (22):13027–33. Gramatica, Paola. 2007. "Principles of QSAR Models Validation: Internal and External." *QSAR & Combinatorial Science* 26 (5):694–701. US EPA. 2017. Estimation Programs Interface Suite™ for Microsoft® Windows, v 4.11. United States Environmental Protection Agency, Washington, DC, USA.

TH166

Cyanobacterial oligopeptides of environmental concern and (co)production dynamics

R. Sanches Natumi, E. Vonwyl, Eawag Swiss federal Institute of Aquatic Science and Technology / Department for Environmental Chemistry; E.M. Janssen, Eawag Swiss federal Institute of Aquatic Science and Technology / Department of Environmental Chemistry

Our ecosystems and drinking water resources are not only vulnerable towards anthropogenic pollutants. Natural toxins present an additional threat for which we still lack comprehensive risk assessment and management plans. Among the natural toxins from various kingdoms, those produced by aquatic organisms have a direct entry into our water resources. More frequent and intense surface water blooms of cyanobacteria have triggered particular scientific interest in their secondary metabolites as potential aquatic toxins. The variety of cyanopeptides is well documented since the 1990s and the growing publication record reflects an increasing scientific awareness. Cyanopeptides can be grouped into structural classes characterized by indicative monomeric building blocks. Microcystins are by far the most intensively studied class of cyanopeptides. While it is known that many cyanopeptides are produced simultaneously from one species, the co-production of these potential toxins has not been explored comprehensively for cyanopeptides beyond microcystins. This project focused specifically on the production and co-production dynamics of cyanopeptides under different culturing condition of common cyanobacterial strains. Our targeted LC-HRMS analysis of biomass samples of single strain cultures show that besides microcystins, cyclamides and various cyanopeptolins are co-produced. Our data shows the evolution of the peptide abundance throughout the growth phase of single strains (e.g., *Microcystis aeruginosa* and *Anabaena flos-aquae*) and under different culturing conditions (e.g., N:P ratios and light intensities) by multifactorial analysis. New insights into co-production dynamics offer critical information about cyanotoxin mixtures present during harmful algae blooms and with that critical knowledge towards comprehensive risk assessment.

TH167

Degradation of the carcinogen ptaquiloside under alkaline conditions

D. Lindqvist, L. Rasmussen, Metropolitan University College

The carcinogen ptaquiloside (PTA) is found in several species of ferns worldwide. The distribution and occurrence is well described for genus *Pteridium* (Bracken ferns) where PTA is found in all parts of the fern. PTA is suspected of causing Human gastric cancer. PTA is a nor-sesquiterpene glycoside and is not sorbed by soils to a great extent (log*Kow* of approx. -0.5). Hence, PTA can leach from Bracken stands. Leaching is most prominent during rain but baseline levels are found in streams in Bracken infested areas. Soil contamination and contamination of upper aquifers has been observed on a number of occasions. PTA may contaminate groundwater resources. Dissipation of PTA under environmental conditions is governed by a number of factors: Enzymatic activity; pH (hydrolysis); irreversible sorption/reactions; and sorption to clay minerals. Bacterial activity and hydrolysis are the most important mechanisms causing dissipation of PTA. The purpose of this study was to describe the underlying mechanisms for the hydrolysis of PTA and formation of reaction products under near-sterile alkaline conditions as found in calcareous aquifers. PTA (4,700ppb) was deglycosidated using 0.01/0.10/1.0 M NaOH and 3 different 0.025M buffer systems (approx. pH 7-12; Na_2HPO_4/$NaHCO_3$/H_3BO_3; pH regulated with 0.1M NaOH). Dissipation of PTA and formation of reaction products were monitored up to 200hrs at 25ºC. PTA and the main reaction product pterosin B (PTB) was quantified by LC-MS using SIM- and TIC-modes. Formation of other reaction products was analysed semi-quantitatively using the relative area distribution of the main mass trace. Chemical rate constants are reported for degradation of PTA and species formation. Dissipation of PTA were pH dependent as previously observed. Addition of 0.01-1.0M NaOH results in immediate degradation of PTA and formation of a small amount of PTB plus two more reaction products. The concentration of PTB remained constant over time. PTB is the endproduct of hydrolysis in pure solutions. One reaction products was identified as the bracken dienone (BDE), a ultimate carcinogen. BDE is formed immediately while the unknown is formed from BDE reaching stoichiometric balance within the timeframe of the experiments. Rate of formation were dependent on pH, as well as of the buffer system used. The results indicate that PTA under alkaline conditions will form carcinogenic reaction products that are stable in groundwater.

TH168

Experimental Determination of Octanol-Water Partitioning Coefficients of Natural Toxins

C.D. Schoensee, Agroscope / Environmental Analytics; K. McNeill, ETH Zurich / Institute of Biogeochemistry and Pollutant Dynamics; T. Bucheli, Agroscope ART / Environmental Analytics

The octanol-water partitioning coefficient (K_{ow}) remains one of the key parameters in environmental fate and risk assessment of organic chemicals for regulatory purposes.[1] Based on predicted K_{ow} values, many natural toxins are expected to be mobile in the aquatic environment. If such toxins are not retained well in soils, they may finally end up in drinking water resources. However, for compounds such as natural toxins, with physicochemical and structural complexity due to large numbers of various functional groups, current estimation models for K_{ow} and other phase distribution coefficients show limited applicability.[2] Thus, experimentally determined physicochemical property data are still of great value to regulatory organizations defining thresholds for potential environmental contaminants. In this study, K_{ow} values are experimentally evaluated by both indirect and direct approaches based on well-established OECD methods modified for application in natural toxin analysis. A multi-step molecular size and functional group specific calibration procedure is applied in indirect evaluation of natural toxin partition properties based on chromatographic retention. Direct analysis of partitioning behavior is performed in a miniaturized shake flask system in standard HPLC vials. As HPLC based methods, both approaches show the capability to be largely automatized for more efficient, less error-prone analysis and thus allow the reliable determination of K_{ow} in the for potential aquatic contaminants relevant range of log K_{ow} from -2 to 5. Natural toxins for analysis comprise previously investigated mycotoxins and isoflavonoids as reference compounds in addition to representatives of different compound classes of phytotoxins. In regards to predicted toxicity, persistence and mobility as well as plant occurrence, specific alkaloid subclasses such as pyrrolizidine alkaloids from *Senecio* spp. or quinolizidine alkaloids from *Lupinus* spp. are investigated in more detail. As an indicator for the partitioning of natural toxins from aqueous media to organic matrices, K_{ow} can be seen as first proxy estimating natural toxin mobility in the aquatic environment. Thus, experimental data will help in prioritization of toxins for further research activities, including field studies and lab-based characterization of fate processes e.g., within the current MC-ITN NaToxAq. **[1] ECETOC; Technical Report No. 123, 2013**.**[2] Schenzel, et al.; *Environ Sci Technol*2012**, 46 (11), 6118-26. \n

TH169

Phytotoxins as aquatic micropollutants: a procedure for prioritization

B.F. Guenthardt, Agroscope / Environmental Analytics; J. Hollender, Eawag / Environmental Chemistry; M. Scheringer, ETH Zurich / Institute for Chemical and Bioengineering; K. Hungerbuehler, ETH Zurich / Department of Chemical and Biochemical Engineering; T. Bucheli, Agroscope ART / Environmental Analytics

Phytotoxins are natural toxins produced by plants with widely varying molecular structures and toxic effects. Despite possibly high concentrations of natural toxins in vegetation, crops and freshwater, they are not yet commonly perceived as environmental contaminants of possible concern. This far, environmental exposure and effect studies have only been conducted for a very limited number of phytotoxins, and systematic and larger monitoring campaigns are completely lacking. A crucial challenge is to systematically identify among the plethora of phytotoxins those that actually present a serious risk for the aquatic environment. For this purpose, we ranked 1586 phytotoxins from over 800 plant species compiled in a previously developed database based on three critical properties: toxicity, plant frequency and environmental behavior of the phytotoxins. Toxicity was included as descriptor of the effect and parametrized by both plant, and estimated compound toxicity. Plant frequency, obtained from InfoFlora, was used to estimate the occurrence of all plant species producing a certain secondary metabolite class and serves as an approximation of exposure. To identify the phytotoxins relevant for the aquatic environment we used a procedure, which systematically ranks substances for being persistent and mobile organic compounds (PMOCs) based on volatilization, biotic degradation and hydrolysis as measure of persistence and D_{oc}, the pH dependent K_{oc}, as measure of mobility The PMOC analysis, using predicted phytotoxin properties, showed that approximately 70% of phytotoxins are mobile in the environment. However, over 50% of the secondary metabolite classes are not of priority due to their fast degradation in the environment. Other secondary metabolite classes are simply not enough toxic to be of primary interest or only rarely produced. Finally, the analysis identified several potentially problematic secondary metabolite classes such as saponins, steroids, the terpene classes triterpenoids and diterpenoids, and several alkaloid classes including pyrrolizidine alkaloids, isoquinoline alkaloids, terpenoid alklaoids or steroidal alkaloids. These phytotoxins were characterized as toxic, frequently produced, mobile and persistent and we propose to consider them in further monitoring programs and risk assessments.

TH170

Sorption of pterosin B to soil materials

J. Sandersen, L. Rasmussen, Metropolitan University College

Bracken ferns (*Pteridium* sp.) are considered environmentally problematic due to their content of the carcinogens ptaquiloside, caudatoside and ptesculentoside (‘the ptaquiloside group’). Brackens are classified by WHO/IARC in Group 2B due their carcinogenic properties at cellular level and due to their association with several veterinary diseases. Brackens cause bovine urinary bladder cancers and are suspected of causing Human gastric cancer. Pterosin B is formed from ptaquiloside upon hydrolysis. Similar pterosins are formed from caudatoside and ptesculentoside. The rate of hydrolysis is strongly dependant on pH and temperature. Under environmental conditions - pH5-7 - pterosin B will form as one of the main products of hydrolysis. At lower or higher pH other compounds may form from ptaquiloside. Pterosin B is not toxic, but is interesting as the compound can be used to assess previous presence of ptaquiloside. Studies have shown rapid microbial degradation of pterosin B in upper soil layers, but longer lifetime is expected in sediments with low microbial activity such as aquifers. The purpose of this study is to assess the sorption of pterosin B and to estimate Kd and Koc. Sorption of pterosin B was studied following OECD Guideline 106 and 9 different Danish soils (SOC-%: 0.1-7.4; pH 3.3-7.3). 0.25g of dry soil were equilibrated with 9mL 0.01M $CaCl_2$ over-night. 1mL of pterosin B solution in 0.01M CaCl was added resulting in a C_{INIT} of 0-10 mg L^{-1} (n=5-20). Sorption were studied after a contact time of 24hrs. The aqueous phase were separated by centrifugation and the content of pterosin B quantified by LC-MS-ESI (SIM; 100µL injections; range 0-100 µg L^{-1}; $r^2 \geq 0.999$). C_{SORB} were calculated as C_{TOTAL}- C_{AQ}. Irreversible sorption and microbial degradation were considered insignificant based on previous studies. Pterosin B sorp strongly to the soils tested. This was expected due to the aromaticity of pterosin B and the log*Kow* of 3.3. Kd ranged between 70 and 180 mL g^{-1} for the soils tested corresponding to a Koc values of 300-2,500 mL g^{-1}. The study demonstrates that pterosin B sorp strongly to soil materials, especially to soil organic matter. As Koc values can vary substantially, depending on soil type and properties like soil pH and mineralogy, some variation were expected in the results. Provided low microbial activity, pterosin B will most likely stay in aquifers and can indicate previous presence of ptaquiloside.

TH171

Modelling the fate of natural toxins in the soil using DAISY- a case study of ptaquiloside

D.B. Garcia Jorgensen, University of Copenhagen / Plant and Environmental Sciences; H. Hansen, University of Copenhagen / Department of Plant and Environmental Sciences; M. Styczen, E. Diamantopoulos, P. Abrahamnsen,

University of Copenhagen
Natural toxins are gaining more interest in the scientific community as emerging pollutants. The reason behind is that they are released continuously to the environment and often in high amounts. The related risks to humans depend strongly on the physicochemical characteristics, load and fate of the natural toxins in the environment. The aim of this work is to develop a modelling approach to predict the fate and in particular the leaching of natural toxins in the vadose (soil) zone. For the work, the model code DAISY, a soil-plant-water-atmosphere model, has been used.\nModelling of natural toxin fate presents several challenges compared with xenobiotics: many and partly continuous sources, variable and poorly studied physicochemical properties of the toxins, highly variable temporal and spatial rates of transfer of the toxins from the source plant to soils, - often linked to climate events.\nThis work focused on ptaquiloside (PTA), a hydrophilic and non-sorbing toxin that exhibits a strongly pH and temperature dependent degradation. The carcinogenic toxin is produced by bracken fern (Pteridium aquilinum) that usually forms dense stands. The PTA content in bracken is up to 9800 $\mu g\ g^{-1}$ dry matter. The modelling approach was to parameterize a bracken growth submodule in order to simulate biomass and canopy. Spraying was used as the method to apply the toxin to the canopy, similarly to pesticides as included in DAISY. It is assumed that the toxin is washed off from the canopy with precipitation. The model was improved with new functions to parameterize hydrolysis. Hydrolysis is pH and soil horizon dependent, while microbial degradation rates follow the guidelines by FOCUS.\nMaximum PTA concentration in the leachate at a depth of 2 m were 2.5 and 1 $\mu g\ l^{-1}$ in a sandy loam and sandy soil, respectively. These concentrations are above calculated health thresholds concentrations. Clayey soils presented higher leaching due to macropore transport, as toxins might bypass the biologically active soil layers. Leaching accounts for less than 1% of the total PTA load, being highest in autumn when bracken wilts and the amount of water percolating is highest.\nThe model presents several uncertainties such as the toxin production in the biomass, seasonal variation in toxin concentrations and in particular, the transfer rates from plant to soil. Spraying is not an ideal "dosing" function and might overestimate the leaching, hence the results must be taken with caution.

TH172

Genomic insight into biosynthetic pathway of retinoids by cyanobacteria

L. Sehnal, Masaryk University RECETOX; K. Hilscherova, Masaryk University, Faculty of Science, RECETOX / Research Centre for Toxic Compounds in the Environment   RECETOX

Extensive occurrence of cyanobacterial water blooms associated with the production of wide range of toxic compounds into environment represents one of the biggest worldwide problems in aquatic ecosystems. One group of the recently described cyanobacterial toxic compounds are endocrine disruptive compounds retinoids. It has been documented that cyanobacteria are potent producers of retinoids and they are able to produce these compounds into their surrounding environment. However, our understanding how are retinoids synthetized by cyanobacteria on genomic level remains poor and description of the biosynthetic machinery of these small "dietary" hormones is essential to the elucidation of original significance of these molecules for cyanobacteria. In the animal kingdom, biosynthetic apparatus for retinoids synthesis has already been described. Mayor role in their synthesis from carotenoids play the enzymes aldehyde dehydrogenases (ALDH) and cytochromes (CYP). Our study has been inspired by biosynthetic apparatus of retinoids in animals and provides an evolutionary comparison of all ALDH and CYP from all publicly available genomes of cyanobacteria to well-characterized ALDH and CYP from human and mouse, which are involved in the biosynthesis of retinoids. This comprehensive phylogenetic study describes evolutionary similarity of cyanobacterial ALDH to human and mouse ALDH from family 1. This fact points out to a similar function of these enzymes in the biosynthetic machinery of retinoids. Based on these results, the most related cyanobacterial ALDHs (to human) were selected from different cyanobacterial genomes and heterologously expressed in direct cloning-proficient E. coli strain GB05-dir. Effectivity of expression reflected as the amount of produced retinoids was assessed by *in vitro* bioassay on cell line P19/A15 with endogenous expression of retinoid receptors stably transfected with reporter luciferase gene under the control of retinoic acid-responsive element. The total concentration of produced all-trans retinoic acid was measured by LC-MS/MS. The project is supported by the Czech Science Foundation and National Sustainability Program of the Czech Ministry of Education, Youth and Sports (LO1214 and LM2015051).

TH173

Emerging treatment methods for the removal of cyanotoxins from drinking water with focus on Advanced Oxidation Processes

M. Schneider, RECETOX, Faculty of Science, Masaryk University / Research centre for toxic compounds in the environment RECETOX; L. Blaha, Masaryk University, Faculty of Science / Research centre for toxic compounds in the environment RECETOX

Cyanobacteria form blooms in freshwaters due to environmental pollution and can produce taste and odour compounds, but also substances that have been shown to be toxic to animals, humans and other organisms. Numerous events of cyanotoxin-associated poisonings of pets, livestock, birds, wildlife and humans, and in some cases even subsequent death, occurred – and still occur – globally. These mainly waterborne secondary metabolites can adversely affect the quality of water intended for drinking and recreational purposes. So far, most countries have not yet enforced strict regulations regarding maximum tolerable cyanotoxin levels in drinking water. Some countries adapted the WHO provisional guideline value of 1 µg/L for microcystin-LR or amended it for country-specific regulatory values. Due to their diversity, fluctuating environmental occurrence and concentration, conventional drinking water treatment can result in insufficient removal of cyanotoxins. Advanced Oxidation Processes (AOPs) are emerging treatment methods that have been shown to be very promising for the removal of organic pollutants in general, also providing a potential for the removal of cyanotoxins. AOPs promote the *in situ* formation of highly reactive radicals, mainly hydroxyl radicals, and other mechanisms. Hydroxyl radicals are non-selective and randomly attacking oxidants, usually reacting with rate constants orders of magnitude higher than for other oxidants. So far, most research focuses on treatment of microcystins, but other toxin classes such as nodularins, saxitoxins, cylindrospermopsin and anatoxins have also been shown to be susceptible to be removed by AOP treatment. The most often reported AOPs for the removal of cyanotoxins include ozonation, (photo-)Fenton oxidation, direct and catalyst-enhanced photolysis, and combinations of these or with hydrogen peroxide. Lesser studied, but still very promising AOPs for the removal of cyanotoxins from drinking water are sonolytic and hydrodynamic cavitation, electrochemical oxidation, radiolysis and other novel approaches such as those based on non-thermal plasmas. The present paper summarizes pros and cons of AOP technologies for the removal of cyanotoxins from drinking water and presents the first experimental outcomes of studies characterizing the potential and novel AOPs for the removal of less explored cyanobacterial metabolites and their mixtures. Acknowledgement: Supported by NaToxAq (H2020 MSC ETN project agreement No. 722493).

TH174

An overview of the effects and bioaccumulation of ciguatoxins in fish

m. Dechraoui Bottein, IAEA / Nuclear Sciences and Applications; r. clausing, International Atomic Energy Agency / Nuclear Sciences and Applications; L. Diaz-Asencio, CEAC; L. Ranada, Philippine Nuclear Research Institute / IAEA Collaborating Center on Harmful Algal Bloom   (HAB) Studies, Chemistry Research Section, Department of Science and Technology (PNRI-DOST); J. Diogène, IRTA / Marine Environmental Monitoring Subprogram; G. nuel, Université Pierre et Marie Curie - Paris VI CNRS UMR7599 / Laboratoire de Probabilité et Modèles Aléatoires (LPMA); P. hess, IFREMER; M. Chinain, Institut Louis Malarde

Ciguatera Fish Poisoning (CFP), the most common non-bacterial seafood intoxication globally, results from the consumption of fish contaminated by suites of dinoflagellate derived marine polyether neurotoxins (Figure 1) known as ciguatoxins (CTXs), that target voltage gated sodium (Na_v) and potassium (K_v) channels. CTXs enter the food web through consumption of *Gambierdiscus* spp. dinoflagellates by herbivorous fish. It is well established that the algal CTXs undergo biotransformation in fish organisms (oxidative metabolism) as they pass through the marine food chain, leading to more oxidized (and more toxic) forms of CTXs. Evidence for concentration of ciguatoxin through the food web has relied largely upon correlation observed between toxicity and trophic level of wild-caught fish. However more detailed regional surveys of multiple species do not uniformly show a clear trend between toxicity and trophic level or size. The absence of signs of intoxication of fish with high CTX concentrations (including fish involved in human poisoning or those tested in the context of field surveys) has long confounded our understanding of how such a potent toxin can be accumulated to the high levels at which it is naturally found (at concentration as high as > 10 ng P-CTX-3C equiv. g-1 of flesh). CTXs are suspected to also cause intoxication of fish and marine mammals, however this has never been reported in the field. In this presentation, we will review published and unpublished toxicokinetic and toxicodynamic data of CTXs in fish, including CTX bioaccumulation in field fish collected in ciguatera hot spots, CTX specific binding interactions with native NAv, and development of short and long term experimental models of CTX trophic transfer to fish to assess CTX bioaccumulation and effects. Such observations open promising research prospects aiming at the identification of potential ciguatera disease markers (transcriptomic signatures) in fish following experimental exposure to ecologically relevant doses of CTXs and for the development of predictive models of tissue-specific CTX accumulation for human risk assessment of seafood.

TH175

AFLATOXIN CONTAMINATION IN IMPORTED NUTS FOR DIRECT HUMAN CONSUMPTION: THREE YEARS (2013-2105) OF OFFICIAL CONTROL RESULTS IN ITALY

R. De Pace, P. D'Agnello, Institute of Experimental Zooprophylactic Puglia and Basilicata / Chemestry

Aflatoxins (AFs) are the most toxic group of mycotoxin and secondary metabolites of various species of *Aspergillus* that can occur in all agricultural commodities under appropriated field or storage conditions. These molecules can cause important health problems and have high potential toxic effects. A validated Enzyme Linked Immuno Assay (ELISA) to monitoring the presence of aflatoxin

B1 (AFB1) and total aflatoxin (AFT), as a screening test, was used in order to analyze imported nuts, from non-European countries, intended for direct human consumption. The percentage of AFs positive samples (only pistachios and almonds), taken during the three years from 2013 to 2015, under the national programs of official control, amounted to 9% for B1 and 10.5% for AFT, and were confirmed by HPLC (High Performance Liquid Chromatography). The results demonstrate that imported nuts from Turkey are more broadly contaminated and that ELISA is a sensitive screening method to monitoring residue levels. The aflatoxins levels in pistachios exceeded even more than five times the maximum permitted limits set by European Commission in Reg 165/2010 and referred to the edible part of the tree nuts. The higher incidence of AFs in imported shelled pistachios is probably due mostly to an easier aflatoxin contamination following the fact that pistachios hulls with intact cuticles are more resistant to the *A. flavus* colonization. The paper should be of interest both for readers in the areas of hazard analysis for monitoring purpose, and for other researchers in mycotoxin field, due to the great utility of low-cost, rapid and reliable methods of analysis

TH176

Impact of climate change drivers on toxin contamination and genotoxicity in Mytilus galloprovincialis: combined effects of warming, acidification and harmful algal blooms.

A.R. Braga, Biology Department CESAM, Aveiro University; C. Camacho, IPMA, I.P.; V. Pereira, R. Marçal, A.M. Marques, Biology Department CESAM, Aveiro University; S. Guilherme, Biology Department CESAM, Aveiro University / Biology; A. Marques, IPMA, I.P.; A. Gago-Martínez, University of Vigo; M. Pacheco, Biology Department CESAM, Aveiro University / Dept of Biology; P. Costa, IPMA, I.P.

Mytilus galloprovincialis under current environmental conditions and at conditions simulating scenarios of climate change, namely warming, acidification and the combination of these two factors were exposed to paralytic shellfish poisoning (PSP) toxin-producing *Gymnodinium catenatum*. Shellfish toxicity derived from accumulation of algae toxins was assessed in mussels at the four treatments as well as the damage at DNA level via the comet assay. Mussels were acclimated for 21 days and then exposed to *G. catenatum*, during 5 days (uptake), followed by 10 days with non-toxic diet (elimination). The analyses of PSP toxins in the mussels were carried out by Liquid Chromatography with Fluorescence detection. The highest PSP content was observed at day 5 in mussels in the actual conditions (1493.8 ± 202.4 µg STXeq. kg^{-1}), which exceeded the international seafood safety limits (800 µg STXeq. kg^{-1}). Significantly lower PSP content was observed in mussels under climate change scenarios. The lowest levels (661.9 ± 22.8 µg STXeq.kg^{-1}) were found in warm-acclimated mussels, followed by acidification (761.2 ± 62.3 µg STXeq.kg^{-1}). However, interaction of both parameters did not reveal an additive effect. Lower toxin elimination was observed in warm-acclimated mussels. Genotoxicity was assessed in gills and hepatopancreas of mussels sampled at the end of each period. In mussels not exposed to toxic algae, the comet assay revealed highest damage levels in mussels under combined effects of warming and acidification at the end of the experiment (i.e after 36 days). When mussels were exposed to *G. catenatum*, DNA damage in both gills and hepatopancreas significantly increased at an earlier stage, *i.e* just after the uptake period. The treatments representing the acidification scenario and the interaction of warming with acidification revealed higher DNA damage than the actual conditions, highlighting a synergistic impact. DNA damage decreased in all treatments at the end of the elimination period, although reduction was subtle in mussels under interaction of warming and acidification. This is the first study assessing the impact of the combined effect of warming, acidification and biotoxins in shellfish. In conclusion, it was provided evidences that changes of global conditions may lead to lower PSP contents, but also to slower elimination rates and to a synergistic effect on DNA damage implying possible consequences for the mussels populations.

TH177

Interest of bivalves for the biosurvey of cyanotoxins in aquatic ecosystems

E. Lance, University Reims Champagne Ardennes / Biology and Biochemistry; A. Lepoutre, UMRI 02 INERIS-URCA-ULH SEBIO; Z. Amzil, IFREMER / Laboratoire Phycotoxines; M. Bormans, UMR CNRS Ecobio / UMR Ecobio; l. brient, University of Rennes 1 / UMR 6553 ECOBIO

The eutrophication of aquatic ecosystems, associated to climate change, enhance the frequency and the severity of cyanobacterial proliferations. Cyanobacteria are photosynthetic organisms producing endotoxins such as neurotoxins, hepatotoxins, dermatotoxins, and cytotoxins, threatening target organisms and humans. The bioaccumulation of the hepatotoxins microcystins (MCs) and their effects on organisms is overall quite well documented. However, the neurotoxin β-methylamino-L-alanine (BMAA), suspected to be a causative agent in the human neurodegenerative disease amyotrophic lateral sclerosis (SLA), is less studied. The bioaccumulation of BMAA has recently been demonstrated with highly selective analytical methods in various marine organisms (zooplankton, mussel, oyster, fish), but very rarely in freshwater organisms. Bivalves are known to consume phytoplanktonic species such as cyanobacteria or diatoms, both known to produce BMAA, and can be used as sentinel organisms to reveal the environmental contamination. A dual approach, in the laboratory and *in situ*, is used to evaluate the pertinence of the bivalves *Anodonta anodonta*, *Dreissena polymorpha* and *Mytilus edulis* as bioindicators of the contamination of fresh and estuarine waters by MCs and BMAA. The laboratory approach consist in the evaluation of the kinetics of BMAA and MCs accumulation and detoxification in bivalves at various times and concentrations of exposure.The *in situ* approach consist in the evaluation of the MCs and BMAA accumulation in caged bivalves along a river continuum from withholding freshwaters to interconnecting estuarine and coastal areas used for mussel aquacultures. First results show MC and BMAA accumulations in laboratory-exposed *D. polymorpha* and *A. anodonta*, with varying kinetics. Freshwater and marine bivalves also accumulated MCs *in situ* and a MC transfer from fresh to estuarine waters occurred, highlighted by an accumulation in the marine bivalve *M. edulis.* The results of this project will facilitate the long-term tracking of the contamination of ecosystems by cyanotoxins, which will provide an advance in the knowledge about the ecodynamic of cyanotoxins and the mains conditions of human exposure.

TH178

Tetrodotoxin an Emerging Threat to Humans in the Mediterranean Area: First Detection in Italian Mussels

C. Dell'Aversano, University of Napoli Federico II, Department of Pharmacy / Pharmacy; L. Tartaglione, F. Varriale, University of Napoli Federico II / Department of Pharmacy; A. Penna, University of Urbino / Department of Biomolecular Sciences; M. Giacobbe, Institute for Coastal Marine Environment, CNR; S. Pigozzi, A. Milandri, Fondazione Centro Ricerche Marine; P. Bordin, L. Bille, Istituto Zooprofilattico delle Venezie; A. Turner, Plymouth University / Food Safety

Tetrodotoxin (TTX) is one of the most potent neurotoxins, originally found in ovary and liver of pufferfish (*Tetraodontidae*) [1]. Successively, TTX was isolated from other marine and terrestrial animals, as xanthid crab, trumpet shellfish, blue-ringed octopus, gastropods, starfish, and frogs. The wide distribution of TTX in genetically unrelated organisms has made TTX origin for long time controversial, with different kind of bacteria being identified as TTX-producing organisms [2]. Even *Alexandrium tamarense* – one of the paralytic shellfish poisoning toxins (PST) producing organisms – was proposed as potential biogenetic source of TTX [3]. Although fatal human poisonings following consumption of TTX-contaminated seafood have been reported so far only in Japan, the accumulation of TTX in fish, oysters and mussels collected in Europe (Spain, Portugal, UK, Greece) has been recently reported. So, in the frame of a collaborative study on evaluation of PST-related risk in the Mediterranean area, mussels collected in the Siracuse bay (Sicily, Italy) over a three year period (2015-2017), were analyzed by hydrophilic interaction liquid chromatography coupled with both high resolution and tandem mass spectrometry detection (HILIC-HRMS and HILIC-MS/MS). Both techniques highligted the presence of high PST contamination levels, with samples collected in 2016 containing up to 10851 µg STX eq/kg. Unexpectedly, together with PST, tetrodotoxin was detected in Sicilian mussels. Although this was the first report of TTX in Italy, contamination levels found in mussels (0.8-6.4 µg/kg) were well below the regulatory limit of 2 mg TTX eq/kg established for TTX in Japan. Interestingly, much higher contamination levels of TTX (413 µg/kg) have been detected in mussels collected in 2017 in the NE Adriatic coasts of Italy (Lagoon of Marano), in the frame of the monitoring programme for marine biotoxins regulated in the EU.
[1] Wu Z, Xie L, Xia G, Zhang J, Nie Y, Hu J, Wang S, Zhang R. 2005. A new tetrodotoxin-producing actinomycete, Norcardiopsis dassonvillei, isolated from the ovaries of puffer fish Fugu rubripes. Toxicon 45:851-859. [2] Yasumoto T, Yasumura D, Yotsu M, Michishita T, Endo A, Kotak Y. 1986. Bacterial production of tetrodotoxin and anhydrotetrodotoxin. Agric. Biol. Chem. 50:793–795. [3] Kodama M, Sato S, Sakamoto S, Ogata T. 1996. Occurrence of tetrodotoxin in Alexandrium tamarense, a causative dinoflagellate of paralytic shellfish poisoning. Toxicon. 34:1101-1105.

TH179

The first report on neurotoxic anatoxin-a occurrence in cyanobacterial blooms in the Czech Republic

L. Blahova, Research Centre for Toxic Compounds in the Environment (RECETOX); K. Hilscherova, Masaryk University, Faculty of Science, RECETOX / Research Centre for Toxic Compounds in the Environment RECETOX; O. Lepsova Skacelova, Faculty of Science, University of South Bohemia; L. Blaha, Masaryk University, Faculty of Science / Research centre for toxic compounds in the environment RECETOX

Prototypical cyanotoxins such as microcystins have been extensively studied all around the world but there is still a lack of research on the occurrence, levels and risks of other toxic metabolites produced in harmful blooms of cyanobacteria. In this paper we present the results of the first survey focusing on less explored cyanotoxins, namely anatoxin-a, in the samples from the Czech Republic. Levels of cyanotoxins were analyzed in freeze-dried biomass collected during 2012-2015 in various reservoirs in the country. The focus was on blooms (total 34 samples) dominated by potential producers of anatoxin-a such as Dolichospermum sp. (syn. Anabaena sp.), Aphanizomenon sp. as well as blooms formed by less common cyanobacteria. The multi-target UPLC-MS/MS methodology was applied that allowed to analyze in parallel all major cyanobacterial toxins (microcystin-LR,

-RR, -YR, -LF, -LW, -LA, -LY, -WR; nodularin; cylindrospermopsin and anatoxin-a) as well as other bioactive metabolites of cyanobacteria (isomers of lipopeptide puwainaphycin F). The quality of the anatoxin-a analysis was assured by the use of D5-phenylalanine internal standard. Cylindrospermopsin (CYN) has been confirmed (4.25 microgram/g d.w.) in a single bloom from the pond Pisecensky (South Moravian region close to Slovakian and Austrian borders) dominated by invasive species Cylindrospermopsis raciborskii. The other species found in CYN-positive bloom were Cuspidothrix issatchenkoi, Sphaerospermopsis aphanizomenoides (formerly known as Anabaena aphanizomenoides), Pseudoanabaena limnetica and Planktolyngbya limnetica. For the first time we have identified anatoxin-a in total 3 samples from the Czech Republic (concentration ranging 0.34 - 2.82 microgram/g d.w.), all originated from South Bohemian region around the town of Trebon (sampling years 2013 and 2015). Three species of Dolichospermum sp. were found in all three anatoxin-a positive samples (different species in different samples - D. planctonica, D. smithii and D. flos-aquae). The phytoplankton communities of the anatoxin-a positive blooms were fairly rich in composition containing also Aphanocapsa sp., Aphanizomenon sp., Microcystis sp., Woronichinia sp., Sphaerospermopsis sp. In addition to CYN and anatoxin-a, the paper discusses concentrations of other above mentioned cyanotoxins and bioactive metabolites and their risks.

TH180

Toxic cyanobacteria succession during a drier summer in a water reservoir in Sicily, Southern Italy. Implications for monitoring programs and risk assessment.

M. Manganelli, Istituto Superiore di Sanità / Dip. di Ambiente e Salute; P. Zuccarello, Università Di Catania / Laboratorio di Igiene Ambientale e degli Alimenti (LIAA), Dip. G.F. Ingrassia; M. Stefanelli, INAIL / Laboratorio VIII Biotecnologie, Dip. Innovazione tecnologiche e sicurezza degli impianti, Prodotti e Insediamenti Antropici; S. Vichi, Istituto Superiore di Sanità / Dip. di Ambiente e Salute; G. Oliveri Conti, M. Ferrante, Università Di Catania / Laboratorio di Igiene Ambientale e degli Alimenti (LIAA), Dip. G.F. Ingrassia; E. Testai, Istituto Superiore di Sanità / Dip. di Ambiente e Salute

Sicily, in the middle of the Mediterranean Sea, is the largest Italian island and is characterized by a dry hot climate. During the forties-sixties of the last century several water reservoirs have been built for drinking and irrigation water, and some of them have been interested by harmful cyanobacteria blooms. However, monitoring programs have been discontinuous, and no recent data are available for most of them. Therefore, a quarterly two years survey of main lakes used for drinking and irrigation water supply started in 2016, with a complete (chemical, physical, microbiological and microscopic) analysis of samples, according to the Italian D.Lgs 152/2006, coupled to a water toxicity assessment through a Vibrio fischeri ecotoxicological test (ISO 11348-3:2007). Lake Disueri (37°11'26"N 14°17'16"E) was the only one in which a persistent bloom occurred during 2017 summer. After the July sampling when a *Microcystis* sp. bloom was first detected, the frequency of sampling was increased, to assess the risk of the exposed population and wild and domestic animals. Between mid-Jul and mid-Sept the composition of the cyanobacterial community changed dramatically. In July the bloom was dominated by *Microcystis* sp. and *Cylindrospermopsis raciborskii* (in the order of 10^8 and 10^7 cell/L, respectively), the only two species detected. By mid-Aug these two species disappeared and have been substituted by *Anabaenopsis* sp. and *Plankthotrix rubescens*, which in mid-Sept were still growing (10^7 and 10^6 cell/L, respectively). Disueri Lake is among the largest lakes, with a surface of 1,85 km^2 and a maximum and an average depth of 31 and 15.2 m. However, due to landfill and increasing and persistent dryness, depth can be reduced as much as to 1 m. Cyanobacteria are not always present (from Nov 2016 until Jul 2017 no species has been detected) and sediments represent probably the source for the inocula triggering the blooms, as well as a sink-source for nutrients, in the agricoltural area of the water catchment. Results on chemical (nutrients and cyanotoxins) and molecular analysis will be discussed with a risk-based approach, to determine the risk for health for the population and to plan future management strategies.

TH181

Cyanobacteria taste and odor compounds; a study in freshwaters of Greece

T. Kaloudis, C. Avagianos, EYDAP SA / WATER QUALITY CONTROL; J. Diaz-Alvarez, M. Panou, Aristotle University of Thessaloniki / School of Biology; S. Zervou, NCSR Demokritos / Institute of Nanoscience and Nanotechnology; M. Pisania, EYDAP SA / WATER QUALITY CONTROL; C. Christophoridis, T.M. Triantis, NCSR Demokritos / Institute of Nanoscience and Nanotechnology; A. Hiskia, NCSR Democritos; S. Gkelis, Aristotle University of Thessaloniki / School of Biology

Cyanobacteria as well as eukariotic algae produce a wide range of volatile metabolites with several of them being odorous, causing taste and odor (T&O) incidents in freshwaters and in finished drinking water. Classes of cyanobacteria T&O include terpenoids, ionones, amines, carbonylic and sulfurous compounds, as well as their transformation products. T&O are hazardous for tourism and recreational activities in lakes, they can result in production losses in aquaculture and they can degrade drinking water quality, making it unucceptable by consumers. The aim of this study was to investigate the profiles of volatile and odorous compounds in natural freshwaters of Greece as well as in cyanobacteria strain cultures isolated from Greek lakes. To do this, analytical workflows combining targeted and non-targeted analysis based on automated HS-SPME-GC/MS for fast and sensitive detection of a wide range of T&O compounds were developed. Samples of lakes and water reservoirs of Greece were collected for T&O analysis according to specified procedures. Samples of cyanobacteria cultures (50 strains) isolated from 15 Greek freshwater bodies were also taken for T&O analysis. Results showed that a wide range of T&O compounds were present in natural water samples and cyanobacteria strains. Examples of compounds (odors) iclude trimethylamine (fishy), dimethyl-and trimethyl-sulfide (septic), methanethiol (septic), b-cyclocitral (tobacco), a-and b-ionones (floral). Interestingly, results showed that in surface water bodies of Greece geosmin and MIB have a minor role, while other T&O compounds having characteristic odor (e.g. fishy, swampy) may be more important. Based on the analysis of T&O in cyanobacteria strain cultures, T&O profiles of cyanobacteria strains were developed. It is concluded that non-targeted HS-SPME-GC/MS analysis is an effective and efficient technique for wide-range screening of cyanobacteria T&O compounds in water. Volatile and odorous metabolite profiles of cyanobacteria strains can be useful in interpreting T&O incidents in natural surface waters and water reservoirs. To better understand and anticipate T&O incidents, monitoring should be extended to compounds beyond geosmin and MIB. *Acknowledgement* - The authors thank CYANOCOST – COST Action ES 1105 www.cyanocost.net. C. Christophoridis acknowledges the program of Industrial Scholarships of Stavros Niarchos Foundation

TH182

Determination of multi-class cyanotoxins in fish tissues

C. Christophoridis, National Center for Scientific Research / Institute of Nanoscience and Nanotechnology; I. Argyropoulos, NCSR Demokritos / Institute of Nanoscience and Nanotechnology; T. Kaloudis, EYDAP SA / WATER QUALITY CONTROL; T.M. Triantis, NCSR Demokritos / Institute of Nanoscience and Nanotechnology; A. Hiskia, National Center for Scientific Research / Institute of Nanoscience and Nanotechnology

The analysis of cyanotoxins in aquatic organisms, particularly in fish, has lately received increasing interest, due to environmental concerns and public health issues. This study presents the development and otpimiation of novel, sensitive and accurate analytical methods for the simultaneous determination of multi-class cyanotoxins i.e Cylindrospermopsin (CYN), Anatoxin-a (ANA-a) and 12 Microcystins (MCs), in freshwater fish tissues. For the efficient extraction of selected cyanotoxins from fish tissue (muscle and liver), prior to LC-MS/MS analysis, several combinations of extraction solvents at different pH were tested. Additionally, various treatment techniques, i.e. protein precipitation using addition of salts and hexane extraction of lipids, were also tested in order to eliminate matrix effects and to maximize the recovery of the target compounds. Different SPE materials were evaluated for the maximum preconcentration of the compounds and in order to further eliminate matrix interferences. The effect of matrix components was evaluated by comparing LC-DAD and LC-MS/MS chromatograms under identical chromatographic conditions. Finally two extraction/clean-up methods were developed, i.e. one for the maximum recovery of selected MCs and one for CYN and ANA-a, offering maximum recoveries for the selected toxins. The developed methods were applied on fish samples, collected from Greek Lakes. The optimized method for MCs provided maximum recoveries 87% and 81%, for MC-RR and MC-LR, respectively. These compounds did not co-elute with several matrix components after the selected pretreatment/clean-up method, therefore matrix effect was minimal. CYN and ANA-a co-eluted with several matrix components, which induced increased matrix suppression, decreased sensitivity and affected method trueness. The use of the optimized methods, including several clean-up steps, significantly improved the recoveries, reaching 58% for ANA-a. Nevertheless, the use of isotopically-labeled surrogate standards, especially for CYN, would significantly improve the efficiency of the method. The diversity and accumulation of toxins in fish collected by Greek lakes, is presented in relation to the risks associated to human consumption. *Acknowledgments*: The authors would like to sincerely acknowledge COST Action ES1105 “CYANOCOST” and the program of Industrial Scholarships of Stavros Niarchos Foundation

TH183

Effects of Asparagopsis armata exudate on the fatty acid profile of two marine invertebrates

C.O. Silva, Polytechnic Institute of Leiria; T.F. Simoes, S.C. Novais, Polytechnic Institute of Leiria / MARE IPLeiria; M.F. Lemos, Instituto Politécnico de Leiria / MARE IPLeiria

Invasive alien species represent a worldwide threat to the integrity of native communities, which increase due to human activities and global changes is now presenting concerns as chemical pollutants do, and are thus often referred to as biological pollution. The red seaweed *Asparagopsis armata* exhibits a strong invasive behavior and it is included in the list of the “Worst invasive alien species threatening biodiversity in Europe”. This alga has been shown to produce a large diversity of halogenated volatile organic compounds with potent biological effects. It can be found on tide pools during the low tide, where its exudate released can represent a threat to the organisms present in just a few hours, leading to a reduction in abundance of native species. Marine organisms, in particular invertebrates, have proven to be a major source of unique fatty acids (FAs). Membrane lipids, especially FAs have great structural diversity and high biological specificity, essential for every living cell, as sources of energy, as membrane constituents, or as metabolic and signaling mediators. FAs have long been used as food-web tracers, and, more recently, changes in FA profile have also been exploited to better understand how contaminants affect organisms in aquatic food-webs (Silva et al. 2017). In this study, the potential impact of *A. armata* exudates in the FA profile of two marine invertebrates was assessed. For that, after calculating the lethal concentrations of the alga exudate, *Gibbula umbilicalis* and *Palaemon serratus* were exposed for 168 hours to non-lethal concentrations of this exudate. Consequently, the trends in changes of FA abundance and composition were evaluated separately in the body tissue of the snails and in the shrimps' hepatopancreas. Results showed different FA profiles between invertebrates but for both species the profile was influenced by exudate concentration exposure, with significant alterations being observed for several saturated FA and polyunsaturated FA like Acid Arachidonic (ARA), EPA or DHA. These alterations can represent an impact on these organisms' cell function since some of these FA have important communication and signaling roles within and between cells. Fatty acid profile showed to be a sensitive and highly informative parameter to address effects of macroalgae exudates toxicity in cohabiting invertebrates, thus constituting a promising tool for understanding biological pollution effect assessment in these coastal organisms.

TH184

Impacts of Asparagopsis armata on marine invertebrates: behavioral and biochemical responses

C.O. Silva, Polytechnic Institute of Leiria; C.E. Silva, S.C. Novais, Polytechnic Institute of Leiria / MARE IPLeiria; M.F. Lemos, Instituto Politécnico de Leiria / MARE IPLeiria

The introduction of non-native seaweeds outside their native distributional range, through human activities, has been causing documented negative effect on native species. The red algae *Asparagopsis armata*, with its invasive strategy including specialized cells capable of producing compounds with potent biological effects, is capable of inducing significant changes in terms of native community composition. This is specially occurring in rocky pools (intertidal zone) where *A. armata* releases several compounds that in these enclosed and extreme conditions are often adverse for other organisms such as other seaweed, vertebrates, and invertebrates leading to severe consequences for coastal ecosystems. The main objective of this study was to evaluate the potential impact of *A. armata* on marine invertebrates by exposing the common prawn *Palaemon serratus* and the marine snail *Gibbula umbilicalis* to the exudate of this macroalga. The seaweed collected at the coast of Peniche, (Portugal) was left in laboratory tanks, for 12 hours, in the dark at 20ºC±1. Afterwards the media was collected and filtered for further testing. After assessing the lethal concentrations of the algae exudate, animals were exposed for 168 hours to non-lethal concentrations of this exudate and subsequently analyzed for biochemical biomarkers responses associated with detoxification (glutathione S-transferase, GST), antioxidant defenses (catalase, CAT; superoxide dismutase, SOD), oxidative damage (lipid peroxidation, LPO; DNA damage), neurotoxicity (acetylcholinesterase, AchE) and energy metabolism (lactate dehydrogenase, LDH; Isocitrate dehydrogenase, IDH; electron transport system activity, ETS; content in lipids, proteins and carbohydrates). Also, behavioral endpoints were performed using the flipping behavior for *G. umbilicalis* and the avoidance behavior for *P. serratus.* The biomarker responses analysed on invertebrates showed an impairment of their physiological status after exposure to this algae exudate, with both species suffering from disruptions in their neuronal and energy metabolism functions, as well as from oxidative stress inducing damage in their macromolecules. Behavioural responses were also highly sensitive to *Asparagopsis* exudate exposure. These results represent an important step in the research of toxic exudates released to the environment and can serve as warning indicators of prospective effects of this macroalga on the invaded ecosystems under a global change scenario.

TH185

Assessing consumption risks through cadmium-contaminated shellfish amplified by ocean acidification

W. Chen, Kaohsiung Medical University / Dept Biomedical Science and Environmental Biology; H. Lin, National Taiwan University; S. Chen, Chung Shan Medical University / Public Health

The purpose of this study is to assess the human health risk of Taiwan population through consumption of cadmium-contaminated hard clam and oyster amplified by ocean acidification. This study employed forecasted ocean surface pH from the coupled model intercomparison project phase climate model in 2100 based projected emission scenarios representative concentration pathways 8.5, and Cd distribution as 0.001 – 2 μg L^{-1} in Taiwan coast to estimate potential Cd accumulation of shellfish. A gender-specific physiologically-based pharmacokinetic model was developed to assess urinary and blood Cd concentration via daily shellfish consumption. The dose-response function was used to account for the prevalence of renal dysfunction and osteoporosis in response to human accumulated Cd in urine and blood. Results showed that median Cd accumulations under current and 2100 ocean acidification scenario were, respectively, 0.0009 and 0.0010 μg g^{-1} for hard clam, whereas 0.0186 and 0.0210 μg g^{-1} were estimated for the oyster. The urinary Cd concentration in female had potential 80% higher for ocean acidification scenario than that for non-ocean acidification. However, results revealed that exceedance risks of renal dysfunction and osteoporosis for gender-specific consumption only and whole groups under 2100 ocean acidification were no significant difference with that of current non-ocean acidification scenario. This study concluded that ocean acidification was not likely to increase synergistically the renal dysfunction and osteoporosis of human health risk through shellfish consumption.

TH186

Cyanobacterial toxins - a threat to the human respiratory tract?

B. Kubickova, Masaryk University, Faculty of Science; P. Laboha, Masaryk University Faculty of Science / Research Centre for Toxic Compounds in the Environment RECETOX; J. Hildebrandt, Universität Greifswald / Animal Physiology and Biochemistry; K. Hilscherova, Masaryk University, Faculty of Science, RECETOX / Research Centre for Toxic Compounds in the Environment RECETOX; P. Babica, Masaryk University Faculty of Science / Research Centre for Toxic Compounds in the Environment RECETOX

Anthropogenic eutrophication and climate change increase the occurrence and intensity of toxic cyanobacterial blooms. Cylindrospermopsin (CYN), a (sub-)tropical cyanobacterial toxin of emerging concern, is detected in temperate climates with increasing frequency, thus driving the scientific effort to investigate health risks linked to CYN-producing blooms. Exposure to CYN occurs primarily orally, causing hepatotoxic effects. However, extrahepatic manifestations of CYN toxicity have also been reported and adverse respiratory conditions have been frequently linked to cyanobacterial blooms. Detection of cyanobacterial toxins in aerosols and dust particles raises the question of potential associated hazard of human exposure via inhalation. The susceptibility and vulnerability of human bronchial epithelia to CYN were investigated *in vitro*. To assess inhalation toxicity on airway epithelia, monolayers of immortalized human bronchial epithelial cells HBE1 and 16HBE14o- were exposed to a concentration range of 0.1-5 μM CYN. Cytotoxic endpoints were assessed as morphologic alterations, resazurin reduction capacity, esterase activity, membrane integrity and by real-time cell analysis. Both cell lines were sensitive to CYN. Depending on the endpoint assessed, EC50 values ranged between 0.8-2.1 μM (HBE1) and 1.6-4.8 μM (16HBE14o-). To evaluate alterations of other cellular events by sub-cytotoxic concentrations of CYN, phosphorylation of regulatory switches, mitogen-activated protein kinases (MAPKs) ERK and p38, was evaluated. After prolonged exposure (8-48 h), stress-activated MAPK p38 was hyperphosphorylated in both cell lines, while elevated phosphorylation levels of ERK following CYN treatment were detected only in 16HBE14o- cells. This study suggests possible hazards of cyanotoxin inhalation which might have a severe impact on the integrity of airway epithelia and epithelial cell signalling, including chronic inflammation due to extended p38 hyperphosphorylation. Further research of CYN-induced toxicity and underlying mechanisms is needed, as well as more data on environmental concentrations of cyanotoxins in aerosols and exposure assessment. The research was supported by the Czech Science Foundation Grant No. GJ17-25279Y and from H2020-MSCA-ITN-2016 Project No.722493 NaToxAq.

TH187

Effects of microcystin-LR and cyanobacterial LPS in human airway in vitro models

O. Brózman, Masaryk University, Faculty of Science, RECETOX / Research Centre for Toxic Compounds in the Environment RECETOX; B. Kubíčková, P. Laboha, Masaryk University Faculty of Science / Research Centre for Toxic Compounds in the Environment RECETOX; K. Hilscherova, Masaryk University, Faculty of Science, RECETOX / Research Centre for Toxic Compounds in the Environment RECETOX; P. Babica, Masaryk University Faculty of Science / Research Centre for Toxic Compounds in the Environment RECETOX; J. Hildebrandt, Universität Greifswald / Animal Physiology and Biochemistry

Intensified occurrence of toxic cyanobacterial blooms is becoming a major human health threat. Microcystin-LR (MCLR) is probably the most frequent and abundant cyanotoxin detected in the environment, known to induce primarily hepatotoxic effects. However, MCLR has been reported to induce *in vivo* adverse effects also on lungs and respiratory system following both inhalational exposure as well as oral or intraperitoneal administration of the toxin. Therefore, we investigated effects of MCLR in human bronchial epithelial cell lines (HBE1, 16HBE14o-, BEAS-2B). Cyanobacterial lipopolysaccharides (LPS) represent another bioactive component of cyanobacterial biomass, which is likely to expose human beings simultaneously with MCLR, thus we studied also effects of LPS isolated from a culture of cyanobacterium *Microcystis aeruginosa* PCC7806. Dose- and time-dependent formation of MCLR-protein adducts was observed in the exposed human bronchial cells. Several genes from OATP family previously implicated in the cell uptake of MCLR were found to be expressed in HBE1 and/or 16HBE14o- cells. Nevertheless, MCLR (up to 20 μM and 48 h) did not induce significant cytotoxic effects. MCLR targets protein phosphatases (PP1/PP2A), which are the major regulators of MAPKs ERK and p38. Although protein adducts with the molecular weight corresponding to MCLR-PP2A complex were detected, MCLR did not alter phosphorylation of MAPKs ERK1/2 and p38 in bronchial cell lines. Short

exposures to LPS (10 ug/mL) also did not significantly decrease cell viability and neither MCLR nor LPS affected gap junctional intercellular communication in bronchial cells lines. Regardless MCLR cell uptake, the toxin was relatively less cytotoxic to human bronchial epithelial cells when compared to the effects of other cyanotoxins (e.g. cylindrospermopsin), or in comparison with other cell types (e.g. hepatic or neural cells). Further experiments should focus on more detailed characterization of MCLR uptake and on long term effects of MCLR and LPS on inflammation-related endpoints. Inhalation toxicity of other hazardous cyanobacterial blooms components and their complex mixtures, such as extracts and LPS isolated from different cyanobacterial strains and natural water blooms, should be also investigated in the future. The research was supported by the Czech Science Foundation Grant No. GJ17-25279Y and H2020-MSCA-ITN-2016 Project No.722493 NaToxAq.

TH188

Estrogenic and retinoid-like activity in stagnant waters

M. Smutna, Masaryk University, Faculty of Science, RECETOX / RECETOX Research Centre for Toxic Compounds in the Environment; T. Prochazkova, Research Centre for Toxic Compounds in the Environment, Faculty of Science, Masaryk University / Research centre for toxic compounds in the environment; J. Priebojová, Masaryk University, Faculty of Science, RECETOX / Research centre for toxic compounds in the environment RECETOX; E. Sychrova, Masaryk University, Faculty of Science, RECETOX / Research centre for toxic compounds in the environment; L. Sehnal, Masaryk University Faculty of Science RECETOX / RECETOX Research centre for toxic compounds in the environment; K. Hilscherova, Masaryk University, Faculty of Science, RECETOX / Research Centre for Toxic Compounds in the Environment RECETOX

Cyanobacterial water blooms represent worldwide problem in many freshwater as well as marine ecosystems as producer of substantial amount of various bioactive compounds, some of which can cause adverse effects and pose risk to both aquatic organisms and human. Recent investigations indicate that cyanobacterial metabolites could include compounds with estrogenic and/or retinoid-like activity. Endocrine disruptive compounds can cause disturbance in hormone signalling and adverse effects in biota even at very low concentrations. Retinoids play a role as important signalling molecules which control vital cell processes like morphogenesis, development, reproduction or apoptosis. This study focused on freshwater ponds and reservoirs affected by water blooms and determined the estrogenic and retinoid-like activity of water by *in vitro* bioassay as well as concentrations of main estrogenic and retinoid compounds by LC-MS/MS analyses. Water samples from freshwater reservoirs and ponds with water blooms elicited estrogenic activity up to 2 ngEEQ/L. This activity could be only partly explained by the concentrations of analysed estrogenic hormones, alkylphenols or phytoestrogens. Other compounds might play a role in these effects as well. Maximal detected retinoid-like activity in water samples reached 256 ng REQ/L. We analysed the presence of nine retinoic substances, where 4-keto all trans retinoic acid and retinal were the most common forms detected in the samples. Retinoid-like activity was almost fully explained based on concentration and relative potency of individual analysed retinoids. However, results also suggest that still other compounds with retinoic acid receptor-mediated modes of action are present. Our study highlights the ability of common species of cyanobacteria to produce retinoids naturally and excrete them directly into the environment in concentrations that can reach hazardous level for vertebrates in aquatic environment such as fish or amphibians. The SOLUTIONS Project is supported by the Seventh Framework Programme (FP7-ENV-2013) of the European Union under grant agreement no. 603437.

TH189

Excitatory effects of 2,4 - diaminobutyric acid on leech Retzius nerve cell membrane potential

S. Spasić, Faculty of Medicine, University of Belgrade / Institute for Pathophysiology; M. Stanojević, V. Nedeljkov, Faculty of Medicine, University of Belgrade / Institute for Pathophysiology Ljubodrag Buba Mihailović; M. Prostran, Faculty of Medicine, University of Belgrade / Institute for Pharmacology, Clinical Pharmacology and Toxicology; S. Lopičić, Faculty of Medicine, University of Belgrade / Institute for Pathophysiology Ljubodrag Buba Mihailović

Neurotoxicity of 2,4 – diaminobutyric acid (DABA), a non-protein amino acid, was first shown after isolation from Lathyrus and related seeds, but mechanisms of neurotoxicity were never completely explained. DABA is also produced by *Cyanobacteria* in aquatic and terrestrial ecosystems. In the light of scarcity of electrophysiological studies and ubiquitous presence of DABA-producing *Cyanobacteria*, we investigated the effect of DABA on the cell membrane potential of Retzius leech neurons. Experiments were conducted on Retzius nerve cells of isolated segmental ganglia of the leech *H. sanguisuga*. Classical intracellular recording technique was performed. Cell membrane potentials were recorded using glass single-barrel microelectrodes and amplified with a high input impedance amplifier. DABA was administered in concentrations of 1, 3, 5 and 10 mM over a period of three minutes each. Input membrane resistance was investigated using current clamp technique by injecting hyperpolarizing current pulses through the recording electrode via a Wheatstone bridge unit. Application of 1mM DABA solution depolarized membrane potential by 5.01±0.43 mV (n=6, p< 0.01), while 3 mM DABA produced depolarization of 9.84±1.38 mV (n=7, p< 0.01). Rapid and substantial depolarization of membrane potential by 39.63±2.22 mV (n=9, p< 0.01) was induced by 5 mM DABA, and administration of 10 mM DABA caused membrane depolarization of 47.05±4.33 mV (n=6, p< 0.01). DABA had several times higher efficacy than Glutamate and β-N-methyalmino-L-alanine (BMAA) on our model. After washout, cells exposed to 1 or 3 mM DABA fully recovered, but only half of the cells treated with 5 mM DABA showed recovery. After application of 10 mM DABA there was no recovery. Applied in concentration of 5mM, DABA induced a decrease of the input membrane resistance by 8.09±1.51 MΩ (n=7, p< 0.01). DABA elicits substantial dose-dependent membrane depolarization. Decrease of input membrane resistance indicates that this effect is a consequence of increased membrane permeability. At higher concentrations DABA induces irreversible functional changes of neurons, confirming neurotoxic effect. As DABA is often produced together with BMAA and other cyanotoxins, some of their previously attributed neurotoxicity could possibly be due to effects of DABA and/or their synergy. **Keywords:** 2,4-diaminobutyric acid, Cyanobacteria, Retzius nerve cells, neurotoxicity

TH190

Generating ecotoxicity information on microcystins and prymnesins: A different approach

J.M. Lazorchak, U.S. EPA / Office of Research and Development; H. Haring, Pegasus c/o U.S. EPA Cincinnati; N. Dugan, U.S. EPA / Office of Research Development; J. Allen, U.S. EPA / Office of Research and Development; T. Sanan, United States Environmental Protection Agency / National Risk Management Research LaboratoryWater Supply and Water Research DivisionTreatment Technologies Evaluation Branch; C.T. Nietch, U.S. EPA / Water Supply and Water Resources Division

There is a lack of information for estimating safe levels for aquatic life concerning the toxicity of natural toxins produced by cyanobacteria and algae. Current literature indicates that LC50s for Microcystin LR range from 1 – 21 mg/L. There is even less ecotoxicity information available for prymnesin which is produced from the estuarine algae *Prymnesium parvum*. This flagellated alga has invaded freshwater systems in the U.S. and has caused numerous fish kills recorded in inland Texas lakes and blooms in 10 other states. Given the uncertainty with the purity of existing toxin standards and the cost of using them to conduct toxicity studies, a new approach is proposed using pure cultures and ambient bloom samples. Herrera, Echeverri and Ferrao-Filho (2015) conducted toxicity tests on several different cladoceran species using lyophilized phytoplankton samples collected from hyrdroelectric/drinking water reservoirs in Brazil. They found that reservoir samples with higher microcystin contents were the most toxic ones and that different cladocerans had different sensitivities to microcystin. In this study we have taken a similar approach but have used laboratory cultures of a toxin-producing strain of unicellular *Microcystis aeruginosa*, non-toxin producing filamentous strain of *Anabaena flos-aquae* and *P. parvum.* Each culture was centrifuged to separate cells from their respective culture media, then resuspended in moderately hard water. The *M aeruginosa* cells were then frozen/thawed 3 times at -80 C. The A flos aquae cells were not lysed. Forty-eight hour acute tests were conducted with *Ceriodaphnia dubia*, *Hyalella azteca* larval *Pimephales promelas* and *Neocloeon triangulifer* on both strains. A similar procedure was also used on lake water samples collected during peak bloom conditions. Current results show microcystin concentrations of 74 ug/L did not cause any significant acute toxicity to any of the four test species. The filamentous non-toxin producer *A flos-aguae* caused significant mortality to *N triangulifer* and *H azteca* (only when tested in Moderately Hard Reconstituted Water but not in Reformulated Moderately Hard Reconstituted Water) which are both grazer feeders. Mechanism of effect is undetermined at this time. August 2017 Lake Harsha bloom sample (300,000 cells/ml > WHO high risk probability value) was not acutely toxic to any of the 4 test species. Additional *P. parvum* acute results and microcystis chronic results will also be presented.

TH191

Proteomic analysis of rice plant exposed to long-term microcystin-LR exposure

J. Jiang, Nanjing Insitute of Environmental Sciences, MEP

Irrigation with cyanobacterial-blooming water containing microcystins (MCs) poses potential threat to the growth of agricultural plants. Rice (*Oryza sativa* L.) is an important grain crop and is widely grown for domestic consumption in China. However, large amounts of rice field in the middle part of China has been irrigating with cyanobacterial-blooming water. So far, the molecular mechanism of MCs-induced inhibition in the photosynthesis and growth of rice remains unclear. In the present study, rice plants were exposed to 1.0 μg/L and 50 μg/L of microcystin-LR (MC-LR) in the hydroponic nutrient solution for 34 days. The proteomic profiles of rice leaves after exposure were analysed using tandem-mass-tag labelling and LC-MS/MS analysis. The results showed that a total of 298 differentially expressed proteins were found, 89 differentially expressed proteins of them in 1.0 μg/L MC-LR treatment group, and 289 differentially expressed proteins in 50.0 μg/L MC-LR treatment group. Different response characteristics of protein expression were found in rice leaves exposed to low-concentration (1.0 μg/L) and high concentration (50.0 μg/L) of MC-LR,

respectively, and the different biological pathways involved in the mechanism of MC-LR-induced toxicity to rice were revealed using GO Term and KEGG analysis. Exposure to 1.0 μg/L and 50 μg/L of MC-LR could disturb the photosynthetic and ribosome pathways in rice leaves, causing the adverse effects on the normal growth and photosynthesis of rice. The significant alterations of the biological processes induced by the exposure to 50 μg/L of MC-LR were the inhibition of ribosome, porphyrin and chlorophyll metabolism, photosynthesis and terpenoid backbone biosynthesis related pathways, and the induction of thiamine, inositol phosphate metabolism, vitamin B6 metabolism and flavonoid biosynthesis related pathways in rice leaves. These results provided evidence of the molecular mechanisms underlying adverse effects in terrestrial plants exposed to water containing microcystins (MCs). **Keywords:** rice, microcystin-LR, photosynthesis, proteomics
Acknowledgments This research was financially supported by the National Natural Science Foundation of China (Grant number 21407056).

TH192

Probabilistic human health risk assessment for dietary exposure to aflatoxin in Taiwan

M. Ling, X. Wang, National Taiwan Ocean University; K. Lien, National Taiwan University

Aflatoxins (AFs) are secondary metabolites naturally occurring in many different kind of food, including peanuts, spices, rice, tree nuts and maize. As both genotoxic and carcinogenic substance, aflatoxins could cause severe adverse health effect. AFs have been classified as group 1 carcinogens by International Agency for Research On Cancer (IARC), because of sufficient evidence provided by cancer studies in humans and experimental animals. The purpose of this study is to evaluate the probabilistic risk of people in Taiwan who accidentally consuming aflatoxin contaminated peanut and peanut products. Concentration data (1.84 ± 4.03 ppb) are gathered from Taiwan Food and Drug Administration (TFDA) between 2005 and 2015, along with consumption rate data (from Nutrition and Health Survey in Taiwan) of five age-group (1-2 baby, 3-9 toddler, 10-17 teenager, 18-65 adult and above 65 elder) in two sub-populations (whole group and consumer only) are essential parameters for exposure analysis. Based on benchmark dose lower confidence limit 10% (BMDL10) (170 ng/kg bw/day) suggested by European Food Safety Authority (EFSA), calculated Margin of Exposure (MOE) value is below 10,000. As the result, it isn't fit the recommended standard by EFSA. According to cancer potency from Joint FAO/WHO Expert Committee on Food Additives (JECFA), estimated population risk ranged from 0.0007 to 0.2713 cancers per 100,000 population per year. This study has calculated the risk of total aflatoxins contaminated peanut and peanut products by MOE approach and population risk method. From the result of population risk for primary liver cancer (Hepatocellular Carcinoma, HCC), it is obvious that aflatoxin isn't the major cause of HCC. Despite the low cancer risk, MOE calculation indicates a possible health problem for Taiwan population. Further studies could focus on the prevention and reduction of AFs in order to reduce AFs occurrence in foodstuff, especially reducing risk for high exposure and vulnerable groups.

TH193

Organ distribution of the environmental neurotoxin β-N-Methylamino-L-alanine in the freshwater mussel Dreissena polymorpha

A. Lepoutre, UMRI 02 INERIS-URCA-ULH SEBIO; E. Faassen, RIKILT; A. Geffard, Université de Reims Champagne Ardenne; E. Lance, University Reims Champagne Ardennes / Biology and Biochemistry

Among toxins synthesized by phytoplankton, BMAA (β-N-Methylamino-L-alanine), a hydrophilic non-proteinogenic neurotoxic amino acid, has the ability to accumulate in marine and freshwater food webs, as well as that in vertebrates' brain. This toxin could promote long-term human neurodegenerative pathologies such as amyotrophic lateral sclerosis (ALS). Human exposure could occur during the ingestion of BMAA-containing food, as this neurotoxin has been detected in animals destined to human consumptions like fish, mussel and oysters. However, BMAA is an emerging toxin from which little data of toxicology or occurrence in the environment are available. In a context in which human activities are promoting the development of phytoplankton, it is important to gather information about this toxin. The zebra mussel *Dreissena polymorpha* is a freshwater filter-feeder bivalve, known for its ability to bioaccumulate substances present in the water column, and therefore could be in contact with BMAA *in-situ*. This freshwater musselhas already been used in in biomonitoring studies in order to detect heavy metals, pesticides as well as parasites and could potentially be used to biomonitor BMAA. It has already been showed that *Dreissena polymorpha* could bioaccumulate BMAA, but further information is needed to understand how this toxin is distributed in individuals. The study of BMAA has long been an analytical challenge: diverse extractions methods are available in order to study this hydrophilic compound. Through the use of polar solvents like trichloro acetic acid (TCA), it is possible to determine the "free BMAA" fraction and a hydrolysis of the whole sample will inform about the "total BMAA". As it was discovered that after a hydrolysis step, more BMAA could be release compared to untreated sample, a hydrolysis of the precipitate obtained during extraction will informed about the "precipitated bound fraction" and an hydrolysis of the supernatant will informed about the "soluble bound BMAA". Here, through and exposure of zebra mussels to 2.5 µg of dissolved BMAA/individual/day, for 21 days followed by 21 days of depuration, we studied the organ distribution of the BMAA among: hemolymph, gills, digestive gland, gonad, mantle, foot and muscles. Results will be discussed in terms of the distribution of various fraction (i.e., total, free, soluble-bound and precipitated-bound) according to the organs.

Developments in the use of bioassays for chemical and environmental risk assessment (P)

TH194

Responses to PFOA and PFBS exposure in the sediment dwelling invertebrate Dendrobaena veneta (Annelida)

P. Irato, L. Guidolin, University of Padua, Department of Biology; F. Manea, Regional Agency for the Environment, ARPA Veneto, Verona / Lab. Operative Service - Verona; G. Santovito, N. Tormen, University of Padua, Department of Biology; S. Trabucco, University of Padova / Department of Biology; A. Vantini, Regional Agency for the Environment, ARPA Veneto, Verona / Lab. Operative Service - Verona; L. Tallandini, University of Padova / Department of Biology

The aim of this work was to study, in the sediment-dwelling Invertebrate Dendrobaena veneta (Annelida), bioaccumulation patterns and cellular and biochemical responses in coelomocytes (mortality and lysosomal membrane stability), and at tissue level (GPX and MTs), following the exposure to two perfluorinated alkyl acids (PFOA and PFBS) for short (72 h) and longer (14 and 28 days) times. The exposures were carried out in soil microcosms prepared with glass containers filled with 300 ml of soil humidified at 30% with PFOA or PFBS spiked water.\nAs for the 72 h tests the exposures were performed, at two different nominal concentrations, i.e. 30% of 1x or 10x MAC-EQS fw values (Maximum Acceptable Concentration-EQS calculated by the Italian Working Group for the derivation of Environmental Quality Standard (EQS)) while for the long time the PFOA, or PFBS, nominal concentrations were 30% of 5x MAC-EQS fw values.\nDifferent accumulation patterns were observed for PFOA and PFBS, with PFOA no longer accumulating between 14 and 28 days, while PFBS continues to be accumulated up to 28 days.\nSignificantly higher coelomocyte mortalities than in the controls, with both compounds, were detected after the 14 and 28 days exposures. As for the lysosomal membrane stability significant decreases were detected both after the short and the long-time exposures. In the soft tissues preliminary data don't show significant differences between control and treated organisms regarding the GPX activity. A significant MT total decrease was detected after PFOA exposure, both at 14 and 28 days while after PFBS exposure only at 14 days. As for MT, because it has been reported that PFASs seem to increase ROS levels, we determined not only the total protein concentration but also the oxidized fraction (MTox). A significant increase in the MTox fraction in PFOA treatment after 28 days and in PFBS after 14 days was observed.\nOur results show, for this invertebrate organism, a higher PFBS bioaccumulation than PFOA and significant exposure effects to the two PFASs both in coelomocytes, the main immunodefensive system cells of the organism, and in the soft tissues. Further studies are planned to explore the mechanisms underlying these results.\n\n

TH195

Toxicity of Per- and Polyfluoroalkyl substances on Chironomus dilutus for use in a relative toxicity model

C.J. McCarthy, CH2M / Environmental Services; M. Stanaway, B. Muckey, Test America; C. Salice, Towson University / Environmental Science & Science Dept.; D. Wright, CH2M

Per- and polyfluoroalkyl substances (PFASs), including perfluorooctane sulfonate (PFOS) and perfluorooctanoate (PFOA) are commonly elevated in soil and groundwater. High detection frequency and concentration has resulted in identification of PFAS as compounds of interest and as emerging contaminants due to their regulatory uncertainty. Published toxicological research to date relates to PFOS and PFOA only and for a limited number of organisms. The lack of robust and defensible ecotoxicity data on other PFASs hinders risk assessment and leads to unsupported risk management decisions. Given this gap in understanding of the additional compounds, the Strategic Environmental Research and Development Program (SERDP) is funding research of these additional PFAS and classes of organisms. This discussion will summarize the first phase of a SERDP research grant to address these needs. Tests were conducted with a common aquatic test species to identify patterns of relative toxicity between the PFASs. *Chironomus dilutus* tests included a 96-hour reference toxicant test, a 10-day range finding test, and a 20-day definitive bioassay. For shorter duration *Chironomus* tests, the main endpoint of interest was survival while for longer-duration tests (20 days), the more sensitive growth endpoint was measured.Opportunistic measurements of deformation were also included to enhance the understanding of potential toxicity to these compounds. Test results will ultimately be used in conjunction with concurrent tests being conducted with avian and reptilian model species to the same chemicals to develop a relative toxicity model. Endpoints measures from the aquatic species tests will be used to identify clear patterns of relative toxicity of the tested PFASs. Results will inform and prioritize PFAS testing on avian and reptilian species. In addition, the relative potency patterns observed after aquatic testing will be reassessed upon completion of the upper trophic level exposure studies. Once all phases of toxicity testing are complete, the results will be used to help develop a risk management framework for addressing potential environmental management issues of PFAS.

TH196

Interpretation of bioassay results in the context of the soil quality TRIAD approach.

N. Pucheux, INERIS; S. ANDRES, INERIS / Toxicological Ecotoxicological Assessment of chemical Substances; P. Pandard, INERIS / Expertise and assay in ecotoxicology unit

The recently standardized method ISO 19204 "Soil quality – Procedure for site specific ecological risk assessment of soil contamination (Soil quality TRIAD approach)" describes in a general way the application of three combined lines of evidence (chemistry, ecotoxicology and ecology) along a tiered approach. Regarding the ecotoxicological component, the TRIAD approach consists in carrying out a battery of bioassays on soil samples and to scale the results from all bioassays to calculate an ecotoxicological combined risk score In order to evaluate its applicability and the relevance of the proposed tools, INERIS used the soil quality TRIAD approach on an applied case: an open mine operated for 60 years and which the activities stopped about 100 years ago (the "TRIPLE" project 2016-2017). Among the conclusions, it was noticed that the selection of the control soil may have a significant influence on the expression of the results and therefore on the risk assessment. This impact is particularly obvious for the assessment of a heterogeneous site and for the first TIER (screening level) of the TRIAD method. This statement is illustrated by observing the consequences on the assessment conclusion when the results of bioassays are expressed according different control/reference soils.

TH197

Estimating the hazardous concentrations of nonylphenol for soil ecosystem protection with probabilistic approach

J. Kwak, J. Moon, D. Kim, Konkuk University; R. Cui, Konkuk University / Department of Enviromental Sciences; Y. An, Konkuk University / Department of Environmental Heath Science

Nonylphenol is known as a xenoesetrogen but is still used for pesticides, detergents, surfactant cleaners and packaging. However, few researches on soil ecotoxicity of nonylphenol were reported. In the present study, we conducted a battery of bioassay, generated soil ecotoxicity data and then estimated soil hazardous concentration for nonylphenol for soil ecosystem protection based on species sensitivity distribution (SSD). For the acute assay, eight soil species from six different taxonomic groups (magnoliopsida, liliopsida, chlorophyceae, secernentea, clitellata and collembola) were tested. Also, for the chronic assay, five soil species from four different taxonomic groups (magnoliopsida, liliopsida, chlorophyceae and collembola) were investigated. Finally, acute and chronic hazardous concentrations for HC_5, HC_{10}, HC_{20}, HC_{50} were suggested for protection of soil ecosystem. These values can be applied to environmental risk assessments for nonylphenols. *This study was funded by the Korea Ministry of Environment (MOE) as the Environmental Health Action Program (1485014458)*

TH198

Organophosphate Triesters and Selected Metabolites Enhance the Binding of Thyroxine to Human Transthyretin In Vitro

K.L. Hill, Intrinsik / Department of Biology; R.J. Letcher, Environment and Climate Change Canada / Ecotoxicology and Wildlife Health Division; T. Hamers, VU University Amsterdam, Institute for Environmental Studies (IVM) / Department of Environment and Health; J. Kamstra, NMBU / BaSam; W. Willmore, Carleton University / Department of Biology

The toxicological properties of organophosphate (OP) triesters that are used as flame retardant and plasticizer additives are currently not well understood. However, there is increasing evidence that suggests OP triesters can affect the thyroid system. Perturbation of thyroid hormone (TH) transport is one mechanism of action that may affect thyroid function. The present study applied an *in vitro* competitive protein binding assay with thyroxine (T4) and human transthyretin (hTTR) to determine the potential for the OP triesters, TDCIPP (tris(1,3-dichloro-2-propyl) phosphate), TBOEP(tris(butoxyethyl) phosphate), TEP (triethyl phosphate), TPHP (triphenyl phosphate), *p*-OH-TPHP (*para*-hydroxy triphenyl phosphate), and the OP diester DPHP (diphenyl phosphate), to competitively displace T4 from hTTR. Enhancement of T4 binding to hTTR, rather than the hypothesized competition, was observed for the six OP triesters and DPHP and in a concentration-dependent manner. For example, T4-hTTR binding was significantly increased at concentrations of TBOEP as low as 64 nM, and up to 184 % of controls at 5,000 nM. A plausible explanation of these results, which to our knowledge has not been previously reported, may be allosteric interactions of the OP esters with hTTR allowing T4 to access the second site of the TH binding pocket. It is plausible that OP triester and diesters can covalently bond to residues of serine, lysine or tyrosine on the surface of hTTR, resulting in a conformational change in the dimer-dimer interface and allowing for both TH binding pockets to be accessible for T4. These *in vitro* results suggest a novel mechanism of OP ester toxicity via T4 binding enhancement, and possible dysregulation of T4-hTTR interactions.

TH199

In Vitro and In Silico Competitive Binding of Brominated Polyphenyl Ether Contaminants With Human and Gull Thyroid Hormone Transport Proteins

K.L. Hill, Intrinsik / Department of Biology; . Mortensen, NTNU University / Department of Biology; D. Teclechiel, Accustandard; W. Willmore, Carleton University / Department of Biology; I. Sylte, The Arctic University of Norway / Department of Medical Biology; B.M. Jenssen, Norwegian University of Science and Technology / Biology; R.J. Letcher, Environment and Climate Change Canada / Ecotoxicology and Wildlife Health Division

Tetradecabromo-1,4-diphenoxybenzene (TeDB-DiPhOBz) is a highly brominated additive flame retardant (FR). Debrominated photodegradates of TeDB-DiPhOBz have been shown to be enzymatically hydroxylated *in vitro* in herring gulls (*Larus argentatus*) liver assays, including one metabolite identified as 4''-OH-2,2',2'',4-tetrabromo-DiPhOBz. Chemically related methoxylated tetrabromo- to hexabromo-DiPhOBzs are known contaminants in herring gulls from the Laurentian Great Lakes of North America. To our knowledge, nothing is currently known about the biological effects of these polybrominated (PB) DiPhOBz-based compounds. The present study investigated the potential thyroidogenicity of 2,2',2'',4-tetrabromo-DiPhOBz along with its methoxy- and hydroxy-analogues, using an *in vitro* competitive protein binding assay with human thyroid hormone (TH) transport proteins transthyretin (TTR) and albumin (ALB). *Para*-OH-tetrabromo-DiPhOBz was found to be capable of competing with thyroxine (T_4) for the binding site on human TTR and ALB. The *para*-MeO-tetrabromo-DiPhOBz and the tetrabromo-DiPhOBz were much less competitve. *In silico* analyses were also conducted using a 3D homology model for gull TTR, to predict whether these tetrabromo-DiPhOBz-based compounds may also act as ligands for an avian TH transport protein despite evolutionary differences compared with human TTR. This analysis found all three tetrabromo-DiPhOBz analogues to be potential ligands for gull TTR, and with similar binding efficacies to THs. Overall, the results indicated both species- and structure-related differences in binding affinities of these ligands, and suggest there is potential for these exogenous chemicals to interact and possibly influence vertebrate thyroid hormone-dependent function.

TH200

Phosphine changes cytochrome c oxidase in Sitophilus oryzae

K. Kim, H. Kim, H. Jeon, Y. Choi, Y. Kim, S. Lee, Kyungpook National University

Phosphine resistance in the stored product insect pests has been reported over the world. In South Korea, *Sitophilus oryzae* has been developed phosphine resistance after the severe use of phosphine. In this study, how *S. oryzae* survived under the recommended dose of phosphine and we assessed the biochemical and molecular mechanisms for referring phosphine resistance. Three strains of *S. oryzae* were prepared as control groups (C), medium-resistant group (MR), and strong resistant groups (R) for this study. One of target sites of phosphine is cytochrome c oxidase (COX) and we analyzed the enzyme activities within the three stains. The highest COX activities were found in R groups with about 1.5-fold increase when compared to the controls. IC50 values on the COX activity by ethyl formate, one of COX inhibitors, was 2.82, 3.71 and 4.55 (mM) for C, MR, and R strains. Lineweaver-Burk plot for COX using ethyl formate exhibited different modes from R strains to C strains. And six genes *cat*, *jhip*, *voltage*, *casp*, *wnt7*, *wnt11* were analyzed using RT-PCR for comparing gene expression and *cat* gene was dramatically down-regulated in the R strain. *Jhip* gene expressing juvenile hormone inducible protein was differently expressed in the two phosphine-resistant strains. wnt 7 gene was also up-regulated in the R strain, but it was not so big different. Three biomarker enzymes such as acetylcholinesterase, glutathione *S*-transferase, and carboxylesterase activities were also determined within the three strains. Only glutathione *S*-transferase activity decreased in the R strain. Taken together, phosphine resistance in *S. oryzae* may be related to the changes in COX enzyme and up-regulation of *jhip* gene expressing juvenile hormone inducible protein.

TH201

Effects of additives in mobile phases in simultaneous analysis of glutathione and glutathione disulfide by HPLC-MS/MS

S. Baik, KIST Europe / Environmental Safety Group; Y. Jung, KIST-Europe; Y. Kim, KIST Europe / Environmental Safety Group

Glutathione is an important non-protein compound and existed in both internal and external of cells. Regarding toxicological effects induced by oxidative stress, ratio of reduced form (GSH) to oxidized form (GSSG) of glutathione is one of important biomarkers. Among all available assays to detect and quantify GSH and GSSG, using high-performance liquid chromatography (HPLC) coupled with mass spectrometry (MS) is very essential with the development of highly selective and sensitive analytical methods. GSH and GSSG are usually analyzed in positive ionization in electrospray, and formic acid can be a general additive in mobile phases for a better protonation in positive ionization mode in MS. In this work, we investigated the effects of additives other than formic acid for a better understanding to enhance the ionization of GSH and GSSG in the gas field of MS source. With the presence of formic acid only, as a result, protonation of GSSG was very limited. However, using formic acid and ammonium acetate together in mobile phases delivered the enhancement of protonation for both GSH and GSSG. Furthermore, as increasing the concentration of ammonium acetate from 5mM to

50mM, sensitivities of GSH and GSSG were increased from 0.0034 to 0.0072 and 0.028 to 0.046, respectively. Detection limits of both GSH and GSSG were also significantly lowered as using higher concentration of ammonium acetate. This indicates that producing ammonium adducts followed by dissociating ammonium ions from adducts to protonate GSH and GSSG is important mechanism for protonation of these compounds with using ammonium acetate as a mobile phase additive. This enhanced methodology was applied to zebrafish liver cells (ZFL) to investigate recoveries of both GSH and GSSG and achieved more than 100% recovery for GSH and around 100% recovery for GSSG. The achievement of higher recovery of GSH than 100% was because ZFL itself might not be oxidized to form GSSG. In addition, same method was also applied to ZFL exposed to different concentrations of a target chemical as well as 6 mg/L of H_2O_2, a negative control. The lowest concentration of GSSG in this work was 5.0 ng/ml, higher than its detection limit, 2.0 ng/ml. This is meaningful because it could not be achieved by other conventional methods and assays with higher detection limit than its original concentration. Therefore, we can conclude that our method could avoid underestimation to quantify biomarkers such as GSH and GSSG.

TH202

Rapid analysis of bivalves' xenometabolome using High Resolution Mass Spectrometry

D.A. Muñoz, Water and Soil Quality Research Group, Department of Environmental Chemistry, IDAEA-CSIC / Water Quality; M. Olmos, IDAEA-CSIC / Water and Soil Quality Research Group; M. Rambla-Alegre, IRTA; S. Monllor, N. Guillem, Institute of Environmental Assessment and Water Research IDAEA CSIC / Department of Environmental Chemistry; J. Diogène, IRTA / Marine Environmental Monitoring Subprogram; M. Farre, IDAEA-CSIC / Environmental Chemistry; M. Lopez de Alda, Institute of Environmental Assessment and Water Research; D. Barcelo, IIQAB-CSIC / Department of Environmental Chemistry

A wide variety of contaminants are released to the environment every day from residential, commercial and industrial uses. They are simultaneously present at different levels in aquatic ecosystems making a “cocktail” of hazardous substances. These xenobiotics interact with wild organisms and may be bioaccumulated. They can have negative implications from an environmental point of view, affecting wild life, but also they may be of great concern from a human health perspective, when they accumulate in highly consumed organisms like bivalves. Given that it is unrealistic to assess every possible combination of chemical substances accumulated by organisms, the major challenge now is to develop systematic ways of addressing these chemical mixtures, and to identify priority mixtures of potential concern. For this purpose, the profiling of the xenometabolome, or range of xenobiotics and their metabolites in an organism exposed to environmental contaminants, seems to be the way forward. In the present work, for the profiling of the xenometabolome, a fast analytical method has been developed for the extraccion and identification of priority contaminants in bivalves from Ebro Delta, Spain. A literature research was done in order to gather all the information available regarding the Ebro Delta and possible sources of contamination. Taking into account this information a mixture of compounds representative of the different contaminant sources identified in the area was selected. This mixture including pesticides, plasticizers, antibacterials, preservatives, stimulants, and pharmaceuticals was used for a recovery study with three different bivalves types of high commercial interest such as mussel, oyster and cockle. QuEChERS (Quick, Easy, Cheap, Effective, Rugged and Safe) was used for the extraction and purification of the samples. Then the purified extracts were injected in Orbitrap-Q-Exactive for identification and quantification of the priority contaminants. Twenty compounds including endocrine disrupting compounds, pesticides, and pharmaceuticals were extracted with recoveries ranging from 40.54 to 105.51 %. Quality parameters such as method detection and quantification limits, accuracy, and precision were studied. Besides, non-target analysis of other relevant contaminants that may be present in bivalves’ xenometabolome is ongoing.
Acknowledgements CTM2015-73179-JIN (AEI/FEDER/UE); GLOBAQUA (603629); 2014 SGR 418; Bekolut and Merck.

TH203

River ecosystem: an ecosystem approach to evaluate the ecological risk linked to the human health protection

L. Mancini, Istituto Superiore di Sanità / Department of Environment and Health; C. Puccinelli, Italian Institute of Health ISS / Department of Environment and Health; L. Tancioni, University of Rome Tor Vergata / Biology department; M. Carere, Italian Institute of Health ISS; E. D'Ugo, Italian Institute of Health ISS / Department of Environment and Health; S. Caciolli, Italian Institute of Health ISS / Department of Environmental and Health; R. Giuseppetti, F. Chiudioni, Italian Institute of Health ISS / Department of Environment and Health; S. Marcheggiani, Istituto Superiore di Sanità / Department Environment and Health

The anthropogenic pressure on river ecosystems can induce changes on their structural and functional characters as well as an increasing risk for human health. Over the last years an ecosystem approach mainly based on multilevel bioindicator methods has been adopted for assessing the risk for human health. The samples processing plays a key role in the environmental analysis. Samplings were carried out in an area characterized by strong anthropogenic pressures (Tiber River Central Italy).The water samples were processed using the technique of filtration and concentration, as suggested within the European project µAQUA FPVII; at the same time, tests were carried out using raw water samples. The approach of this study is based on the following parameters: biological community (diatoms, macro invertebrates, macrophytes and fishes fauna); chemical–physical parameters, a set of ecotoxicological bioassays (*Vibrio fischeri*, *Daphnia magna and Vicia faba*), microbiological analysis(*Salmonella spp*, *Staphyloccocus spp*, *Clostridium spp*. and *Campylobacter spp)*and virological analysis that included *viral Hepatitis* HAV and HEV, Norovirus NoGI and NoGII, Reovirus, Enterovirus: A, B and C, Adenovirus: ADV40 and ADV41. The results of this study showed that the pre-concentration of larger amount of water improves both the contaminants detection within aquatic ecosystems and the ecological risk evaluation. The ecotoxicological analysis is an important aspect in the integrated approach to evaluate the ecological risk linked to the human health protection. The integrated approach adopted has been a useful tool to describe the ecological status of surface waters and the related risk for human wellbeing, providing a complete and organic vision of the qualitative state of the ecosystem. In conclusion these results highlighted the different levels of alteration and the potential need for the primary prevention and restoration, confirming that an ecosystem approach plays a key role in the ecological and human health risk assessments.

TH204

INTEGRATED EXPOSURE AND EFFECT DATABASE TOOLS TO SUPPORT HAZARD AND RISK ASSESSMENT

K. Tollefsen, NIVA / Ecotoxicology and Risk Assessment; Y. Li, G. Løvoll, J. Vedal, L. Xie, NIVA   Norwegian Institute for Water Research; Y. Song, NIVA Norwegian Institute for Water Research / Ecotoxicology and Risk Management; K. Petersen, Norwegian Institute for Water Research

Use of available exposure and effect data are key to performing hazard and risk assessment of pollutants, and compiling different sources of data are often done in a case-by-case manner. Processing data is thus cumbersome and time-consuming, whereas the availability of data is a large source of uncertainty in resulting assessments. The NIVA Risk Assessment database (NIVA RAdb^{TM}) has been developed as a module-based tool to facilitate the assembly, organisation, integration, visualisation and quality assurance of available exposure and effect information in order to speed up and perform consistent handling of relevant data. The NIVA RAdb^{TM} compile available experimental and predicted (computational) effect data that range from molecular and cellular responses characterising the mode of action (MOA), typically derived from high-throughput and high-content (*in vitro*) bioassays, to (apical) adverse data derived from whole organism bioassays of potential regulatory relevance. These effect data are assembled within the context of Adverse Outcome Pathways (AOPs) by anchoring data to initial cellular responses referred to as molecular initiating events (MIE), to downstream key events (KE) at the cellular/organ level and finally to adverse outcomes (AO) at the individual or organism level. The resulting multi-level assemblies of data can be used in hazard assessment to identify the MOA of one or more stressors, to link molecular responses to higher organisation level effects and to identify potential stressors among large assemblies of pollutants that can give rise to a given AO. The NIVA RAdb^{TM} also support risk assessment by calculating risk quotients (RQs) of single pollutants and mixtures of these on basis of exposure (typically measured or predicted environmental concentrations) and effect data (typically NOEC, ECx, PNEC or EQS values) and can identify risk drivers (most toxic chemicals), relevant toxic endpoints (i.e. MIE, KE and AO) and susceptible species for a given exposure scenario. Recent development includes integration of non-chemical stressors such as ionizing and non-ionizing radiation.Examples on uses on specific exposure scenarios will be presented to show the utility of the databases and the tools developed. **Acknowledgements:** RCN projects 221455-EDRISK (www.niva.no/edrisk), 268294 MixRisk (www.niva.no/mixrisk), 223268 CERAD (www.nmbu.no/en/services/centers/cerad), and EU-FP7 project SOLUTIONS (http://www.solutions-project.eu/project/).

TH205

Assessing exposure risk for marine bivalve Mytilus posed by microplastic polystyrene particles

C. CHEN, Y. Yang, National Taiwan University / Bioenvironmental Systems Engineering; H. Lin, National Taiwan University; C. Liao, National Taiwan University / Department of Bioenvironmental Systems Engineering

BACKGROUND: Microplastics (MPs) are one of the most widespread emerging pollutants in aquatic ecosystems, posing impacts on marine organisms. However, little is explored for potential risks of environmentally relevant concentrations of MPs in organisms. **OBJECTIVE:** To evaluate the immunotoxic risks of marine bivalve *Mytilus* posed by environmentally relevant concentrations of polystyrene microplastics (PS-MPs) and MPs based on bioassay results from related published literature. **METHODS:** We used Hill-based dose-response model to simulate the effects of PS-MPs on the lysosomal destabilization and phagocytosis in bivalves. The predicted no-effect concentrations (PNECs) causing 1% inhibition of immune functions were also estimated. Moreover, a risk-based probabilistic model was used to characterize the potential hazards of marine bivalves in response to predicted environmental concentrations (PECs) of PS-MPs or MPs by quantifying exceedance risks (ERs) and hazard quotients (*HQ*s) in five plastic-filled gyres.

RESULTS: We found that PNECs for inhibition of lysosomal membrane stability and phagocytic cells were 0.04 and 0.07 µg mL^{-1}, respectively, implicating that phagocytosis is a more sensitive endpoint for immune responses in bivalves. In addition, our results demonstrated that the North Pacific Ocean appeared to be the greatest risks among global oceans. **CONCLUSIONS:** The strict thresholds estimated by applying a environmental risk assessment framework could be recommended as a criteria for environmental management of PS-MPs or MPs. Potential effects of PS-MPs/MPs on marine organisms at higher trophic levels should also be taken into consideration. *Keyword*s: Polystyrene microplastics; Bivalve; Environmental risk assessment; Predicted no-effect concentration; Predicted environmental concentration; Hazard quotient

TH207

Innovative Design of Nationwide Dutch Water Quality Monitoring

M. de Baat, University of Amsterdam / IBED-FAME; Y. Coolen, D. van der Pouw Kraan, R. Rood, University of Amsterdam / Department of Freshwater and Marine Ecology; M. Kraak, University of Amsterdam / IBED-FAME

According to the European Union Water Framework Directive (EU-WFD), chemical surface water quality is assessed by analysing the concentration of 45 priority compounds. However, the analysed chemicals are often absent and biological effects are thus caused by (un)known (mixtures of) compounds. Alternatively, water quality can be assessed by observing adverse effects of surface water on test organisms. Therefore, the present study aimed to innovate surface water quality assessment by applying an innovative design in a nationwide monitoring campaign in The Netherlands. To this purpose bioassays with two aquatic invertebrate species, *Daphnia magna* and *Chironomus riparius*, were employed and the performance of passive sampling techniques to include time integrated compound concentrations was explored. *D. magna* neonates and *C.riparius* larvae were exposed to surface water samples from 34 locations. Daphnids were additionally exposed to POCIS passive sampler extracts from 7 of these locations. For the daphnids, none of the surface water samples or passive sampler extracts caused significant mortality after 48h of exposure. In contrast, for the chironomids, three surface water samples caused significantly lower larval survival compared to the controls. The use of *C. riparius* bioassays thus allowed for differentiation between water quality of the sampling locations. Possible explanations for the observed chironomid mortality include insecticide sorption to the provided food, which may lead to increased exposure resulting in higher mortality. A possible culprit compound could be the neonicotinoid imidacloprid, which was detected at two locations with observed chironomid mortality. Moreover, toxicity of imidacloprid to *C. riparius* is 500 times higher than to *D. magna*. This could thus explain the high mortality at these greenhouse locations. It is concluded that the use of bioassays with multiple test species provides better insight into surface water quality, and is therefore a valuable addition to regular water quality monitoring.

TH208

Smart Monitoring: Application of innovative tools in nationwide water quality assessment

M. de Baat, M. Kraak, University of Amsterdam / IBED-FAME; R. van der Oost, Waternet / Onderzoek en Advies; P. de Voogt, University of Amsterdam / IBED; P. Verdonschot, University of Amsterdam / Department of Freshwater and Marine Ecology

The European Union Water Framework Directive requires member states to assess chemical water quality of surface waters by monitoring the presence of 45 priority substances. However, these substances are often banned and their concentration in surface waters is strongly decreased, frequently to levels below the limits of detection. Simultaneously, industries have switched to a myriad of alternative compounds that have serious impacts on water quality, most of which are not listed as priority substances. Consequently, a large portion of the observed toxic effects of surface waters cannot be attributed to compounds measured by the water authorities. Hence, there is an urgent need for an effect-based monitoring strategy that employs bioassays to identify environmental risks. Therefore, the aim of the present study was to implement innovative tools in a smart, integrated monitoring strategy, applied in a nationwide water quality assessment campaign in The Netherlands. The Smart Monitoring strategy combines passive sampling (PS) with a battery of bioassays to investigate ecotoxicological risk to aquatic biota. At 47 locations silicone rubbers and Polar Organic Chemical Integrative Samplers (POCIS) were exposed to surface water for 6 weeks. Alongside the PS a 7-day *in-situ* daphnid test was performed at all locations. Subsequent to field exposure, accumulated compounds were extracted from the PS after which a battery of 3 *in-vivo*, 5 antibiotic SCAN and 10 *in-vitro* Chemical Activated LUciferase gene eXpression (CALUX) bioassays was exposed to the re-dissolved extracts. The bioassay battery was selected such that it can identify the risk posed by a wide range of chemical pollutants and their transformation products, while simultaneously allowing for more targeted identification of groups of compounds that cause specific effects. Bioassay responses were compared to effect-based trigger values to identify potential ecotoxicological risks at the investigated locations. Subsequently, the SIMONI model was applied to rank sites based on ecotoxicological risk, rather than on the presence of priority compounds. It is concluded that the Smart Monitoring strategy allowed prioritization of sites based on ecotoxicological risks, identified the presence of hazardous compounds, regardless of being listed as priority compounds, but meanwhile could prevent costly chemical analysis at sites with low ecotoxicological risks.

TH209

Passive sampling in effect-based monitoring of two European rivers - explicability of in vitro toxic potentials by detected chemicals

J. Novak, Masaryk University / RECETOX; Z. Tousova, B. Vrana, Masaryk University, Faculty of Science, RECETOX / RECETOX Research Centre for Toxic Compounds in the Environment; F. Smedes, RECETOX / Environmental chemistry and modelling; R. Grabic, University of South Bohemia in Ceske Budejovice / South Bohemian Research Center of Aquaculture and Biodiversity of Hydrocenoses; S. Ait-Aissa, INERIS / UMRI SEBIO ECOT; M. Smutna, Masaryk University, Faculty of Science, RECETOX / RECETOX Research Centre for Toxic Compounds in the Environment; K. Hilscherova, Masaryk University, Faculty of Science, RECETOX / Research Centre for Toxic Compounds in the Environment RECETOX

EU commission Water Framework Directive considers employment of passive sampling and use of effect-based tools in the monitoring of aquatic pollution. A combination of both approaches was used for monitoring of two rivers differing significantly in pollution levels. The Bosna, moderate-sized river in Bosnia-Herzegovina, which is burdened by untreated wastewaters, was sampled by semipermeable passive sampling devices (SPMD) and POCIS samplers. The Danube, the largest river in the EU with relatively low pollution level, was sampled using a mobile dynamic passive sampling device with silicone rubber (SR) and SDB-RPS Empore™ (ED) disc samplers. Both sampler sets consisted of partitioning sampler for non-polar chemicals (SPMD, SR) and adsorption sampler for the polar-ones (POCIS, ED). For the partitioning samplers, concentrations of collected chemicals in river water were derived using dissipation of performance reference compounds. For the adsorption samplers, the sampling rates were either taken from literature (POCIS) or calculated from correlated levels of chemicals that were detected both in adsorption (ED) and partitioning samplers (SR). The samples were analyzed for aryl hydrocarbon-, estrogen- and androgen receptor-mediated effects using *in vitro* bioassays. The effects were expressed as bioanalytical equivalents (BEQ_{bio}) of respective model compounds in water. The BEQ_{bio} levels were significantly higher in extracts from POCIS and ED samplers showing that the polar chemicals were responsible for most of the detected effects. Chemical analyses detected 103 and 209 chemicals in the Bosna and the Danube samples, respectively. The passive sampling allowed detection of chemicals at pg/L concentrations. The levels of chemicals with known biological potency for the studied endpoints were used for modeling of bioanalytical equivalents (BEQ_{chem}). The comparison of bioassay- and chemical analysis-derived equivalents showed that the detected chemicals explained mostly a low fraction of the BEQ_{bio}. Only in the case of estrogenicity in extracts of the samplers collecting polar chemicals, the BEQ_{chem} was comparable with the BEQ_{bio} levels. Both sampler combinations proved to be suitable for the detection of a large set of chemicals even at trace levels and for the complementary assessment of the biological potentials of the environmental mixtures. The SOLUTIONS Project was supported by the 7th Framework Programme EU (FP7-ENV-2013) with grant agreement no. 603437.

TH210

Testing of realistic contaminant mixtures with the harpacticoid copepod species Nitocra spinipes using passive sampler extracts

S. Moeris, Ghent University (UGent) / Laboratory for Environmental Toxicology and Aquatic Ecology; J. Koch, GhEnToxLab (Ghent University) / Applied Ecology and Environmental Biology; K. De Schamphelaere, Ghent University (UGent) / Applied Ecology and Environmental Biology

The use of passive sampling as a tool in environmental monitoring has gained wide acceptance within the past decades. More recently the possibilities of combining passive sampling and biotesting gained higher attention and researchers focus on reconstituting environmentally realistic contaminant mixtures in aquatic biotest systems. Equilibrium based samplers (e.g. silicone rubber sheets) can mostly be used as passive dosing devices in biotest systems without prior treatment but have the disadvantage that only one single concentration level can be tested. For integrative samplers (e.g. Speedisks™) an extraction is needed before spiking of biotest medium and the downside of this approach is that an extraction always changes the natural mixture composition due to compound specific partition coefficients. The advantage on the other hand is that the extraction of the samplers is well established, efficient and easy to combine with chemical analysis. Thus, even though this approach does not reconstitute the natural concentration profile quantitatively it allows for testing of environmentally realistic contaminant mixtures in terms of qualitative chemical composition. In the current study we extracted Speedisk™ passive samplers that were deployed for 8 weeks in two Belgian harbours and one location next to one of the harbours to spike a 7-day larval development test with the harpacticoid copepod *Nitocra spinipes* following ISO 18220. In order to fractionate the compounds on the Speedisks™ we followed two different procedures: a sequential and a parallel extraction approach using three solvents: acetonitrile, ethyl acetate and dichloromethane. We exposed 80 larvae divided into 8 replicates in a fully randomized setup including controls and solvent controls to each of the Speedisk™ extracts and counted larvae and copepodites after

5, 6 and 7 days to calculate the larval development ratio. Results showed no statistically significant developmental effects for all tested extracts. The tested concentrations after solvent spiking in our test system were slightly below environmentally realistic contaminant concentration levels. Overall the larvae showed to be unaffected by the exposure to the Speedisk™ extracts and we expect no direct effects of environmentally realistic contaminant mixtures on the development of *N. spinipes.*

TH211

Passive dosing of polar and non-polar substances using Oasis HLB® - Pre-equilibration of media for transferring complex mixtures.

D. Kämpfer, Institute for Environmental Research (RWTH Aachen University) / Institute for Environmental Research (Bio V); T. Seiler, RWTH Aachen University / Ecosystem Analysis; H. Hollert, K. Smith, RWTH Aachen University / Institute for Environmental Research; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics

The passive dosing approach is currently focused on hydrophobic organic substances. Dosing phases such as polydimethylsiloxane (PDMS) silicone are suitable for this task, but also limit the approach to specifically this group of substances. However, metabolism, volatilization and sorption – particularly medium sorption – are also highly relevant for polar substances. Therefore, a requirement for the passive dosing of these substances also exists. To overcome the current limitations of silicone, the applicability of Oasis HLB for dosing polar and non-polar substances was tested in a neutral red assay. The findings show a high affinity of Oasis HLB for non-polar, polar as well as some ionic substances. It could also be shown, that the adsorption based accumulation of the substances was reversible and – due to the high surface area and the wettable pores – relatively fast. This demonstrates its suitability for dosing a broad range of substances. With respect to combining equilibrium passive sampling and dosing for the recreation of field mixtures in toxicity test, pre-equilibration of the cell culture medium with Oasis HLB was successfully tested and compared with the direct passive dosing using Oasis HLB. On the one hand, the medium pre-equilibration approach enables one to control the role of temperature on the equilibrium state. On the other hand, due to the fact that the dosing phase is not directly introduced in the assay, maintaining of the test concentration over the test duration is diminished for some compounds. In summary, the application of Oasis HLB as a passive dosing phase was successfully established and medium pre-equilibration for re-establishing field mixtures in an exposure medium was tested. This opens up the possibility of recreating broad mixtures sampled with Oasis HLB at natural ambient concentrations in toxicity and other tests.

TH212

Passive dosing strategy for in vitro test systems: static concentration generator and continuous release

F. Begnaud, Firmenich / DRAP; C. Debonneville, Firmenich / Research and Development; V. Laubscher, F. Berthaud, Firmenich SA / DRAS; H. Schug, Eawag - Swiss federal Institute of Aquatic Science and Technology / Environmental Toxicology; C. Kropf, University of Bern / Centre for Fish an Wildlife Health; K. Schirmer, Eawag / Environmental Toxicology; H. Segner, University of Bern / Centre for Fish and Wildlife Health; S. Gimeno, Firmenich / Product Safety and Regulatory Affairs

The ability to generate a true solution of a chemical substance at controlled concentrations is essential to generate meaningful aquatic toxicity information. This is especially critical for *in vitro* test systems, and becomes highly challenging when dealing with hydrophobic ($logK_{ow}$> 3.5) and volatile substances like some fragrances. Historically, solvents were used to enhance the solubility, but their potential impact on results (e.g. could impact transmembrane permeation, increase bioavailability, disrupt enzymes, generate oversaturated solutions) triggered the search for alternative solutions. Passive dosing has proven to be effective to generate solutions of truly dissolved substances at controlled and constant concentrations. To increase the robustness of *in vitro* alternative approaches, involving permeation and biotransformation, to the fish bioconcentration test, we set up a global strategy to prepare solutions of hydrophobic substances using customized PDMS-reservoirs. These tube-shaped reservoirs were used either in static mode to prepare the test solutions for the *in vitro* biotransformation test with rainbow trout S9 or hepatocytes, or in dynamic mode to maintain a constant concentration in a selected compartment of the permeation setup. The strategy was applied for each fragrance tested to determine the appropriate loading conditions of the tubes to reach a defined concentration in the test media at a controlled temperature, and when necessary for the tube to act as infinite reservoir for continuous enrichment. Specific handling tools and concentration models were used to improve the throughput of the tubes preparation. We present here this strategy and corresponding results for selected fragrance compounds with varying hydrophobicity.

TH213

Identification of Gestagen(s) and Corticosteroid(s) from Danube River wastewater sample by using LC-HRMS and non-target screening approach

M.A. Hashmi, M. Krauss, Helmholtz centre for environmental research - UFZ / Effect-Directed Analysis; B. Escher, Helmholtz Centre for Environmental Research GmbH - UFZ / Cell Toxicology; W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis

Biologically active substances (generally termed as endocrine disrupting chemicals (EDCs)) are present in untreated municipal wastewater, which may cause deterioration of freshwater ecosystem due to their potential to disrupt the endocrine system of aquatic organisms. Untreated municipal wastewater is directly discharged into Danube River, Novi Sad, Serbia and the objective of this study is to identify compounds responsible for gestagenic and corticosteroid effects in Danube river water by using non-target screening. Water sample from Danube River were extracted on-site using large volume solid phase extraction (LVSPE) and was pre-screened on genetically modified bioassays for agonistic and antagonistic hormonal activity for progesterone and glucocorticoid receptors (PR and GR). The extract was cytotoxic on both nuclear hormone receptors and in order to remove the cytotoxicity, sample was fractionated by using reversed phase-high performance liquid chromatography (RP-HPLC) by using C-18 silica based column. Two minute fractions were collected (total 30 fractions) and applied on respective bioassays and identified one agonistic active fraction for both PR and GR. Second step fractionation was performed on the only active fraction by using aminopropyl column with gradient elution with methanol:water (30:70) with 0.1% formic acid. One to two minute fractions (total 28 fractions) were collected and biological analysis of these sub-fractions revealed again one active fraction with reduced potency as compared to F18 (parent fraction). For unraveling the compounds responsible for gestagenic and corticoid activity, non-target screening is being performed by using LC-HRMS.

TH214

Mixture Risk - Development of an effect-based chemical risk assessment strategy for sites contaminated with complex mixtures of organic and inorganic contaminants

G. Nilen, B. Holmes, M. Larsson, Orebro University / Man-Technology-Environment research centre (MTM); N. Scherbak, Orebro University / School of Science and Technology, Life Science Centre; M. Engwall, Orebro University / Man-Technology-Environment research centre (MTM); S. Keiter, Orebro University / MTM Research centre

Environmental contamination is usually comprised of a mixture of pollutants, each of them bearing the potential of causing different toxic responses towards humans and wildlife. Recent risk assessments still generally rely on chemical analyses only; however, such investigations do not provide information regarding the interactions between chemicals including their integrated toxicity. The limited knowledge of the risks associated with mixture toxicity is the starting point for this study, and is part of the EnForce project that aims to investigate the toxic responses of mixtures of pollutants and integrate those results into risk assessment. Particularly, per- and polyfluorinated alkyl substances (PFAS) are of major concern as they are extremely persistent and able to alter the toxicity of other pollutants. However, preliminary results showed that selected PFASs were not able to alter mechanism-specific toxic effects *in vitro*, while they decreased gene expression of the same mechanism using an *in vivo* model with zebrafish embryos. Moreover, so far no vertebrate based test system exists to quantify the toxic response of PFASs; thus, one objective of the project is to develop a bioanalytical tool for measurement of PFAS contamination. The toxic effects of environmental samples will be assessed by a combination of biotests and chemical analysis. For the identification of non-target pollutants, effect-directed analysis will be used consisting of fractionation, chemical analysis and biotests. In addition, the project aims to fill crucial gaps in the knowledge regarding molecular and mechanism-specific effects of mixture toxicity. The results will be used to perform comprehensive risk assessment of contaminated sites and will be communicated with industry partners and stakeholders.

TH215

Analyzing chemical pollutants in water samples from an urban river and wastewater effluent in Hyderabad (India) and their ecotoxicological effects using effect-directed analysis (EDA)

J. Daniel, RWTH Aachen University; P. Böhm, RWTH Aachen University / Department of Ecosystem Analysis; J. Ahlheim, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; A. Dayakar, Gamana NGO Hyderabad; A. Sathish Lekha, J. Vijayan, I.M. Nambi, Indian Institute of Technology Madras / Department of Civil Engineering; V. Schiller, RWTH Aachen University / Department of Ecosystem Analysis; M. Krauss, Helmholtz centre for environmental research - UFZ / Effect-Directed Analysis; S.E. Crawford, RWTH Aachen University / Institute for Environmental Research, Dept. of Environmental Analysis; S. Schiwy, RWTH Aachen University / Department of Ecosystem Analysis; T. Schulze, W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; H. Hollert, RWTH Aachen University / Institute for Environmental Research

In India, surface water contamination in urban areas is a common issue. One major source of pollution may result from the discharge of treated and untreated wastewater, both domestic and industrial in receiving environments. This contamination composed of a complex mixture containing e.g. polychlorinated biphenyls (PCBs) and polycyclic aromatic hydrocarbons (PAHs) from industries or pharmaceuticals from residential waste may pose a risk not only to the environment but also human health. Previous studies have reported a strong presence of multi-resistant bacteria in the Musi River, which might be due to large pharmaceutical production located in Hyderabad. A cooperation between the Department of Ecosystem Analysis RWTH Aachen (ESA), the Helmholtz Centre for Environmental Research Leipzig (UFZ) and the Civil Engineering Department from the Indian Institute of Technology Madras (IITM) was formed to evaluate the water quality in the Musi River, an urban river in Hyderabad (Telangana state, India) to aid sustainable water management. To assess the ecotoxicological state of the Musi River, water samples (40-100 L per sampling site) were extracted using a novel device for onsite large-volume solid phase extraction (DOI: 10.1016/j.scitotenv.2016.12.140). Two samples were taken along the Musi River, one from a tributary, one from a wastewater treatment plant effluent and another sample from an industrial effluent. Currently, these samples are screened for their toxicity using the water extracts in a set of different bioassays to select samples that are suitable for an effect-directed analysis (EDA) study. Considered endpoints are algae growth inhibition, acute daphnia immobility and mechanism-specific endpoints such as estrogenic activity and genotoxicity. In combination with target and non-target chemical analysis, the overall goal of the EDA study is to identify the main toxic drivers in one sample. Preliminary results show an adverse effect on the water flea *Daphnia magna* and the freshwater algae *Pseudokirchneriella subcapitata*. Estrogenic activity was induced in four out of five samples in the lyticase yeast estrogen screen. Further evaluation of the data and investigation on genotoxicity using the AMES assay is needed to make a well-founded decision on which assay, and sample are most suitable for EDA. Results from this work will provide insight into the composition of chemical pollutants in an Indian urban river and their ecotoxicological effects.

TH216

Ecotoxicological assessment of water samples from an urban river, wastewater treatment plant effluent and industrial effluent in Hyderabad (India) using a set of different bioassays

P. Böhm, J. Daniel, RWTH Aachen University / Department of Ecosystem Analysis; J. Ahlheim, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; A. Dayakar, Gamana NGO Hyderabad; A. Sathish Lekha, J. Vijayan, I.M. Nambi, Indian Institute of Technology Madras / Department of Civil Engineering; V. Schiller, RWTH Aachen University / Department of Ecosystem Analysis; M. Krauss, Helmholtz centre for environmental research - UFZ / Effect-Directed Analysis; S.E. Crawford, RWTH Aachen University / Institute for Environmental Research, Dept. of Environmental Analysis; S. Schiwy, Institute of Environmental Research-RWTH Aachen / Department of Ecosystem Analysis; T. Schulze, W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; H. Hollert, RWTH Aachen University / Institute for Environmental Research

Contamination of surface water is a common issue in urban areas of India. Large proportions of urban river water may consist of untreated wastewater, both domestic and industrial. The city of Hyderabad (Telangana state, India) has large industrial clusters including pharmaceutical, dye and battery factories that have the potential to affect surrounding waterbodies. Recent studies on antibiotic resistances proposed pharmaceutical industries as a potential cause for antibiotic resistances in bacteria in surface waters of Hyderabad. Daily contact of cattle and cattle herding shepherds as well as monsoon flood events are only two examples in which the river pollution is not only an environmental risk but also a human health issue. To work towards a more sustainable water management in urban areas of India, a cooperation between environmental engineers from the Indian Institute of Technology Madras, ecotoxicologists from the Department of Ecosystem Analysis (ESA), RWTH Aachen University and environmental chemists from the Helmholtz Centre for Environmental Research Leipzig (UFZ) has been formed. For a complementary assessment to previous studies, water samples were extracted from an urban river (Musi river), it's tributary, the effluent of a wastewater treatment plant and industrial wastewater, in Hyderabad in June 2017. The samples were taken using a novel device for onsite large-volume solid phase extraction (DOI: 10.1016/j.scitotenv.2016.12.140) with a defined extracted water volume between 40 and 100 L per sample over 4 to 8 hours. The resulting water extracts will be investigated through non-target and target chemical analysis as well as effect-directed analysis (EDA). A bioassay battery to investigate the toxicological effects of the samples included: algae (*Pseudokirchneriella subcapitata*) growth inhibition, daphnia (*Daphnia magna*) immobilization, fish embryo toxicity (*Danio rerio*), endocrine disruption (lyticase Yeast Estrogen Screen; ER-CALUX), genotoxicity (Ames fluctuation; micro nucleus test), neurotoxicity (*D. rerio*) and dioxin-like activity (micro EROD), these tests are currently ongoing. Preliminary results indicate adverse effects on *P. subcapitata*, *D. magna*, as well as endocrine disruption in the lyticase Yeast Estrogen Screen in four out of five samples. The combined results of this work will provide a comprehensive ecotoxicological characterization of an urban Indian river and potentially raise awareness of possibly related risks.

TH217

NAWA SPEZ 2015: Ecotoxicological risks in five small Swiss streams within agricultural catchments

M. Langer, Centre Ecotox EAWAG-EPFL / Aquatic Ecotoxicology; M. Junghans, Centre Ecotox EAWAG-EPFL; S. Spycher, Eawag Swiss federal Institute of Aquatic Science and Technology; M. Koster, Amt für Umwelt, Thurgau / Gewaesserqualitaet; C. Baumgartner, AquaPlus; E. Vermeirssen, Ecotox Centre Eawag-EPFL / Aquatic Ecotoxicology; I. Werner, Ecotox Centre Eawag-EPFL / Department of Anatomy Physiology and Cell Biology

The Swiss National Monitoring of Surface Water Quality (NAWA) is occasionally complemented by focused studies on relevant topics. The latest focus study evaluated pesticides in small streams in catchments with intensive agricultural land use. For this purpose five small streams were sampled from the beginning of March to the end of August using half-day composite samples and an analytical method covering 213 active substances. The chemical analysis was complemented with several biological investigations. Measured concentrations of pesticides were used to determine the risk of pesticide mixtures. Using acute and chronic effect-based water quality criteria (QC), we calculated risk quotients (RQ). In a second step RQs of individual compounds were summed to provide separate mixture RQmix for plants, invertebrates and fish. In all the tested water bodies a chronic mixture risk with RQmix > 1 was determined. There was a chronic mixture risk at three sites for almost the entire sampling period. Consequently this resulted in no recovery time for aquatic organisms. An acute mixture risk was determined for four streams, with two of them showing high risks with RQmix greater than 10. In addition, it was investigated to what extent the predicted mixture risk of herbicides was consistent with endpoints that are determined in the combined algae test. This test provides information on the inhibition of photosystem II (PS II) and algae growth. The resulting risk corresponded very well with the calculated mixture risk for PS II inhibitors. In one stream PS II inhibiting plant protection products dominated the mixture risk for plants. However in two other streams metazachlor and nicosulfuron dominated the mixture risk for plants. Both herbicides do not interfere with photosynthesis. For the detection of insecticide effects, invertebrate communities were sampled at all sites. Also an *in situ* biomonitoring with gammarids was carried out in the Eschelisbach, the site with particular high RQmix for invertebrates. Increased gammarid mortality was observed at the beginning of June 2015. This was consistent with the time course of the acute mixture risk, which during this period was dominated to more than 78% by the insecticide chlorpyrifos-methyl. The SPEARpesticide index also indicated a poor condition for invertebrates in the Eschelisbach. In this study, the biological investigations proved to be a valuable link between the chemical compound risk assessment and stream biology.

TH218

An ecotoxicological assessment of Lake Mondsee, Austria: a two year survey

S. Goncalves, Department of Biology & CESAM - University of Aveiro / Department of Biology and CESAM; C. Venâncio, Department of Biology / Biology; S. Loureiro, Universidade de Aveiro / Biology; R. Vogt, J. Wanzenböck, University of Innsbruck / Research Institute for Limnology Mondsee, 5310 Mondsee; A.M. Soares, University of Aveiro / department of Biology & CESAM; I. Lopes, University of Aveiro / Department of Biology & CESAM, 3810-193 Aveiro

Lake Mondsee is a recreational area in Austria for both bathing and water activities/sports. Nearby exists a wastewater-treatment plant (WWTP) which can represent a point emission for the lake's contamination. Accordingly, an ecotoxicological assessment of Lake Mondsee was carried out. Water (W) and sediment (S) samples were collected from Lake Mondsee and the reference site, Lake Irrsee, on three different seasons: 1) summer 2015 (preliminary assessment of W and S samples' toxicity), 2) spring 2016 (possible best-case scenario, since lake was frozen for the winter) and 3) summer 2016 (worst-case scenario, tourist activities peak). The WWTP inflow and outflow, plus pre-thickening (PS) and thickened sludge (TS) were also collected. The toxicity assessment for summer 2015 was made by performing 48-h population growth (*r*) assays with the rotifer *Brachionus calyciflorus* (W samples) and the 15-min luminescence inhibition assay with the bacterium *Vibrio fischeri* (all samples). Regarding the W samples, results showed no luminescence inhibition for *V. fischeri* and average *r* inhibition rate (%) of *B. calyciflorus* was below 26%. The WWTP inflow samples presented high toxicity to *B. calyciflorus* (EC_{50} > 60%). Samples of S, PS and TS were extremely/high toxic to *V. fischeri.* The samples collected during spring 2016 and summer 2016 were analysed through a battery of assays, with species belonging to different trophic levels. In addition to the two above mentioned tests: the 72-h growth inhibition test with *Raphidocelis subcapitata* and the feeding inhibition test with *Daphnia magna* with the W samples were performed and the 6-day mortality and growth assessment with *Heterocypris incongruens* for S, PS and TS samples. Regarding spring 2016, the average *r* inhibition rate (%) of *B. calyciflorus* was lower than 30% for most of the W samples and average did not surpass 12%. The Microtox® tests showed high toxicity only for all W, S, PS and TS samples. Samples collected in the summer 2016, showed similar results for the growth inhibition for *R. subcapitata* and the Microtox® test. The *r* inhibition rate (%) of *B. calyciflorus* and the 6-day mortality and growth assessment with *H. incongruens* showed some variation. No evidences of the influence of the WWTP present at lake Mondsee were retrieved, since W and S samples from both Lakes Mondsee and Irrsee showed similar toxicity. Further chemical analysis is necessary to clarify the high toxicity observed in the sediments.

TH219

Availability of estrogens applied onto 96-well plates in the LYES

M. Ragulan, Swiss Centre for Applied Ecotoxicology Eawag-EPFL; E.

Vermeirssen, Ecotox Centre Eawag-EPFL / Aquatic Ecotoxicology; D. Olbrich, Swiss Centre for Applied Ecotoxicology EAWAG - EPFL; I. Werner, Ecotox Centre Eawag-EPFL / Department of Anatomy Physiology and Cell Biology; E. Simon, Centre Ecotox / Aquatic Ecotoxicology

Many *in vitro* bioassays are run on 96-well plates and typically, reference compounds, standards and samples are added to the wells of a plate using solvents (e.g. ethanol). These solvents are then evaporated, media with cells are added to start the assay and subsequently compounds redissolve and become available to cells in the bioassay. However, there is scant information on the kinetics of the redissolving behaviour of test substances on 96-well plates. Furthermore, a redissolving step can be circumvented by adding samples and standards directly to the assay, either dissolved in water or medium (or DMSO). In this study we compared the availability of four estrogenic compounds (E2, E1, EE2 and BPA) on 96-well plates in the lyticase-based yeast estrogen screen assay (LYES; this test was recently adopted as an ISO standard). Two-fold dilution series of compounds were added directly to the wells via medium (aqueous; i.e. as suggested in the LYES ISO protocol) or using ethanol (ethanolic) which was evaporated before medium was added. We tested different redissolving times by shaking the plates, using shaking times between 0 to 120 min. After redissolving, medium was transferred to new wells for further testing (redissolved) and emptied wells were given fresh assay medium and yeast cells and were also tested (rest). We evaluated the recovery of test substances in “redissolved” and “rest” wells. Results revealed that, for all test substances: 1) less activity was observed after ethanolic application compared to aqueous application, while their relative potency towards the reference substance remained comparable; 2) only about 50% (for the compounds E2, E1, EE2 and BPA) of the nominal activity appeared in the “redissolved” wells and ca. 50% of activity remained in the “rest” wells; and 3) shaking times beyond 10 min did not further enhance redissolving. The fact that less activity was observed following ethanolic application compared to aqueous application may be because: 1) a fraction of the compounds remain sorbed to the wells and never become available to the cells or 2) compounds are partially evaporated along with the ethanol. To gather more information on these aspects – and to determine the actual concentrations in the two application methods – LC-MS/MS measurements of E2, E1, EE2 and BPA were performed in parallel to the bioassay. These data are currently being evaluated.

TH220

Mutagenic and ontogenetic responses in freshwater guppy Poecilia vivipara chronically exposed to waterborne sodium dodecyl sulfate (SDS)

L.C. Penha, Federal University of Maranhão - UFMA / Departamento de Ciências Biológicas; D.B. Boaes, Federal University of Maranhão - UFMA / Instituto de Ciências do Mar-ICMar; M. Jorge, Universidade Federal Maranhão - UFMA / Oceanografia e Limnologia; R.L. Santos, Federal University of Maranhão - UFMA / Instituto de Ciências do Mar-ICMar

The deposition and persistence of some chemicals in aquatic environments is a constant threat to aquatic organisms. Sodium dodecyl sulfate (SDS) is a surfactant widely used as an emulsifier in household products and is constantly present in the environment in small concentrations. Several studies have indicated acute effects of high SDS concentrations on animals’ behavioral, reproduction and cell division. However, little is known about chronic effects of SDS in aquatic animals. Thus, the present study evaluated the mutagenic (nuclear abnormalities) and ontogenetic (embryo formation) responses in freshwater pregnant female of guppy *Poecilia vivipara* chronically exposed (90 days) to waterborne sodium dodecyl sulfate (0.3 and 0.6 mg/L). During the exposure time, newborn fish was fixed and destined for morphological analyzes. After exposure, females' blood was analyzed for nuclear anomalies (micronucleated cells, nuclear buds, binucleated cells and cells with nucleus presenting apoptotic fragments), and non-parity females were submitted to cesarean section for embryo classification of development stage (less developed until completed newborn fish). The results demonstrated that there were no external deformities in the newborn fish during the exposure. However, there was a decrease in the number of fry on the females exposed to both concentrations of SDS in relation to the control, as well a delay in the development of the embryos of the exposed females, indicating ontogenetic effects even at low concentrations of SDS (0.3 mg/L). Regarding the nuclear anomalies, both SDS concentration caused significant increments in the frequency of all anomalies when compared to the control group (p = 0.01). The major concern about nuclear anomalies is the permanent damage they can cause and the consequently genotoxic and mutagenic damages. These results indicate that freshwater *Poecilia vivipara* chronically exposed to SDS does not appear to be protected by European Directive (73/405/EC) that allowing the concentration of 0.5 mg/L of anionic surfactants (such as SDS) in drinking water and 1 mg/L in the freshwater used for other purposes.

TH221

DETERMINATION OF IZMIR BAY POLLUTION BY USING GENETIC BIOMARKERS IN THE MUSSEL (MYTILUS GALLOPROVINCIALIS) TAKEN FROM THE NATURAL ENVIRONMENT

. Ãakal Arslan, University Ege / Hydrobiology; H. Parlak, Ege University; M. Boyacioglu, Ege Univercity; M.A. Karaaslan, University of Ege; G. G!lsever, Ege University

Izmir Bay, which is surrounded by many agricultural and industrialized cities like Izmir and Manisa, has been polluted nearly 50 years. Aquatic ecosystems were effected very badly due to much kind of pollutants such as heavy metals, polycyclic aromatic hydrocarbons (PAH), Polychlorinated Biphenyls (PCB) and Pesticides. Besides agricultural and industrial activities, heavy marine transport and redging activities in the harbor activities are also disturbed Izmir bay. Authorities have decided to take serious action when the effects of pollution were being unbearable in 1980_s as all the city smell very badly. Micronuclei (MN) tests is a system of mutagenicity testing used for determining the pollution and chemicals causing changes in DNA fragments such as micronuclei in the cytoplasm of interphase cells. Damage caused on the DNA by genotoxic pollutants is the first consequence occurring in the aquatic organisms. In this study, we aimed to investigate the genotoxic effects of Izmir Bay by detecting the MN frequency changes in the gills of mussels (*Mytilus galloprovincialis*). According to our results MN frequency of 10 station varied between ‰ 39,33 - ‰ 5,60 and Binucleated cells frequencies were 0,17 – 5,27. Pollution of Izmir Bay is a long story and cannot be healed in short time but there are some signs that it is in the healing trend. **Key Words:** Izmir Bay, Pollution, micronucleus, *Mytilus galloprovincialis*

TH222

Bioassays stress the ecotoxicological differences between polymers and plastics additives in the marine environment

r. beiras, University of Vigo / Toralla marine sciences station (ecimat); L. Mantilla-Aldana, P.C. López, University of Vigo / Ecology and Animal Biology; T. Tato, Universidade de Vigo / ECIMAT; L. Vidal-Liñán, IEO

Commercial objects made of plastics are composed of two very different components with dissimilar ecotoxicological properties, namely the polymer matrix and the chemical additives used to provide the final physical and chemical properties demanded by the consumers. Most conventional polymers are made of innocuous monomers (olefins, terephthalates), they are inert under environmental conditions and, according to standard ecotoxicological bioassays using early life stages (ELS), do not pose any ecotoxicological risk to marine organisms, with the possible exception of mechanical damage. In contrast, many common plasticizers (e.g. orthophthalates), flame retardants (polybrominated and organophosphorus chemicals), UV filters (benzophenones and other aromatics) and biocides (triclosan) have shown sublethal toxicity for the reproductive and endocrine systems of aquatic organisms. Those potential effects are difficult to test in laboratory since they may result of very long exposure times and plastic-organism interactions not considered in standard toxicity tests. Using ELS of marine organisms, we have adapted standard bioassays with ELS, tested ‘virgin’ microparticles of conventional polymers (PE, PS, PVC) and did not find any relevant short-term toxicity. In contrast, when microparticles obtained from commercial plastic objects are used events of acute toxicity are found, pointing at the additives as the causal agents of the toxicity found. Ongoing experiments explore the kinetics of additive leaching and resulting toxicity in order to assess the relevance of the results under environmental conditions. In addition, some commonly used chemical additives of plastics were also tested and some of them did show acute toxicity at levels not far above those found in polluted coastal waters. The overall experimental evidence obtained so far using standard bioassays with ELS of marine invertebrates point at certain chemical additives as ecotoxicologically unacceptable and stresses the need of finding non-toxic alternatives useful for the industry.

TH223

EFFECTS OF POTASSIUM BROMATE ON THE EMBRYOLOGICAL DEVELOPMENT OF THE SEA URCHIN Arbacia lixula (Linnaeus, 1758)

. Ãakal Arslan, University Ege / Hydrobiology; DŽ. Avşar, Les Établissements Scolaires Tevfik Fikret; G. Kenanoğlu, Turkish Education Foundation İnanç Türkeş private high school; M.A. Karaaslan, University of Ege; S. tez, Ege University

Potassium bromate is a powerful oxidizing agent that chemically ages flour much faster than open air. Potassium bromate bleaches dough, which makes for the formation of tiny, thin-walled bubbles as the bread rises. The product is fluffy, soft and unnaturally white. In this investigation, the embryotoxic, spermyotoxic effects of Potassium bromate analyzed during the development of the sea urchin *Arbacia lixula* from the post-fertilization to pluteus stage (72-h). Moreover, effects of Potassium bromate on fertilization success were observed. Sea urchin sperms and embryos were exposed to Potassium bromate concentrations ranging from 5 to 50 mg /L. The effects on developing embryos were evaluated by scoring normal plutei and developmental defects such as: retarded plutei, skeletal malformations, blocked gastrula/blastula and dead embryo/larvae. A dose–response-related reduction (approximately 35%) was observed in fertilization success. But was not observed significant increases in the number of larvae with skeleton malformations at the pluteus stage of the exposed sperms which is showed that potassium bromate not effected the offspring quality of *A. lixula*. The EC50 (Effective Concentration 50) levels were determined as 104.64 mg/L for fertilization succes. Following exposure to Potassium bromate, a concentration-related increase in the number of larvae with skeletal malformations at pluteus stage observed. The EC50 for 72 hours was determined as 7.893 mg/L for embryotoxicty. **Key Word:** Sea urchins, Embryotoxicity, Fertilization success, Potassium bromate

TH224

Effect of thermal stress on endocrine disruption in Daphnia magna

J. Na, Korea University; H. IM, J. Jung, Korea University / Environmental Science and Ecological Engineering

Endocrine disrupting chemicals (EDCs) include various types of natural (17β-estradiol, estrone) and synthetic (nonylphenol, bisphenol-A) compounds presenting inhibition or mimicking of the reproductive action of endocrine system in animals and humans. Recently, several study reported that daphnid species, which reproduce by parthenogenesis, may generate male offspring in response to EDCs. In addition, it was demonstrated that variation of water temperature is able to change reproduction, growth, and survival of aquatic organisms and population number. This study aims to evaluate the effect of thermal stress on endocrine disrupting effect of EDCs using *Daphnia magna*. Short-term screening (STS) assay was performed to identify the endocrine disruption effects using adult (10-17 days old) daphnids. Animals were exposed to two temperatures of 20 ºC and 25 ºC, and reproduction, growth, male production and survival rates were evaluated. This study can give a insight into the endocrine disrupting effects of EDCs on aquatic organisms under influence of thermal effluents discharged into streams and rivers.

TH225

Microplate Alga Growth-Inhibition Bioassay

I. Iturria, O. Jaka, C. Martí, A. Alzualde, BioBide; A. Muriana, BBD BioPhenix S.L. / RD

The development of new chemical compounds is a long and costly process that may span up to 10 years. However, the success rate of new chemical families has decreased exponentially in the last decades mainly due to compounds toxicity detected in later phases of the R&D process. The OECD publishes a series of guidelines to define the toxicity evaluation assays required for regulatory purposes. The 201 guidelines, in particular, describes the alga growth inhibition test for the evaluation of aquatic toxicity. Nevertheless, the procedure is tedious and time-consuming, so it´s not suited for high throughput screening of toxicity on early development phase. Given so, there is a need for new fast and cost-effective assays with an increased throughput to assess the aquatic toxicity of a compound in early phases of the development In this work, we present a miniaturized version of the OECD 201 alga growth inhibition test. The miniaturized test is carried out in 96 well plates and the biomass measurement is performed on a plate reader. The methodology makes possible to test ten concentrations of a compound and a negative control on the same plate. The biomass measurement by fluorescence read produces a sensitive and reproducible measurement of alga concentration in an efficient manner, with a significant time and human labor reduction. The alga microplate assay was validated with environmentally relevant reference compounds (such as pesticides or flame retardants) and the resulting IC_{50} values-were compared to the OECD 201 results.

Challenges, methodological developments and practical solutions for Social Life Cycle Assessment in industry and policy (P)

TH226

Applying Social-LCA and Social Hot Spot Analysis including a SDG Evaluation to Product Assessments with SEEBALANCE®

P. Saling, BASF SE / Sustainability Strategy; A. Alba Perez, T. Gruenenwald, P. Koelsch, BASF SE / CDS/S

Social criteria and objectives – such as education, health or working conditions – are becoming increasingly important which is why these factors are also addressed by the SDGs (Sustainable Development Goals). For this reason, social aspects also have an increasing impact on marketing and management decision-making processes.\n The SEEBALANCE® methodology, measures the ecological and economic consequences of alternate products or processes. The Eco-Efficiency Analysis is integrated to an overall result together with the Social Analysis (Figure 1).

TH227

Piloting Responsible Research and Innovation in Industry

E. Yaghmaei, I. Van de Poel, Delft University of Technology / Values, Technology & Innovation; A. Porcari, Airi - Italian Association for Industrial Research; E. Mantovani, E. Borsella, Italian Association for Industrial Research

There is now only limited experience with Responsible Research and Innovation (RRI) in industry and there is also limited evidence of the added value of opening up the innovation process in industry for social engagement and gender considerations. In the PRISMA project (http://www.rri-prisma.eu), we overcome these current limitations by carrying out eight RRI pilot projects in a real-world industry context. To establish the added value of the RRI approach and the gender dimension in and for industry, we assess the pilot projects on a number of product and process RRI dimensions and compare the pilots on the relevant RRI dimensions with similar projects in the same companies in which the RRI approach has not been followed. We focus on implementing RRI for some of the major technological challenges in the EU including nanotechnology, synthetic biology, Internet of Things (IoT) and self-driving or automated cars.

TH228

Sustainable Guar Initiative - an integrated approach of social and environmental LCA

P. Martz, LOréal Research & Innovation / LOREAL; P. Arsac, N. Zaaraoui, LOREAL; A. Wathelet, Solvay SA / LCA; J. Viot, F. Laurent, Solvay SA; M. Vuaillat, S. Causse, EVEA

Sustainable Guar Initiative (SGI) is a three-year long integrated program aiming at developing sustainable guar production within the Bikaner district in Rajasthan, India. This desert district is one of the largest producers of guar and guar gum in India. SGI was set up by Solvay, L'Oréal, HiChem and the NGO TechnoServe, and is based on 4 themes: (1) Agronomy: enhancing sustainable practices for rain-fed guar production, (2) Environment: groundwater-neutral approaches and best practices in guar farming, along with tree plantation, (3) Social impact: gender approaches, nutrition, health & hygiene and (4) Market improvement: traceability, supply chain and market access. Guar gum is extracted from guar seed and can be used as such, or functionalized. It is for example used as a bio-based thickening agent in personal care products. To confirm and consolidate the relevance of the program and to identify potential improvement opportunities, an environmental and social Life Cycle Assessment (LCA) has been conducted, comparing the guar production before and after the SGI. The social LCA has been conducted following already available guidances, including UNEP-SETAC Guidelines for Social Life Cycle Assessment of Products and WBCSD Social Life Cycle Metrics for Chemical Products. Methodological developments have furthermore been undertaken in order to fully take into account the smallholders. The Master`s thesis of Diana Indrane on "Integrating Smallholders within the Handbook for Product Social Impact Assessments" has been completed with some developments, related to: (1) Goal and scope: better identification of relevant stakeholders and social aspects, (2) Inventory: improvement of data quality among the social aspects, stakeholders or life cycle steps and (3) Performance assessment: common rating system enabling aggregation related to inventory from multiple sources. Besides environmental LCA, the poster will focus on social LCA. Methodological challenges encountered will be presented and solutions to tackle them will be detailed as long with other limits related to these new developments.

TH229

How the social pillar can be properly integrated into sustainability evaluation methodology? Evidence from bio-based products case study

P. Falcone, E. Imbert, A. Tani, V. Tartiu, P. Morone, Unitelma Sapienza University of Rome

Abstract Along with environmental and economic assessment, social sustainability of the bioeconomy have become a growing challenge, with important effects for the market uptake of bio-based products. In recent years social and socioeconomic aspects have progressively been included in both the discourses and sustainability analyses concerning the bioeconomy. Yet, when it comes to bio-based products the situation still lags behind (Siebert et al. 2017[1]), given that bio-based products involve longer and more complex value chains (Bell et al. 2014[2]) that make the assessment of social and socio-economic impacts extremely challenging. Furthermore, the success of a sustainable bioeconomy depends on stakeholders' acceptance – especially consumers and manufacturers – leading to a growth in demand for such products. The choice of 'what is to be measured' is the critical point in S-LCA, and, by employing recognised participative techniques, the stakeholders' involvement can be used to shape the final sustainability criteria and regulatory recommendations. Against this background, our study aims at investigating to the social dimension of the transition towards bio-based products, by identifying and validating the main social impact categories pertaining to the bio-based products realm. In doing so, we employ a robust three-step methodological framework encompassing: impact categories identification, stakeholders mapping, and social impact categories validation. In order to operationalize the methodological framework, empirical data is gathered by means of in-depth literature review, stakeholders' interviews, and focus groups. By providing empirical evidence on the social dimension, which incorporates different visions of the stakeholders involved in the bio-based value chains, our study paves the way for further developments concerning the integration of social assessments within bioeconomy context. **Keywords: bio-based products, social assessment, stakeholders analysis** <br clear="all" /> [1] Siebert A., Bezamaa O'Keeffea, S., Thränab D., (2017) Social life cycle assessment indices and indicators to monitor the social implications of wood-based products. Journal of Cleaner Production, Available online 9 March 2017 [2] Bell, G., et al., (2014). IEA Bioenergy Task42 Biorefining. Wageningen: IEA Bioenergy.

TH230

Methodological considerations for applying social LCA to modelled future European energy systems in the REFLEX project

N. Brown, KTH royal Institute of Technology / Sustainable Development, Environmental Science and Technology; E. Ekener, KTH royal Institute of Technology; M. Fuss, KIT Karlsruhe Institute of Technology / Institute for Technology Assessment and Systems Analysis ITAS; L. Xu, KIT Karlsruhe Institute of Technology

A methodology has been developed for the social assessment from a life cycle perspective of supply chains for future energy systems for the European Union (modelled techno-economically as part the Horizon 2020 project REFLEX). It has been developed in light of previously published work aiming at life-cycle based social and/or environmental assessment of single energy technologies and energy systems with a future perspective, and in careful collaboration with the handful of REFLEX partners responsible for energy systems models. The functional unit for the assessment of the future scenarios is the provision of energy services in the EU in 2050 (the final year of the scenario and modelling). The system boundary for the energy system proposed shall encompass all energy flows and associated supply chains required for the production and delivery of heat (in all sectors), electricity and mobility services. Foreground LCI data for the capital equipment (primarily plant and vehicles) and fuels are taken from output data from energy systems models. Background LCI data about separate future energy technologies (as required by the identified foreground processes noted earlier) start from existing LCI data for current systems (from EcoInvent) to which changes are made based on certain assumptions about future developments of the technology in question. Thanks to the availability of price data for material in the latest version of EcoInvent, LCI data acquired can be converted into monetary values. Finally, the cost data can be inputted into a multi-region IO-table linked worker hours model such as social hotspots database. This yields the country specific sectors required for the material in the energy system. The social impacts are then be evaluated with social theme tables for each country specific sector. It is proposed that IO-tables used should be adapted in order to reflect the development of the energy system in the future. Since the energy models and scenarios used in REFLEX are preserving in nature, it is suggested as a simplification that social impacts for the future system may be estimated based on the social performance data for the present. However, if doing so it is necessary to be clear about how such results should and should not be interpreted. The methodology will be operationalized in the coming year as part of the REFLEX project.

TH231

Social Life Cycle Assessment of the water system in Mexico City

M. Garcia, Instituto de Ingenieria, UNAM / Ingenieria Ambiental; L. Güereca, Engineering Institute Universidad Nacional Autónoma de México / Environmental Engineering

One of the main elements of the sustainability of water systems in the cities, is to guarantee a decent job that promotes the welfare of workers in accordance with the objectives of sustainable development in Agenda 2030. Mexico City is one of the most populated cities in the world and is considered as the main political, economic and cultural centre of Mexico. However, it has been several water sustainability problems in the social aspect as risks to the health of workers of the water system. The activities of operation that they perform, are also subject to the infrastructure operating problems such as the lack of maintenance, professional development, and aging of the labour force. The objective of this research was to carry out an assessment of damages to human welfare of the workers, through a holistic and systemic approach to assess the impacts of each of the processes of the water system in relation to working conditions. The evaluation of the social impacts of the water system was based on methodological guidelines of S-LCA edited by UNEP/SETAC/LCI and other instruments of social impact assessment. Were considered five stages of the water system: water abstraction and treatment, distribution, storage, waste water collection and wastewater treatment. The evaluation used the method of impact assessment with a nominal scale between 0 and 1, divided into five ranges of social performance: W*ithout information*, *Bad, Medium*, *Good* and *Very Good*. The results were that **Water abstraction and treatment** had a *good performance* as well as the **Distribution**. The **Storage** was the stage with the lowest value of social performance with a level of *Medium*. While, in the stages of the wastewater management, the stage of **Waste water collection** obtained a performance of *Medium* and the stage of **Wastewater Treatment,** with a performance of *Good*. Any stage of the system has reached a *Very good level* in social performance. In conclusion according to the methodology used, which adopts a scale of 0 to 1, where 0 is the worst and 1 the best; it determines a score of 0.6 for all the analysed system, which places the system analysed in *Good* social performance, but is identified as priority needs like decrease of overtime in the drinking water and wastewater treatment plants; improve security conditions in the facilities in order to reduce the risks to health. The welfare of workers requires attention on these points to get closer to the definition of decent work.

Improvements in environmental exposure assessment: Development and application of tools across industry sectors, regulatory agencies, and international boundaries (P)

TH232

Environmental Risk Assessment for some additives used in hydrocarbon extraction activities into the sea

S. Santoro, National Research Council of Italy (CNR); S. Giardina, Ministry for the Environment, Land and Sea; M. Orrù, National Center for Chemical Substances - National Institute of Health; D. Romoli, Italian National Institute for Environmental Protection and Research

Concerning the oil and gas offshore platform activities, the Italian Ministry of the Environment, Land and Sea has adopted a new approach to decide for the release/renewal of the authorisation to discharge the Produced Formation Water (PFW),a by-product of both oil and gas extraction, into the sea. This approach aims at assessing more deeply the possible environmental impact of the additives used in hydrocarbon extraction activities. In this context, we present the application of the environmental risk assessment methodology, set out by REACH Regulation on chemicals, for some additives (e.g. Dietylene glycol) used in oil and gas platform activities. This approach allowed to determine specific concentration limits eligible for seawater discharge of these additives. By applying this methodology we compared the Predicted No-Effect Concentration (PNEC) with the predicted environmental concentration (PEC) related to the release of the substance in the seawater. The work already concluded on Dietylene glycol showed that the concentration below which the risk is considered adequately controlled is 730 mg/l for constant/frequent release and 5900 mg/l for intermittent release. These limits have been included by the Ministry of Environment as a binding condition for granting the authorisation of discharge to the platform manager. **Session: 3.12** Improvements in environmental exposure assessment: development and application of tools industry sectors, regulatory agencies and international boundaries **Authors:** Serena Santoro - National Research Council (CNR) - Institute of Atmospheric Pollution Research /Italian Ministry of the Environment, Land and Sea Silvia Giardina – Italian Ministry of the Environment, Land and Sea – General Directorate for environmental assessments and authorisations Maria Antonietta Orrù – National Institute of Health - National Center for Chemical Substances Debora Romoli - Italian National Institute for Environmental Protection and Research.

TH233

Multidisciplinary approach for discussing the rice crop specific needs in Southern Europe in the view of the Plant Protection Products assessment: conclusions from an ad hoc workshop

A. Carone, Dow AgroSciences Italia srl; A. Whyte, Dow AgroSciences Ltd; A. Alix, Dow Agrosciences / Risk Management; G. Azimonti, ICPS International Centre for Pesticides and Health Risk Prevention; R. Cabella, INAIL - Dipartimento di Medicina, Epidemiologia, Igiene del Lavoro e Ambientale; G. Canha, Lusosem S.A.; C. Civitella, T. Corinti, ICPS International Centre for Pesticides and Health Risk Prevention; M. Corvaro, Dow AgroSciences Ltd; N. Dalla Valle, Dow AgroSciences Italia srl; F. Dias, Cooperativa Agrícola Montemor; E. Garcia, I. Gonzalez, Dow AgroSciences Iberica, S.A.; M. Guarise, Dow AgroSciences Italia srl; P. Havens, Dow AgroSciences LLC; M. Luini, F. Marchetto, ICPS International Centre for Pesticides and Health Risk Prevention; G. Meregalli, Dow AgroSciences Italia s.r.l. / Ecotoxicology; M. Osuna Ruiz, CYCITEX; A. Ritter, Waterborne Environmental, Inc.; M. Rubbiani, ISS Istituto Superiore di Sanità; C. Thomas, Centre Français du Riz; I.S. Travlos, Agricultural university of Athens; S. Ullucci, ICPS; W. Williams, Waterborne Environmental, Inc.; V. Zaffagnini, C. Vaj, Dow AgroSciences Italia s.r.l.

In Europe and in the context of Regulation (EC) 1107/2009 for placing into the market of plant protection products rice as a crop is an anomaly and has created several difficulties in its evaluation. For regulators, there is a need for comprehension of the unique agronomic practices, application techniques, water management and environmental concerns, both from an ecotoxicology and environmental fate perspective considering the majority of rice cultivated within the Europe is grown in paddy fields. This gap in knowledge caused issues in the evaluation of rice as a representative use in the process of European authorization of active substances for plant protection products and raised questions over the suitability of environmental indicator species and risk assessments within the context necessary. Rice is a major crop in many Southern Zone Countries and the difficulties gaining an understanding of rice practices, compounded by uncertainty with changing regulatory requirements and a lack of transparency in evaluation procedures has hampered the process of active substances approval. Such a complex framework could dissuade active substance renewal by agrochemical manufactures or indeed inhibit innovation. In conjunction, an increase in weed resistance to plant protection products currently available has forced Member States to continually rely on Emergency Uses Permit year on year to support the rice growing community. Weed and pest tolerance to agrochemicals is increasing across Europe in all crops, in conjunction with the lack of technical tools available for weed and pest control. Rice farmers recognise the urgent need for active substances to be placed on the market with different modes of action to combat resistance and safeguard the production. The above mentioned topics have been deeply discussed among experts of different disciplines from the rice producing European Countries in an ad hoc workshop facilitated by Dow AgroSciences in July 2017. The outcome of the discussion highlighted the need of a bottom up multidisciplinary approach, with farmers, local networks, users and research institutes facilitating an environment to coordinate a strategy for implementation, with a Member State authority championing this venture through zonal steering groups. The main conclusions of the workshop will be presented and discussed in the poster.

TH234

The Water Column Monitoring Program in Norway: when regulation and science meet

D. Pampanin, International Research Institute of Stavanger; S.J. Brooks, NIVA

Norwegian Institute for Water Research; B. Grøsvik, Institute of Marine Research; E. Lyng, International Research Institute of Stavanger; R.C. Sundt, Statoil
Oil and gas companies operating on the Norwegian Continental Shelf (NCS) are required to carry out environmental monitoring to obtain information on the actual and potential environmental impacts of their activities and to give authorities a better basis for regulation. Scientists, operators and regulators have worked jointly for two decades in this program, for developing, communicating and implementing knowledge, methods and tools to manage the offshore produced water discharges. A multidisciplinary approach (e.g. chemistry, biology, modelling and risk assessment) has been developed to monitor the discharge and reduce risk. Potential impacts from no-targeted chemicals have also been in the aim of this extensive monitoring program for anticipating negative effects at the ecosystem level. In 2015, new guidelines were published as result of research activity performed since 1995. The new requirements have been applied for the first time in the 2017 Water Column Monitoring program. This holistic approach shows a significant improvement in the scientific outcomes of the monitoring, in a cost-efficient way. The surveys included the use of species from various trophic levels and the analysis of both chemical and biological parameters. Three regions were selected: Tampen, Southern North Sea and Egersundbanken (reference area) and in additional the near platform effect (Statfjord A) was assessed. The study design included the use of a predictive discharge model (Dose-related Risk and Effect Assessment Model, DREAM). This model calculates the fate of the discharge in 4 dimensions (including time) to predict environmental concentrations, risk and effects. Biological and chemical data confirmed the accuracy of the study design and provided information on the actual impact of the discharge on the ecosystem. Data showed a general decrease in the actual impact in comparison to previous years. This is a great achievement, that demonstrates the importance of collaboration between researchers, operators and regulators. It is worth to notice that while developing the Water Column Monitoring program, scientists in Norway prioritise a RRI (Responsible Research and Innovation) approach.

TH235

DAPHNE: a supporting tool for pesticides risk assessors and stakeholders

A. Linguadoca, F. Galimberti, S. Ubbiali, ICPS International Centre for Pesticides and Health Risk Prevention / Public Health; L. Menaballi, ICPS International Centre for Pesticides and Health Risk Prevention; S. Ullucci, ICPS International Centre for Pesticides and Health Risk Prevention / Public Health

DAPHNE (DAtes and PHeNological Estimation) is a tool created to contribute supporting the Environmental Risk Assessment (ERA) of pesticides. The rationale behind its development is based on correlating crop phenological stages (BBCH) to specific dates for representative geographic areas is often a crucial step both for the exposure and (higher tier) effects assessment. However, currently there is no source of information clearly addressing this issue at the national, Zonal or EU scale. Data from a number of field efficacy trials were collected in a database that could realistically represent reference scenarios and typical Italian crops. These data included information on BBCH and related date, agronomic and pedoclimatic conditions. The dataset was primarily used to extrapolate BBCH vs date curves for selected crops. These interpolation curves are meant to be used to reduce the degree of uncertainty in both exposure and ecotoxicological higher tier effects evaluation. Among the potential applications, correlating dates and BBCH would help to: - harmonize the application date selection to parametrize the application scheme implemented in the SWASH model, in order to predict pesticide’s loading in surface water due to drift, drainage and run-off; - provide data to substantiate the geographic and temporal representativeness of higher tier ecotoxicological studies. Hence, to support the risk assessment process where a weight of evidence approach is envisaged.

TH236

The applicability of the assessment entity concept in the REACH registration of complex mixtures. A case study for fragrance substances.

K. Jenner, Givaudan / Global Regulatory Affairs & Product Safety; G. Kreutzer, Givaudan SA; S. Kern, Givaudan Schweiz AG; M. Pacella, M. Torres Sanchez, Givaudan Suisse SA

The assessment entity (AE) concept was developed by ECHA together with industry for when more than one fate/hazard profile is relevant for a substance. The tool was introduced in IUCLID 6 and aims to assist users in documenting complex assessment cases in a transparent and systematic way. The assessment entities (AEs) may be imported in Chesar 3 for the purpose of exposure assessment. The relevance and applicability of the AE concept to multi-constituent substances is illustrated by a fragrance ingredient case study. For the purpose of the worker and consumer safety assessment, the traditional whole substance approach was used. However, to assess environmental exposure and risk, a constituent block approach was used because the substance consists of components with different environmental fate properties (e.g. water solubility, log Kow, adsorption coefficient) and ecotoxicity profiles (e.g. acute EC50/LC50 values). The use of whole substance testing versus constituent data is explored. The adaptation of standard tests, such as the determination of water solubility, will be presented showing how the different HPLC partitioning characteristics of the components and the use of two analytical detection methods was exploited to obtain water solubility information for the individual blocks from a test performed on the whole substance. The case study also demonstrates the combined applicability of experimental data, QSAR and read-across in the assessment of the aquatic toxicity of the individual constituents and impurities in order to derive appropriate PNECS for each assessment entity.

TH237

Canada's Approach to Determining Causes of Impairment at Federal Contaminated Sites

M.H. Henning, D. Pelletier, Ramboll EH; M.T. Sorensen, Ramboll / Senior Science Advisor

Canada's Federal Contaminated Sites Action Plan (FCSAP) was developed to reduce risks to human health and the environment from--and to reduce the financial liabilities associated with--federal contaminated sites. Given FCSAP's common use of ecological risk assessment (ERA) as a site management tool, a Focus Group developed guidance for conducting ERAs under FCSAP. One element of that guidance is a technical guidance module on conducting causality assessment. Causality assessment has the overarching goal of differentiating ecological impairment due to chemical stressors from natural variability and from impairment due to other stressors, such as biological and physical stressors. Costly remediation and litigation decisions often hinge on an assumption of causality. It is therefore essential that ERAs objectively examine all plausible causes of observed impairment and attempt to establish cause-and-effect relationships between stressors and responses. The FCSAP causality assessment frameowrk is based on U.S. Environmental Protection Agency's CADDIS guidance, though it is simplified in an effort to facilitate its use at FCSAP sites, which tend to be limited in complexity and size. The adaptable and iterative process described in the guidance module consists of four main steps: listing all candidate causes, integrating causality into study design and sampling; analyzing data for causality; and weighing the evidence on various candidate causes and drawing conclusions. Systematic analysis and documentation of the strength of evidence for and against candidate causes is a fundamental element of causality assessment. Each potential cause of impairment is transparently evaluated with respect to co-occurrence, temporality, consistency of association, biological gradient, complete exposure pathway, plausibility, specificity, and predictive performance. Multiple lines of evidence on each candidate cause are then evaluated for both consistency and coherence of evidence. The guidance and the presentation close with an overview of case examples of the use of causality assessment at contaminated aquatic sites from the peer-reviewed literature.

TH238

Improving "man via the environment" exposure assessment for lead: a case study with lead battery manufacturing and recycling uses

S. Navis, Arche consulting / Laboratory of Aquatic Ecology, Evolution and Conservation; F. Verdonck, ARCHE; K. De Brouwere, VITO NV / Health; L. Geerts, VITO NV; J. Chowdhury, International Lead Association / Senior Scientist -Environment; L. Allen, S. Binks, International Lead Association

Current chemical safety assessments for metals under REACH typically include a generic, worst-case assessment of local and regional risks to human populations resulting from exposure via the environment. As a result of comprehensive biological monitoring campaigns undertaken by European Member States in the past and supplemented modelling data, a large body of data is available on lead body burdens in the general European population (children and adults). However, little information is reported for blood lead in populations surrounding lead manufacturing facilities. Moreover, whereas lead in food and beverages is the primary expected source of exposure (with soil and dust also contributing to children’s exposure due to play habits), it is difficult to apportion the source of this lead exposure to specific uses. Under REACH authorization processes (as part of a socio-economic analysis), it becomes more important to estimate the contribution of a specific use and specific exposure pathways. Consequently, there is a need to better define the contribution of lead exposures resulting from battery manufacturing and recycling operations in the EU. This paper presents the development of conceptual model to assess risk in humans indirectly exposed to lead via the environment using a tiered approach that utilizes the European Union System for the Evaluation of Substances (EUSES) and other advanced tools such as Merlin-Expo for risk assessment. As a first step, local site-specific and regional environmental exposure scenarios and assessments are updated. Next, lead specific empirical bioaccumulation and transfer factors are derived based on a comprehensive literature survey. These parameters are used to describe additional pathways missing in EUSES, such as deposition on crops and soil/dust ingestion. Such pathways have been demonstrated to be dominant sources of lead exposure in humans and thus included in a so called EUSES MvE$^+$. Based upon the results of this EUSES-like screening exercise, higher tier approaches are developed for selected exposure pathways and/or scenarios. Ultimately the results of the environmental exposure modeling have been used in a comparison of predicted blood lead levels with biomonitoring data in the process of risk characterization and documentation as needed for REACH authorization purposes

TH239

Validation of the industrial Simple Treat model for a site-specific setting

J.C. Otte, M. Alter, A. Boehm, H. Elpel, I. Lemche, S. Pawlowski, BASF SE

The multimedia model Simple Treat is applied in the chemical legislation in Europe for decades and recently also in other legislations around the globe. It evaluates the distribution and elimination of chemicals by sewage treatment plants (STPs) in a municipal environment. It has been shown that the specific settings of industrial STPs differ largely compared to the standard settings of the municipal model of Simple Treat in terms of inter alia biological oxygen demand, hydraulic retention time and temperature. Recently, the model has been adapted to predict the chemical fate in industrial STPs (iTreat; Struijs et al. 2016, Chemosphere 159, 619-627) but failed to show site specific validation due to lack of appropriate data. Therefore, the iTreat model was parametrized to the specific parameters of an industrial STP at a specific site. Time series of measured elimination of 22 substances in the waste water stream of this specific STP were gathered and compared to the calculated elimination rate of the parametrized iTreat model. The measured elimination rates were also compared to the non-parametrized model of iTreat and to the municipal models of Simple Treat 4.0 and Simple Treat 3.1. Overall, the iTreat model was successfully adapted to model chemical fate and behavior in an industrial, site specific STP. The elimination rates of the parametrized iTreat model were generally in better agreement to the measured elimination rates than for all other models investigated. The biodegradation rate constant of substances turned out as a sensitive parameter when predicting the elimination rate with either model. In detail, substances with low biodegradation rate constants (e.g. the rate constant of $0.1*h^{-1}$) in the dataset show far more realistic elimination rates in the parametrized iTreat model than for the non-parametrized iTreat or the two municipal models. Compared to the municipal model of Simple Treat 3.1, the parametrized iTreat shows up to two-fold higher elimination rates which reflect the measured elimination. The application of site specific degradation rate constants (derived from biodegradation tests with adapted activated sludge of the respective site) for the site-specific model of iTreat will also be discussed in this poster contribution. Taken together, the validation exercise was successful and the parametrized iTreat model is applicable to other substances being produced at this site where measured data is not available.

TH240

Combination of remote sensing and coarse statistical data for determination of precise spatial distribution of a pesticide load onto soils at a national scale

V. Kodes, Czech Hydrometeorological Institute / Section of water quality; L. Brodsky, Mapradix Ltd.; T. Herza, Hydrosoft Veleslavin Ltd.

Objective of the Study In order to calculate an annual pesticide load over a certain area, one needs detailed data on pesticides' application that are hard to find in a real world. One way is to collect desired data from the farmers, but this is feasible just in relatively small areas. Due to missing detailed data, we computed more precise pesticide load using data aggregated at a certain spatial unit (in case of the Czech Republic districts) and maps of crops derived from a remote sensing imagery. Material and Methods Data on annual pesticide usage for 77 districts in the Czech Republic and remote sensing multispectral data (IRS AWiFS and multitemporal images Envisat MERIS, Landsat 7 – LEC, lately Landsat 8 – LDCM and Sentinel-2) together with a custom database of plant protection products were used. Crop cover (12 classes) grids of 100 m cell size (lately 14 classes, 25 m cell size) were derived from remote sensing images; the crops were linked to plant protection products (PPP) and active substances. Then redistribution of pesticide usage from districts to grid cells was carried out using established link between a crop, PPPs and respective active substances. Results The grid of pesticide usage on perennial crops is produced before the end of spring every year in order to provide data needed for monitoring of pesticides that starts regularly in April. The grid of pesticide usage on all the other crop classes is produced regularly in November. The results are published on WWW and annually updated in order to provide water managers with information necessary for a meaningful design of pesticide monitoring in the Czech Republic. Conclusion The product provides more detailed information on a spatial load of pesticides than other publicly available data on pesticide usage and it is very welcome by interested water managers. Further enhancements are planned in the future as new remote sensing sensors become available.

TH241

A Bayesian approach to estimate biodynamic model parameters: bioaccumulation of PCB 153 by the freshwater crustacean Gammarus fossarum.

A. Ratier, Irstea Lyon; C. Lopes, Université Lyon 1, UMR CNRS 5558 / Laboratory of Biometry and Evolutionnary Biology; H. Budzinski, University of Bordeaux; P. Labadie, UMR CNRS EPOC Universite Bordeaux / UMR 5805 EPOC; L. Peluhet, CNRS / UMR EPOC LPTC; N. Delorme, H. QUEAU, Irstea Lyon / UR MALY Laboratoire Ecotoxicologie; O. Geffard, Irstea / UR MALY Laboratoire Ecotoxicologie; M.P. Babut, Irstea / Water

The first step to evaluate the effects of contamination on organisms is to study toxicokinetics. The bioaccumulation of contaminants by aquatic species is a variable phenomenon, since it depends on the characteristics of the environment, the properties of the contaminants and the species. Different toxicokinetic models have been developed to describe the accumulation of contaminants in aquatic food webs. In these models, the organism is often considered as a single compartment: the bioaccumulation is then described as the balance between uptake and elimination processes. The absorption process can involve both dissolved or trophic route. The diet of aquatic organisms is known to be an important route of bioaccumulation of contaminants. The elimination process includes excretion, metabolism and dilution by growth. To date, there are few models focusing on persistent organic contaminants. Furthermore, estimating models' parameters is generally done through a frequentist approach in two steps: first by estimating parameter(s) related to depuration and then estimating parameter(s) related to accucumulation. The problem by doing this is that depuration during the accumulation phase is neglected, while this process occurs in the two phases. The aim of our study is to propose a Bayesian framework to estimate the parameters of a biodynamic model together by considering simultaneously accumulation and depuration data. The posterior distribution obtained for all parameter will enable a more accurate assessment of bioaccumulation uncertainty. We illustrate our approach with the freshwater benthic invertebrate *Gammarus fossarum* exposed for 7 days to a sediment spiked with PCB153 and transferred to a clean media for 7 more days. The PCB153 concentrations in *Gammarus fossarum* increased from an initial concentration of 0.32 to 12.36 $ng.g^{-1}$ ww (wet weight) at the end of accumulation step. When gammarids were transferred into a clean media, the PCB153 concentration in organisms decreased to 6.41 $ng.g^{-1}$ ww at day 14. The bioaccumulation model assuming first-order kinetics was fitted to the data using Bayesian inference. The inference process quickly converged and thin posterior distributions were obtained for each parameter, meaning that data brough enough information to estimate precisely each parameter. The median model predictions and their 95% credibility intervals showed a good fit of the model to the data.

TH242

Bioaccumulation and biotransformation of Hexabromocyclododecane (HBCD) by the freshwater crustacean Gammarus fossarum: a Bayesian approach to estimate biodynamic model parameters.

A. Ratier, Irstea Lyon; C. Lopes, Université Lyon 1, UMR CNRS 5558 / Laboratory of Biometry and Evolutionnary Biology; H. Budzinski, University of Bordeaux; P. Labadie, UMR CNRS EPOC Universite Bordeaux / UMR 5805 EPOC; L. Peluhet, CNRS / UMR EPOC LPTC; N. Delorme, L. Garnero, Irstea Lyon / UR MALY Laboratoire Ecotoxicologie; O. Geffard, Irstea / UR MALY Laboratoire Ecotoxicologie; M.P. Babut, Irstea / Water

Toxicokinetic models are used to describe how organisms bioaccumulate chemicals or other substances according to uptake and elimination processes. They provide a theorical framework for understanding phenomena, testing hypotheses, and predicting some outputs of interest. In these models, the absorption process can result from dissolved or trophic routes. The elimination process includes excretion, biotransformation and dilution by growth. To date, models exist to describe the accumulation of persistent - i.e. not or poorly metabolized - compounds by various aquatic organisms. However, taking into account biotransformation remains problematic despite its potential importance. It is a key process that can limit the bioaccumulation of parent compounds while generating potentially hazardous metabolites. It varies considerably among species and contaminants. The aim of our study is to propose a Bayesian framework to estimate the parameters of a biodynamic model taking into account biotransformation, by considering simultaneously accumulation and depuration data. We illustrate our approach with the freshwater benthic invertebrate *Gammarus fossarum* exposed to a sediment spiked with Hexaromocyclododecane (HBCD) for 9 days and transferred to a clean media for 9 days. HBCD is a brominated flame retardant which has been detected in various environmental media and has been shown toxic for aquatic life. Previous studies have shown an isomerization of HBCD from sediment (γ-HBCD) to fish (α-HBCD). The bioaccumulation model assuming first-order kinetics was fitted to the data using Bayesian inference.This poster will present the first results about this experiment. We will discuss about the posterior distributions obtained for each parameter and the fit of the model to the data.

TH243

Chemical Exposure Disparities by Demographic Traits in the US Population 1999-2014

V. Nguyen, University of Michigan / Department of Computational Medicine and Bioinformatics; J. Colacino, University of Michigan / Department of Environmental Health Sciences; O. Jolliet, University of Michigan

Identifying individuals or populations at high risk for adverse health outcomes due to chemical exposure requires understanding how chemical exposure patterns vary by inherent traits. Currently, we lack of comprehensive screening to study the thousands of chemicals populations are exposed to on a daily basis. The purpose of this study is to develop a systematic approach that quantifies chemical exposure disparities for a broad set of chemicals by demographic traits in order to identify populations at high risk for exposure. We used the National Health and Nutrition Examination Survey (NHANES) datasets to collect information on chemical biomarker measurements and demographic traits for the years 1999-2014 (n = 74,942), focusing on 229 chemical biomarkers from 16 different classes of chemicals. Poverty income ratio (PIR) was used as a surrogate variable for socioeconomic status, while cotinine levels was used as a proxy for smoking habits. We evaluated the association of each individual biomarker and various demographic factors (age, gender, race/ethnicity, PIR, and smoking status) by using generalized linear model while controlling for relevant confounders and covariates.

Our findings show that race/ethnicity, gender, and socioeconomic status can be statistically significant predictors of chemical exposure. More specifically, parabens, which are chemicals used in personal care products (PCPs), and 2,4- and 2,5-Dichlorphenol, which can be a products of photo-degradation of triclosan, a common antibacterial and antifungal agent, were observed to elevated in African Americans when compared to White Americans. In addition, higher levels of parabens were observed in women, while men had showed higher concentrations of N,N-Diethyl-meta-toluamide (DEET). This could possibly be from women using cosmetic PCPs more frequent and in larger amounts, and men using insect repellant slightly more frequently than women do. Finally, individuals of higher socioeconomic status had higher levels of benzophenone-3 (used in sunscreen products) , parabens, and triclosan, which could possibly be explained by more accessibility to PCPs. In this study, we have identified inherent and demographic traits associated with elevated biomarker concentrations. We hypothesize that this is due to use patterns of consumer product, particularly PCPs. This could support research findings emphasizing the importance of near-field chemical exposures.

TH244

Occupational exposure to flame retardants among Canadian e-waste dismantlers

L.V. Nguyen, University of Toronto - Scarborough / Department of Physical and Environmental Sciences; V.H. Arrandale, Cancer Care Ontario; M.L. Diamond, University of Toronto / Department of Earth Sciences

The amount of e-waste produced globally is growing dramatically. National numbers suggest that the amount of e-waste dismantled across Canada increased seven times in the period of 2002-2012 from 10,250 to 71,300 tonnes/y. One hazard associated with e-waste dismantling is flame retardants (FRs) which are added to electronic and electrical products to meet flammability standards. Little is known about exposure of workers to FRs in e-waste dismantling facilities in high-income countries such as Canada. Here, we have undertaken the first study to report on concentrations and profiles of selected FRs in indoor air and dust at an e-waste dismantling facility in Southern Ontario, Canada, and to estimate occupational exposure of dismantlers at the facility to these FRs. Sampling was conducted daily over a total of five days in February 2017. Thirty-three dust samples were collected using vacuum cleaners and air samples were collected using polydimethylsiloxane passive air samplers (PDMS-PASs) co-deployed with active low-volume air samplers (LV-AAS). A Micro-Orifice Uniform Deposition impactor (MOUDI) was used to obtain particle size distribution of air samples. Post-deployment, samples were extracted and analysed for 12 target FRs, including novel brominated flame retardants (NFRs), polybrominated diphenyl ethers (PBDEs) and organophosphate esters (OPEs), using gas chromatography mass spectrometry (GC-MS). The most abundant FRs in air and dust samplers were the now-banned BDE-209, accounting for ~70 - 98% of all target compounds. The median air concentrations of $\sum_4$ PBDEs ranged from 1930 ng/m^3 to 2900 ng/m^3. Preliminary estimates made using air concentrations measured here suggest that the total daily inhalation intake of all 12 FRs was ~17 µg/day FRs among e-waste dismantlers. Results for the MOUDI samples showed that triphenyl phosphate (TPhP) and other replacement FRs were most abundant in PM10, PM2.5 and ultrafine particles in the air of the dismantling facility. Levels of FRs in air collected from this Canadian e-waste recycling facility suggest opportunities for inhalation exposure to flame retardants among e-waste dismantlers in Southern Ontario, Canada.

TH245

Global approaches to environmetal exposure - assessment of e-wastes

D. Purchase, Middlesex University / Department of Natural Sciences, Faulty of Science and Technology; L. Bisschop, Erasmus University Rotterdam / Department of Criminology; C. Ekberg, Chalmers University of Technology / Division of Energy and Materials, Department of Chemical and Chemical Engineering; P. Fedotov, Russian Academy of Sciences / Vernadskii Institute; H. Garelick, Middlesex University; N. Kandile, Ain Shams University / Department of Chemistry, Faculty of Women; R. Luque, University of Córdoba / Chemistry; O. Popoola, Yaba College of Technology / Department of Chemical Science; H. Ruedel, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Department of Environmental Monitoring; A. Serpe, University of Cagliari / Department of Civil Engineering, Environmental and Architecture; K. Surati, Sardar Patel University / Department of Chemistry; B.P. Wilson, Aalto University / Department of Chemical and Metallurgical Engineering

Obsolete or end-of-life electrical and electronic equipment waste streams continue to grow exponentially, creating a worldwide pollution problem. E-waste comprises a heterogeneous mix of hazardous and non-hazardous metals, metalloids, glass, plastics, flame-retardants and valuable materials (e.g. gold, silver, copper, palladium, platinum and indium). In developed countries, e-waste management is resolved using two major strategies: either by internal recycling/disposal or via exportation to developing nations. For developing countries, the management of e-waste is complicated by illegal waste shipments and further exacerbated by weak environmental regulations coupled to inadequate technology and organizational structures. Rudimentary methods such as dismantling, chipping, melting and burning are often used by the informal sector to recover valuable materials from different e-waste components. These unofficial recycling practices contribute to the release of toxic metals and persistent pollutants that affect both the environment and human health. As a result, e-waste issues are complex, multi-faceted and can only be successfully tackled via a multidisciplinary, trans-boundary approach that involves all stakeholders that include amongst others: manufacturers, scientists, economists, policy makers, waste professionals and consumers. The e-waste project [*"The Environmental and Health Challenges of E-waste and its Management: an Emerging 21st Century Global Concern*" (#2014-031-3-600)], supported by the International Union of Pure and Applied Chemistry (IUPAC), brings together multidisciplinary global expertise to explore different aspects of the e-waste challenge: chemical analysis of contaminants, policy and governance, environmental and health impacts, development and advances in treatment technologies including e-waste valorisation. This presentation makes use of studies from the around the world to highlight the following: i) discrepancies in the provision and enforcement of regulations between developed and emerging countries; ii) complexity in the analysis of e-waste contaminants in environmental and biological samples; and iii) lack of harmonisation of tools or indices to assess risk in environment and health, particularly in soil. It is proposed that a harmonised approach should be taken to use appropriate speciation analysis (e.g. to assess bioavailable or bioaccessible fractions) to evaluate e-waste contaminant risk.

TH246

Droplets deposition pattern from a prototype of a fixed spraying system in a sloping vineyard

S. Otto, Italian National Research Council; G. Innerebner, A. Schmid, C. Roschatt, Laimburg Research Centre; D. Loddo, University of Padova / DAFNAE; M. Venturelli, Edmundo Mach Foundation

In Italy quality vines are sometimes grown in small fields on steep slopes where spray-gun application of pesticides is used, a technique that is very costly and labor intensive. A possible alternative is the use of a fixed spraying system, and first researches are in progress. A fixed spraying system prototype was built in a vineyard at Laimburg Research Centre and a trial was performed with the aim of measuring the deposition pattern of droplets on the row and between rows with water sensitive papers, also in comparison with a precise low-drift air-blast sprayer. Results show that a fixed spraying system has the potential to apply plant protection products without generating drift problems, with a performance similar to a low-drift sprayer, becoming an opportunity for vineyards on very steep slopes.

TH247

Sensitive Arsenic Speciation by Capillary Electrophoresis Using UV Absorbance Detection with On-Line Sample Preconcentration Techniques

H. Lee, J. Kwon, Seoul National University; D. Chung, Seoul National University / Chemistry

The World Health Organization (WHO) guideline states that the total arsenic concentration in drinking water must not exceed 10 ppb. However, arsenic toxicity varies significantly, with inorganic arsenic species being more toxic than organic species. Arsenic speciation is therefore important for the evaluation of health risks from arsenic-contaminated drinking water. Capillary electrophoresis (CE) provides the necessary high performance separation for the determination of arsenic species in water, but its sensitivity with absorbance detection is far below than needed. Using a coated capillary, several on-line sample preconcentration techniques such as large volume sample stacking with an electroosmotic flow pump, field amplified sample injection (FASI), transient isotachophoresis (tITP), electrokinetic supercharging (EKS) combining FASI and tITP, and counter flow (CF)-EKS were therefore investigated. With CF-EKS using phosphate and N-cyclohexyl-2-aminoethanesulfonate respectively as leading and terminating electrolytes, standard samples of arsenite, arsenate, monomethylarsonic acid, and dimethylarsinic acid were preconcentrated from 6,300- to 45,000-fold. The limits of detection obtained with UV absorbance detection were 0.08–0.3 ppb As. For a spring water sample spiked with the four arsenic species, LODs of 2–9 ppb As were obtained, which are lower than the WHO guideline of 10 ppb total As.

TH248

Determination of background levels of free cyanides in surface waters

B. Knopf, Fraunhofer Institute for Molecular Biology and Applied Ecology IME / Environmental Specimen Bank and Elemental Analysis; D. Hansknecht, Fraunhofer IME Institute for Molecular Biology and Applied Ecology / Environmental Specimen Bank and Elemental Analysis; H. Ruedel, Fraunhofer IME - Institute for Molecular Biology and Applied Ecology / Department of Environmental Monitoring; A. Mieth, J. Ohlemacher, CyPlus GmbH; K. Kreuzer, Evonik Performance Materials GmbH

Natural background concentrations of cyanide can originate from the degradation of plants and microbes such as algae. Besides, cyanides may also be emitted from anthropogenic sources. Recently, environmental quality standards (EQSs) for free cyanide were proposed under the European Water Framework Directive (WFD). The EU Joint Research Centre, for example, has proposed an annual average EQS of 0.5 µg/L free cyanide. Since there is a lack of reliable data on background concentrations of free cyanide in surface waters it is not clear whether the proposed EQS values can be practically implemented. To this end a project was initiated to implement and test a method that allows reliable measurements of free cyanide background concentrations in surface waters. Current methods for the measurement of free cyanide in waters only achieve limits of quantifications (LOQs) of about 1 µg/L. Here an existing continuous flow analysis (CFA) method was selected using a system with a special cuvette installation allowing a higher sensitivity. The protocol was validated and accredited according to standard ISO/IEC 17025. With this system an LOQ of 0.15 µg/L can be reached under optimal conditions while an LOQ of about 0.3 µg/L is achieved during routine operation. Previous to field testing it was verified that samples can be stabilized for at least 24 h by adjusting the pH of the sample to 12 and storage in the dark at 4°C. Samples spiked with low concentrations of a cyanide standard were used as positive controls. The field validation results were satisfactory, confirming that the protocol is fit for purpose. Finally, samples from several sites of a small stream with low anthropogenic influences (River Lenne) were taken and analyzed. Free cyanide concentrations of up to 0.4 µg/L were detected. There were significant differences in free cyanide concentrations between the spring, with levels mostly below the limit of detection (LOD, 1/3 of the LOQ), and downstream sampling points where free cyanide concentrations were at least 50% higher, possibly due to degraded plant biomass in the water. This first measurements revealed that background concentrations of free cyanide in the tested surface waters can be below the proposed EQS of 0.5 µg/L. However, the analysis of further parameters (e.g. geographical regions, seasonality) is necessary to create a reliable database on the range of free cyanide background levels as basis for EQS implementation.

TH249

Application of equilibrium and kinetic passive sampling method to quantify integrative chemical profile in a small river and the outflow of WWTP

Y. Jeong, H. Kwon, H. Jeon, KIST Europe / Environmental Safety Group; A. Meyer, E. Fünfrocken, H. Beck, Saarland University; K. Smith, RWTH Aachen University / Institute for Environmental Research; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics

Monitoring water quality is challenging as most of organic contaminants present at trace levels and chemical profile is fluctuating. Current legislative requirement of water monitoring (i.e. EU Water Framework Directive) specifies once-a-month monitoring for priority substances, which is efficient approach in water management. However, the capturing variable contaminant level is also critical for eco-toxicological risk assessment. The present study reports on how to exploit equilibrium and kinetic passive sampler in parallel and evaluate dynamic chemical profile in order to satisfy regulatory requirements and environmental risk assessment. Furthermore, partitioning kinetics of a range of organic contaminants toward each passive sampler type were discussed. The equilibrium sampler tends to be biased toward present chemical profile, which is rather close to instant sample, whereas kinetic sampler provides a time-weighted average concentration (C_{TWA}) over the full sampling period. PDMS sheets with two different thicknesses (76 and 203 µm), as an equilibrium passive sampler, were deployed without the application of performance reference compounds. From the concentration ratio from two PDMS sheets, true *in situ* concentration ($C_{in\ situ}$) of a range of target compounds was determined. In parallel, two types of polar organic chemical integrative sampler (POCIS) were selected as a kinetic passive sampler. The one is typical POCIS with Oasis HLB® sandwiched between polyethersulfone membranes and the other is modified POCIS with polytetrafluoroethylene membrane to reduce membrane sorption artefact, which has been often discussed as one of limitation of POCIS. River Ellbach and the outflow of wastewater treatment plant located south-western Germany were selected as sampling sites and duplicate passive samplers were deployed for two weeks. After sampler recovery, targeted analysis via LC-MS/MS analysis was followed. Based on earlier results, both sampler types performed well and 24 contaminants were detected in total including 8 priority substances in EU WFD. C_{TWA} values can be used as a representative values for the comparison with environmental quality standards and $C_{in\ situ}$ values can be regarded as variable exposure level. Complementary use of kinetic and equilibrium passive sampler enabled us to comprehensively identify multiple aspect of water quality. Studies on coupling passive sampling with passive dosing mode for risk assessment are ongoing.

TH250

Improvement of relationship between water pesticide contamination and land used at a large scale using the Polar Organic Chemical Integrative Sampler

M. Bernard, Irstea; S. Lissalde, University of Limoges; G. Guibaud, University of Limoges / Research Group on Water Soil and Environment GRESE; S. Boutry, Irstea Bordeaux / UR EABX; M. Saut, J. Rebillard, AEAG Toulouse; N. Mazzella, Irstea Bordeaux / UR EABX

The water quality monitoring of organic micropollutants, is generally achieved by collecting grab water samples. Some disadvantages have been highlighted with this technique, such as the limits of quantification and the lack of spatial and temporal representativeness. Currently, grab sampling only provides a snapshot of the contamination, *e.g.* leading to a partial picture during a flood event. To overcome such issues, an alternative sampling strategy can be the use of passive samplers which allow *in situ* pre-concentration of analytes and offer an integrative capacity of several weeks. This study focuses on the Polar Organic Chemical Integrative Sampler (POCIS), widely used for polar pesticide sampling. The interest and relevance of POCIS has already been demonstrated to estimate pesticide loads in small watershed monitoring. Our study aims to show that POCIS provides more comprehensive information during a large scale deployment, and then a better correlation between water quality and urban or various agricultural land uses. A selection of 46 polar pesticides was investigated in 51 monitoring network stations of the Adour Garonne basin (SW of France). These stations were selected in function of the agrochemical pressures and the land uses, assuming different contamination profiles in the different sub-watersheds. Six sampling periods of 14-days were performed over 2016. Firstly, a low loss of POCIS (*i.e.* < 10%) was recorded, proving a good implementation despite the complexity of field conditions, especially for large scale deployment. Secondly, this study demonstrated that the use of POCIS can provide valuable and unprecedented knowledge about pesticide contamination of the Adour Garonne basin. With the large amount of data collected during this 1-year monitoring, correlations between the targeted pesticides and the various land uses over this large watershed (116 000 km²) were established. For example, principal component analysis and Spearman correlations revealed the relationships between pesticide (Metolachlor, Tebuconazol…) contaminations and typical agricultural activities (corn, sunflower and wheat crops) for some sub-watersheds. Similarly, the presence of Norflurazon and Dimetomorph was correlated with vineyards, which is consistent with their use. Additionally, cartographic projections of the contamination levels for the 6 sampling period highlighted seasonal variations which are characteristic of some pesticide uses.

TH251

Development and calibration of o-DGT for pesticides, hormones and pharmaceuticals

B. Bonnaud, Irstea Bordeaux; C. Miège, A. Daval, M. Gregson, Irstea Lyon; N. Mazzella, Irstea Bordeaux / UR EABX

The sampling of micropollutants is a challenge due to their weak concentrations and their temporal variability. These last years, passive samplers have been developed with the advantage to improve the temporal representativeness by measuring “Time Weighted Average (TWA) concentrations”. For the passive sampling of moderately hydrophilic organic contaminants, Polar Organic Chemical Integrative Sampler (POCIS) is the most used and investigated device to date. However, POCIS has some drawbacks since sampling rates are highly affected by water flow velocity, leading to possible bias for TWA concentration estimates. An alternative to POCIS is the Diffusive Gradient in Thin-film technique for organic contaminants (o-DGT). Unlike POCIS, the presence of a diffusive gel may reduce the influence of the water boundary layer, and then hydropdynamic effects on sampling rates. Our objective is to develop the o-DGT for a reliable sampling of a wide range of 60 pesticides, 20 hormones and 38 pharmaceuticals in water selected to cover a wide range of physico-chemical properties (hydrophobicity, ionisability, size, functionnal groups, ...). For that purpose, we first chose the best diffusive gel (e.g. agarose or polyacrylamide) by determining diffusion coefficients for all the compounds, with the comparison of 2 methods: slice stacking and diffusion cell. The slice stacking consists in contaminant diffusion from 1 spiked gel to 5 clean gels let in contact for 30 minutes. The concentration in each gel disk was determined over the time, allowing the calculation of diffusion coefficients according to Fick’s second law. Diffusion coefficients obtained with this method are congruent with those found in literature. Those obtained with the diffusion cell method are similar than those obtained by slice stacking except for ionic compounds, which also exhibited lower affinity with gels than water, in comparison to neutral compounds. The second step consisted in membrane selection, necessary to protect diffusive gel, and that needs to exhibit the lowest possible compounds retention. Three types of membrane (cellulose, polyethersulfone and nylon) were tested with different pore sizes (0.45 and 5 µm). Finally, to estimate the sampling rates, and to validate diffusion coefficients, a calibration experiment with micropollutants at environmental concentrations was conducted.

TH252

Evaluation of Translocation of [14C]Radiolabeled Plant Protection Product in Tomato Fully Grown in a Greenhouse

S. Freedlander, Smithers Viscient, LLC / Environmental Fate and Metabolism; S. RAO, Gowan Company / Regulatory; K. Malekani, S. Kang, Smithers Viscient / Environmental Fate and Metabolism

Once applied to a plant, pesticide residues have the potential to move to other plant tissues via phloem and xylem. This translocation can affect pollinating insects and consumers of the plant tissues. The objective of this study was to evaluate the translocation of a pesticide through phloem and xylem to various tomato tissues (flower, leave, stem, and root) when applied to leaves and soil. A suspension concentration (SC) formulation was prepared with ^{14}C radiolabeled active ingredient. The study was conducted with three groups of tomato plants. Group I was an untreated control group. Group II and Group III tomato plants were treated with formulation containing [^{14}C]active ingredient by foliar application and soil drench, respectively, in a single application at a rate of 0.50 kg a.i./ha. Leaves, stems, and flowers were harvested at 1, 2, 4, 6, and 8 weeks after application (WAP) and root tissue was harvested at 8 WAP. At each sampling interval, the collected tissues were analyzed for total radioactive residue (TRR) by combustion analysis and autoradiography by phosphor-imager analysis. The TRR in all tissue types from the soil drench group was higher than in corresponding tissues from the foliar application group. The autoradiographs of all tissue types from the soil drench group were comparatively darker than in corresponding tissues from the foliar application group. Although both basipetal movement (downward from leaf application site) via phloem and acropetal movement (upward from both leaf and root application sites) via xylem were observed, results indicate the movement of radioactive residues is much faster through xylem. Select tissue samples were extracted and analyzed by HPLC-RAM, which shows that the majority of translocated radioactive residues by phloem was metabolites of the active ingredient. In conclusion, evaluation of translocation during a conventional plant metabolism study can provide valuable information to better assess the potential effects of plant protection products on pollinating insects.

TH253

An Examination of Microbial Biomass in Sediments and the Impact of Seasonal Variation

K. Malekani, Smithers Viscient / Environmental Fate and Metabolism; S.P. McLaughlin, Smithers Viscient / Department of Environmental Fate; K. Campbell, Smithers Viscient / Environmental Fate Metabolism

Microbial biomass is an important measure of the health and viability of a sediment just as it is for soils. It is also a parameter used to assess how viable the microbial population is for a sediment as used in guidelines like the OECD 308 Guideline ‘Aerobic and Anaerobic Transformation in Aquatic Sediment Systems’ and the EPA Guideline OCSPP 835.4300 ‘Aerobic Aquatic Metabolism’. Although there is no strict recommendation for the level of microbial biomass that should be contained in sediments used for testing, like there is for soils (i.e., 1% of the soil organic carbon content, OC), it is still a useful parameter to assess viability of the sediment and to compare different sediments used for testing Microbial biomass in sediments is typically determined by fumigation/extraction procedure prior to test initiation (post-handling/pretreatment), near test initiation and near test termination. The current presentation will focus on the initial microbial biomass as an indication of health and viability of sediments at the time of their collection. A collection of initial sediment microbial biomass values has been summarized and presented based on their time of collection during the year. An example of one loamy sediment (Taunton) and one sandy sediment (Weweantic) collected during all four seasons, over a two-year period produced microbial biomass values (expressed as % OC) shown below. **2016 Taunton Weweantic 2017 Taunton Weweantic** Winter 0.47 0.11 Winter 0.81 0.05 Spring 0.32 1.1 Spring 0.76 0.82 Summer 0.63 0.28 Summer 0.51 0.41 Fall 0.40 0.22 Fall 0.60 [a] 0.71 [a] late summer Additional biomass results will be presented, discussed and correlated to other sediment parameters, including texture, pH, and % OC. Conclusions from several sediments used in recent years will be extrapolated from trends in the data set concerning seasonality, environmental conditions and sediment characteristics.

TH254

Use of scanning electron microscope (SEM) in evaluation of hypopharyngeal glands development in Honey bees (Apis mellifera L.)

A. Drzewiecka, M. Napora-Rutkowska, Institute of Industrial Organic Chemistry Branch Pszczyna / Department of Toxicological Studies; E. Kulec-Ploszczyca, P. Parma, Institute of Industrial Organic Chemistry Branch Pszczyna / Department of Ecotoxicological Studies; N. Lemańska, Institute of Industrial Organic Chemistry, Branch Pszczyna; M. Frontczak-Baniewicz, R. Strzalkowski, Mossakowska Medical Research Centre, Polish Academy of Sciences / Electron Microscopy Platform; A. Daniel-Wójcik, Institute of Industrial Organic Chemistry Branch Pszczyna / Deparment of Toxicological Studies

The hypopharyngeal glands (HPG) of Honeybees consist of many acini connected with collecting duct, arranged in the form of long paired cords lying on the both sides of the head. They played important role in maintaining healthy colonies i.e. through production of "milk" containing proteinic substances to feed larvae and queen. The aim of this study was to check the possibility of using scanning electron microscope (SEM) to evaluate the development of hypopharyngeal glands of bees, considering reliability, work-, time-consuming and cost-effectiveness of the method, including collecting of material. The study was conducted on Honey bees (*Apis mellifera* L.) subjected to chronic toxicity studies performed according to the EFSA guidelines (EFSA Journal 2013;11(7):3295). Animals were treated with four different chemicals in 4 to 5 concentrations. The left HPG were obtained from 5 bees per test item (in the highest concentration, which did not cause mortality below 50%) and the negative control. The specimens were fixed in 2,5 % paraformaldehyde in phosphate buffer, then postfixed in 1% OsO_4 and dehydrated with grades series of ethyl alcohol followed by acetone. Next specimens were critical-dried (CPDS, Critical Point Drying System), then coated with gold particles before observation in JEOL JSM-6390LV. The images and linear measurements (small and big axis of symmetry) from ten acini as well as number of acini per 1 mm^2 were taken from each samples. The analysis of the results showed decreases and increases of acini and their number per 1 mm^2, depending on the test item, however, these differences were not always statistically significant. It turns-out, that images were very valuable, allowing visual comparison of acini. Data obtained from the studies indicate that SEM can be useful tool for evaluation of hypopharyngeal glands development of Honey bees.

TH255

Comparison of International Quality Assurance and Quality Control Standards for High Resolution Mass Spectrometry Dioxin Analysis

D. Thal, E. Ogburn, Environmental Standards Inc; R. Vitale, Environmental Standards; D. Blye, Environmental Standards, Inc.

The analysis of polychlorinated dibenzo-p-dioxins and polychlorinated dibenzofurans (PCDD/Fs) in environmental media, foods and tissues by high resolution gas chromatography-high resolution mass spectrometry (HRMS) is frequently used as the reference methodology against which other candidate analytical approaches are measured. Official methods based in this technology, employing isotopically labeled standards for recovery correction have been established in the EU, the USA, Japan and other nations for decades and international standards for such methods have been established by ISO (Standards 13914 and 18073, for example). To identify achievable best practices and to understand differences in precision, accuracy and qualitative certainty for data produced from wide-ranging sources, an examination of the requirements of these methods was conducted. A review identifying critical differences and areas of agreement with regard to qualitative criteria, precision and accuracy will be presented, with perspectives on the impact differences may have on data uses by researchers conducting analyses under different protocols.

TH256

New Mass Spectrometry Techniques for the Measurement of Persistent Organic Pollutants.

P.D. Jones, University of Saskatchewan / School of Environment and Sustainability; J. Giesy, University of Saskatchewan / Department of Veterinary and Biomedical Sciences and Toxicology Centre

Recent development of new mass spectrometry technology and instrumentation has increased the amount and quality of analytical information that can be obtained from samples. In particular, dramatic increases in mass resolution have made possible unequivocal identification of contaminants even in complex mixtures and matrices. In the area of POPs analyses of PCDD/Fs and PCBs are of concern due to small concentrations that need to be quantified and the presence of a wide range of other chlorinated chemicals that might potentially interfere. The recent release of a GC/OrbiTrap system brings levels of mass resolution not previously available for analysis of POPs by GC chromatography coupled with ultra-high resolution mass spectrometry (GC-UHRMS). Here we report use of GC-UHRMS for identification and quantification of PCDD/Fs and PCBs. The methods developed are based on standard US-EPA methods (Methods 1613 and 1668) but are enhanced by use of new capabilities provided by image current detection and high mass resolution (> 100,000 FWHM). Robustness of the PCDD/F analyses were demonstrated by excellent calibration characteristics and ability to detect all 2,3,7,8-substituted congeners even in an extract of used motor oil. Reanalysis of fish tissues previously analyzed by a magnetic sector instrument demonstrate accuracy of identifications and quantifications and provide validation of the method. Analyses were also conducted to determine the potential for a ‘mutiplex‘ analysis of various POPs where the resolving power (>120,000 FWHM) of the MS system was able to eliminate potential interferences from a variety of ‘non-target’ organochlorines. These multiplexed analyses significantly reduce the time and cost of sample extract preparation and clean-up. In addition, the relatively simple auto-tuning and mass calibration algorithms available for the advanced mass spectrometry systems greatly simplify these otherwise challenging analyses.

TH257

Influence of water temperature and salinity on impact of Hazardous and Noxious Substances (HNS) in the marine environment

M. vannoni, D. Doran, C. Garcia, J. Uzyczac, M. Kirby, D. Sheahan, Cefas

It is recognised that Hazardous and Noxious Substances (HNS) transported at sea present a broad range of potential marine spill scenarios due to wide range of fate and effects of the many types transported in bulk through national and international waters. To improve preparedness of response and to provide better advice during marine incidents there is a need to improve our knowledge of the marine hazards/risks associated with the highest priority chemicals. Harbours and ports are typically located in transitional and coastal waters and so ship traffic is more concentrated around these areas and so potential collisions are more likely. These areas are characterised by a range of salinity from 10 - 20 ppt within estuaries up to 34 ppt in coastal and offshore waters. Similarly, seawater temperatures vary significantly with seasons (from 6?C to 20°C in the UK). Based on these levels of salinity and temperature, preliminary studies were performed to investigate the toxicity of HNS associated with different salinities (from 20 to 30 ppt) and for temperatures (10 - 25?C). Different chemicals were chosen for the tests. Aniline and zinc sulphate were chosen as they are high priority HNS Chemicals due to their relatively frequent transport in bulk quantities. Additionally, benzalkonium chloride (a surfactant and biocide) and the biocide and disinfectant sodium hypochlorite were also investigated as they are transported in moderate quantities, have different chemical properties and modes of toxicity. Toxicity tests were performed with a micro crustacean, *Tisbe battagliai*, and two seaweeds, *Ceramium tenuicorne* and *Fucus vesiculosus*. Our results show that in most cases, chemical toxicity is positively correlated with temperature (higher toxicity with increasing temperature) and negatively with salinity (lower toxicity with increasing salinity). This means that chemical spills are likely to have more impact in the summer in temperate regions and in lower salinity coastal or estuarine areas. These are also the areas that due to the presence of cities and port and harbour facilities have higher marine traffic and potentially greater risk of marine incidents.

TH258

Using Correlations of Biological Toxicity Equivalent Quotients and Toxicity Equivalent Quotients to Derive Threshold Values for Dioxin-Like Compounds in Sediment

J.D. Ouellet, RWTH Aachen University / Department of Ecosystem Analysis; M. Brinkmann, University of Saskatchewan / School of Environment and Sustainability and Toxicology Centre; C. Gembé, Institute for Environmental Research RWTH Aachen University / Department of Ecosystem Analysis; S. Buchinger, Federal Institute of Hydrology / Department G Biochemistry Ecotoxicology; G. Reifferscheid, German Federal Institute of Hydrology; H. Hollert, RWTH Aachen University / Institute for Environmental Research

Sediments can act as a sink and source of pollution in aquatic environments, particularly with respect to persistent organic pollutants (POPs) which bind to sediments and then can be released into the aquatic environment if, and when, the sediments are disturbed (e.g., dredging, floods, storm events). Among POPs, dioxin-like compounds (DLCs), which consist of a variety of contaminants that share similar structures and can bind to the arylhydrocarbon receptor (AhR) in cells, are of particular concern. In addition to chemical analyses, which are often expensive and unnecessary if the contamination is low or below threshold concentrations, measurement of the induction of ethoxyresorufin-*O*-deethylase (EROD) activity using the rat hepatoma cell line (H4IIE) has been identified as a potential bioanalytical screening tool for the presence of DLCs in the environment. In the presented project, the bioanalytical component involved the use of a 96-well plate-reader–based assay to measure EROD induction with the rat hepatoma cell line H4IIE. The micro-EROD assay can be used to determine the cytochrome p450 subfamily 1a (CYP1A)-inducing potential of a variety of substances, including extracts of sediment samples. For this project, micro-EROD assays and chemical analyses were performed on extracts of 22 sediment samples collected from waterbodies in Germany. We investigated the correlation of biological toxicity equivalent quotients (BEQs) determined from H4IIE micro-EROD to toxicity equivalent quotients (TEQ) determined from chemical analysis of the sediment extracts for PCDD/Fs and DL-PCBs. Correlation analysis indicated strong significant relationships between BEQs and TEQs for PCDD/F (r^2=0.940, $p<0.001$) and DL-PCBs (r^2=0.924, p=0.003). From these correlations, threshold values can be established and the assay used as a pre-screening tool to identify samples that would require additional chemical analyses.

TH259

Measuring bioconcentration of cationic surfactants in fish

A. Kierkegaard, C.L. Chen, Stockholm University / Department of Environmental Science and Analytical Chemistry ACES; M. McLachlan, Stockholm University / Environmental Science and Analytical Chemistry (ACES); J.M. Armitage, University of Toronto - Scarborough / Physical and Environmental Sciences; J.A. Arnot, ARC Arnot Research & Consulting / Adjunct Professor, Department of Physical & Environmental Science/Department of Pharmacology and Toxicology; S. Droge, University of Amsterdam/IBED Institute / IBED

Measuring the bioaccumulation of cationic surfactants in fish is challenging. Their disposition in water depends on both pH and alkalinity. Many cationic surfactants have a tendency to sorb extensively to surfaces, making it difficult to generate and maintain constant concentrations in aquarium water. They can also sorb extensively to the surface of fish, making it difficult to separate internal exposure from external exposure. They display a partitioning behaviour that is similar to biomolecules, making it challenging to separate them from major matrix components in fish tissue samples. They can also be toxic to fish, which constrains the concentrations that fish can be exposed to. We are currently working to define the cationic surfactant property space that is amenable to measurement of bioconcentration factors in fish. We will exploit this property space to measure the bioaccumulation behaviour of a range of cationic surfactants. These data will be used to evaluate the BIONIC model, a mechanistically based model employing in vitro assay derived key input parameters (membrane-water partition coefficients and intrinsic hepatic clearance). The BIONIC model can in turn be used to estimate bioaccumulation of cationic surfactants in the property space that is not amenable to measurement. Our first experiments are being conducted with a series of methyl alkyl amines, including primary, secondary, tertiary and quaternary amines ranging in chain length from C9 to C16. The test chemical mixture is infused continuously into the water inflow of a flow-through aquarium using a syringe pump. To determine the concentrations of the test chemicals in aquarium water, 400 uL of aquarium water is transferred with an auto-pipette to a vial containing 600 uL of methanol, and this mixture is analysed with LC-MS/MS. This method allows measurement in the high ng/L concentration range with a precision of 2-8%. Concentrations in the aquarium were maintained at a constant level for more than a week, whereby the ratio measured:nominal decreases with chain length. To determine the concentrations in fish tissue, methanol extracts are cleaned up on a weak cationic exchange SPE column followed by large-volume injection. This method allows quantification in the low ng/g range. The results of the first bioconcentration experiments will be presented.

TH260

Acetylcholinesterase inhibition: a comparison of available methods for determination of acetylcholinesterase in muscle tissue of Limanda limanda.

J. Uzyczak, Centre for Environment, Fisheries and Aquaculture Science (Cefas) / Environmental and Animal Health

Acetylcholinesterase inhibition (AChE) has been used as a biomarker of the effects of organophosphate and carbamate compounds. AChE is present in most animals and is responsible for the rapid hydrolytic degradation of the neurotransmitter acetylcholine (ACh) into the inactive products choline and acetic acid. The role of AChE is to regulate the nervous transmission by reducing the concentration of ACh. When AChE is inactivated by an organophosphorus or carbamate ester, the enzyme is no longer able to hydrolyse ACh and the concentration of ACh remains high. Continuous stimulation of the muscle or nerve then occurs, resulting in tetany and eventually paralysis and death. The ICES/IOC International workshop on Biological Effects of Contaminants, that took place in Bremerhaven, Germany during March 1990, provided on opportunity to test AChE inhibition as an index of marine contamination. An official ICES Technique in Marine Environmental Sciences (TIMES No.22 Biological effects of contaminants: Cholinesterase inhibition by organophosphate and carbamate compounds) is available and recommended for contaminant monitoring programmes in the marine environment. This method was published to improve and standardise the comparability between results from different laboratories and/or countries. However, the method has not been updated since 1998 and does not provide enough details on different marine species, preparation and handling of samples or storage conditions. The search continues for new monitoring tools, improvement and harmonisation of existing methods, which may be used as specific markers for contaminant effects on the marine environment. Four different methods were assessed using the muscle tissue of *Limanda limanda* collected from sampling areas in the North Sea as a part of the Clean Seas Environmental Monitoring Programme (CSEMP)in the UK. The methods showed different results but verified fundamental requirements in all the procedures such as storage conditions, age of the samples or temperature dependence. More work needs to be done to standardise different approaches and come up with a more detailed and accurate guideline. This should help to obtain precise, consistent and comparable results across the national and international laboratories and therefore provide a real evaluation of the status of the marine environment.

TH261

Environmental emission to surface water for analogous exposure path. A reflection on the matter for biocides, human and veterinary medicines.

A. Haro-Castuera, R. Carapeto García, Spanish Medicines Agency / Veterinary medicines; G. Cortés Ruíz, C. Rubio Montejano, Spanish Medicines Agency / Department of Veterinary Medicines

Once emission has happened for one active substance the chain of events affecting the Environment follows its path. But how we study them depends upon the approach, dictated by legislative frames, subsequent guidance and, eventually, inertia and tradition. One remarkable example is the case of insecticides. While sharing the same active substance, different products authorized under different regulation can be applied differently. Then, to be marketed, scientific evidence of safety is mandatory according to different legal frameworks. The analysis of risk for the Environment share some principles making emphasis in different areas. Here, we will review emission paths and key risk elements as a thought starter pursuing harmonizing approaches and resource sharing between assessment schemes.

Hazard and exposure assessment of chemical mixtures: steps towards increasing the realism of chemical risk assessment (P)

TH263

Using microarthropod community assays in metal mixture testing

J. Renaud, CFE - Centre for Functional Ecology; T. Natal da Luz, University of Coimbra / Department of Life Sciences, University of Coimbra; S. Siciliano, University of Saskatchewan / Department of Soil Science; J. Sousa, University of Coimbra / Department of Life Sciences

Due of anthropogenic activities metal contaminated sites are a constant environmental concern and because of the persistence of these metal elements, will continue to be a pressing issue for many years to come. In terms of legislation and environmental risk assessment, much effort has been undertaken to understand the effects of metals and, more recently, the effects of metal mixtures. However, most legislation still focuses on single metal elements, disregarding mixtures. Similarly, research has been predominantly focused on single metals while mixture studies have mostly focused on binary and tertiary mixtures to determine metal interactions and deviations from concentration addition, which is the most commonly accepted model and that considered in legislation. These studies provide valuable information on the metal mixtures but are performed with few standard test species and use mixture ratios optimized for the goal of modelling mixture interactions, which many times lack environmental relevance. In this presentation we take a different approach and test three complex five element metal mixture ratios (Pb, Cu, Ni, Zn, Co) using a natural soil microarthropod community. These metal mixture ratios were selected base on environmental and legislative relevancy, two ratios

produced from guideline values (Canadian soil quality guideline and EU REACH PNEC values) and a ratio based on the average concentrations in a contaminated site (Sudbury) for each metal. Each mixture was tested with 11 doses in toxic units estimated from *Folsomia candida* reproduction EC50 for each metal in the mixture. The community results from this experiment were transformed to similarity matrixes using the Bray-Curtis coefficient and used to calculate dose response curves. This approach assumes that community changes are promoted by increasing metal contamination. These community dose response curves allowed the estimation of microarthropod community EC values, which provide valuable insight on the adequacy of current guideline values and in developing site-specific risk assessments and remedial goals with community endpoints. Produced EC values from this simplified community experiment are currently under validation in a terrestrial model ecosystem experiment, for these same mixtures.

TH264

Alteration of stress-related and thyroid hormone related genes in zebrafish larvae after the administrations of lead acetate, and mixtures of lead acetate and BDE-209

K. Chan, The Chinese University of Hong Kong / Life Sciences; C. Leung, The Chinese University of Hong Kong / School of Life Sciences; Z. Zhou, J. Yang, The Chinese University of Hong Kong / School of Life Science

The expression profile of oxidative stress-related genes (*sod1, sod2, sod3a, ccs, cat, gr, gst*), and thyroid-related genes (*ttr, trβ, dio1, dio2, nis)* in zebrafish larvae was examined upon lead acetate treatments to have a complete analysis of lead toxicity on larvae. Up-regulation of s*od1, sod3a, gr, gst*, and down-regulation of *ttr* and *nis* were evidenced in the experiment.The expression profile of *ttr, trβ, dio1, dio2, nis, sult1-st1, sult1-st2, sult1-st3, ugt1ab, ugt2a1*, and the above oxidative stress-related genes, was also studied to unveil the impacts of co-exposure of lead acetate and BDE-209 on thyroid hormone metabolism and oxidative stress balance. Transcripts of *gr* and *gst* were induced upon both individual exposures and co-exposure, suggesting that both lead and BDE-209 are capable of disrupting cellular oxidative stress balance. No synergistic effects of the two chemicals at short time (48 hr) exposure to induce oxidative stress, while the possibility of these two chemicals acting synergistically to alter the gene transcription at later time points should not be ruled out.

TH265

Assessment of the toxic interaction of lanthanides on aquatic organisms

A. Romero, Universite de Lorraine / LIEC, CNRS, UMR 7360, Université de Lorraine, Campus Bridoux, Bâtiment IBISE, 8 rue du General Delestraint, 57070, Metz, France; E. Joonas, M. Muna, National Institute of Chemical Physics and Biophysics; D.A. Vignati, CNRS / LIEC UMR7360; L. Giamberini, Université de Lorraine, CNRS UMR 7360 / LIEC, CNRS

The relevance of lanthanides (LNs), in agricultural, industrial and, especially in high-tech applications has increased in the last decades. As consequence, more LNs are expected to enter into the environment and accumulate in the ecosystem. Although no great accumulations have so far been recorded, alterations in the LN natural cycles have already been observed. Most of the available data on LN toxicity focuses on the effect of single elements, however they are commonly found as a group in nature. LNs are expected to have cumulative toxic effects on organisms, owing to their similar chemical properties, but studies as mixtures, more representative of real scenarios, are required to support this hypothesis. In this research, we evaluated the toxic interactions of binary and ternary mixtures of cerium (Ce), gadolinium (Gd), and lutetium (Lu), representative of heavy, middle and light LNs, respectively, on seven aquatic species belonging to different trophic levels. From the seven organism studied (*A. fischeri, R. subcapitata, C. vulgaris, B. calyciflorus, H. incongruens, D. magna* and *D. rerio*) potential toxic effects were observed only in five; and the inhibitory LN effects were consistently concentration-dependent only for *A. fischeri, R. subcapitata* and *B. calyciflorus*. Bioavailable LN concentrations significantly decreased during all tests and the major decline took place at the beginning of the tests, but the extent of the decrease varied across test media. Thermodynamic speciation calculation highlighted important differences: in distilled water and 1% NaCl, LNs were predicted to occur mainly as free ions; in more complexes media, LNs appeared as free forms and with complexes with groups $LNSO_4^+$, but these species lowered with the decrease in the ionic radii, and, in detriment of these complexes, species with $LNCO_3^+$ and $LN(OH)^{+2}$ groups increased. The two multi-toxicity approaches used in this study (concentration addition and toxic unit calculation) showed more than additive effect for the mixtures to the bacterium *A. fischeri* and the algae *R. subcapitata*; whereas less than additive toxicity was instead observed for the rotifer *B. calyciflorus*. Obtained results highlight that it should not be assumed LN toxicity as additive as so far, and predicting the response of aquatic organisms exposed to mixtures should be further research to better understand their toxic interactions in real scenarios.

TH266

Predicting the chemical and biological effects of tertiary metal mixture (Ni, Cu, Cd) to aquatic plant, Lemna gibba under different dissolved organic carbon concentrations

S. Martinez, CONICET PRIET UNLU; Y. Gopalapillai, Environment and Climate Change Canada; M. Saenz, PRIET CONICET, National University of Luján; B. Hale, School of Environmental Sciences, University of Guelph; W.D. Di Marzio, CONICET-PRIET / PRIET

Toxic effects of single metals on aquatic environments are well established. In nature, organisms are exposed to a mixture of them at different bioavailability conditions. However, this situation is not always well studied. Here *Lemna gibba* were exposed to Ni, Cd and Zn individually and as ternary mixtures. The influence in the uptake and toxicity of dissolved organic carbon (DOC) as an environmental ligand was studied. Two sets of tests were performed:0.5 mg/L and 10 mg/L of DOC. The metal concentrations at the mixture tests were chosen by an incomplete factorial design, resulting in controls plus 20 test cases. Frond number inhibition (%FNI) and root growth inhibition (%RGI) were calculated at the end of 7-days tests. Determinations of internal dose [M_{tiss}] and external dose [M_{diss}] were also conducted for all chronic tests. Single metal toxicity thresholds (IC25) were obtained for the three metals, resulting %RGI a more sensitive endpoint than %FNI in all cases. For the test with 0.5 mg DOC/L, Cd presented the higher toxicity, based on %RGI, when concentration expressed as M_{diss} ($IC25_{CdDiss}$ = 20.8 µg/L) being 6 times more toxic than Ni and 30 times than Zn. When concentration expressed as M_{tiss},Cd was also the more toxic metal ($IC25_{CdTiss}$ = 76.67 µg/g dry weight) being 10 times more toxic than Ni and 26 times than Zn. For the test with 10 mg DOC/L, Ni was the most toxic when dose expressed as M_{diss} but Cd when expressed as M_{tiss}. At the end of assays, for both DOC concentrations, [Cd_{Tiss}], [Ni_{Tiss}] and [Zn_{Tiss}] were higher in the single metal exposure compared to the mixtures. For the mixtures exposures, the %RGI responses ranged from 17 to 94 % in the lower DOC concentration test and from 15 to 97 % in the higher. Concentration addition (CA) based on M_{tiss} and M_{diss} was evaluated. Multiple regression analysis was used to fit the observed metal mixture toxicity data to either M_{tiss} or M_{diss}. The concentrations of the three metals in mixture expected to result in 50% RGI or FNI were calculated by solving the regression for Cd while holding the concentrations of Ni and Zn constant for each of the 20 cases. To determine whether CA was the appropriate model for the mixtures' toxicity, those combinations of concentrations were used to calculate the 'sum of toxic units' (ΣTU). Deviations from CA approach were observed as relative results according with the metal under analysis.

TH267

ISOLATION AND CHARACTERIZATION OF HEAVY METAL RESISTANT BACTERIA IN SOIL SAMPLES FROM MAMBILLA ARTISANAL MINING SITE, NIGERIA

O. Otitoju, federal University Wukari / Department of Biochemistry; T. Silas, federal University Wukari; A. Martins, S. Asemave, federal University Wukari / Biochemistry

Incidence of soil contamination by heavy metals is widely increasing with the spread of industries. Artisanal mining of blue sapphire gemstone on the Mambilla plateau has been on the increase in recent years. Therefore, the present study was aimed at characterizing and determining resistance to lead, mercury and copper by selected bacteria strains isolated in soil from Mambilla Plateau artisanal mine and to explore their bioremediation capacity. Bacteria were isolated from soil samples obtained from different locations at the Mambilla artisanal mining site, Nigeria. Five (5) distinct bacteria were isolated through gram-staining and some biochemical tests and they were identified as *Staphylococcus aureus*, *Escherichia coli*, *Bacillus sp*, *Enterobacter aerogenes* and *Pseudomonas aeruginosa*. Out of the five (5) bacterial isolates, three (*Staphylococcus aureus*, *Pseudomonas aeruginosa, Escherichia coli*) were selected and grown on nutrient agar plates incorporated with heavy metals namely Lead, Mercury, and Copper. These isolates showed multiple resistance to these metals. *Staphylococcus aureus* exhibited maximum tolerance to Lead, Mercury and Copper at concentrations of 0.15, 0.25 and 0.10g/100ml respectively. *Pseudomonas aeruginosa* showed maximum tolerance to lead, Mercury and Copper at concentrations of 0.20, 0.20 and 0.10g/100ml. *Escherichia coli* exhibited maximum tolerance to Lead, Mercury and Copper at concentrations of 0.25, 0.15 and 0.15g/100ml respectively. The isolates also exhibited high level of resistance to these metals with MICs ranging from 0.15-0.30g/100ml. Copper was the most toxic metal with MIC of 0.15g/100ml while Mercury was the least toxic with MIC of 0.30g/100ml. Antibiotic sensitivity test showed that the 3 bacterial isolates were multi-antibiotic resistant. The results of the present study showed that *Staphylococcus aureus*, *Pseudomonas aeroginosa* and *Escherichia coli* are capable of utilizing heavy metals. Therefore, these bacterial isolates could be a potential agent for bioremediation of heavy metal contaminated environments. However, some species of these bacteria are opportunistic pathogens.

TH268

The exceptions to the rule? Metal bioaccumulation in macroinvertebrates from metal polluted sites with a good ecological status

B. Slootmaekers, R.M. Town, Systemic Physiological and Ecotoxicological Research (SPHERE), University of Antwerp / Department of Biology; R. Blust, L. Bervoets, University of Antwerp / Department of Biology (SPHERE Research Group)

Surface waters are continuously facing a variety of anthropogenic stressors, including pollution, habitat degradation, and loss of connectivity. In such complex and dynamic environments it is challenging to unambiguously establish the effects of trace metal contamination on the resident organisms. The European Water Framework Directive (WFD) obliges member states to set specific water quality guidelines for surface waters, in order to reach a good ecological water quality status for all water bodies. Nevertheless, many rivers and streams are still experiencing trace metal concentrations that exceed the current Environmental Quality Standards (EQS). In combination with other stressors, this situation may lead to an unfavorable shift in the composition of the ecological community due to a variety of direct and indirect effects. The range of concomitant contributing processes means that it is not straightforward to predict the way in which an aquatic environment and community will respond to the presence of a stressor(s). To gain insights into the contributing factors, we are investigating eleven sites for which apparently contradictory effects are observed. That is, based on monitoring data (www.vmm.be/geoview) gathered by the Flanders Environment Agency (VMM), the sites that have an exceedance of the EQS, yet a good ecological quality is observed as expressed by the Multimetric Macroinvertebrate Index of Flanders (MMIF). We hypothesize that the macroinvertebrate communities at these locations have (i) adapted to high trace metal concentrations and/or (ii) experienced a lower metal bioavailability due to the water chemistry. To sort out the involved processes, we will systematically characterize the bioaccumulation and exposure patterns of trace metals in a suite of macroinvertebrate taxa collected at these sites and determine the trace metal concentrations in the different ecological compartments (water, sediment and biota). The results, together with general water quality parameters (pH, conductivity, temperature, DOC and macronutrients) will identify whether the ecological quality is primarily governed by chemical or biological factors, or a combination of the two. The outcomes of our research will provide mechanistic insights into the determinants of ecological quality and facilitate development of a more differentiated basis for the setting of EQS.

TH269

Effects of heavy metal mixtures on bioaccumulation and defence mechanisms in common carp, Cyprinus carpio

G. Castaldo, University of Antwerp / Biology; G. De Boeck, University of Antwerp / Biology SPHERE; R. Blust, University of Antwerp / Department of Biology (SPHERE Research Group)

The aquatic environment is continuously under threat because it is the final receptor and sink of waste streams. This environment receives a huge number of different compounds including heavy metals that can harm the health of aquatic organisms. The main goal of the present study is to better understand the effects of waterborne heavy metals and their mixtures on a freshwater fish. Common carp were exposed to sub-lethal concentrations of Cu and Zn and different combinations thereof for a period of one week at a temperature of 20°C. Our aim is to assess the effect of sub-lethal concentrations of Cu and Zn on fish survival rate, determine the bioaccumulation of heavy metal in the gills and assess changes in gene expression of enzymes related to ionoregulation, oxidative stress and defence mechanisms. Preliminary results indicate that metal accumulation induced expression of metal binding and stress proteins, and metal specific compensatory effects were seen in genes related to ionoregulation and oxidative stress. Further analysis will determine whether antagonistic, additive or synergistic effects occurred.

TH270

Silver nanoparticles exposure inhibits glycans synthesis and induces cytotoxicity in human cell line

K. Shimizu, Toyo University; M. Horie, Advanced Industrial Science and Technology; S. Kashiwada, Toyo University / Graduate School of Life Sciences

Silver nanoparticles (SNPs) are used in industrial products worldwide. Hence, there are concerned about environmental pollution risks. Although silver nanoparticles have been reported having induction of cytotoxicity and ROS accumulation, there is limited information of the toxic mechanisms. In our previous study using embryos of medaka, we have revealed that glycans are one of the toxic targets of silver nanocolloids (SNCs). SNCs is a kind of SNPs and nano-sized particles composed of aggregated silver ions; SNCs keep balance with dissociated silver ions. Glycans have roles of cell-protective, stabilizing and barrier function, we assumed SNP would disrupt glycans function. Beyond medaka research as a vertebrate model in nanotoxicology, in order to evaluate toxic effects of SNPs on humans, we evaluated cytotoxicity of SNPs using human cell line. In this study, we employed four different SNPs including SNCs to compare their different toxicities using three human cell lines. Of SNPs, one was coated with sulfur and diameter was *ca.* 30 nm. Another one's coating material and diameter are unknown. Of SNCs, one SNCs was non-coated and its diameter was *ca.* 30 nm. The another SNCs was coated with nitrogen and diameter is *ca.* 20 nm. We used three kinds of human cell lines; lung cancer-derived A549, epidermal-derived HaCaT, and monocyte-derived THP-1 because we supposed SNPs have a chance to contact to alveolus of lungs, epidermis and blood. To evaluate cytotoxicity, each cells were exposed to SNPs or SNCs (10 μg/mL) and incubated for 24 hours, and then we measured survival rate, membrane damage, inflammatory response, ROS accumulation, caspase-3 induction, intracellular ion concentration, and gene expression. In results, SNPs suppressed survival rates. SNPs and SNCs exposures exhibited membrane disturbance and inflammatory response. However, ROS accumulation and caspase-3 induction were observed in only SNCs exposure. Measurement of concentration of intracellular silver found that higher silver concentration in SNCs exposure rather than SNPs exposure. Finally, to investigate effects of SNP/SNC exposure on glycans, we measured glycan-relative genes (ALG2, B4GALT2 and GNS) expressions. Tested gene expression levels were all suppressed by SNPs and SNCs exposures. Since this study demonstrated that SNPs inhibited glycan synthesis in medaka *in vivo* and human *in vitro* models, toxic effects of SNPs on glycan is probably universal among vertebrate organisms.

TH271

Mixture toxicity of ZnO and silver nitrate to Daphnia magna

M. Baek, KIST Europe; Y. Seol, University of Science and Technology; H. Kwon, Y. Kim, KIST Europe / Environmental Safety Group

Zinc oxide nanoparticles (ZnO NPs) and silver nanoparticles (AgNPs) as Engineered nanomaterials (ENMs) can be mainly found in numerous materials or consumer products. These applications of metal (oxide-) nanoparticles indicate that single nanoparticles released into the aquatic environment may lead to mixture forms of by biological system. In this study, the acute toxicity tests using *Daphnia magna* were conducted for examining the single- and mixture toxicity. The methodological approaches for mixture toxicity (Mixture I – 5:5; Mixture II – 7:3 and Mixture III – 3:7) were conducted as three binary mixtures of $AgNO_3$ and ZnO based on the estimated toxicity data (i.e., EC_{50} values) of single substance. To compare with control response and mixture results, the mode of action in mixtures, the effects of mixture were analyzed using the MIXTOX models. The EC_{50} values of $AgNO_3$ and ZnO were 0.0009 mg/L (with a 95% CI of 0.0007-0.0011 mg/L) and 2.2884 mg/L (with a 95% CI of 1.3702-3.2066 mg/L), respectively. Among the 3 mixtures, mixture III was the highest toxicity at the low concentration. With reference at the concentration addition (CA) and independent action (IA) model of all mixture types, the negative ***a*** points for dose-ratio deviation (DR) in both model indicated an increased toxicity when the mixture effect was caused mainly by ZnO, and the positive b_{DR} points of both model indicates a decreased toxicity (antagonism) when the mixture effect was due mostly to $AgNO_3$. In the end the MIXTOX model was applicable for the prediction of combined effects of toxic compounds. Keywords: ZnO, $AgNO_3$, MIXTOX model, nanoparticle

TH272

How relevant is mixture toxicity of herbicides in surface water?

R. Sur, Bayer AG - Crop Science Division / Environmental Safety; A. Weyers, Bayer Ag / EnSa. Ecotoxicology; M. Ebeling, Bayer AG Crop Science Division / Ecotoxicology - Terrestrial Vertebrates Expert Team; D. Baets, Bayer AG Crop Science Division / Sustainable Operations

The relevance of mixture toxicity of herbicides in surface water based on long-term and high-resolution monitoring data has been assessed in an intensively used catchment in Belgium under real agricultural conditions with significant diffuse and point source entries. Twelve herbicides and one metabolite were monitored in a watershed of 992 ha size for 3.5 years with (sub-)daily sampling intervals. Mixture toxicity was evaluated using hazard quotient (HQ), hazard index (HI) and maximum cumulative ratio (MCR) calculations based on regulatory acceptable concentrations and daily averaged measurements of the site-specific cumulative herbicide exposure. Combined effects of two or more herbicides on algae and Lemna were only relevant in < 2% of samples. Mixture toxicity can therefore be considered of relatively minor relevance and does not seem to pose a real-world concern. A single substance risk assessment would have been sufficient in the vast majority of situations to assess the risk rather than a cumulative risk assessment. Further analysis of the time course of exposure revealed that cumulative effects predominantly occurred in narrow time intervals during the application season in combination with high rainfall intensity causing run-off entries into surface water. Hence, the minor cumulative effects observed can be managed by effective mitigation measures such as vegetated filter strips, conservation tillage practices or green cover crops in addition to reducing point source pollution.

TH273

Simplify: reasonable approaches to Mixtox assessment for plant protection products

A. Weyers, Bayer Ag / EnSa. Ecotoxicology; K. Bender, Bayer Ag / Crop Science Ecotoxicology; M. Ebeling, Bayer AG Crop Science Division / Ecotoxicology - Terrestrial Vertebrates Expert Team; A. Gladbach, Bayer Ag / Crop Science, Environmental Safety Assessment

Interest in regulatory implementation of risk assessment of mixtures has increased and several guidance documents describe the process. Our suggestions here deal with mixtures of PPP that require an environmental risk assessment (ERA) for cumulative exposure. Depending on the regulatory context, this may include PPP with multi active substances, relevant co-formulants, adjuvants, safeners or metabolites. While publications on mixture toxicity understandably tend to focus on "interesting", complex, (or critical) cases, for regulatory purposes when a mixtox ERA is required its implementation should be efficient and identify non-critical areas as early as possible. Simple exclusion criteria and harmonised approaches are usedto avoid a waste of resources both for industry and regulators. ERA is a tiered process, where lower tiers (steps) are designed to identify and exclude uncritical scenrios so that only potentially critical scenarios need more detailed evaluations. Therefore a guiding principle in the regulatory process is that in a first tier some over-conservative assumptions can be made, if they allow to correctly identify scenarios of low risk. If formulation studies are available, the measured toxicity of the mixture (and exposure to it) should be used in ERA. When a calculation of cumulative risk is needed based on active substance endpoints, risk indicators that have already been calculated for single substance ERA such as toxicity exposure ratios or risk quotients should be used to describe the cumulative risk. In a first simple step different endpoints, species and PECs in time and space can be mixed, to show that a given scenario is of low concern. After that first step, a mixture toxicity assessment would describe the cumulative risk more precisely at a given time and place for a defined species and the same endpoint.

TH274

Sublethal toxicity of pesticide mixtures on early life stages of non-target aquatic organisms

E. Rozmankova, RECETOX, Faculty of Science, Masaryk University / Research centre for toxic compounds in the environment RECETOX; B. Morin, J. Cachot, University of Bordeaux / EPOC; P. Gonzalez, University of Bordeaux / UMR EPOC CNRS 5805; L. Blaha, Masaryk University, Faculty of Science / Research centre for toxic compounds in the environment RECETOX

Pesticides are widely used throughout the world in many agricultural and domestic activities. By their presence in the environment, they can have an impact on non-target organisms. Moreover, due to the persistence of some products and the formation of active metabolites, more or less complex mixtures of pesticides can be found in the environment. Thus, the aim of this study was to evaluate the effects of one herbicide (S-metolachlore and his two metabolites) and an insecticide (imidaclopride) on the embryo-larval development of two non-target aquatic organisms. These pesticides are the most abundant representatives of their groups in the Arcachon Bay in France. We were focused on environmentally relevant concentration in this Bay (1 µg/L for herbicides and 0.2 µg/L for insecticide and 2-3 higher concentrations) and their sub-lethal effects on the oyster (*C. gigas*), which is widely present in the Bay due to oyster farms and the zebrafish (*D. rerio*), chosen as a prototypical (eco)toxicology model organism. Firstly, the embryos were exposed to the separate substances, then the mixtures for 5 days (zebrafish) or 2 days (oyster). The malformations, the locomotion activity and target gene expression levels were assessed to understand the mechanisms of possible sublethal toxicity of the selected pesticides. According to our results, no malformations and no effect on locomotion activity for the tested concentrations were observed for zebrafish. On the contrary, the effects on the malformations and the locomotion activity of the oyster larvae is already induced at low concentrations of the selected pesticides. The first results of the gene expression show an overexpression of some of the selected genes of zebrafish (12S, TR-beta – known to be related to the thyroid disruption) caused by one of the metolachlor metabolites. In conclusion, an indication of a novel mode of action of the chronic pesticide toxicity has been detected in zebrafish. The embryo-larval stages of oyster seem highly sensitive even to low environmental concentrations of pesticides.

TH275

Including multistress in risk assessment of pesticides. Current state of knowledge, based on a literature review and evaluation of tank mixture applications in a spraying schedule for strawberries.

P. van Vliet, Board for the Authorization of Plant Protection Products and Biocides; G. Arts, Wageningen Environmental Research (Alterra) / Environmental Risk Assessment; W. Beltman, Alterra Wageningen UR; H. Holterman, Wageningen University & Research / Agrosystems Research; L. Wipfler, Alterra Wageningen UR / Environmental Risk Assessment Team; J. Van de Zande, Wageningen University and Research / Agrosystems Research

This study presents a number of stepping stones towards answering the question if the current product by-product and active substance-by-active substance evaluation provides sufficient protection in the context of the authorisation of plant protection products (PPPs) in the Netherlands. This report is based on a literature review and an evaluation of tank mixture applications in a spraying schedule for strawberries. The topic of tank mixes has been identified by Ctgb (the Board for the Authorisation of Plant Protection Products and Biocides in The Netherlands) as an important knowledge gap. We have quantified the environmental risk for an intensively cultivated crop with sequential applications of products and mixtures of products based on a realistic application schedule and spray drift on surface water in a ditch, the corresponding exposure profiles and the effects based on the Regulatory Acceptable Concentrations of the used active substances. This study shows that the actual strawberry crop scenario is not protective for invertebrates and fish in surface water. Therefore, for the risk assessment of PPPs it needs to be considered that PPPs are part of a crop protection programme and thus should be evaluated in this context. Keywords: multistress, pesticides, environmental risk, aquatic Poster presentation\n

TH276

MODELLING ACUTE AND CHRONIC RISKS OF PESTICIDES RESIDUES IN SOUR CHERRIES

I. Simion, Gheorghe Asachi Technical University of Iasi Romania; R. Hlihor, Gheorghe Asachi Technical University of Iasi, Romania / Environmental Engineering and Management; P. Manuela Olga, Phytosanitary Office / Department of Environmental Engineering and Management; M. Rosca, Gheorghe Asachi Technical University of Iasi Romania; P. Cozma, Gheorghe Asachi Technical University of Iasi Romania / Environmental Engineering and Management; M. Gavrilescu, Gheorghe Asachi Technical University of Iasi Romania / Department of Environmental Engineering and Management

To destroy or prevent insects, rodents, and weeds that might harm agricultural crops, and to control and mitigate plant diseases, farmers started to use pesticides, which are highly toxic chemicals or mixtures. Due to their persistence in the environment and ability to bioaccumulate in living organisms, pesticides generate environmental and human health impacts and risks, which are in a complex relationship. The present study proposes a modelling strategy to assess acute and chronic risks of pesticides residues in sour cherries, considering different age groups and cluster models according to EFSA PRIMo model revised version 2. We initially applied 8 fungicides and 5 insecticides in four treatments during the phenological growth stages of sour-cherries according to Good Agricultural Practices (GAP), in double doses, while ensuring a buffer zone between the plants subjected to the experiments. We followed variation of environmental conditions: temperature, humidity, rainfall patterns and pesticide dissipation in time considering each treatment. The results of pesticides concentration at harvest allowed us to model the pesticides risks to human health. Based on our assessment, it appears that acute and chronic risks of pesticides residues in sour cherries are low. Sour cherries dietary intake of pesticides residues poses an acute risk for children lower than 64.6% and lower than 22.5% for adults. The highest chronic risk level reaches 2.4% for adults and 9.5% for children. Our study suggests that the risk assessment estimates are strongly influenced by age and dietary preferences.

TH277

Environmental and Human Cumulative Risk Assessment of Pesticides Using Local Monitoring Data: A Case Study from the Pucara River Basin, Bolivia

L. Herrero Nogareda, University of Copenhagen / Department of Plant and Environmental Sciences; M. Álvarez Caero, H. Antezana Fernández, Universidad Mayor de San Simon / Facultad de Ciencias y Tecnología; N. Cedergreen, University of Copenhagen / Department of Plant and Environmental Sciences

In Bolivia, pesticides are widely used. There is, however, a lack of regulation of their imports and farmers ignore the toxicity of these compounds. Thus, cases of pesticide intoxication and pollution of natural ecosystems are common. In this study, we performed a cumulative risk assessment of measured concentrations of atrazine, 5 organochlorines and 5 organophosphates (many of them banned) in water and sediment samples from the Pucara river basin. Samples were obtained from 11 sampling points in the river basin. Pesticides from water samples were extracted by liquid-liquid extraction and from sediment samples by Soxhlet extraction. They were then quantified by gas chromatography. The toxicity data of each pesticide were obtained from online databases. Combining the exposure and toxicity data, the environmental risk (sum of toxic units (SumTU)) was characterized for four taxa: algae, the crustacean *Daphnia magna*, and fish for water concentrations, and the midged larvae *Chironimus riparius* for the sediment concentrations. The human risk (hazard index (HI)) for chronical and acute exposure was calculated for children and adults who would drink the water from the river. Pesticide concentrations exceeded the current risk safety thresholds for the environment and the human health at approximately 50% and 20% of the sampling points, respectively. The thresholds were especially surpassed at the discharge zone of the river basin, where SumTU reached values of approximately 0.5 for *D. magna*, fish and *C. riparius*, and HI for chronic exposure reached values of 4.70 and 1.57 for children and adults, respectively. The results suggest that pesticide pollution likely impaired the stream system biota in multiple points, while water was not acceptable for a human daily intake in two sampling points, especially for children. The detected pesticides that caused more concern were heptachlor (banned) and dimethoate (approved). Based on these results, risk management strategies should be highly prioritized in order to reduce the risk posed to the ecosystem and the human health in the studied area. However, further studies should analyse the pesticide levels in other times of the year to see if the present results are representative. It is also necessary to analyse a higher number of pesticides which include an even representation of herbicides, fungicides and insecticides, to be sure that risk is not underestimated.

TH278

Developing a strategy to improve the environmental risk assessment of difficult to test multi-component substances: a new HESI Emerging Issues Committee

D.T. Salvito, Research Institute for Fragrance Materials, Inc. / Department of Environmental Science; M.R. Embry, ILSI Health and Environmental Sciences Institute (HESI)

An international workshop was held in 2016 to address challenges in assessing ecological risk of complex mixtures of substances (e.g., multi-constituent substances (MCS), unknown variable composition and biological substances (UVCBs)). International regulatory frameworks (specifically REACH, Canada's DSL Categorization and Chemicals Management Plan and USEPA's TSCA PMN process) have highlighted the complexities of registering, characterizing fate and exposure, and assessing the risk of complex chemical mixtures, whether resulting from manufacturing environments or plant-derived materials. Several industrial sectors (e.g., petrochemical, personal care) have developed frameworks and methodologies to characterize and analyze these complex substances, and best practices and key research needs were identified at the workshop to support

environmental risk assessment. Bridging from the workshop discussions and conclusions, a new HESI Emerging Issues Committee was formed in late 2017, with the overall mission to develop a tiered approach to UVCB and MCS ecological risk assessment. Initial objectives of this committee are to identify and develop models and methods, develop best practices and guidance, and engage with multi-stakeholder collaborative research projects. This presentation will highlight the initial goals and strategy of this multi-stakeholder, collaborative group.

TH279

Environmental Risk Assessment of Technical Mixtures under REACH

E. Hassold, W. Galert, German Environment Agency - UBA / IV 2.3 Chemicals; W. Drost, Federal Environment Agency (UBA) / Chemicals

A large proportion of substances regulated under REACH end up in mixtures. During their processing and downstream-use substances are blended together in formulations, are reformulated for different uses and end up in a variety of products. REACH addresses the safe use of single substances in technical mixtures, but not explicitly the risks arising from joint effects and exposures of the components. In contrast to other substance-oriented regulations, not the authorities, but the registrants and down-stream users have to assess and guarantee the save use of the registered substances, formulations and products. Recently, some attempts have been made by industry organizations with the concept of LCID/SUMI to improve the assessment and communication of safe use conditions for technical mixtures. However, essential improvements are needed. The development of sound prioritization criteria is essential for a mixture assessment. But a sole consideration of the classification and labeling status is insufficient as it only applies for a minority of substances and further hazardous substances may be missed. All substances with ecotoxicity data need to be considered together with information on uses and exposure. The methods for an assessment of mixtures including component-based approaches, whole mixture testing, identification of main drivers and tiered assessment strategies are in general available. These need to be transparently addressed, so that further calculations by other down-stream user are possible. The quality and availability of hazard and exposure data is central and data (eventually considering an anonymization of CBI) needs to be communicated between registrants and down-stream user in the supply-chain to reach the respective “mixture evaluator”, i.e. the formulator. Communication formats (e.g. extended SDS + SUMI) as well as central data bases seem promising and need to be built up. Indeed, detailed guidance and assistance is needed for formulators of mixtures to enable the assessment of a safe use of mixtures. An implementation in guidance documents needs to involve all stakeholders (authorities, industry, academia) and approaches should be followed and evaluated with respect to their feasability and a sound risk assessment in case studies.

TH280

Natural complex mixtures: Ecotoxic behaviour, what we know and what is next?

R. SAMSERA, CEHTRA SAS; N. DELPIT, Laboratoires des Pyrénées et des Landes; P. Bicherel, KREATiS; J. Rivera, A. Barret, C. durou, CEHTRA SAS; P.C. Thomas, CEHTRA SAS / Ecotoxicology and Risk Assessment

With June 2018 coming, the registration steps of chemicals under REACH review program come to an end but with much more to follow. But now it is time to consider the overall tasks we performed to meet that 2018 deadline. All the different classes of organic substance under REACH were considered during the Phase-in period: monoconstituents, multiconstituents, & UVCBs. Amongst these substance types several families in particular presented challenges to testing especially in ecotoxicology and environment. One of the most challenging groups was fragrances. \n Fragrance chemicals can fall under multiple categories: natural, synthetic, monoconstituent, multiconstituent or considered as UVCBs. One group of fragrances that fell under the title of multiconstituent/UVCB were known to be particularly difficult to assess: Essential oils (EO). EO are complex mixtures, with differing but often low solubility, high volatility and are known to contain some toxic constituents such as terpenes. We met some even more complex substances in this family: gums, resinoids and concretes, sub-categories of essential oils. They were as complex as EO but their composition was mostly unknown and their physical state leads to further difficulties for ecotoxicity testing: they were (mostly) solid(ish), extremely viscous resins, with a frozen honey-like texture. We tested hypotheses and performed ecotoxicology and e-fate studies on all the different classes of these substances (gums, resinoids, concretes and everything in between) to optimize our testing strategies for such compounds: i.e. necessitating avoidance of some studies using alternative approaches. We will present our hypothesis and overall conclusions on the probable next steps for these complex substances.

TH281

Testing chemical mixtures: how to determine the effects concentration(s)?

G. Deviller, DERAC / TERA PRAPS HSE

When the properties of a mixture cannot be estimated from the related properties of its components then testing on the mixture is required under most chemical regulations. However, the available standard methodologies to assess the environmental fate and toxicity have been developed for single substances and are often not directly applicable to mixtures. The first issue is related to the identification of the relevant constituents to monitor during the tests (e.g. composition main constituents, bioavailable fraction…) which may differ according to the substance regulatory frame(s). Second, the development of a specific and quantitative analytical method for each relevant constituent could be technically challenging because (1) all analytical standard substances might not be available since some constituents of the mixture are produced by reaction and (2) the different chemical nature of the constituents may require different type of analytical techniques that might not be (all) available in (the same) GLP testing laboratory. Once the analytical issue has been resolved, a methodological issue may arise during toxicity tests if one or more constituent's concentrations vary during the exposure time. Indeed, if the constituents are found to be all stable, then the effect concentrations (e.g. ECx or NOEC) can be based on the nominal concentration of the mixture. But, in case the constituents have different degradation patterns during the test then, how the recommendation for single substances to base the effect concentrations on the measured concentration should be applied for these mixtures? This presentation is intending to discuss these issues and to bring some elements of response based on case studies.

TH282

Deriving USEtox human non-cancer toxicity Effect factors from the REACH database for thousands of chemicals using R-Studio program

F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Resources Bioeconomy Unit; L. Ceriani, M. Fuart-Gatnik, M. Pavan, S-IN Soluzioni Informatiche Srl; A. Gissi, European Chemicals Agency; E. Saouter, EU Commission JRC / Sustainable Assessment UNit

The Product and Organisation Environmental Footprint (PEF/OEF) methods form a core part of the European Commission (EC) Recommendation “on the use of common methods to measure and communicate the life cycle environmental performance of products and organisations” (2013/179/EU). Based on these methods, the potential impact on humans and ecosystems of chemicals emitted during the life cycle of a product is assessed *via* the USEtox model. To this aim, USEtox requires the most up-to-date input data for a chemical. Focusing on human health (HH), the EC Joint Research Centre has recently obtained from the European Chemicals Agency (ECHA) genotoxicity and repeated dose toxicity (RDT) data (41’381 test results, as of March 2017) generated under the REACH Regulation. Based on these data, data-selection criteria were defined to automatically derive non-cancer HH toxicity effect factors (using R-studio) for thousands of chemicals in USEtox. Genotoxicity data were not retained in the assessment, being associated with qualitative outcomes, and rules for cancer HH effect factors were not derived, since the USEtox ED50, and cancer-TD50 endpoints are not commonly provided under REACH. According to the USEtox methodology, specific fields of the REACH data, included in the RDT endpoint study records (ESRs) via the oral and inhalation route, were used to define selection criteria for non-cancer HH effects, in particular: reliability, adequacy, type of information, test guideline, GLP, species, duration of exposure, route of administration, effect level qualifier, effect level, unit, effect level based on, basis for effect levels. A tiered approach for selecting good quality data was also proposed, based on four quality levels, where studies of the highest quality (key studies, Klimisch 1/2) were included in the first two levels. The main challenges that were identified include the way of data reporting and detailing in ESRs, especially for some critical fields, e.g. endpoint, unit text, duration of exposure, thus leading to the use of either general rules for computing reasons, or other fields in their substitution. Nonetheless, the final effect value per chemical (e.g. NOAEL), automatically derived from REACH data based on the developed criteria, coincided with the critical endpoint value chosen in the ESR in the majority of cases. Based on this work, RDT REACH data for thousands of chemicals can automatically be selected and used for their life cycle assessment in USEtox.

TH283

Deriving USEtox aquatic freshwater toxicity Effect factor from OpenFoodTox database (EFSA) using R-Studio program.

E. Saouter, EU Commission JRC / Sustainable Assessment UNit; F. Biganzoli, EU Commission Joint Research / Directorate D Sustainable Resources Bioeconomy Unit; L. Ceriani, S-IN Soluzioni Informatiche Srl; J. Dorne, European Food Safety Authority EFSA / Scientific Committee and Emerging Risks Unit Department of Risk Assessment; J. Richardson, EFSA / AMU; D. Versteeg, EcoStewardship LLC

Product Environmental Footprint (PEF) and Organisational Environmental Footprint (OEF) form a core part of the Commission Recommendation “on the use of common methods to measure and communicate the life cycle environmental performance of products and organisations” (2013/179/EU). There are currently 25 PEF and 2 OEF pilots testing the method and developing Product Category Rules (PCRs). The potential impact of chemicals emitted during the life cycle of a product is assessed via the USEtox multimedia fate model [3]. This model requires ecotoxicity data to freshwater aquatic life. For PEF/LCA, those data are required for thousands of chemicals using the most up-to-date information [4,5]. The EFSA OpenFoodTox database was used to extract the information required to calculate effect factor for Plant Protection Products. EFSA has populated a chemical hazards database to hold summary hazard data from EFSA’s chemical risk assessments in food and feed (Barbaro et al. 2015; Dorne et al. 2017). The data are freely accessible via the EFSA website OpenFoodTox but also accessible via downloadable Excel files. From the OpenFoodTox database, 2695 ecotoxicological observations were available for about 451 chemicals. After selecting the appropriate data, species geometric means have been calculated for each taxonomic groups. The following final calculation have been performed for thousands of chemicals: - Acute and Chronic species geometric means with standard deviation and number of individual test available per species - Arithmetic average of all the log of the species geometric mean with standard deviation and count of species as well as count of SSD group for each chemical - Lowest Acute and Chronic species geometric means with standard deviation Comparison of the chemical toxicity based on the above calculations and chemical ranking based on GHS and CLP.

TH284

Bioassays for assessing effects of overall migrate from food contact materials

K. Groh, Food Packaging Forum Foundation; J. Muncke, Food Packaging Forum Foundation / General Management

Food contact articles (FCAs) are made from highly diverse materials, and they are chemically complex. FCAs can transfer their chemical constituents, the so-called food contact chemicals (FCCs), into foods. Exposure to FCCs is assumed to be highly relevant in the context of human exposure to (synthetic) chemicals. To assess the risk to human health from chronic ingestion of FCCs, basic information on migrating chemicals must be available, such as their chemical identity. However, this is often not the case for all migrating FCCs, especially the non-intentionally added substances (NIAS), as some or most NIAS typically remain unknown, depending on the type of FCA. Furthermore, the current approach to chemical risk assessment is focused on single substances, while it is known that many FCCs migrate simultaneously, forming the 'overall migrate' and resulting in typical and predictable mixture exposure scenarios. One alternative approach to estimating chemical hazards of FCAs is to assess biological effects of the overall migrate. In addition to assessing mixture toxicity this approach also includes effect-assessment for unknown NIAS which otherwise remain unassessed. We review this approach, discuss benefits and disadvantages, and highlight future research needs.

TH285

A unique index to characterize the global noxiousness of stable and radioactive substances for both human health and ecosystems

K. Beaugelin-Seiller, Institut de Radioprotection et de Sûreté Nucléaire / SRTE; R. Gilbin, Institut de Radioprotection et de Sûreté Nucléaire / PSE-ENV/SRTE; S. Reygrobellet, Institut de Radioprotection et de Sûreté Nucléaire / PSE-ENV/SEREN; J. Garnier-Laplace, Institut de Radioprotection et de Sûreté Nucléaire / PSE-ENV

Inspired by methods and tools developed in the field of life cycle analysis (LCA), we constructed a series of indices to appreciate the harmfulness of radioactive materials and wastes for human and environment health. Six impact categories were considered: human cancer and non-cancer effects on one hand, and ecotoxicity on the other hand, both considering chemotoxicity and/or radiotoxicity. For ecosystems, a comparative toxic unit has already been defined from which we derived our noxiousness index. It is based on the concept of Potentially Affected Fraction (PAF), used here as a damage indicator at the ecosystem level. This concept expressed initially the toxic pressure on the environment due to one chemical. It has been enlarged to mixtures of substances as multi-substances PAF (ms-PAF), and yet applied to a mix of stable and radioactive substances. Combining ecotoxicity data and a simplified model of exposure of fauna and flora, we proposed a chemotoxicity index and a radiotoxicity index, which definitions ultimately allow the addition into a single index. According to acknowledged practices in LCIA and corresponding available data, we suggested declining this approach to human health, taking into account exposures resulting from both ingestion and inhalation pathways. This led to eight basic indexes, which may be aggregated on substances, effects categories or exposure pathways to produce intermediate indexes. The principle of additivity that underlies the whole proposed approach may authorize their complete aggregation in order to produce a unique index also for human health. Different source terms may be then easily directly compared in terms of human and ecological noxiousness.

TH286

Solution-focused application of mixture modelling and chemical footprints

M.C. Zijp, RIVM / Centre for Sustainability Environment and Health; J. van Gils, DELTARES; A. van Wezel, KWR Watercycle Research Institute / Chemical Water Quality and Health; D. De Zwart, DdZ Ecotox / Centre for Sustainability Environment and Health; D. van de Meent, Association of Retired Environmental Scientists ARES / Environmental Science; L. Posthuma, RIVM / Centre for Sustainability, Environment and Health

Status reports of the Water Framework Directive suggest many cases of non-compliance according to formal criteria for Good Ecological and Good Chemical Status as well as for River-Basin Specific Pollutants. This signals problems for both the assemblages of species and the functions we try to protect. Multiple stressors, including 100k chemicals and their mixtures, are the causal agents. In the FP7-project Solutions, a modelling train is developed with the purpose to support derivation of water quality management plans that provide best value for money regarding chemicals and their mixtures, i.e., help to identify the largest potential risk reduction per euro spent. Thereby, the challenge is to focus on those chemicals that matter most, which can result in prioritization to sites (where are largest impacts to be expected), to times (when are largest impacts to be expected, e.g., crop-growing season & pesticides), to affected species groups (which species (groups) are most sensitive to the present impact) and to compound (which chemical (groups) contribute most to local impacts). Collaboration with the FP7-project MARS (on multiple-stress effects on ecological status) forms a multi-stress, multi-chemical integrated context. In SOLUTIONS, the modeling train aims to result in chemical footprints (ChF). ChFs were developed to summarize and communicate predicted mixture risks in simple units. In the current presentation, ChFs are evaluated in terms of the net contributions to predicted mixture impact of emissions in one region, e.g. a sub-catchment, on the mixture toxic pressure in the total river downstream, including the estuary. Hence, it expresses potential transfers in burden from one region to the other. Expanding the information that can be obtained by single-chemical risks, chemical footprints indicate which combination of chemicals and locations require priority for abatement. We present ChF-analysis results for the river Rhine. The river basin was divided in more than 800 sub-basins and chemical footprint information resulting from the emissions of a large amount of chemicals are derived from hydrology driven spatially and temporally explicit modelling for the whole catchment. Subsequently, abatement priorities are proposed, based on the ChF results, that act on the most severe combinations of chemical and region of emissions. The effects of local risk management are expressed in reduced toxic pressure in all downstream sections of the river.

TH287

One-week observation of phthalate metabolites in urine from 12 Korean adults: Exposure levels, profiles, and source identification

J. Lim, Hanyang University; S. Kim, Seoul National University Graduate School of Public Health; I. Lee, Seoul National University / Department of Environmental Health Sciences; A. Lee, Soonchunhyang University; S. Kim, Y. Kho, Eulji University; J. Park, SOON CHUN HYANG UNIVERSITY / College of Natural Sciences; K. Choi, Seoul National University / Environmental Health Sciences; H. Moon, Hanyang University / Marine Sciences and Convergent Technology

The esters of phthalic acid (phthalates) are representative endocrine disrupting chemicals (EDCs) to cause a variety of adverse health effects to humans. Phthalates have been primarily used as additives of the plastic products, cosmetics and personal care products (e.g., DEP). Phthalates are metabolized and eliminated in urine within few hours after human exposure. Due to a short half-life of phthalates, the urinary phthalate metabolites are utilized as an indicative for integrative exposure to phthalates from multiple sources and pathways. In this study, all of the urine samples for 7 days (n=401) were collected from 12 adults to identify the concurrent exposure levels, profiles and exposure pathways of phthalates for different population groups. Eighteen phthalate metabolites include mono-ethyl phthalate(MMP), mono-ethyl phthalate (MEP), mono(2-ethyl-hexyl) phthalate (MEHP), mono(2-ethyl-5-oxohexyl) phthalate (MEOHP), mono(2-ethyl-5-hydroxyhexyl) phthalate (MEHHP), mono-(2-carboxymethylhexyl) phthalate (MCMHP), and mono-(2-ethyl-5-carboxypentyl) phthalate (MECPP) were analyzed. Among 18 phthalate metabolites, MEHP, MEOHP, MEHHP, MCMHP, MECPP, MiBP, MEP, and MBzP were detected in almost all of the urine samples (detection rate >97%). However, MCHP, MiPrP, MiNP, MOP, and MPeP were rarely detected in all of urine samples (< 10%). Total concentrations of phthalate metabolites ranged from 3.12 to 6300 ng/mL with a median concentration of 104 ng/mL. Five DEHP metabolites concentrations were the highest (median: 63 ng/mL). MiBP (median: 8.4 ng/mL), MnBP (6.8 ng/mL) and MEP (5.2 ng/mL) showed relatively higher concenrations than other phthalate metabolites. Our finding suggests the highest burden of DEHP from multiple sources. In the present study, we defined the peak showing the concentration higher than summation of average and double values of standard deviation as a specific source input associated with phthalate exposure. Tracking the exposure source of phthalates suggests that the major contribution of the phthalates exposure pathways was different depending on chemical properties (e.g., molecular weight) and usage of phthalates. The exposure of lower-molecular-weight phthalates such as DEP and DMP was associated with the consumption of cosmetics and personal care products, whereas the urinary DEHP exposure levels varied with the dietary intake. The present study provides an important information for intervention study to reduce phthalates from humans.

TH288

Integrating chemical monitoring data with high-content effects data to prioritize contaminants and hazards in chemical mixtures

D. Martinovic-Weigelt, University of St. Thomas / Biology; A.C. Mehinto, Southern California Coastal Water Research Project / Toxicology; N. Vinas, US Army Engineer Research and Development Center; A. Schroeder, University of Minnesota-Crookston / Math, Science and Technology; E.M. Curran, University of St. Thomas; C. Lai, University of St. Thomas / School of Engieenering; Y. He, University of St. Thomas / School of Engineering; M.L. Ferrey, Minnesota Pollution Control Agency / Environmental Outcomes

Determining ecological risks associated with exposures to complex chemical mixtures in the environment is challenging. Bioeffect-based monitoring tools that can measure integrated biological activity of mixtures have been proposed as one of the solutions. A limitation of these is that they typically do not provide insight into which chemicals are causing the observed biological responses. Utility of methodologies that integrate chemical monitoring with bio-effects data to prioritize chemicals and hazards in complex mixtures will be discussed. More specifically, outcomes of a Minnesota streams case study are used to critically evaluate approaches where: 1) prior knowledge regarding toxicity of detected chemicals is combined with empirical, in situ bio-effects assessment, and 2) where in situ chemical occurrence data and in situ bio-effects data are integrated directly (without the prior knowledge of toxicity of individual chemicals). Samples from 50 randomly selected locations in Minnesota were analyzed for 146 chemicals of emerging concern. Concurrently, at 10 sites, exposures of fathead minnows to stream water were conducted (48h, custom 60K feature microarray platform, liver, N=70). Site chemistry and public bio-effects data for individual chemicals were integrated to prioritize chemicals and predict biological targets of detected chemicals for each site. Partial least-squares (PLS) regression and association rule learning (AR) were used to identify associations between in situ chemistry and in situ transcriptomic effects. At most sites, both prior knowledge-based predictions and fish transcriptomics, indicated activation of estrogen receptor alpha and peroxisome proliferator-activated receptors; their predicted chemical initiators were bisphenol A, caffeine, carbamazepine, and triclosan. Some chemicals (triclosan) were indicated by both knowledge-supervised and direct data integration approaches, but iopamidol (detected at 78% of MN sites) and metformin were only indicated by PLS and AR. Estrogenic effects remain of special concern as all methodologies indicated disruption or estrogen receptor signaling. Collectively, analyses indicated that high frequency of non-detects in the chemistry data, wide datasets, and relatively small sample sizes pose challenges for utility of PLS and AR, whereas poorly populated knowledge bases, lack of fish-specific data and poorly characterized in vitro models are problematic for knowledge-based predictions.

TH289

CENTRAL ASIA POLLUTION: OBSOLETE TAILINGS, OBSOLETE PESTICIDES, OBSOLETE GASOLINE AND HUMAN HEALTH DISORDERS

I. Hadjamberdiev, Toxic Action Network Central Asia / Chief Scientific Officer; A. Rispekova, Asian Medical Institute named Tentishev; V. Didenko, Asian Mediacal University named Tentishev; I. Kniazev, Asian Medical Institute named Tentishev; B. Hadjamberdiev, Toxic Action Network Central Asia

We study the radioactive and toxic wastage health impact: in areas of tailings and obsolete pesticides, and severe air-environment pollution by obsolete gasoline. They all have the same targets - immunity, genetic, endocrine system. The old uranium tailings of former USSR military industry in Central Asia (CA): in North Tadjikistan (tremendous Degmay); in Kyrgyzstan – 29 tailings (high concentration in MailuuSuu river cost), in Uzbekistan 11 tailings and mines. Total radioactive wastages volume of three CA countries – 700 mln ton. There are 268 warehouses of obsolete pesticides and abandoned airfield used for agro pulverization. These warehouses and tailings could pollute distant areas of CA by infiltration by rivers. Additional toxicant is gasoline from noncurrent cars. These three health danger impacts have common target – immune system and genetic system of human. We made map of most danger areas (it has been using irrigation water and under ground water stream, air condition, etc). After that, real level of pollutants was analyzed: Uranium, DDT-DDE sum - in drinking water and meat; PHB and benzapyrene in room air. There were used research tools (GIS System programs; Manual International Chemist Analytic Association; Manual - Disease Mapping and Risk Assessment; etc). Numerous health disorders (in children and pregnant women) of polluted areas were found: abnormal high level of goiter and chronic lung diseases; disorders of immune status in 1,2,3- levels; lower clearance via renal; higher permeability of cell membrane (liver, erythrocytes); leucocytes chromosomal disruption tests. Close correlation between two (or three) toxicant levels and health markers of abnormality was found. Mutual, interconnected impact of different pollutants causes synergetic effect on human's health marker. We offered results of the research and recommendation for five local and two national governments. Specific actions were: 1) to installing special mark: danger-uranium, or/and DDT, or/and bad air; 2) resettled seventeen families for most dander points; 3) implementing order of pregnancy hospitalize-time would be in comparatively clear town district.

TH290

Evaluating HPC ingredients in WWTPs & surface water of the Songhua Catchment using monitoring & high tier modelling tools

J. Kilgallon, Unilever / Safety and Environmental Assurance Centre SEAC; A. Franco, Eurisko Research; C. Hastie, Unilever / SEAC; J. Hodges, Unilever; O. Jolliet, University of Michigan; C. Wannaz, The Unversity of Michigan, Ann Arbor / SPHEHS; C. Sparham, Unilever RD Colworth / Safety and Environmental Assurance Centre SEAC; L. Speirs, Unilever; Z. Zhang, Harbin Institute of Technology; C. Lindim, Stockholm University / SEAC

Introduction: Ingredients commonly used in home and personal care (HPC) products can enter the aquatic environment after use if they are not completely removed during wastewater treatment. We investigated the occurrence and fate of a range of widespread used ingredients in HPC products in wastewater treatment plants (WWTPs) and surface waters of the Songhua River catchment (China) using a high tier modelling framework and monitoring. The aim of our study was to advance understanding in the occurrence and fate of ingredients found in HPC products in the Songhua catchment, in particular 1) to assess spatial trends in the catchment, and 2) to evaluate and improve modelling predictions. **Methods:** A monitoring campaign was carried out by IRJC-PTS, in the Songhua catchment (China) undertaken from June-July 2017, sampling WWTPs and watersheds. Emission estimates were generated for each ingredient based on product sales data for China and were input into the modelling framework. The hydrobasins hydrological dataset has been integrated within the Pangea multiscale multimedia modeling framework, using the hydrological flow between each basin and its downstream basin to parameterize the transfer rates from the corresponding water compartments. **Results:** Initial monitoring results for the Songhua catchment indicate the concentration of HPCs are dominated by LAS in WWTPs and rivers. Modelled influent concentrations show good agreement with measured concentrations for most ingredients, demonstrating emission estimates are reasonable. WWTP median measured removal rates range from 90.6% for TCS up to 99.8% for LAS. In the freshwater compartment there is good agreement, with the model overpredicting concentrations for most ingredients. **Conclusion:** Our combined modelling and monitoring approach is advantageous for assessing exposure, as monitoring data can be used to evaluate model predictions and refine parametrization while modelling provides feedback to improve the representiveness of sampling. This method enables a more detailed analysis of the key sources of uncertainty and variability at each step of the modelling framework (i.e. influent, effluent and river concentrations). Further work to understand the uncertainties in both monitoring and modelling will be carried out in a monitoring campaign in November 2017.

TH291

Mesocosm experiment evidences complex responses of biofilm communities along a gradient of chemical pollution

l. sabater, CSIC-IDAEA / Department of Environmental Chemistry; A. Ginebreda, Institute of Environmental Assessment and Water Research IDAEA CSIC / Department of Environmental Chemistry; O. Pereda, University of the Basque Country; F. Romero, ICRA Catalan Institute for Water Research; D. Barcelo, IIQAB-CSIC / Department of Environmental Chemistry

Freshwater ecosystems are receivers of chemical pollution, which has been recognized as one of the major causes of river impairment. Wastewater treatment plants (WWTP) are point sources of contaminants in natural receiving waters but information about their effects in the ecosystems is still scarce. Ecosystems are known to react to any environmental change by initiating a series of ecological changes to recover its ability to respond to subsequent changes. The objective of this study is thus to verify how robust and resilient is an ecosystem to WWTP effluents using a mesocosm experiment which have been revealed as particularly convenient tool to study biological communities' responses. We exposed twenty-four artificial streams to a range of WWTP effluent concentrations, from no effluent to pure effluent and under controlled conditions of light and water flow during 34 days, followed for 22 days of recovery under no effluent conditions. We analysed river biofilm inhabiting in sediments and cobbles surface because of its major role in ecosystem functioning. To assess impact and recovery we measured Chlorophyll-a content, Chlorophyll-a fluorescence measurements, extracellular enzyme activities (APA and LAP), algal community and metabolism at weekly scale. Pollution load associated to the WWTP effluent was characterized analyzing physical-chemical parameters (pH, dissolved oxygen, conductivity and temperature), nutrients, organic matter, heavy metals and microcontaminants in the water phase. Our preliminary results indicate a complex response of stream ecosystem functioning in front of the WWTP effluent. We notice changes on the system balance and the final return to equilibrium. *Acknowledgements* - The research leading to these results has received funding from the European Comunities 7th Framework Programme under Grant Agreement No. 603629-ENV-2013-6.2.1-Globaqua

TH292

Risk assessment of chemical mixtures in the Erft river basin

S. Rhiem, North Rhine Westphalian State Agency for Nature, Environment and Consumer Protection (LANUV NRW) / Department 32 Ecotoxicology; B. Guhl, North Rhine-Westphalia State Environment Agency (LANUV NRW) / FB 32; U. Rose, M. Trimborn, Erftverband

Mixture toxicity was assessed using extensive chemical monitoring data from the river Erft, its tributaries, and ten municipal waste water treatment plants (WWTPs) discharging into the Erft river basin. A Toxic Units (TU) approach was applied assuming concentration addition and using acute toxicity endpoints for algae, macrophytes, daphnia and fish. Substances with high TU as well as the taxonomic groups displaying highest added up TU response (SUM TU) were determined. The chemical inventories of WWTP effluents were analysed to gather information on non-detects i.e. potentially ecotoxicologically relevant substances which are present in surface waters in concentrations below the analytical limit of detection (LOD). Additionally, single substance risk assessment was performed by determining risk quotients (environmental concentration/ PNEC). Risk quotients larger than one indicate a possible risk for aquatic organisms. SUM TU were mainly explained by few compounds which varied between sampling sites and dates (e.g. seasonal use of pesticides). Overall, WWTPs increased mixture toxicity in the receiving surface waters. For most samples highest SUM TU could be calculated for macrophytesand algae. As a substance highly toxic for algae Triclosan generated high TU. It was detected in nearly all WWTP effluents but in surface waters it was only rarely present in concentrations above LOD. Triclosan can be considered to be a non-detect substance contributing to mixture toxicity even at concentrations below LOD. Pesticides often generated high TU but due to the seasonal application substance patterns varied strongly between sampling dates and different locations. Highest pesticide concentrations in surface waters were measured during heavy rainfall which caused run-off from arable land. Single substance risk assessment identified mainly Triclosan, Ibuprofen and Diclofenac as substances with a possible risk for the aquatic organisms. In waterbodies strongly influenced by WWTP discharges Diclofenac and Ibuprofen were nearly ubiquitous and caused high chronic toxic stress to fish. It was concluded that a combination of single substance risk assessment and mixture toxicity assessment is a suitable tool to evaluate complex monitoring data. Monitoring of substances with high TU (contributing mainly to mixture toxicity) could help to identify surface water for a more extensive monitoring and support specific management planning.

TH293

Assessing groundwater toxicity of emerging contaminant mixtures

M.D. Pavlaki, University of Aveiro / Department of Biology; J.F. Mousinho, University of Aveiro / Department of Biology and CESAM; A.R. Silva, University of Aveiro / Dept.of Biology & CESAM; S. Goncalves, Department of Biology & CESAM - University of Aveiro / Department of Biology and CESAM; R. Morgado, University of Aveiro / Department of Biology and CESAM; S. Loureiro, Universidade de Aveiro / Biology

Groundwater is one of the most important natural resources, as globally it comprises the primary available source of freshwater. Groundwater aquifers consist in an important drinking source in many parts of the world and a point of supply for irrigation in agriculture, among others. Additionally, groundwater aquifers are considered valuable in sustaining ecosystems´ health and functioning. The Groundwater Directive (2006/118/EC) was created to protect groundwater bodies from contamination but to date it does not consider a diverse array of emerging contaminants used in great quantities by society. These emerging contaminants can often occur in mixtures rather than alone, therefore understanding and predicting the toxicity of such complex mixtures, will eventually lead the way to developing new strategies for setting adaptations in regulations. Additionally, adapting surface water protocols to groundwater contamination scenarios might lead to erroneous results due to water different composition. The present work was performed in the context of the European Research Project WE-NEED (Water JPI-WATERWORKS2014 ERA-NET) focused on developing new management strategies to sustainably exploit groundwater resources. A thorough identification of emerging contaminants took place in two well-characterized case-studies, the Bologna and Cremona aquifers. For that, four priority contaminants identified in the two aquifers were chosen as model chemicals and synthetic water was built to mimic groundwater composition from the two aquifers, Bologna and Cremona. The acute toxicity of complex mixtures in these synthetic groundwaters was tested in *Daphnia magna* and deviations from the Concentration Addition reference model were predicted. Based on this step, the toxicity of three emerging compounds and their mixtures (binary and ternary) were assessed in the two synthetic groundwaters using adapted standardized protocols for *Daphnia magna* (OECD 202) and *Danio rerio* (OECD 236). Using a stepwise modelling approach, information from the binary mixtures was used to predict the effects of the ternary mixtures. Deviations from the Concentration Addition reference model indicate interaction between the contaminants in *D. magna* and *D. rerio.*

TH294

Mixture effects of Dibutyl phthalate and Sodium dodecyl sulphate on a mesozooplankton community from the Swedish west coast

C. Jönander, University of Gothenburg; I. Dahllöf, University of Gothenburg / Biological and Envrionmental Sciences; T. Backhaus, University of Gothenburg / Department of Biological and Environmental Sciences

In a typical coastal marine environment chemicals are not present on their own but in mixtures. A chemical monitoring survey detected more than 60 organic chemicals along the west coast of Sweden in 2012, many of which exceeded their environmental thresholds. A total of 33 chemicals was found to co-occur in the water column of the coast of Stenungsund, an area with multiple harbours and home to Sweden's biggest chemical industry cluster. Dibutyl phthalate (DBP) and Sodium dodecyl sulphate (SDS) were amongst the compounds with the highest risk quotient (concentration / ecotoxicity). We therefore investigated their single substance and mixture toxicity to natural mesozooplankton communities, which constitute an important link between primary producers and higher trophic levels like fish. Structurally diverse communities generally possess a large resilience capacity, and it is thus essential to identify sensitive species and structural changes caused by chemical exposure. Potentially, structural changes on this level could indirectly affect even higher levels of biological complexity. We used copepod egg production, hatching success and feeding rate as our primary ecotoxicity parameters, complemented with dead/alive staining of zooplankton with neutral red after the exposure. Additionally, we analysed the community structure before and after chemical exposure by image analysis, comparing images of the exposed samples and untreated controls to a manually classified reference library of mesozooplankton taxa. Single substance experiments show toxic effects on the zooplankton communities by decreasing copepod egg production and hatching success in a concentration-dependent manner, with first effects becoming visible at concentrations of 0.20 µmol/L (SDS) and 0.32 µmol/L (DBP), respectively. The evaluation of structural endpoints as well as the mixture experiments are currently (Nov. 2017) ongoing and will be presented on the poster.

TH295

Analysis of the Mixture Toxicity Burden in 17 Rivers in North Eastern Australia - Implications for the Great Barrier Reef.

F. Spilsbury, University of Gothenburg / Dept of Biological and Environmental Sciences; M.S. Warne, Coventry University / Centre for Agroecology, Water and Resilience; T. Backhaus, University of Gothenburg / Department of Biological and Environmental Sciences

The Great Barrier Reef (GBR) is a protected ecosystem, listed as a UNESCO World Heritage site since 1981. It runs for approximately 3000km along the coastline in north-eastern Australia. A total of thirty-five major river basins discharge to the GBR and many transport large loads of pesticides, suspended sediment, nutrients from agricultural land. Over the past 6 years an extensive program has been conducted by the Queensland Government to monitor concentrations of 51 pesticides and their breakdown products in 17 rivers that discharge to the GBR. To explore the potential impact that the pesticides pose to the riverine environments and to the GBR we analysed the risk posed by the individual pesticides and their mixtures. Australia currently does not have water quality guidelines for 17 of the 38 pesticides detected. For those, we calculated ecotoxicity thresholds using a simplified version of the Australian methodology for determining water quality guideline values, based on species-sensitivity distributions. In all rivers, multiple pesticides were routinely detected at concentrations greater than their level of reporting. All rivers had at least one sample where the combined toxicity was greater than 1 toxic unit (TU), i.e. exposure situations where the total pesticide concentration exceeded acceptable levels. In a number of rivers more than 50% of samples had a combined toxicity greater than 1 TU. Average TU's per river ranged from 13.47 to 0.10, with substantial fluctuations over the seasons but without clear trends between years. The patterns indicate that specific events such as severity of wet/dry seasons and cyclone events impact the combined toxicity found. We also found land use patterns affected the combined toxicity in the river ecosystems. In each of the rivers, 90% of the expected mixture toxicity was caused by only between 2 and 6 pesticides, although the individual pesticides that dominated the combined toxicity differed between rivers.

TH296

Physiological and transcriptomic responses in the tropical coral Stylophora pistillata to inorganic sunscreen exposure.

A. Tagliati, Heriot Watt University / Institute of Life and Earth Sciences; S.J. Hennige, The University of Edinburgh / School of Geosciences; T.F. Fernandes, Heriot-Watt University / Institute of Life and Earth Sciences

Between 16000 and 25000 tons of sunscreen are used annually in tropical countries with tourism linked to coral reef areas considered to be one of fastest growing tourism sector worldwide. Sunscreens are complex mixture of UV filters, emollients, and emulsifiers, and at least 25% of the cream is washed off from the skin during swimming or bathing. Given the lipophilic characteristics and the resistance against degradation of these compounds, sunscreen products can reside in coastal waters and potentially bioccumulate in aquatic animals. Therefore sunscreens may pose a major threat for marine organisms in the shallow water near tourist beaches. Organic UV-filters have been documented to cause bleaching both in adult and larval stages of corals but only few studies have addressed the impact of inorganic sunscreens. A common component of inorganic sunscreen are titanium dioxide nanoparticles ($nTiO_2$) which are widely used as UV-filters in the cosmetic industry due to their transparency and broad spectrum protection along with absence of skin irritation. Results from previous experiments on corals' symbiotic algae *Symbiodinium* indicate that sunscreen toxicity is likely driven by the oil components in a sunscreen formulation and it is independent of the tested concentrations of the UV-filter $nTiO_2$ in the cream. Thus in the present study the tropical coral *Stylophora pistillata*, a common model coral species, was exposed to increasing concentrations of custom-made sunscreen formulations with and without the UV filter $nTiO_2$ to characterize the responses of the chemical mixture either containing or not nanoparticles in it. A series of short-term (5 days) experiments was carried out to analyse effects of these sunscreens on corals, by studying coral photophysiology, coral respiration, symbiont density and chlorophyll content. The expression of genes involved in thermal stress (HSP70), carbon acquisition (intra and extracellular carbonic anhydrase) and calcium and ATP exchange (CA-ATPase) were also analysed to characterize *Stylophora pistillata* transcriptomic response to sunscreen exposure. Results from this work will be presented and compared to other studies carried out with organic sunscreens. Results from the present studies are essential to understand how the exposure to inorganic sunscreens affects reef-building corals, and they will contribute to the development of effective conservation programs and support eco-tourism

economies.

TH297

Effect of antibiotic mixtures on the growth of Anabaena flos-aquae

K. Budin, Environment Department, University of York / Environment; L. Carter, University of York / Environment Department; A. Agatz, IBACON GmbH / Environment Department; J. Wilkinson, The University of York / Natural and Built Environments; K. Selby, Environment Department University of York; A. Boxall, University of York / Environment Department

Antibiotics can be released to the environment, following use in both human and veterinary medicine. As a wide range of antibiotics active ingredients are in use, the natural environment will be likely exposed to mixtures of these compounds. The environmental risks of these mixtures are however poorly understood. In this study, the toxicity of single and mixture of six major human antibiotics from different classes; amoxicillin (AMO), oxytetracycline (OXY), clarithromycin (CLA), meropenem (MER), ciprofloxacin (CPO), cephalaxin (CEP) to the blue green algae, *Anabaena flos-aquae* was assessed. All antibiotics showed high toxicity to the cyanobacteria with EC50 concentration ranging from 0.001 to 0.08 mgl^{-1} (CLA, 0.001mgl^{-1}; CEP, 0.003 mgl^{-1}; CIP, 0.008 mgl^{-1}; OXY, 0.006 mgl^{-1}; MER, 0.02 mgl^{-1} and AMO, 0.03 mgl^{-1}). Use of these toxicity data alongside predictions of surface water concentrations, using simple models, resulted in risk characterisation ratio values of 30.2, 2.5, 2.4, 1.9, 1.4 and 0.2 for AMO, OXY, CLA, MER, CIP and CEP respectively, suggesting that five of the six compounds may be adversely affecting the aquatic environment. The mixtures toxicity studies are ongoing bit once the data available; these will be used to evaluate the concentration addition (CA) and independent action (IA) for estimating the mixture toxicity. The best performing model will then be used alongside exposure modelling approaches to explore the risks of mixture for different scenarios.

TH298

Exposure to mixtures of Persistent Organic Pollutants (POPs) can inhibit the transactivation activities of Aryl hydrocarbon Receptor (AhR) in vitro

Q.T. Doan, Université de Liège (ULiege) / Département des Sciences des Denrées alimentaires; M. Muller, University of Liege / GIGA-R, Laboratory for Organogenesis and Regeneration; H. Berntsen, NMBU-School of Veterinary Science / Department of Production Animal Clinical Sciences; K.E. Zimmer, NMBU-School of Veterinary Science / Department of Basic Sciences and Aquatic Medicine; S. Verhaegen, E. Ropstad, NMBU-School of Veterinary Science / Department of Production Animal Clinical Sciences; L. Connolly, Queens University of Belfast / Institute for Global Food Security, School of Biological Sciences; M. Scippo, University of Liege / Department of Food Science, FARAH

All living organisms are exposed to persistent organic pollutants (POPs) not as individual compounds but as mixtures of chemicals. However, to assess the toxicity of POPs, scientific studies usually focus on the effect of one single compound at a time and do not address the cocktail effect of the mixtures, where different chemicals can act additively, antagonistically or synergistically to produce adverse effects. This study aims to determine, *in vitro,* the effect of a mixture of POPs at the level of the Aryl hydrocarbon Receptor (AhR) function. AhR is a key receptor involved in an organism's response to POPs as xenobiotics. In this study, three different luciferase reporter cell lines (rat hepatoma H4IIE, human mammary gland carcinoma T47-D and human hepatoma Hep G2) were used to screen AhR transactivation activities (i.e. agonistic and antagonistic) of 29 compounds listed as POPs under the 2001 Stockholm Convention. Their mixture, prepared according to their individual concentrations found in human blood, was also tested for the same activities. We show that these compounds have species- and tissue-specific effects and that the rat cells DR (Dioxin responsive)-H4IIE are more sensitive than the two human cell lines (DR-T47-D and DR-Hep G2). Only 6 out of them showed AhR agonistic activities. PBDE-153, PBDE-154, PCB-138, and PCB-118 were able to activate the AhR in DR-H4IIE cells only, γ-HCH was active in DR-T47-D only, while PBDE-99 was found to be an AhR agonist in both cell lines. No agonistic effect was seen for DR-Hep G2. In contrast, 19 out of the 29 compounds showed AhR antagonistic activities in DR-H4IIE, while 10 and 6 compounds displayed AhR antagonistic activities in DR-T47D and DR-Hep G2 cells, respectively. Not surprisingly, the mixture of the 29 compounds also showed an AhR antagonistic activity in all cell lines. In DR-H4IIE, AhR inhibition was observed with concentrations of the POP mixture corresponding to 75 times the blood level and above, which could be plausibly reached in humans after a food contamination incident. The IC50 for the POP mixture was 262.6 ± 104.6 times the background blood level, which corresponds to an interpolated antagonistic equivalent of 0.165 µM BDE-47, while only 0.0047 µM BDE-47 presents in the mixture at the IC50. In addition, the isobole coefficient of the mixture is 0.3 (< 1) according to additive mixture effect model. This indicates that AhR antagonistic effects are significantly enhanced in real mixtures.

TH299

Ecotoxicity of biofuel-mixture DnBE and 1-Octanol on aquatic organisms Danio rerio and Daphnia magna

M.D. Esser, Institute for Environmental Research RWTH Aachen; S. Heger, Institute for Environmental Research, RWTH / Institute for Environmental Research; M. Du, Institute for Environmental Research, RWTH Aachen / Department of Ecosystem Analysis; H. Hollert, RWTH Aachen University / Institute for Environmental Research; A. Schaeffer, RWTH Aachen University / Chair of Environmental Biology and Chemodynamics

The worldwide demand for fuels is increasing, but currently used fuels are based on fossil resources. A possible alternative for diesel is the biofuel mixture of 1-Octanol (1-Oct) (80%) and Di-n-butyl ether (DnBE) (20%). These fuels are based on the raw material lignocellulose and CO_2-neutral. However, the use of this fuel mixture induces a risk of environmental contamination, especially for aquatic ecosystems. This study focuses on the ecotoxicological evaluation of this mixture and the investigation on a possible interaction of the two substances. Acute embryotoxic and developmental effects were investigated in the fish embryo toxicity test (FET) with *Danio rerio* (OECD 236). The acute immobilization assay (OECD 202) was performed to determine the acute toxicity of this mixture for *Daphnia magna*. To interpret the results for possible interactions between the two substances, the investigation of DnBE and 1-Octanol as single substances was necessary. In the acute immobilization test, the EC_{50} values were 14.7 mg/L for 1-octanol and 17.3 mg/L for DnBE. Both biofuels led to teratogenic and lethal effects in the FET ($LC_{50, DnBE}$: 24.7 mg/L; $LC_{50, 1\text{-}Oct}$: 11.3 mg/L). Especially in the study of DnBE was a low hatching rate, while edemas were often observed at the pericardium of the developing larvae. Testing the mixture in the FET revealed a LC_{50} of 14.7 mg/L. The acute immobilization test resulted in a EC_{50} of 25.6 mg/L. The determined EC_{50}/LC_{50} values in both bioassays suggest an additive mode of action of the compounds. The comparison of the determined values with data of other biofuels (2-methylfuran, 2-methyltetrahydrofuran) showed a higher toxicity of the mixture. Diesel showed a higher toxicity on *D. magna* than the tested mixture. For the comprehensive ecotoxicological assessment of the mixture and there compounds as single substances further tests are required. Especially regarding to the possible impairment of the slippage of *D. rerio*. Preventing of slippage can cause the larvae to be no longer viable, resulting in a misinterpretation of the detected LC_{50}. In future, further ecotoxicologically relevant endpoints should be investigated. This work was performed as part of the Research Cluster "Tailor-made fuels from biomass" funded by the Excellence Initiative by the German federal and state governments to promote science and research at German universities.

TH300

Single and combined effects of propiconazole and ZnO (bulk and nano form) on various biomarkers and reproduction in Enchytraeus albidus

N. Ćurčić, . Lonńarí, University of Osijek / Department of Biology; D. Hackenberger, Department f Biology, University of Osijek / Department of Biology; L. ZeliDŽ, University of Osijek / Department of Biology; B. Hackenberger, Department f Biology, University of Osijek / Department of Biology

Organisms are often exposed to a combination of chemicals. These chemicals may and may not interact with each other and affect their toxicity to an organism. Nanoparticles have been recognised as an emerging contaminants over the last decade. However, the majority of research have dealt with the effect of a nanoparticle as a single stressor. In this study we have investigated whether a combination of fungicide (propiconazole - PCZ) and nanoparticle (zinc oxide - ZnO) have different effect on oxidative stress and reproduction of *Enchytraeus albidus* than each of these compounds applied separately. Propiconazole was tested as a commercial formulation and ZnO was tested and used in a nano and bulk form. In a preliminary experiment an EC50 value for reproduction for each compound was calculated (480 mg/kg ZnO and 40 ug/g PCZ). In the second experiment encyhtreids were exposed to five concentrations in following ratios: 100% PCZ, 75% PCZ/25% ZnO, 50% PCZ/ 50% ZnO, 25% PCZ/75% ZnO, 100% ZnO. 100% is an EC50 value. The time of exposure in both experiments was 21 days after which adult enchytraeids were removed from soil, following another 21 days for juveniles to hatch, according to OECD protocol. Adult enchytraeids were used for subsequent measurement of biomarkers of oxidative stress (AChE, CAT, GST) and metallothionenin content. The results showed a slightly different response of measured biomarkers to nZnO than to a bZnO in both single exposure and when combined with propiconazole as well as the effect on reproduction.

TH301

Mixture toxicity of abamectin and difenoconazole to zebrafish embryos (Danio rerio)

A. Madeira Sanches, University of Sao Paulo - USP / Núcleo de Ecotoxicologia e Estudos Ambientais NEEA; E. Freitas, University of Sao Paulo - USP / Hydraulic and Sanitation; I. Domingues, University of Aveiro / CESAM Department of Biology; M. Daam, CENSE & New University of Lisbon, Lisboa; L.P. Figueiredo, University of São Paulo USP; E. Espindola, University of São Paulo USP / Hydraulics and Sanitation

There are many agrochemicals used in agriculture, among them the insecticide abamectin and the fungicide difenoconazole. They are widely used compounds in strawberry crops in regions of tropical climate, although they are compounds classified as extremely toxic and very dangerous to the environment. The use of fish as test organisms stands out in ecotoxicology due to its representativeness and critical role in aquatic environment. Due to ethical issues and to reduce costs, space and waste generated, alternative methods such as assays using fish embryos are currently widely used. The FET - Fish Embryo Toxicity Test is an example of a standardized test that use *Danio rerio* embryos. Considering the ecological risks

inherent to the use of the insecticide abamectin and the fungicide difenoconazole, the main objective of this research was to evaluate the effect generated in *Danio rerio* embryos exposed to pesticide mixtures and evaluate the effects produced by the interaction of these compounds. For this, *Danio rerio* embryos were exposed for 96h to the binary mixtures of abamectin and difenoconazole following the recommendations of OECD TG 236. The concentrations used were 0.5; 1.1; 2.4; 5.3 and 11.7 mg L^{-1} of abamectin and 0.2; 0.5; 1.0; 2.3 and 5.0 mg L^{-1} of difenoconazole. The factorial design was used combining all possible concentrations, and in total 35 treatments plus the control were performed. The exposures were performed in 50 mm Petri dishes using three plates per treatment. In each plate containing 15 mL of solution were arranged 5 eggs totalizing an n = 15. Survival data were recorded every 24 hours and the results were analyzed in the Mixtox program. It was observed that, in acute exposures, the binary mixtures of abamectin and difenoconazole promotes in *Danio rerio* embryos the antagonistic effect in the lower concentrations, but in higher concentrations the produced effect is synergic. This means that, at lower concentrations the interaction of abamectin + difenoconazole seems to decrease the toxicity of pesticides to *Danio rerio* embryos, but the toxicity of the compounds is potentiated at higher concentrations of the mixtures. Similar results have been obtained in other studies with cladocerans exposed to mixtures of these same compounds, but complementary studies are necessary to better understand the toxicokinetic of these pesticides mixtures.

TH302

Cocktail-effect of persistent organic pollutants on selected bioreporter-systems and zebrafish embryos

N. Pagano, RWTH Aachen University; G. Nilen, B. Holmes, M. Larsson, M. Engwall, Orebro University / Man-Technology-Environment research centre (MTM); H. Hollert, RWTH Aachen University / Institute for Environmental Research; S. Keiter, Orebro University / MTM Research centre

There is an ever-increasing number of chemicals including pharmaceuticals and industrial pollutants that are released into the aquatic environment, leading to the exposure of fish and other aquatic organisms. Moreover, at the present time environmental risk assessment is mainly based on chemical analysis, only. However, "compound-by-compound" based assessments seriously run the risk of underestimating the risk of chemicals as the true exposure scenario for humans and wildlife is known to be far more complex. Under regular environmental conditions organisms can be exposed to multiple chemicals associated with different risks and specific effects, e.g. teratogenicity, immune toxicity and suppression, genotoxicity, and endocrine disruption. Moreover, it has been repeatedly demonstrated that pollutants and the underlying toxic responses may interact and generate effects that are different from the toxicity of the individual chemicals. Thus, understanding the effects of mixture toxicity, generally referred to as “cocktail effects”, represent one of today´s greatest challenges in environmental but also in human toxicology. The aim of the present study is to investigate embryotoxic and teratogenic, but also mechanism-specific effects using zebrafish embryos. They will be exposed to selected priority pollutants and their mixtures (e.g. polychlorinated biphenyls, heavy metals, polycyclic aromatic compound). These chemicals represent highly relevant chemicals which can be found in great levels in the environment. First results indicate that beside biological interactions heavy metals may alter the toxicity of organic pollutants. This study is part of the EnForce project (https://www.oru.se/enforce), which aims at the development of an effect-based risk assessment in cooperation with different stakeholders and several industrial partners.

Emerging technologies and related raw materials requirements scenarios: the role of life cycle thinking (P)

TH304

Environmental impact assessment of carbon fibers reinforced composites pyrolysis process

E. Neri, Alma Mater Studiorum - University of Bologna; F. Passarini, Alma Mater Studiorum - University of Bologna / Dept. of Industrial Chemistry; B. Berti, I. Vassura, L. Giorgini, G. Zattini, Toso Montanari Department of Industrial Chemistry, University of Bologna; C. Tosi, I. Montanari, CURTI S.p.A., Energy Division

The end-of-life management of carbon fibers reinforced composites (CFRCs) has been investigated by comparing the environmental sustainability of Curti S.p.A. company’s pyrogasification process with waste-to-energy (WtE) and landfill disposal. The determination of environmental loads was carried out through the Life Cycle Assessment (LCA) methodology, modeling and analyzing each scenarios through SimaPro software and Ecoinvent database. CFRCs are highly engineered materials, with high calorific power and excellent mechanical properties. From their recovery, it is possible to obtain a secondary raw material that can be used in application requiring lower performance than originals, or the recovery of thermal/electrical energy. The market still offers few CFRC recovery technologies. Therefore the most developed ones have been chosen to compare with landfill disposal, even though nowadays it would be avoided, for waste with a LHV>13 MJ\kg [1]. The pyrogasification process involves a first pyrolysis and a subsequent gasification of the waste within the same reactor. This system allows the quantitative recovery of carbon fibers (CF) contained in the initial composite and generates hot gaseous compounds that are burnt and released into the atmosphere. Since the plant is at a pilot scale, a heat recovery system has not been designed yet for the combustion of fumes. Considering the impact assessment results, pyrogasification has proved to be the most sustainable treatment due to the quantitative recovery of carbon fibers, which avoids the consumption of material and energy deriving from the production of virgins carbon fibers. The worst scenario is WtE, mostly considering the damage to human health category generated by emissions in air and water. The impact of landfill disposal is intermediate, due to the good stability of CFRCs: having a slow degradation, their disposal in landfills does not cause an high impact, except for land occupation. The LCA study made it possible to carry out a preliminary assessment supporting the pyrogasification pilot plant design, to identify critical aspects and strengths of each scenarios. [1] Legislative Decree n°36 of 13 January 2003; Implementation of Directive 1999/31/EC on landfill of waste, Official Journal of the Italian Republic, 2003. [2] Legislative Decree n°205 of 3 December 2010; provisions implementing Directive 2008/98/EC of the European Parliament and of the Council of 19 November 2008 on waste, Official Journal, 2010.

TH305

Critical raw materials in a new building integrated photovoltaic system

D. Garrain, I. Herrera, Y. Lechón, CIEMAT / Energy Dpt   Energy Systems Analysis Unit

REELCOOP, an EU-FP7 funded project which stands for REnewable ELectricity COOPeration (www.reelcoop.com), aims to develop and test novel prototypes of renewable electricity generation technologies. One of the prototypes is a solar photovoltaic (PV) ventilated façade (6 kW) and involves the development of c-Si solar cells, as well as the study of the ventilation effect in PV façades. PV solar panels have particular metals or rare earths that are potentially included in the category of ‘critical raw materials (CRMs)’. This work aims to identify the potential CRMs in this prototype and to define several ways to improve the sustainability from a life-cycle approach, including aspects like substitution or recycling of these materials.

TH306

Environmental sustainability assessment of a biological Active Pharmaceutical Ingredient: A resource based Life Cycle Assessment

A.G. Renteria Gamiz, Ghent University / Department of Sustainable Organic Chemistry and Tehnology; W. De Soete, Ghent University / EnVOC; B. Heirman, Johnson and Johnson / EHSS Product Stewardship; J. De Graaf, Janssen Biologics / Safety Health   Environment; S. De Meester, Ghent University / Department of Industrial Biological Sciences; J. Dewulf, Ghent University / Department of Sustainable Organic Chemistry and Technology

Most Life Cycle Assessments (LCAs) performed in the pharmaceutical sector have been done on chemically synthesized drugs, leaving biopharmaceuticals aside. The fact that sustainability assessments of biopharmaceutical products and technologies have been rarely done is probably a consequence of the major challenge that building a robust Life Cycle Inventory (LCI) represents for the upstream and downstream processes of biologics. However, the low number of LCAs performed on biopharmaceuticals does not reflect the current reality of the pharmaceutical sector, as its market is rapidly growing at an annual rate of around 9%. Therefore, this study focuses in performing a resource based LCA to measure the environmental impact of a biological Active Pharmaceutical Ingredient (API). A resource based methodology is chosen addressing the task that society faces on supply and efficient use of resources. The API investigated is infliximab, a monoclonal antibody that treats autoimmune diseases. An Exergetic Life Cycle Assessment (ELCA) was conducted, using the Cumulative Exergy Extracted from the Natural Environment (CEENE) method. First results show that the unit operations with the highest impact are: i) The first chromatographic process for purification (Direct Product Capture), since it requires the highest quantity of buffers which are produced using chemicals as well as complex organic compounds such as amino acids. ii) Fermentation, as similar complex components are required for its medium, which are also produced through biotechnological processes. Furthermore, fermentation is the process that takes the longest (several days), leveraging the Heating Ventilation and Air Conditioning (HVAC) system to achieve the clean room conditions needed for the production of biologics. HVAC has shown to be the utility with the highest impact, consuming a significant percentage of the total electricity used in the plant. Performing an LCA on a biologic mainly using primary data has been proven to be a possible task. However, challenges such as the data unavailability of biotechnologies used to produce the nutrients needed throughout the process, as well as the further integration of these technologies into databases should be addressed.

TH307

LCA methodology: a case study of the industrial production of terephthalic acid from renewable sources

M. Volanti, Università di Bologna / Industrial Chemistry Toso Montanari; F. Passarini, Alma Mater Studiorum - University of Bologna / Dept. of Industrial Chemistry; D. Cespi, Environmental management and consulting (EMC) Innovation Lab S.r.l.; E. Neri, Alma Mater Studiorum - University of Bologna; F.

Cavani, Toso Montanari Department of Industrial Chemistry, University of Bologna / Dept. of Industrial Chemistry; P. Mizsey, D. Fozer, Budapest University of Technology and Economics / Department of Chemical and Environmental Process Engineering

The scope of the present study is to investigate the environmental sustainability of different routes of terephthalic acid (TA) production, comparing the results achieved for the traditional way with those of three bio-based routes. The aim of the study is to identify which of the selected pathways has the lowest environmental load. Below the four routes selected are briefly described: Traditional way: p-xylene is obtained from catalytic reforming of crude oil as part of extracted BTX (benzene, toluene and xylenes isomers); GEVO's process: isobutanol from the fermentation of biomass is converted into hydrocarbons, iso-octene and p-xylene; From HMF and ethylene: it involves the production of HMF (5-hydroxymethylfurfural) from starch, its conversion to DMF and the Diels-Alder reaction with bio-ethylene to obtain p-xylene; Alternative pathway from p-cymene: it consists in the oxidation, using O_2 in the presence of a catalyst, of p-cymene (derived from orange peels) to obtain TA. The first two routes are already set at industrial level; the others are still under development. Therefore, in order to estimate the energy requirements of the scenarios, a simulation of the chemical processes was carried out using ChemCad software. The first three syntheses are based on the production of p-xylene, subsequently oxidized to TA through the Amoco process. On the contrary, the last route is proposed as an alternative way, since it uses p-cymene as a precursor of TA. In order to perform the comparison, a LCA (Life Cycle Assessment) methodology has been used as a scientific tool of analysis and evaluation of the potential impacts of each scenario. LCA allows the evaluation of the environmental weight associated with the entire life cycle of a product, in our case 1 ton of terephtalic acid. The model was analyzed by the use of SimaPro software and the results indicate that bio-based processes are sustainable only if they use organic residues as raw materials. Both analysis methods used, CED and ReCiPe, have shown that only the alternative way from orange peels has lower environmental impacts than the traditional way from crude oil. For the other processes, the cultivation and transformation of biomass present an environmental weight that does not justify their use as starting materials for p-xylene, and then TA, production. Thanks to this work it was also possible to make some considerations about energy requirements and their weight in the life cycle of the processes.

TH308

Environmental assessment of vanadium redox flow batteries

C. Minke, Technische Universität Clausthal / Energy Research Center; J.F. Peters, Karlsruhe Institute of Technology KIT / Helmholtz Institute Ulm HIU; M.J. Baumann, Karlsruhe Institute of Technology KIT / Institute for Technology Assessment and Systems Analysis; M. Weil, Karlsruhe Institute of Technology KIT / Institute for Technology Assessment and Systems Analysis ITAS

The rising production of electric energy from renewable sources requires electrical energy storage systems to compensate for the fluctuations in energy generation. The vanadium redox flow battery (VFB) is a suitable technology for stationary energy storage on a broad scale. Due to the independent scalability of system power and energy capacity, residential applications as well as industrial applications can be realized with VFB systems. Especially the innovative concept of extruded graphite composite bipolar plates leading to large-area electrodes (about 2.7 m²) paves the way to megawatt applications. Thus, a sharp increase in research and development may be observed in the past decade with strong efforts towards cost-effective VFB systems [1]. In a holistic approach besides of technical optimization and economic feasibility the environmental impact of technologies for sustainable energy systems has to be assessed. A research gap on environmental assessment of VFB has been identified. The most cited LCA has been published in 1999 [2]. To date the VFB technology has been further developed and commercialized. Based on preceding detailed studies on the production of VFB components and systems an update of their environmental assessment with primary LCI data is developed [1]. A focus is set on the potential for recycling of battery components at their end of life. In a recent study it has been quantified for a small 5 kW-4 kWh-VFB-system with only 18wt.-% associated with a potential of 16% ecological impact reduction by recycling [3]. In the present study recycling paths for all VFB components are assessed in detail considering substantial differences in design of small systems for residential application (kilowatt-size) and large systems for industrial application (megawatt-size). 1. Minke C, Kunz U, Turek T (2017) J Power Sources 361: 105–114. 2. Rydh CJ (1999) J Power Sources 80: 21–2 3. Unterreiner L, Jülch V, Reith S (2016) Energy Procedia 99: 229–234.

TH309

Towards the Life Cycle Assessment of engineered nanoparticles production: a comparison between batch and continuous flow synthesis

F. Grimaldi, University College London / Department of Chemical Engineering; M. Pucciarelli, University College London / Chemical Engineering; A. Gavriilidis, University College London / Department of Chemical Engineering; P. Lettieri, University College London / Chemical Engineering

The aim of this study is to provide an assessment of continuous micro/milli-flow technologies applied to Engineered Nanoparticles (ENPs) production, benchmarking them against the conventional batch productions. As the quantity of ENPs produced and their applications are steeply growing, an increasing attention is being paid to the quality of the product and the efficiency of the synthesis. Continuous micro/milli-flow (CF) synthesis is considered to be the natural evolution of the currently most diffused conventional batch synthesis, as it intensifies the production in terms of productivity, energy and chemicals use, product quality and functionalisation. Given the importance and potentiality of the transition, an evaluation of these new technologies is needed. Life Cycle Assessment is the chosen methodology for addressing this objective, as it looks deep into the sustainability, efficiency and environmental impact of the system considered, in order to identify the key parameters for a deliberate green process design. The application of the LCA methodology to ENPs production presents many challenges: to date, few LCA studies within the manufacturing process of ENPs have been carried out. In this work, a wide selection of ENPs production is modelled and analysed, notably Au, Ag, SiO_2 and iron oxide. The CF syntheses are evaluated on a lab scale, performing a hot-spot analysis and benchmarking them against the equivalent batch syntheses. The output of the assessment permits valuable considerations on the best equipment materials, solvents, stabilizers, type of heating and mixing for maximising the efficiency of the process, even at the very early stages of its development. The results obtained highlight a general favourable tendency toward the CF system as a greener and more efficient way of ENPs synthesis than the correspondent batch production.

TH310

LCA of nanomaterials production for the emerging technology: the case of printing batteries

I. Sánchez, LEITAT / Sustainability Division; C. Hidalgo, Leitat Technological Center / Sustainability Division; M.R. Riera, LEITAT / Sustainability Division; S. Vazquez, ACONDICIONAMIENTO TARRASENSE (LEITAT TECHNOLOGICAL CENTER) / Sustainability Division

BASMATI is an ambitious project which main goal is to develop active nanomaterial and electrochemical inks for printing technologies to transfer and up-scale to pilots at SMEs and industry facilities. This project is co-financed by European Union under The Framework Programme for Research and Innovation HORIZON 2020. The project is developed by 10 different partners, where LEITAT is in charge of the Life Cycling Assessment. For that task, the environmental impact has been performed during the whole life cycle considering synthesis, formulation and disposal of printed batteries. The LCA has been focused on quantifying the potential impacts of Cu, LFP ($LiFePO_4$) and NMC (Ni-Mn-Co) nanoparticles synthesized, the inks which contain these nanoparticles and the batteries printed with these inks. The LCA applied in the study is based on the standard ISO-framework for LCA (ISO 14040:2006 and ISO 14044:2000). Calculations have been done using the software GABI, and taking as a basis the GABI Database, Ecoinvent Database and the ILCD impact assessment method. The functional unit has been defined as "a printed flexible battery to be used for power source" and the scope has been based on the "cradle to grave" approach. Primary data have been prioritised and secondary data from databases and literature have been used when needed. The inventory process has been completed with the information of inputs and outputs gathered from partners. Then, the impacts related to inventory flows has been calculated for ten impact categories. Results show that the impact of nanoparticles synthesis is mainly dominated by raw materials. Moreover, for the inks comparative, and results it can be seen that inks with NMC nanoparticles have higher impacts in most of the impact categories. Specifically, NMC inks represent the highest weight due to raw materials used during NMC synthesis. In addition, the highest impacts in climate change and resource depletion are dominated by Cu Inks. Also, LFP ink represents the highest impact in Ozone depletion category due to LFP synthesis. After that, the manufacturing of the printed lithium-ion batteries has been analysed. The stack and interdigitated battery has been chosen as demonstrators to develop the LCA. Landfill and recycling have been assumed as end of life scenarios. Finally, the conclusions take into consideration the new generation of technologies and their environmental performance.

TH311

Life Cycle Assessment (LCA) applied to new and advanced material solutions in Concentrated Solar Thermal technology

A. Claret, Leitat Technological Center / Sustainability Division; S. Vazquez, ACONDICIONAMIENTO TARRASENSE (LEITAT TECHNOLOGICAL CENTER) / Sustainability Division

The benefits of high efficiency concentrated solar power (CSP) and photovoltaic (PV) are numerous: environmental protection, zero-carbon process, energy security and economic growth. CSP has advantages in front of PV: possible 24h continuous electricity production, electricity and heat generation, heat for distributed in cogeneration plants. Nonetheless, the energy efficiency and the energy production cost of this promising technology have to be improved. The aim of IN-POWER project is to develop high efficiency solar harvesting CSP architectures based on holistic materials and innovative manufacturing processes while reducing the environmental impact associated to CSP architectures and the energy production cost. To achieve this objective IN-POWER develops a set of advanced solutions: Polymeric Smart light mirrors with high optical and mechanical performance. An optimized and lighter mirror support structure. High-operational-temperature absorber coating in new vacuum-free-designed receiver. A novel modular solar field architecture and design reducing the land use requirements by 4 times.

High-operating-temperature thermal storage materials for TES increasing up to 3 times the thermal capacity. All these solutions are being assessed through a comprehensive LCA, considering the entire life cycle of materials and components, from raw material extraction until the end-of-life. A comparative analysis is being prepared between baseline scenario (with reference materials) and the scenario with the IN-POWER innovative materials. Along the project different candidate materials and approaches are being assessed as a decision making process looking for high performance component but environmentally friendly. Some improvements are being made such as: use of aluminium instead of silver in the mirror reflective coating, to meet the European requirements for use of non-critical materials; the reduction of materials weight; and the increase in materials robustness. The expected results are to: Obtain a complete environmental profile of IN-POWER CSP architecture. Calculate the environmental impacts associated to: new polymeric materials for mirrors; high absorber coating; high thermal storage capacity materials; polymeric composite for CSP structure. Evaluate the benefits of IN-POWER materials compared with reference materials. Evaluate the benefits of IN-POWER CSP architecture compared with current mature Parabolic Trough Collector technology

TH312

Environmental impact and social influence of an Advanced Adiabatic Compressed Air Energy Storage (AA-CAES) located in Eisenerz, Austria. The case of RICAS2020 PROJECT.

A. Claret, Leitat Technological Center / Sustainability Division; G. Ferrer, Leitat Technological Center / RD Safety Sustainability Division; M.R. Riera, LEITAT / Sustainability Division; S. Vazquez, ACONDICIONAMIENTO TARRASENSE (LEITAT TECHNOLOGICAL CENTER) / Sustainability Division

European society has a **highly dependency on electric power**. In 2009 European Union fixed that at least 20% of EU gross final energy consumption have to come from renewable energy sources until 2020. The increasing use of renewable energy sources to produce electricity has generated a **worldwide challenge** for the electric grid, where the peak production of energy is usually not in phase with the peak demand; the **developing of large scale electric storage systems**. The innovative **AA-CAES** developed within the **RICAS2020 project** can solve this problem. In a CAES the air is compressed in a storage unit when electric energy overproduction is available, and by the inverse process, is reintroduced in the grid when required in the high demand periods. Additionally, AA-CAES collects the heat produced by compression in a specific Thermal Energy Storage (TES) and returns it to the air when the air is expanded to generate power, delivering **higher efficiencies via a zero CO_2 emissions process**. RICAS2020 is being assessed under the **Environmental and Social LCA**, in order to define improvement measures to guarantee its sustainable performance. **The scope of the LCA** covers the **construction** and the **operation stage of** the **AA-CAES**. Regarding the construction stage are being assessed: the site excavation methods and the manufacture of materials needed for the construction of the Cavern and TES. Respect to the operation stage, the impact of machinery used (turbines, compressors, coolers) are being considered. The **main goals** of this assessment are to: - **Quantify the environmental and social impacts** and benefits of AA-CAES, compared to current energy storage technologies: Pumped Hydroelectric Energy Storage and Batteries. - Identify **which of the candidate materials** involved in the construction of the Cavern and TES **have the most suitable environmental profile**. **Preliminary results of the environmental LCA** of candidate materials for TES have been obtained so far. 12 scenarios have been generated, by combining 3 different structural configurations (an excavated "trough" in the rock, steel container, concrete structure) and 4 different storage materials (rocks from the excavation site, gravel, alumina or silica ceramic spheres). Results have shown that worst cases are the scenarios that include alumina ceramic spheres as storage material, due to the high impact of aluminium oxide. The best scenario is the use of rocks from the excavation site without including structural material.

TH313

Upgrading wastewater treatment technologies in the framework of current renewable energy policies - an environmental assessment

A. Petit-Boix, University of Freiburg / Chair of Societal Transition and Circular Economy; M. Rufí-Salís, Universitat Autònoma de Barcelona / Institute of Environmental Science and Technology (ICTA); G. Villalba, X. Gabarrell, J. Rieradevall, Universitat Autònoma de Barcelona / Institute of Environmental Science and Technology & Department of Chemical, Biological and Environmental Engineering; E. Moliné, Depuración de Aguas del Mediterráneo; M. Suárez-Ojeda, Universitat Autònoma de Barcelona / Department of Chemical, Biological and Environmental Engineering

A large number of wastewater treatment plants (WWTP) use anaerobic digestion to treat surplus sewage sludge, which produces methane-rich biogas. This biogas can be used for cogeneration with the ultimate goal of turning WWTPs into energy self-sufficient facilities. For this reason, current innovation projects focus on (i) improving the energy efficiency of the plant and (ii) updating the technologies used in WWTPs or proposing new processes that increase biogas production. However, we need to clearly define whether these technological updates and innovations result in net environmental benefits or generate tradeoffs. Here, renewable energy policies should align with the environmental goals of energy self-supply in order not to discourage the investment in this type of infrastructure and its potential environmental benefits. Thus, we question whether upgraded or new wastewater treatment technologies generate larger environmental impacts when renewable energy policies are not favorable to the self-supply of energy through cogeneration. In this case, our study focuses on a conventional WWTP in the city of Rubí (Barcelona), which currently only removes organic matter. This facility considers an upgrade that consists of a new wastewater treatment scheme, i.e., (i) a first stage of modified enhanced biological phosphorus removal with struvite recovery and (ii) a second stage of autotrophic biological nitrogen removal in two-steps. A life cycle assessment will determine the environmental impacts and benefits of this upgraded system with respect to the conventional setup. Policy tradeoffs will also be assessed. This wastewater treatment plant originally benefited from cogeneration, but eventually decided to use biogas flare systems due to the taxes on self-supply of electricity imposed by the Royal Decree 900/2015 approved by the Spanish Parliament. As a result, we need to determine if applying technologies that generate more biogas will result in larger impacts than the current scenario if the biogas cannot be used for cogeneration.

TH314

Anticipatory life cycle assessment of sol-gel derived anti-reflective coating for greenhouse glass

N. Tsoy, CML Leiden University / CML; J. Quist, Delft University of Technology / Technology, Policy, and Management; A. Wypkema, M. Mourad, TNO / Materials Solutions; V. Prado, CML Leiden University

Technological innovation is crucial for sustainable development but at the same time it can cause unfavorable consequences to environment and society. Environmental assessment of the technologies is usually performed when they have already been launched in the market with a low possibility to transform their development towards better environmental performance. Anticipatory Life Cycle Assessment (LCA) has been evolved to assess the environmental impacts of the technological innovation using an explorative approach. Here, we show the application of Anticipatory LCA in the assessment of the anti-reflective greenhouse glass coatings. The light intensity is essential for the crop growth in the greenhouses. A portion of the light is lost when it reaches the greenhouse glass surface due to the glass reflection. The anti-reflective glass coatings can be applied on the surface of the greenhouse glass to allow more sunlight to enter the greenhouses. As a result, more yield of crop could be obtained under coated glass. The Netherlands Organization for Applied Scientific Research (TNO) is developing a novel anti-reflective coating for greenhouses which is expected to have higher light transmittance than the conventional coatings by 2.5%. In this study, three reference coatings in addition to the novel coating were assessed. The functional unit was the mass of tomatoes obtained under a certain area of a greenhouse with uncoated/coated glass during 30 years. The novel coating is being synthesized in the laboratory scale, and thus, in LCA, What-if scenarios were used to scale-up the coating system to pilot and industrial scales. The laboratory parameters, e.g. the amount of electricity used to produce the coating and the solution volumes, were optimized using literature and expert consultation. The comparative analysis showed that the new coating is competitive to the conventional coatings, and this could be due to the simplicity of the coating method applied by TNO. Also, it was revealed that the coatings do not bring significant environmental benefits rather they bring economic benefits in terms of increased yield of tomatoes. Finally, the sensitivity analysis showed that electricity used for the production of glass has higher impacts than transmittance or degradation time of the coatings.

TH315

Combine process simulation analysis with Life Cycle Assessment method in polyurethane rigid foam production

A. Bordignon, M. Fermeglia, Università di trieste; A. Bortoluzzi, S. Rondinini, C. Locatelli, A. Vertova, Università di Milano

Process simulation is a computational technique used in several sectors of process engineering. It is very useful at design stage, for defining the feasibility of a new process, as well as in the process optimization stage, where the optimum value of the production is pursued and, last but not least, in the process analysis stage for understanding the potential improvement of an existing process. It consists in the complete simulation using algebraic and differential equations of the chemistry and the physics of the process as well as the material and energy balances of the unit operations involved. The Life Cycle Assessment is a methodology aiming at analysing the overall life cycle of products, processes or service. In this work, we will present the analysis of the complete life cycle of polyurethane rigid foam from cradle to grave in tight connection with process simulation methods, thus combining the characteristic independent variables of the production process, such as pressure drop, energy consumption and other, with impact assessment. This combination will allow us to identify the best solution for the production of polyurethane rigid foam both in terms of end of life scenery and environmental impact.

TH316

Life Cycle Assessment of CO2-based Methanol Production using Captured CO2 from Fossil Fuel Power Plants

C. Lee, University College London / Department of Chemical Engineering; R.

Catlow, Cardiff University / School of Chemistry; P. Lettieri, University College London / Chemical Engineering

As a way to address the climate–related CO_2 emissions from fossil fuel power plants, photocatalytic methanol production using a novel form of CO_2 conversion process has been investigated within the research project "Low Carbon Fuel". The primary goal of this study is to evaluate the environmental performance of photocatalytic CO_2 conversion in comparison to conventional fossil-based technologies for power generation and methanol production. Life cycle assessment (LCA) is used to determine and compare the environmental performance of the methanol production systems. In the LCA study, cradle to gate system boundaries are used because the downstream processes and properties of methanol are similar for CO_2-based and fossil-based systems. Since the main environmental motivations for carbon utilisation are reducing CO_2 emissions and establishing an alternative carbon source, this study compares the CO_2-based and fossil-based methanol production systems with respect to global warming and fossil resource depletion. The CO_2-based methanol production system consists of the following three stages: CO_2 source including CO_2 capture, electricity compensation, and CO_2 utilisation for methanol production. The fossil-based methanol production system serves as benchmark and is divided into electricity generation and fossil-based methanol production. The main functions of the CO_2-based and fossil-based systems are production of methanol, and supply of electricity to the UK electricity grid. To quantify the main functions, we choose 14.3 MJ methanol (655 g) as reference for the function 'methanol production'. The second function 'electricity supply' can be quantified through the amount of CO_2 that is captured to produce equivalent amount of methanol. CO_2-based and fossil-based methanol production processes are analysed and compared based on life cycle assessment. Our analysis reveals that CO_2-based methanol production system using photocatalytic CO_2 conversion is not always mandatory to achieve CO_2-based system with lower environmental impacts than the fossil-based system. However, CO_2-based methanol production has the potential to reduce impacts for global warming and fossil depletion if the environmental performance of intermediate reaction and steps are decreased, compared to the corresponding fossil-based route. Furthermore, additional environmental benefits can be obtained from environmentally friendly hydrogen production from photocatalytic water splitting process.

Advancing the Adverse Outcome Pathway Framework - An International Horizon Scanning Approach (P)

TH317

Linking failed swim bladder inflation of larval Japanese medaka (Oryzias latipes) after embryonic exposure to 17α-ethinylestradiol, levonorgestrel and diclofenac, to disrupted βcatenin/Wnt signaling

Z.P. Pandelides, University of Ontario Institute of Technology; M. Overturf, University of Louisiana at Monroe / Biology; E. Ussery, University of Ontario Institute of Technology / Biological Sciences; J. Guchardi, University of Ontario Institute of Technology / Faculty of Science Aquatic Toxicology; D.A. Holdway, University of Ontario Inst. of Tech / Science

Embryotoxicity testing is a high throughput alternative to using the whole fish model. Previous studies in our laboratory have demonstrated that embryonic exposure to pharmaceutical compounds capable of disrupting the endocrine system such as 17α-ethinylestradiol, levonorgestrel, and diclofenac, both alone and in mixtures, can impair swim bladder inflation of Japanese medaka (*Oryzias latipes*). Failure of swim bladder inflation can have serious long-term effects on fish populations and has been correlated with larval medaka mortality. How embryonic exposure to xenobiotic compounds are able to cause swim bladder inflation is not fully understood; however, it is possible that compounds are able to cause their effects through a disruption of embryonic cell signalling pathways. The canonical Wnt pathway plays a crucial role in fish swim bladder development, and the disruption of Wnt signalling during swim bladder organogenesis could lead to improper swim bladder formation. The effects of two Wnt modulators IWR-1 and lithium exposed from 36-101 hours post fertilization on gene expression of medaka embryos was determined. The effects of embryonic exposure to the three pharmaceutical compounds on whole embryo gene expression were then established and compared. It was measured that these pharmaceutical compounds significantly inhibited the expression of genes related to the formation of the three layers of the swim bladder (epithelial, mesenchyme, and outer mesothelium). Both levonorgestrel and 17α-ethinylestradiol also significantly downregulated the expression of *lef1* and *tcf7l1* (β-catenin/Wnt transcription factors), but not the expression of the Wnt ligand *wnt5b*. Thus demonstrating that these compounds may be altering swim bladder inflation through a disruption of β-catenin/Wnt cell signalling during early embryo development.

TH318

Linking mode of action of the model respiratory and photosynthesis uncoupler 3,5-dichlorophenol to adverse outcomes in Lemna minor

L. Xie, NIVA - Norwegian Institute for Water Research; T. Gomes, Norwegian Institute for Water Research (NIVA) / Ecotoxicology and Risk Assessment; K.A. Solhaug, Norwegian University of Life Sciences; Y. Song, NIVA Norwegian Institute for Water Research / Ecotoxicology and Risk Management; J. Moe, NIVA Norwegian Institute for Water Research; K. Tollefsen, NIVA / Ecotoxicology and Risk Assessment

Lemna minor is an aquatic plant commonly used in laboratory phytotoxicity testing due to its rapid reproduction capacity, resource-effective exposure format and central function in the aquatic ecosystem. Several standard methods have already been adopted by international standardization organisations using this species as an ecotoxicological model. Although being highly useful for regulatory purposes focusing on traditional adverse endpoints, these tests provide limited information about the toxic mechanisms and modes of action (MoA) and rarely address complex environmental issues such as exposure to multiple stressors. The present study aimed to use selected functional assays in *L. minor* after exposure to 3,5-dichlorophenol (3,5-DCP) as a model to characterize the toxic mechanisms causing growth inhibition and lethality in primary producers. The results demonstrated that 3,5-DCP caused concentration-dependent effects in chloroplast and mitochondria. Endpoints such as uncoupling of oxidative phosphorylation (OXPHOS), chlorophylls content, reproduction rate and frond size are more sensitive to 3,5-DCP compared to other responses as well as reactive oxygen species (ROS) formation, lipid peroxidation (LPO) and impairment of photosynthesis efficiency. Principal component analysis (PCA) indicated that suppression of photosystem II (PS II) efficiency, electron transport rate (ETR), ROS production and LPO, pigments content and growth were strongly correlated while inhibition of oxidative phosphorylation (OXPHOS) which was more closely correlated with growth parameters. A set of conceptual Adverse Outcome Pathways (AOPs) were developed by using Bayesian network model to decipher the causal relationships between molecular, cellular, and apical adverse effects occurring in *L. minor* to form a basis for future studies with similar compounds.

Acknowledgements: This research was supported by Norwegian Research Council through its Centre of Excellence (CoE) funding scheme (Project No. 223268/F50).

TH319

Development of adverse outcome pathways for oxidative stressor-mediated reproductive effects in aquatic invertebrates

Y. Song, NIVA Norwegian Institute for Water Research / Ecotoxicology and Risk Management; L. Xie, NIVA - Norwegian Institute for Water Research; Y. Lee, Norwegian University of Life Sciences; F. Lyne, Newcastle University; Y. Kassaye, D.A. Brede, Norwegian University of Life Sciences / Centre for Environmental Radioactivity; T. Gomes, Norwegian Institute for Water Research (NIVA) / Ecotoxicology and Risk Assessment; G. Caldwell, Newcastle University; K. Tollefsen, Norwegian Institute for Water Research

Oxidative stress is a common type of stress in living organisms and induced when the production of reactive oxygen species (ROS) overwhelms the endogenous antioxidant defenses. Essential biological macromolecules such as DNA, protein and lipids can be damaged by ROS through oxidation and peroxidation, thus leading to diverse types of adverse effects, such as growth arrest, developmental abnormalities, reproductive failure and lethality. A wide range of oxidative stressors such as ionizing radiation, ultraviolet (UV) radiation, metals and organics are known to induce oxidative stress. The consequences of oxidative stress have been well studied in vertebrates, especially mammlian species. However, for other ecologically relevant environmental species, such as aquatic invertebrates, the knowledge is still limited. The present study was therefore conducted to: develop AOPs for ROS-mediated reproductive effects in aquatic invertebrates based on existing data from the literature; assess the weight of evidence (WoE) of the AOPs based on a combination of literature survey and *in silico* predictions; evaluate the key events in the AOPs experimentally using the forecaster species *Daphnia magna*. An extensive litreature survey to collect existing knowledge on ROS-mediated reproductive effects in aquatic invertebrates, and metals, ionizing and non-ionizing radiations as prototypical stressors. A network of conceptual AOP was assembled first and a linear AOP with the strongest data support was then selected prior to WoE assessment using the Evolved Bradford Hill considerations. The laboratory evaluations of the AOPs were performed using UVB and gamma radiations as prototypical oxidative stressors. *Daphnia magna* was used as the model species. The laboratory studies clearly showed that both UVB and gamma radiation caused dose-dependent reduction in reproduction after the exposure. The reproductive effects were correlated with excessive ROS production, lipid peroxidation, DNA damage, apoptosis, reduced mitochondrial membrane potential and lipid storage, and abnormal ovary structures and oogenesis in *D. magna*, thus verifying several KEs in the conceptual AOPs. This study has for the first time systematically linked excessive ROS production to reproductive effects in aquatic invertebrates using the AOP concept, thus providing mechanistic knowledge for future hazard and risk assessment of oxidative stressor-mediated adverse effects in ecologically relevant species.

TH320

Development of an Adverse Outcome Pathway for cardiotoxicity mediated by the blockade of L-type calcium channels

L. Margiotta-Casaluci, H. Dusza, I. Moreira, Brunel University London / Institute of Environment, Health and Societies; M.J. Winter, The University of Exeter / College of Life and Environmental Sciences; H. Prior, National Centre for the Replacement, Refinement and Reduction of Animals in Research (NC3Rs)

A diverse set of chemical compounds, including some pharmaceuticals and insecticides, have the potential to perturbate the functionality of calcium channels. Among the different types of calcium channels, the L-type calcium channel (LTCC) is responsible for the excitation-contraction coupling of skeletal, smooth, and cardiac muscle. Chemicals that unintentionally block this channel in cardiac cells may impair heart function and health, leading to various cardiac pathologies and predisposing individuals to heart failure. Advancing our understanding of the mechanisms underlying those adverse effects is of paramount importance if we want to develop effective strategies able to accurately predict the cardiac risk posed by chronic exposure to those chemicals. In this presentation, we describe the development of an Adverse Outcome Pathway (AOP) that outlines the series of causally related key events triggered by the blockade of LTCC, and that can ultimately lead to cardiac adverse effects. We discuss the integration of *in silico*, *in vitro*, and *in vivo* evidence to support the AOP development, as well as the application of computational and network biology approaches that may accelerate the identification of relevant key events. Considering the multifaceted role of LTCC in different components of the cardiovascular system other than the heart, we also discuss the importance of applying AOP network considerations to guide a reliable and fit-for-purpose AOP development. This AOP will represent a valuable knowledge base able to guide the identification of key events that are highly predictive of *in vivo* toxicity, and that can be measured *in vitro* without relying on animal testing. The knowledge base will also be used as platform to drive future development projects aimed at incorporating additional layers of complexity in the model, and at driving the transition towards a fully quantitative AOP able to effectively support regulatory decision-making and risk assessment.

TH321

Quantification of AOP by Bayesian network modelling: linking 3,5-DCP exposure to adverse outcomes in Lemna minor

J. Moe, Norwegian Institute for Water Research (NIVA) / Section for Catchment Processes; W.G. Landis, Western Washington University / Institute of Environmental Toxicology; L. Xie, NIVA - Norwegian Institute for Water Research; K. Tollefsen, NIVA / Ecotoxicology and Risk Assessment

AOPs has gained international recognition as a systematic approach for capturing existing toxicological knowledge to transparently link mechanistic data to toxicity endpoints. Nevertheless, most AOPs are qualitative and not directly suitable for quantitative risk assessment. Quantitative AOPs (qAOP) should define the relationships underlying transition from one KE to the next sufficiently well to allow quantitative prediction of the probability or severity of the AO occurring for a given activation of the MIE. We have started developing a Bayesian Network (BN) model to quantify a recently proposed AOP, which links the mode of action of the model respiratory and photosynthesis uncoupler 3,5-dichlorophenol (DCP) to adverse outcomes in the aquatic plant *Lemna minor*. The BN model is based on data from a laboratory experiment exposing *L. minor* to DCP in 8 concentrations with 3 replicates. The measured response variables include OXPHOS (oxidative phosphorylation), ROS (reactive oxygen species), ETR (electron transfer rate), Fv/Fm (maximum quantum yield of photosystem II), LPO (lipid peroxidation) and number of fronds (leaves). The proposed AOPis a network consisting of three chains with the same chemical stressor (DCP) and AO (fronds number). All AOP components are defined in the BN as nodes with discrete states. Each node is quantified by a probability distribution across these states. The causal links (Key Event Relationships) are quantified as conditional probability tables (CPTs), which determine the probability distribution of a child node conditionally on the probability distribution of the parent node(s). The CPTs are calculated directly from the data in this BN version. The BN was run by changing DCP concentration and inspecting the changes in all subsequent nodes. Qualitatively, the model predictions were as expected: increasing the DCP concentration caused reduced OXPHOS, reduced ETR and reduced fronds number. For example, when DCP was increased from 1 to 2 mg/L, the probability of fronds number being in the lowest (worst) state increased from 6% to 30%. Fine-tuning of the intervals of some nodes is needed to make the BN more responsive. Moreover, we will use statistical approaches to obtain more credible CPTs, such as estimation of dose-response curves with uncertainty. Other planned developments include linking the AO to an endpoint with regulatory relevance, and linking the chemical stressor to an Aggregate Exposure Pathway.

TH322

Development of Quantitative Adverse Outcome Pathway (AOP) of Pulmonary Fibrosis with Effectopedia

J. Jeong, University of Seoul; N. Chatterjee, University of Seoul / Environmental Engineering; S. Choi, Unviersity of Seoul / Envirionmental Engineering; J. Choi, University of Seoul / School of Environmental Engineering

Pulmonary fibrosis (PF) is a chronic and progressive lung disease where the scars are formed in the lung tissues and the air sac in the lungs (alveoli) becomes stiff leading to serious breathing problems. Several substances are identified as inducer of PF, but high cost of inhalation toxicity studies refrain to conduct systemic studies of all those substances. Hence, the regulations of these substances become obscure. To solve this problem, Adverse Outcome Pathway (AOP) concept has been emerged. AOP is a framework that organizes existing knowledge about linkage between molecular-level perturbation and an adverse outcome. To facilitate the development of AOP, OECD launches AOP knowledge-base (KB). In recent years, the application of quantitative AOP (qAOP) which provide dose-response and time-course prediction, has been gaining much more attention in regulatory decision-making field. To develop the AOP of pulmonary fibrosis, in one hand, we made preliminary AOP from literatures, which constitutes the PPARγ interaction as Molecular Initiation Event (MIE), Collagen activation, Inflammation and EMT-Fibrosis activation as Key Events (KEs), and Cytotoxicity/Apoptosis and Fibrosis as Adverse Outcome (AO). On the other hand to make qAOP, we conducted cytotoxicity and apoptosis test using human bronchial epithelium cell (Beas2B). Beas2B cell was exposed to CMIT/MIT (a biocide which possess potential risk to respiratory systems) at various doses from 0 mg/L to 2 mg/L for various time for 1 to 72 hr. Cytotoxicity and apoptosis was analyzed using various available assays at mid to high through put condition. While, quantitative analysis between the KEs was performed using Effectopedia platform of AOP-KB. For further study, we are planning to do various dose- and time response test (using qPCR and ELISA) for each potential KEs, so that we can integrate data for building qAOP model with the network between MIE-KEs-AO. Acknowledgement: This work was supported by a grant from the Korean Ministry of Environment through 'Environmental Health R&D Program' (2017001370001).

TH323

Exploring Potential of Knowledge Databases for Adverse Outcome Pathway Discovery

C. Lai, University of St. Thomas / School of Engieenering; Y. He, University of St. Thomas / School of Engineering; D. Martinovic-Weigelt, University of St. Thomas / Biology

Adverse outcome pathways (AOPs) have potential to support and enhance the use of mechanistic data in regulatory decision-making. AOPs organize existing knowledge about relationships (ideally causal ones) between initial chemical-biological interactions (molecular initiating events; MIEs), intermediary key events (KEs), and adverse outcomes (AOs) relevant to risk assessment. Efficient ways of AOP development and weight of evidence assembly are lacking. This study evaluated potential of the existing knowledge databases (Unified Medical Language System - UMLS, and National Library of Medicine – NLM) for AOP discovery and development. UMLS contains more than 68-million relationships among more than 3-million unique biomedical concepts (or terms). The NLM literature database contains more than 100-million relationships among similar 3-million biomedical concepts extracted from the abstracts of more than 16 million biomedical journal papers. First, AOP network was downloaded and parsed from AOP Wiki (https://aopwiki.org/). We found that there are 3,084 relationships among stressors, MIEs (main initial events), KEs (key events), AOs (adverse outcome), stressor-chemicals, and stressor-events. High performance graphic processing unit (GPU) was used to determine which of 3,084 relationships can be found in hundred million of relationships in UMLS and NLM databases. 610 (20%) relationships were found in the UMLS database. About 1,837 (60%) relationships were found in the abstracts of 16 million biomedical papers on NLM. When combining our searches over both the UMLS and NLM databases, 1,983 (64%) relationships from AOP wiki were found; relationships in some sub-categories such as stressor-chemicals had much higher hit ratio - 78%. These findings indicate that AOP-discovery system that uses UMLS and NLM to predict new probable AOP relationships (that can connect to objects in the AOP Wiki) could substantially accelerate AOP development and contribute to weight of evidence analyses. The confidence of the predicted relationships could be calculated based on frequency of the relationships, whereas the quality of the predicted relationships could be further improved by training deep learning models with knowledge curated in databases such as The Comparative Toxicogenomics Database, ECOTOX and iCSS ToxCast Dashboard**.**

Poster Corner Abstracts

Fish model species in human and environmental toxicology (PC)

MOPC01

Fish caging experiment as a tool for in situ assessment of neurotoxic effects of untreated wastewaters

B. Mićić, Petnica Science Center/Faculty of Sciences, University of Novi Sad / Department of Biology and Ecology; D. Tenji, University of Novi Sad Faculty of Sciences / Biology and Ecology; S. Sipos, Faculty of Sciences University of Novi Sad / Biology and Ecology; V. Knezevic, Faculty of Sciences / Department of Biology and Ecology Laboratory of Ecotoxicology LECOTOX; S. Kaišarević, Faculty of Sciences University of Novi Sad / Department of Biology and Ecology, Laboratory of Ecotoxicology (LECOTOX); I. Teodorovic, University of Novi Sad / Department of Biology and Ecology, Laboratory of Ecotoxicology (LECOTOX)

In the framework of FP7 project Solutions, the city of Novi Sad has been selected as a pollution hot spot of the River Danube, mainly due to the direct discharge of untreated sewage into the river. Significant biological effects of untreated sewage were previously observed through an *in vitro* study. To study neurotoxic effects more in details and in realistic ecological context, we conducted fish caging experiment. Common carp (*Cyprinus carpio* (L.), Cyprinidae) has been selected for the experiment, as one of the most common species in the Middle Danube, genetically well described and economically important. Ten fish per cage were exposed in cages for nine days at three sites on the Danube River: upstream from sewage discharge – reference site, 230 m and 7 km downstream from the discharge and at one site on Sava River (downstream from industrial wastewater discharge near the city of Šabac). After the exposure period, brains were isolated and prepared for gene expression and enzyme activity analyses. The expression of genes encoding for five proteins was studied: tachykinin 3a and tachykinin 3b (involved in neuroendocrine regulation of reproduction), GABA a1 receptor (receptor for the inhibitory neurotransmitter GABA and various drugs), synaptotagmin 10 (integral membrane protein of synaptic vesicles with a role in exocytosis) and myelin basic protein (responsible for myelination of axons and neuroprotection). The activity of acetylcholine esterase, enzyme that terminates action potential transmission in chemical synapses of cholinergic type, was also examined. A trend of slight upregulation for the expression of the genes encoding for tachykinin 3a and tachykinin 3b, GABA a1 receptor and synaptotagmin 10 was observed for all three studied sites when compared to the reference site. The expression of the gene encoding for myelin basic protein was similar at reference site and 230 m downstream from the sewage discharge, but this gene expression was significantly downregulated downstream from the industrial wastewater discharges. Based on this result, myelin basic protein might be potential selective biomarker which can be used to differentiate the effects of these two types of chemical pressure. No significant difference was observed in the activity of the acetylcholine esterase between studied sites. The study is part of the SOLUTIONS project, funded by the EU FP 7 (FP7-ENV-2013-two-stage Collaborative project) under grant agreement number 603437.

MOPC02

Toxicity analysis of treated sugar cane vinasse by integrated systems using gills of Oreochromis niloticus as model

A. Marcato, Sao Paulo State University - UNESP / Department of Biology; C.P. de Souza, Sao Paulo State University - UNESP / Biology; J. Evangelista Correia, Unesp - Institute of Biology / Biology; C.S. Fontanetti, Sao Paulo State University - UNESP / Biology

The alcoholic fermentation of sugar cane (*Saccharum* sp.) results in a by-product known as vinasse. This by-product is used as fertilizer because of its richness in organic matter, and also because it promotes improvement in soil fertility, favoring the availability of some elements for the plants. However, the amount of vinasse used in the fertirrigation should not overcome the ion retention capacity of the soil, since the dosages should be directed to the specific characteristics of each soil. When used in unbalanced proportions can impair to the soils and the plants, in addition to being able to reach water resources. Considering studies that prove the toxicity of vinasse *in natura*, the use of treatment systems has become quite interesting. The integration of systems such as natural attenuation, filtration and phytoremediation increase the effectiveness of the treatment, since they are highly effective biogeochemical systems to treat waste water from different sources. Aquatic macrophytes, which not only accumulate pollutants directly in their tissues but also act as catalysts for purification reactions that usually, occur in the rhizosphere of plants, they are part of the alternative treatment for vinasse. Fish are excellent experimental models for aquatic toxicity studies because they warn of the potential danger of chemicals reaching water resources. Therefore, this study aimed to verify the efficacy of sugar cane vinasse treatment in reducing its toxic potential by histological and histochemical tests on tilapia gills. The animals were submitted to two different dilutions of the treated vinasse for 96 hours; after this period the gills were removed and submitted to standard histological routine. Morphological analyzes of the gills revealed that the cellular pattern described for the species was not altered and histochemical tests showed a decrease on number of mucous cells, thus attesting to the decrease on toxicity of the treated vinasse. Thus, it can be inferred that integrated treatment systems were effective in reducing the polluting potential of vinasse, since the animals did not present histological changes.

MOPC03

Assessing toxic effects in the fish Violet Goby (Gobioides broussonnetii - Gobiidae) from one of the most productive estuaries in Brazil.

L. Salgado, Universidade Federal do Paraná / Farmacologia; A.M. Marques, UFPR / Genetics; F. Garrido de Oliveira, UFPR / Pharmacology; S.L. Moretto, M.M. Cestari, UFPR / Genetics; H. Silva de Assis, UFPR / Pharmacology

The Estuarine-Lagoon Complex of Iguape-Cananéia (São Paulo, Southeast Brazil) is among the most productive areas in the South Atlantic. The Ribeira de Iguape River (RIR) is the major freshwater contributor of the estuary. It carries different classes of contaminants from former mining activities, agricultural areas and urban centers through an artificial channel. The disordered human occupation, presence of boats and the disposal of waste and sewage are also sources of pollution throughout this system. The Violet Goby (*Gobioides broussonnetii* - Gobiidae) is a demersal fish of a social and economic importance to traditional fisheries. Over the last decade the regional disappearance of this fish species has been reported, including events of high mortality. The contamination by the RIR has been referred as one of the causes of the decline of that population. Therefore, this study aimed to observe possible toxic effects in *G. broussonnetii*in the studied area. Fishes were sampled near Cananéia, Subaúma and Iguape in winter (2016) and summer (2017). The animals were anesthetized, euthanized and the blood, brain, muscle, liver and kidney were collected for the biochemical and genetic biomarkers analysis. The muscle AChE activities showed similar results among the points and seasons, while cerebral AChE were lower in Subaúma in winter. Hepatic and renal GPx and GST activities, GSH concentrations and LPO damage in liver were similar. However, it was observed genotoxicity in Cananéia and Subaúma points in the liver and blood in summer. In winter, nuclear morphology alterations were identified in erythrocytes more frequently in fishes of Cananéia. The results suggest that contaminants such as metals and HPAs previously reported in the sediments may have been stressing this species. The marked seasonality of the region, which consequently influences the temperature, the rainfall regime and the bioavailability of contaminants may interfere in these responses. No expressive anthropic activity was observed near to Cananéia and Subaúma points. It suggests an influence of the local hydrodynamics by dragging the contaminants of the main sources (RIR and Cananéia city) to these areas once lower impacts were seen in Iguape. This last point is located above the artificial channel in an area of low hydrodynamic. Water and sediment chemical analyzes are being performed in the studied points in order to support a better understanding of these responses.

MOPC04

Does ozonation of the Aachen-Soers WWTP improve the water quality in the field? Caging experiments with juvenile rainbow trout and various biomarkers

Y. Mueller, RWTH Aachen University / Department of Ecosystem Analysis ESA; T. Rosenberger, RWTH Aachen University / Institute for Environmental Research BioV; S. Schiwy, Institute of Environmental Research-RWTH Aachen / Department of Ecosystem Analysis; H. Hollert, RWTH Aachen University / Institute for Environmental Research

The European Water Framework Directive aimed to achieve a good chemical and biological state of all surface waters until 2015. However, the good biological state is only reached by around 20 % of the German surface water bodies. One reason might be the release of a variety of anthropogenic contaminants by waste water treatment plants (WWTPs) into these water bodies. Additionally, these substances are not sufficiently eliminated via conventional waste water cleaning processes. Nevertheless, these chemicals can have adverse effects on the river biota. The implementation of a further treatment step into WWTP could reduce this burden. There are advanced treatment processes, as the ozonation. At the “Aachen Soers” WWTP a large-scale ozonation plant was installed at the end of 2017. Regular operation will start by approximately March 2018. The “Aachen Soers” WWTP is located near the city of Aachen (North Rhine-Westphalia, Germany) and releases its effluent to the Stream Wurm. At medium and low water levels the stream runs around 70 % treated waste water. To elucidate the impact of the additional waste water treatment on this river the *status quo* was recorded before the implementation of this treatment step. After the installation of the ozonation the WWTP as well as the Wurm will be monitored for two years. Beside numerous *in vitro* and *in vivo* experiments also *in situ* experiments were conducted. Caging experiments with juvenile rainbow trout (*Oncorhynchus mykiss*) were performed upstream and downstream the WWTP. The goal was to evaluate the impact of the WWTP outlet on the river. Further, the impact of the upstream burden was part of the study. Several biomarkers were investigated on different organs of the fish. Detoxification enzymes were investigated in liver tissue and acetylcholinesterase was measured in brain tissue. Furthermore, micronuclei formations counted in blood smears to get information on genotoxic effects. To gain information on endocrine effects Vitellogenin levels were measured in blood plasma and mucus to compare the conventional invasive method with a new non-invasive method. Fish were caged during late summer in 2017 to evaluate the *status quo* of the stream and the performance of the "Aachen Soers" WWTP. This project is funded by the Ministry for Climate Protection, Environment, Agriculture, Conservation and Consumer Protection of North Rhine-Westphalia.

MOPC05

Environmental applications for medium-throughput, in vivo androgen disruptor identification with the RADAR assay

A. Tindall, A. Phan, N. Roxane, Watchfrog S.A.; B.A. Demeneix, MNHN / CNRS UMR 7221; G.F. Lemkine, Watchfrog S.A.

Over recent years, it has become evident that environmental contamination with endocrine disruptors is not limited to those acting on the estrogen axis. In contrast, large numbers of chemicals, in particular pesticides, have been identified with *in vitro* models. Two key studies identified 66/200 and 37/134 pesticides tested as anti-androgenic. However, due to the absence of medium-throughput *in vivo* assays for androgen axis disruption, the effects of many of these pesticides have yet to be confirmed *in vivo*. We developed a transgenic medaka line harbouring a portion of the *spiggin1* gene promoter driving expression of GFP. We have previously demonstrated that this line is capable of correctly identifying androgens and anti-androgens, including pesticides, with similar sensitivity to the androgenised female stickleback screen but in a greatly reduced time frame. Using eleuthero-embryonic life stages we developed the Rapid Androgen Disruption Adverse outcome Reporter (RADAR) assay. Extracts of Danube River water from sites upstream and downstream from a major effluent stream from the city of Novi Sad in Serbia was analysed. Comparison of our results to previously published data from four *in vitro* assays carried out on the same extracts indicated that the effect observed *in vivo* was two orders of magnitude higher than the *in vitro effect*, suggesting additional mechanism(s) of action present *in vivo* in addition to androgen receptor activation indicated by the *in vitro* assays. Application of the RADAR assay for effect-directed analysis was demonstrated for the rapid *in* vivo confirmation of the activity of coumarins 47. A number of pesticides originally identified by screening with in vitro models were also tested. The anti-androgenic effects of these pesticides had not previously been confirmed in vivo to our knowledge. Powerful anti-androgenicity was observed with the RADAR assay for some of the tested pesticides, confirming the results of the the in vitro study. The RADAR assay is a reliable medium-throughput tool which can be applied within a variety of environmental scenarios in order to identify androgen axis disruption, such as environmental monitoring, identification of unknown toxicity drivers and testing of pure chemicals in a REACH context. In addition, this model, based on the use of early life stages non-compliant with the EU definition of a laboratory animal, provides ethical advantages in line with the three R's principle of animal replacement.

MOPC06

Evaluation of the toxicity of environmental samples collected near vineyard parcels on rainbow trout larvae (Oncorhynchus mykiss) and liver cell line RTL-W1

S.L. Weeks Santos, EPOC University of Bordeaux; P. Gonzalez, University of Bordeaux / UMR EPOC CNRS 5805; J. Groussin, EPOC University of Bordeaux / UMR EPOC; Q. Papin, University of Bordeaux / UMR EPOC; C. Clérandeau, EPOC University of Bordeaux / EPOC UMR; B. Morin, University of Bordeaux / EPOC; B. Cormier, Université of Bordeaux / EPOC UMR; P. Gourves, University of Bordeaux / UMR EPOC CNRS 5805; J. Cachot, University of Bordeaux / EPOC

Viticulture is one of the agricultural domain that uses the most pesticides. Aquatic ecosystems are usually the final receptacle of all pollutions by leaching, deposition or infiltration; but, because of its capacity of accumulation, sediments represent a reservoir of contaminants larger than the water column itself. The aim of this work was to assess the toxicity of environmental samples, waterborne and sediments, from La Livenne river in hepatic cell lines (RTL-W1) and rainbow trout (*Oncorhynchus mykiss*) larvae. Samplings were done in La Livenne's watershed near Bordeaux (Southwestern France), a region with a strong presence of vineyards, over three campaigns in February, May and August 2017. Waterborne and sediment samples were collected in 4 sites from La Livenne (Menanteau, Parodier, Grand Village and Vignolles) and one site Reguignon from Les Souches (a small stream highly impacted by viticulture activity that rejects in La Livenne). Pollutants from 1 L water column had been extracted by SPE (Solid Phase Extraction) method, and from sediments had been extracted by elutriates. In the first part of the study, RTL-W1 cells were exposed separately to extracts of water and sediment samples from the three campaigns and different toxicity tests were performed as cytotoxicity (MTT test) and ROS (Reactive Oxygen Species) induction. In the second part of the study, newly hatched larvae were exposed for 48h to both water and sediment samples collected in May (during spreading season). Different toxicity criteria as viability, biometry and genotoxicity were studied. Waterborne extractions from Grand Village, Vignolles and Reguignon were cytotoxic but at 10 or 20 times the environmental concentration. No ROS production was observed. In the other hand, cells exposed to sediment elutriates were able to induce ROS; but no cytotoxicity was observed. In the case of rainbow trout larvae, no mortality was observed after 48h of exposure. However, differences in biometry was observed on larvae exposed to Menanteau, Grand Village and Reguignon when compared to non-exposed larvae. In particular, the head size was significantly smaller than control larvae, and yolk sac area was bigger in exposed larvae when compared to control larvae. Our study demonstrated that environmental samples of water and sediments collected close to vineyards are toxic in *in vitro* and *in vivo* assays on rainbow trout.

New Horizons in Particulate Polymer Analysis: Micro- and Nanoplastics and Tire Rubber Detection, Characterisation and Impacts in the Environment (PC)

MOPC07

Optimization and Automation of Raman Microspectroscopy for Microplastic Analysis

P.M. Anger, Technical University of Munich / Chemistry Department, Chair of Analytical and Water Chemistry; L. Prechtl, Technical University of Munich / Institute of Hydrochemistry; R. Niessner, Technische Universität München / Chemistry Department, Chair of Analytical and Water Chemistry; M. Elsner, N.P. Ivleva, Technical University of Munich / Chemistry Department, Chair of Analytical and Water Chemistry

On the one hand, plastics are a most important part of our daily life. Due to their versatile properties, especially their low weight, formability and their low costs they are an ideal packaging material.[1] On the other hand, microplastics (MP) disposed into the environment are increasingly recognized as a problem that needs to be addressed. Raman microspectroscopy (RM) is a versatile tool for MP analytics.[2] Optimization and automation of RM measurements as well as automation of spectral data evaluation are of high importance for monitoring programs and enable quantitative results. We advanced RM-based analysis by optimizing measurement parameters, measurement automatization and automated spectral data evaluation. First, a filter holder was constructed that flattens the filter surface. This filter holder is superior compared to filters deposited on, or glued to, glass slides. However, a flat filter surface is only a first step for successful measurements. For an automated particle recognition we also optimized the contrast between particles and filter. To this end, a variety of polymers (e.g. PE, PP, PS, etc.) of different sizes (10^0 – 10^3 µm) and forms (spheres and irregular shapes) were analyzed on different filter materials (e.g. PC, gold coated PC, nitrocellulose, etc.) under different modes of illumination (bright and dark field, fluorescence mode). We found that reflecting filter materials combined with dark field illumination yield superior contrast. Finally, we tested these optimized parameters with samples of different complexity (incl. environmental samples) and three RM methods (non-, semi- and automated in regard of particle recognition and Raman measurements). Samples with a low loading are accesible via an automated approach, whereas the analysis of more loaded samples is better done with manual particle recognition.[3] The results brought forward in this work aim to catalyse advances towards automated methods for a better assessment of environmental risks arising from MP. In the future, the challenges lie in developing automated methods for various samples, especially for complex samples resulting from sediments. [1] A.L. Andrady, Mar.Poll.Bull., 2011, 62, 1596-1605 [2] N.P. Ivleva, et al., Angew.Chem.Int.Ed., 2017, 56, 1720-1739 [3] P.M. Anger, et al., *in prep.* *Acknowledgement* - The authors thank the German Federal Ministry of Education and Research for funding of the project MiWa and the Raman microspectroscope *alpha 300R* (WITec GmbH).

MOPC08

Preparation of model small microplastics and nanoplastics

G. Balakrishnan Nair, T. Nicolai, C. Chassenieux, IMMM LE MANS / Institute of Materials and Molecules of Le Mans IMMM UMR CNRS; f. lagarde, Institute of molecules and materials of Le Mans / Institute of Materials and Molecules of Le Mans IMMM UMR CNRS

Pollution with plastic debris and plastic fragments has recently been recognized as a major water quality problem in fresh and marine water systems. Degradation of plastic debris in the marine environment leads to the formation of microplastics (< 5mm) and potentially nanoplastics (< 1 µm). Recent investigations show that the microplastics can interact with the marine biota. The impact of the interaction on the exposed organism depends on the nature and size of the particles. To acquire more knowledge on these impacts and to optimize analytical procedures, model particles of different sizes and nature of polymers are necessary. However, in the smallest range (< 10 µm), particles of only a few types of polymers are currently available. For this reason, most toxicity tests were realised using PS beads whereas polyethylene (PE) and polypropylene (PP) fragments are by far the most common in the aquatic environments. In addition, commercial micro and nanobeads have surfactants at their surface which may enhance their toxicitys. Here, we present a simple methodology that allows one to prepare small microparticles of PE with sizes between 0.7 µm to 3 µm. These particles were obtained by dissolving PE pellets in toluene at high temperatures that was emulsified in water by ultrasonication. After removal of the solvents particles were recovered as powders that could be re-dispersed in water. Besides providing particles of small size, the advantage of the presented methodology is that it is possible to produce these particles without any surfactant. However, to obtain significant yields it is necessary to add a surfactant. Several types of surfactant were tested (Tween60, Tween80 and a biosurfactant obtained from an algae culture). The effect of each surfactant on the size, shape and stability of the particles will be discussed. These particles are currently used to optimize strategies of identification by Raman microspectroscopy for particles smaller than 1 µm.

MOPC09

Effects on humic substances and sediments on the sorption of anthropogenic chemicals to different MP particles

S. Huppertsberg, V. Zilles, Hochschule Fresenius University of Applied Sciences; T.P. Knepper, Hochschule Fresenius, University of Applied Sciences / Chemistry and Biology

Plastic products are nowadays omnipresent as they possess excellent characteristics as raw materials. An increased production and usage of plastic products in the last decades led to an emerging pollution of the environment. Slow or no biodegradability facilitates accumulation of plastic materials in the environment. Plastics can be found in various sizes in the environment, large items were found as well as small particles or debris, which are referred as microplastic (MP) in a size range from 5 mm to 1 µm. In aquatic environments organic pollutants may sorb to MP, which can act as vectors for the sorbed pollutants. It is assumed that polymer material, characteristics of the sorbate, embrittlement of MP-particles, biofouling, and presence of humic acids or sediments have, among others, an effect on the sorption. Humic substances are a complex mixture of breakdown products of biologic matter, representing about 50% of dissolved organic in surface waters. The composition and structure of humic substances are not yet fully elucidated due to their heterogeneity. Humic substances contain a high number of electron donors, which can interact with many natural and anthropogenic substances. Sediments consist of inorganic and organic components, which pose alternative sorption sites for solved substances. To investigate the effects of humic acids and sediments on the sorption of anthropogenic pollutants, sorption isotherms of i.e. galaxolide to polystyrene (PS) and other synthetic polymers were modelled in presence and absence of humic acids and sediments. A liquid-liquid extraction of the aqueous phase was performed to determine the equilibrium concentrations of galaxolide. A comparison of sorption isotherms regarding the influence of humic acids and sediments was performed to prove or disprove the following hypothesis. (I) Polymer material does not have an impact on sorption in presence of humic substances, due to the sorption of humic substances to the polymer surface. (II) Polymers do not contribute a significant amount to the overall sorption of organic pollutants as majority of them sorbs to sediment which is present in excess. The results of this study should provide a tool for predicting sorption behavior of MPs in environmental freshwater samples. Therefore, different parameters presumably affecting the sorption were investigated or are still under investigation to identify their impact on the sorption of organic pollutants in freshwater systems.

MOPC10

Micronized tire rubber: abundance and distribution within microplastic litter of the Charleston Harbor Estuary, South Carolina, USA

R. Leads, College of Charleston / Biology; J.E. Weinstein, The Citadel / Department of Biology

Microplastics (2are present on shorelines worldwide. A previous survey of Charleston Harbor, South Carolina, USA reported an average of 591±103 microplastic particles/m^2 in intertidal sediments, with black fragments suspected to be micronized tire rubber making up >90% of the particles at some sites. The objective of the present study was to further characterize the abundance and distribution of microplastics, and in particular, micronized tire rubber, in an effort to identify the sources and pathways into Charleston Harbor. As rivers are thought to be a contributor of non-point and point source microplastics, three major tributaries of Charleston Harbor—the Ashley River, Cooper River, and Wando River—were surveyed. Intertidal sediment (n=6), subtidal sediment (n=3), and sea surface microlayer (n=3) samples were collected from three sites (upstream, midstream, downstream) along each of the rivers and were analyzed for microplastics (63-500 µm). Intertidal sediment microplastic abundance ranged from 0-652 particles/m^2. Subtidal sediment microplastic abundance ranged from 3-4375 particles/kg wet weight. Sea surface microlayer microplastic abundance ranged from 3-36 particles/L. Microplastic abundance in intertidal sediments and subtidal sediments differed significantly among rivers ($p< 0.0001$ (intertidal), $p=0.02$ (subtidal)), while microplastic abundance in the sea surface microlayer did not differ significantly among rivers. Blue microplastic fibers and micronized tire rubber were the two most abundant types of microplastics observed, constituting 26.2% and 17.1%, respectively, of total microplastics collected. Furthermore, micronized tire rubber was collected at every sampling location and in every environmental sample type (intertidal sediment, subtidal sediment, sea surface microlayer). These results suggest that microplastics in Charleston Harbor originate primarily from non-point sources and that micronized tire rubber is a significant contributor of microplastic litter in Charleston Harbor. These results are the first to report on the abundance and distribution of micronized tire rubber as microplastic litter in a southeastern U.S. estuary and contribute to the understanding of the worldwide environmental presence of micronized tire rubber.

MOPC11

Crumb rubber in sports fields - Advances in environmental chemistry

D. Herzke, NILU - Norwegian Institute for Air Research / FRAM Centre Tromso; C. Halsband, Akvaplan-niva; L. Sørensen, A. Booth, SINTEF Ocean / Environmental Technology

In Norwegian coastal communities, rubber microplastic granules (≤ 5 mm in size) derived from discarded vehicle tires are used in large quantities on outdoor synthetic turf sports pitches. Through transport by waste water effluents and terrestrial runoff, these rubber particles are considered a significant source of MPs to the marine ecosystem. In the here presented interdisciplinary project we study the composition, degradation and environmental impacts of these rubber granules from locations in northern Norway and Svalbard. Their persistence and residence time in the Arctic marine environment is unknown. These rubber particles pose a potential health risk for arctic wild life through direct ingestion, especially at the base of the marine food chain, but may also provide an exposure route for toxic additive chemicals present in tires to marine organisms. Furthermore, the rubber particles may act as a vector for other persistent organic and heavy metal pollutants already present in the marine environment. Arctic marine environments present special abiotic conditions for the degradation of these particles, with cold water temperatures and long periods with unlimited sunlight. During a 12 months period, rubber crumbs were placed out in the ocean in stainless steel containers and sub-sampled continuously for the measurement of persistent organic pollutants, metals and additives. Hydrophobic persistent organic pollutants such as PAHs, PCBs, DDTs, bisphenols, as well as metals were measured to establish the adsorption and leaching kinetics in seawater under in situ conditions. Samples were extracted using ultrasound and nonpolar solvents, followed by GPC and SPE clean up. Chemical analyses using pyroGC/MS, GC/MS/MS and LC/HRMS were done in the laboratories of NILU, Tromsø and SINTEF, Trondheim. Exposure experiments with rubber leachate were also conducted and high mortality rates were found for different marine zooplankton species.

MOPC12

Nanoplastics analysis with Nano-FTIR

M. Meyns, Alfred Wegener Institute; S. Primpke, G. Gerdts, Alfred Wegener Institute / Shelf Sea System Ecology

The distribution of microplastic particles in marine environments and their ecotoxicological effects are matters of intensifying research. A significant amount of these particles is generated by degradation and fragmentation processes of larger marine litter. Although experimental proof is scarce, it can be assumed that fragmentation does not stop at the micro scale. This hypothesis is supported by results of our group that were obtained from various environments (e.g. North Sea sediments and arctic ice). A mathematical extrapolation of size dependent particle abundances in the samples returns abundances of up to 3.6×10^5 kg^{-1} for particles with diameters of 0.5 µm. This circumstance raises concerns as particlesviaspectroscopy. With imaging FTIR this is possible only down to particle sizes of ~10 µm. Electron microscopy suffers from sample instabilities and small aliquot sizes and does not provide the opportunity to simultaneously size and identify plastic samples. Nano-FTIR is a novel technique combining the nanoscale local resolution of AFM imaging with near-field infrared measurements resulting in unprecedented material differentiation on a nanometre level. In our proof-of-principle study, we show measurements with defined nanoscale polymers. To demonstrate the detection of nanoplastics in environmental matrices we analysed samples obtained from arctic sea ice. The Nano-FTIR technique is independent from the source of the sample so that it presents a universal tool for application in the analysis of nanoplastics samples from marine but also all other environments.

Environmental fate of emerging contaminants in the water cycle: analytical challenges and engineered solutions (PC)

MOPC17

Neonicotinoid insecticides in surface waters discharging into the Great Lakes of Southern Ontario, Canada

T. Sultana, Trent University / Environmental and Resource Studies; P.A. Helm, Ontario Ministry of Environment and Climate Change / Environmental Monitoring and Reporting Branch; C.D. Metcalfe, Trent University / Water Quality Centre

Neonicotinoid insecticides (NNIs) are a large part of the global pesticide market, as they are very effective at controlling a wide range of insect pests. In Canada, NNIs are used extensively in agricultural regions of southern Ontario to control insect pests on field and greenhouse crops, orchards, nurseries and woodlots etc. Because of their persistence and high solubility in water, there is potential for NNIs to be transported from agricultural fields into surface waters. The objective of this project was to evaluate the distribution of NNIs in surface waters located in areas of intensive agriculture in southern Ontario, Canada that discharge into the Great Lakes basin; specifically into Lake Erie, Lake Saint Clair, Lake Ontario and Lake Huron. Passive sampling with Polar Organic Chemical Integrative Samplers (POCIS) was the principal monitoring technique. To correct for the effect of environmental factors on the rates of uptake of the target NNIs into POCIS, Performance Reference Compounds (PRCs) were spiked into some of the POCIS deployed at each of the monitoring sites. POCIS were deployed for 2 weeks over two different deployment periods and grab samples of surface water were also collected three times over the deployment period. The sites monitored were located in 5 major rivers and 11 smaller creeks in the Great Lakes basin. Static, renewal experiments were conducted to estimate sampling rates of NNIs and to estimate elimination rates of PRCs These experiments were conducted using synthetic water at 15^{0}C over 14 days. Extracts were analyzed by liquid chromatography with tandem mass spectrometry (LC-MS/MS) using an AB Sciex QTrap 5500 instrument with electrospray ionization coupled with an Agilent 1100 HPLC. Nine NNIs were detected, but the concentrations varied widely from 2 ng/L to 4.6 µg/L. Flonicamid was detected at concentrations up to 1140 ng/L in watersheds discharging into Lake Ontario. Imidacloprid at concentrations up to 1731 ng/L and thiamethoxam at concentrations up to 625 ng/L were detected in the watersheds discharging into Lake Erie. Overall, these data indicate that NNIs are widely distributed in surface waters in agricultural regions in Ontario within the Great Lakes basin.

MOPC18

Occurrence and removal of antibiotics in municipal wastewater by conventional activated sludge (CAS) and membrane bioreactor (MBR) systems

N. Tran, National University of Singapore / NUS Environmental Research Institute; K. Gin, National University of Singapore / Civil & Environmental Engineering

This study provided the first and comprehensive data on the occurrence and removal of twenty-one target antibiotics and antimicrobials in a full-scale conventional activated sludge and membrane bioreactor systems in the Southeast Asian region. Nineteen out of the twenty-one target compounds were ubiquitously detected in raw influent samples. Concentrations of the detected ECs in raw influent samples ranged by several orders of magnitude (e.g. from 23.8 to 43,740 ng/L) depending upon the compound and sampling date. The elimination of antibiotics and antimicrobials in full-scale conventional activated sludge (CAS) and membrane bioreactor (MBR) systems at a local WWTP was evaluated and compared. Numerous antibiotics and antimicrobials, such as meropenem (MER), amoxicillin (AMX), ciprofloxacin (CIPX), clindamycin (CLI), azithromycin (AZT), clarithromycin (CLAR), oxytetracycline (OXY), triclosan (TCS), vancomycin (VCM), and chloramphenicol (CAP), were largely removed by both CAS and MBR systems. In contrast, trimethroprim (TMP), lincomycin (LIN) and erythromycin (ERY) appeared to be persistent in the both CAS and MBR systems. Field-based monitoring results showed that MBR outperformed CAS in the elimination of most target antibiotics and antimicrobials. The relationship between molecular characteristics of ECs (i.e. physicochemical properties and structural features) and their removal efficiencies during biological wastewater treatment was also elucidated. Excellent removal efficiencies (>90%) were often noted for compounds with the sole presence of electron donating groups (i.e. phenolic [–OH], beta-lactam ring, amine [–NH_2], methoxy[–O–CH_3], phenoxy [–O–C_6H_5], or alkyl groups). Conversely, antibiotics and antimicrobials with the absence of electron donating groups or the predominance of strong electron withdrawing groups (e.g. halogenated, carbonyl, carboxyl, sulfonamide, etc.) tended to show poor removal efficiencies (< 30%) in biological wastewater treatment processes.

MOPC19

The effect of activated sludge conditions on micropollutants biodegradation and transformation products formation

L. Gusmaroli, G. Buttiglieri, Catalan Institute for Water Research ICRA

Micropollutants such as pharmaceuticals (PhACs) and endocrine disrupting compounds (EDCs) have been detected in all water compartments and the European Union is therefore updating its legislation to limit the release of emerging contaminants. Nevertheless, for several micropollutants, knowledge on (bio)degradation and especially on transformation products (TPs) is missing. This work aims at achieving a better understanding of the mechanisms and the operative conditions regulating PhACs and EDCs removal as well as TPs formation and degradation in conventional activated sludge (CAS) systems to maximize their biodegradation. Short term (6 hours) experiments were performed to assess the biodegradation of a set of micropollutants and the formation of some of their known TPs in CAS at different pH (7 and 8) in aerobic and anoxic conditions at a TSS of 1 g/L. Activated sludge was spiked with a mixture of PhACs (100 µg/L) and EDCs (10 µg/L). The best removal of estrone (E1) was obtained under aerobic conditions at pH 8 (80%), while a 60% removal was observed at pH 7. During the first 30 minutes, E1 concentration rises up to 180%, while estradiol (E2) degradation profile drops, suggesting that E2 is oxidized to E1, in consistency with literature. Almost no biodegradation occurred in anoxic batch tests. Estriol was significantly degraded under all conditions. Glucuronides were also monitored, though never detected. EE2 and bisphenol-A were not significantly eliminated in any of the batch tests, though some removal was achieved under aerobic conditions (20 and 15% respectively). Ibuprofen (IBU) was not degraded under anoxic conditions but proved highly biodegradable under aerobic conditions at both pH 7 and 8, leading to the formation of 2-hydroxil-ibuprofen (2-OH-IBU) and carboxyl-ibuprofen (CBX-IBU). 2-OH-IBU concentration increased up to 60% of initial IBU concentration and CBX-IBU concentration raised up to 21% in aerobic batches at pH 7. Metoprolol removal varied from zero (anoxic conditions, pH 7) to 22% of initially spiked concentration (anoxic conditions, pH 8), but its TPs were detected only at negligiblle concentrations. Sulfamethoxazole (SFX) was not significantly biodegraded by CAS; under aerobic conditions and at pH 7, small amounts of desamino-SFX and acetyl-SFX were produced while 4-nitro SFX was detected at minor concentrations. Venlafaxine and carbamazepine proved to be persistent pharmaceuticals, in consistency with literature.

MOPC20

Ciprofloxacin By-Products in Seawater Environment in the Presence and Absence of Gilt Head Bream

H. Ziarrusta, L. Mijangos, University of the Basque country UPVEHU / Department of Analytical Chemistry; M. Irazola, University of the Basque country UPVEHU / Research Centre for Experimental Marine Biology and Biotechnology (PIE); A. Prieto, N. Etxebarria, University of the Basque Country UPV/EHU / Plentzia Marine Station (PiE-UPV/EHU) & Dep Analytical Chemistry; E. Anakabe, University of the Basque country UPVEHU / Organic Chemistry; M. Olivares, O. Zuloaga, University of the Basque Country UPV/EHU / Plentzia Marine Station (PiE-UPV/EHU) & Dep Analytical Chemistry

The widespread use of pharmaceuticals has caused a growing concern on the presence of pharmaceuticals such as the antibiotic ciprofloxacin (CIPRO) in the aquatic environment, since they may exert adverse effects on non-target organisms, including fish. In order to study the uptake, distribution in different tissues (liver, muscle, brain and gill) and biofluids (plasma and bile), metabolism and elimination of CIPRO in gilt-head bream (Sparus aurata), controlled dosing experiments for 8 days at 200 µg/L concentration were carried out. CIPRO was only observed in bile, probably due to its low octanol-water partition coefficient and the zwitterionic behaviour. CIPRO by-products (BPs) were also identified in seawater environment, both in presence and absence of fish. The analysis done by means of liquid chromatography–high resolution mass spectrometry permitted the annotation of up to 35 BPs of CIPRO in seawater and bile, from which 30 structures were reported for first time. Up to 20 BPs were annotated in the absence of fish. The phase I degradations suffered by CIPRO in seawater were oxidation, methylation, oxidative defluorination (in 3 BPs out of 20), reductive defluorination (1 BP our of 20), dehydrogenation of the piperazinyl ring (in 2 BPs) and the cleavage of the piperazinyl ring with (1 BP) or without (2 BPs) the loss of the primary amine formed during the cleavage. The only phase II transformation of CIPRO observed was BP18 that, apart from the oxidative deamination of the piperazinyl ring, also suffered the glycine conjugation. 14 of the previously observed BPs were plus 10 new BPs were annotated in water in the presence of fish. Compared to the BPs annotated in the absence of fish, oxidative deamination and both glycine and glutamine conjugation gained importance since 4 of the 10 new BPs had suffered both transformation reactions. Although CIPRO metabolites were searched in gilt-head bream liver, brain, muscle, gill, plasma and bile, BPs were only detected in bile. 5 BPs were found and none of them was detected in seawater. While defluorination and oxidative deamination gained importance in bile, neither glycine nor glutamine conjugates were observed in bile BPs. *Acknowledgements* - This work was financially supported by the Ministry of Economy and Competitiveness through the project CTM2014-56628-C3-1-R. H. Ziarrusta is grateful to the Spanish Ministry and L. Mijangos to the Basque Government for their predoctoral fellowships.

MOPC21

Assessment of the occurrence and impact of polar pesticides in irrigation and drainage ditches at the Ebro River Delta cultivated area (NE Spain)

M. Barbieri, Ins of Environ Assessment&Water Resch (IDAEA-CSIC) / Water and Soil Quality Research Group; S. Monllor, N. Guillem, Institute of Environmental Assessment and Water Research IDAEA CSIC / Department of Environmental Chemistry; M. Rambla-Alegre, IRTA; C. Postigo, IDAEA, CID-CSIC / Environmental Chemistry; M. López de Alda, Institute of Environmental Assessment and Water Research IDAEA-CSIC / Department of Environmental Chemistry

The Ebro River Delta, located in northeastern Spain, is one of the largest wetland areas of the Mediterranean region. It is an area of high ecologic and economic values, where wildlife shares the territory with intensive rice growing and other agricultural activities and seafood production. The objective of this work was to investigate the occurrence of different classes of medium to polar pesticides and transformation products in irrigation and drainage ditches at the Ebro Delta in summer, when application of pesticides is more intensive in the area, and to assess the impact that these contaminants may have on local ecosystems and seafood production activities, and eventually on human health. To this end, an analytical method based on on-line solid-phase extraction-liquid chromatography–tandem mass spectrometry (SPE-LC–MS/MS) was developed and validated for analysis of over 50 pesticides, including various neonicotinoid and organophosphate insecticides, as well as herbicides pertaining to the classes of triazines, phenylureas, anilides, anilines, chloroacetanilides, acidic herbicides, oxadiazoles, carboxamides, benzothiadiazines, nitriles, diphenyl ethers, and carbamates in water. This methodology, which offers various advantages for its routine use in the analysis of medium to polar pesticides in the different water compartments, allowed the quantification of most of the target analytes at levels below 10 ng/L, and with a high reliability of results that stems from the use of an automated and highly selective analytical technique and the use of deuterated analogues of the target compounds as surrogate standards for their quantification. Bentazone followed by propanil presented the highest average concentrations in the analyzed samples, being in the µg/L level. Oxadiazon, acetamiprid, imidacloprid, and triallate were also found at

relevant concentrations in the investigated area and time (above 100 ng/L). As expected concentrations were higher in drainage canals than in irrigation ditches. Measured concentrations were used to evaluate the ecotoxicological risk for the aquatic organisms in this area by means of a hazard quotient-based approach. This work was possible thanks to the Government of Catalonia (2014 SGR 418) and the Spanish State Research Agency (AEI) and the European Regional Development Fund (ERDF) through the project BECAS (grant number CTM2016-75587-C2-2-R), and to Merck for the gift of LC columns.

MOPC22

Degradation kinetics and degradation products of diclofenac with persulfate

J.M. Monteagudo, University of Castilla-La Mancha; H. EI-taliawy, Aarhus University / Department of Environmental Science; A. Durán, i. San Martín, University of Castilla-La Mancha; K. Bester, Aarhus University / Environmental Science

Diclofenac concentrations in effluent wastewater are often exceeding local limits or upcoming EU regulations. This study was undertaken to explore the possibilities of removing diclofenac with persulfate in respect to kinetics and reaction pathways. In-situ chemical oxidation of a diclofenac aqueous solution was performed using persulfate anions activated by ultrasound. The diclofenac (DCF) removal reaction by the persulfate process and the role of various intermediate oxidative species of persulfate such as hydroxyl, sulfate, superoxide anion or singlet radicals in the removal process as well as to determine a possible reaction pathway was observed. The removal efficiency was highest at pH values below 4.5. In addition, the production rate of sulfate radicals from persulfate anion was increased with decreasing pH values. A reduction in the reaction rates in the ultrasonic persulfate (US/PS) process was observed with excess persulfate as the reagent decomposed via the non reactive $S_2O_8^{2-}$ (with no generation of the very effective $SO_4^{\cdot-}$). Sulfate and hydroxyl radicals were involved in the main reaction pathway of diclofenac. Diclofenac amide and three hydroxy-diclofenac isomers (3´-hydroxy diclofenac, 4´-hydroxy diclofenac and 5-hydroxy diclofenac) were identified as reaction intermediates. The obtained results demonstrated that the US/PS process could be a potential alternative to remove compounds of emerging concern, such as diclofenac from wastewater.

Mercury Biogeosciences - Fate, Effects and Policy (PC)

MOPC23

Identifying, Characterising and Quantifying Atmospheric Mercury Sources Using Passive Air Sampling Networks

D. McLagan, University of Toronto Scarborough / Chemistry; C. Mitchell, University of Toronto Scarborough / Physical & Environmental Sciences; F. Monaci, University of Siena; Y.D. Lei, University of Toronto Scarborough / Department of Physical and Environmental Sciences; F. Wania, University of Toronto at Scarborough / Physical and Environmental Sciences

The Minamata Convention on mercury (Hg) stipulates that complete emissions inventories should be established. Passive air samplers (PAS) produce time-averaged concentration data over long deployment periods and are therefore particularly well suited for mapping gaseous Hg concentrations, identifying and locating unknown Hg sources, and quantifying emission rates. We used networks of PAS in both the Greater Toronto Area (GTA) in Canada and the Monte Amiata Hg mining district in Italy to illustrate this approach to Hg source characterisation. We used a PAS for gaseous Hg, which incorporates a sulphur-impregnated activated carbon sorbent and a radial diffusive barrier to control uptake kinetics. 145 PASs were deployed across the GTA in July and August 2016 for time periods ranging from 34 to 46 days. In Italy, PASs were deployed at two spatial scales: a 0.56 km^2 square comprising the former Abbadia San Salvatore mercury mine and a 41.6 km^2 square covering the eastern slope of Mt. Amiata. Both squares were divided into a grid of 7x7 cells and a sampling site was selected within each of the 49 cells. The finer spatial resolution grid was sampled twice with one-week long deployments in Oct. 2015 and Jul. 6 2016. The coarser spatial resolution grid was sampled for an entire year (Oct. 2015-Oct. 2016), in four seasonal deployments of approx. 3 months each. Mean gaseous Hg concentrations in downtown Toronto (1.77 ± 0.28 ng m^{-3}) were slightly, but significantly elevated relative to other parts of the GTA (1.42 ± 0.20 ng m^{-3}). Concentrations at sites close to waste/recycling (1.61 ± 0.22 ng m^{-3}) and hospitals/dental facilities (1.63 ± 0.21 ng m^{-3}) were significantly higher than at sites presumably distant from potential sources (1.37 ± 0.20 ng m^{-3}). In the mine area in Italy concentrations reached as high as 12,500 ng·m^{-3} and declined rapidly with distance from the most contaminated site. Concentrations were higher in July than in October. At both spatial scales, concentrations declined less steeply towards the East, consistent with prevailing westerly winds. Atmospheric emission from the mine was estimated to range from 50 to 100 kg annually. Clearly, the PAS's ability to precisely and accurately discriminate small differences in gaseous Hg concentration (< 0.2 ng m^{-3}) across a wide range of concentrations, including at and near global background levels, enables the mapping of the spatial concentration variability and the identification, characterisation and quantification of both fugitive and major Hg emission sources.

MOPC24

Mercury trend as a possible result of changes in cod age distribution

A. Ruus, NIVA / NIVA; D. Hjermann, NIVA Norwegian Institute for Water Research; B. Beylich, NIVA; M. Schøyen, S. Øxnevad, NIVA Norwegian Institute for Water Research; N. Green, NIVA

Mercury (Hg) enters the biosphere from natural and anthropogenic sources. Methylmercury is the most toxic form of Hg and has a high bioaccumulative potential, thus high concentrations of Hg may accumulate in fish tissue. Mercury in Atlantic cod (*Gadus morhua*) is one of many things that are monitored through the Norwegian contribution to the Coordinated Environmental Monitoring Programme (CEMP) carried out by the Norwegian Institute for Water Research (NIVA) by contract from the Norwegian Environment Agency. CEMP is administered by the Oslo and Paris Commissions (OSPAR), and the results from Norway and other OSPAR countries provide a basis for a paramount evaluation of the state of the marine environment. Data on mercury and other contaminants in cod from the Inner Oslofjord (Norway) reach back to 1984. Until 2014, annual median Hg-concentrations in cod from the Inner Oslofjord displayed significant upward long-term (whole time series) and short-term (last 10 years) trends (when 2015 was included, the short term trend was not significant). However, the median length of the cod sampled also increased significantly over time. This is consistent with results of beach seine surveys conducted in the Inner Oslofjord, showing that cod recruitment in the area has been low since the start of the 2000s. To elucidate how length would possibly impact the trend analysis, the Hg-concentrations in the cod muscle were normalized to that of 50 cm cod. No significant long- or short-term trends could be observed for the normalised Hg-concentrations. The results indicated that most of the upward trend in Hg-concentrations in cod muscle from the Inner Oslofjord during the last 10-20 years could be attributed to the sampling of larger fish. This again may result from changes in the population structure (e.g. repeated recruitment failure), or changes in sampling bias. These findings point to the need for uncovering the effect of cod length/age on the mercury concentration trends also on other localities along the Norwegian coast. The results from this analysis is newly finalized and will be presented.

MOPC25

Contributions from biomass burning to mercury emissions at Cape Point, South Africa

V.S. Somerset, CPUT / Chemistry; C. Van der Horst, University of The Western Cape / SensorLab Department of Chemistry; L.G. Martin, South African Weather Service; C. Walters, CSIR / Natural Resources and the Environment

Mercury (Hg) is known to be a persistent and toxic heavy metal that can bio-accumulate in the aquatic environment and lead to serious human health effects. Hg is released into the atmosphere from both natural and anthropogenic sources, wherein the atmosphere it can be present in a gaseous phase or in particulate matter. Researchers working in South Africa have been quantifying the emission of Hg from various sources in southern Africa for the last decade. These studies have shown that the emissions from coal burning are reasonably well documented, with some recent inventories showing Hg results between 40 – 50 t Hg/yr in 2004 to 2006. However, only a few attempts have been made to estimate the mercury emission from biomass burning to the atmosphere in South Africa. This study provides a quantitative assessment of biomass events from 2007 to 2017, for which the gaseous elemental mercury (GEM) measurements in conjunction with the measurement of trace gases (CO, CO_2, CH_4, O_3), meteorological parameters and air mass trajectories were studied.

MOPC26

Building a predictive model for methylmercury photodemethylation in freshwater ecosystems

S. Klapstein, Acadia University / Earth & Environmental Science; D.A. Risk, St Francis Xavier University / Earth Sciences; S.E. Ziegler, Memorial University of Newfoundland / Earth Science; N.J. O'Driscoll, Acadia University / Department of Earth and Enviromental Science

Quantifying why and how some ecosystems are more sensitive to contamination following atmospheric mercury deposition is key to mercury fate modelling. Photodemethylation of MeHg is thought to be one of the main processes that convert MeHg into a less biologically toxic form of mercury [4]. While previous studies highlight the importance of photodemethylation to mercury budgets, few have examined the magnitude and variability of photodemethylation rates as a function of associated dissolved organic matter (DOM). DOM absorbs specific wavelengths of solar radiation and therefore MeHg that is bound to these compounds containing photoreactive functional groups may be more readily degraded than unbound MeHg. Alternatively, DOM may shade much of the water column and inhibit photodemethylation. To address this research gap we have used numerous controlled and semi-controlled experiments that focused primarily on the quantification of the relationships between solar radiation exposures, DOM, and MeHg within six freshwater lake systems in Kejimkujik National Park and National Historic Site in southwestern Nova Scotia. Using incident irradiation values measured from floating instrumentation the incoming UV-A could be modelled with depth in the lakes as DOM concentration changes. From these numbers we were able to apply our photodemethylation rate constants, derived from controlled experiments, to available UV-A to predict the loss of MeHg based entirely on DOM concentration (Figure 1). In the subset of Kejimkujik National Park lakes that were studied, lakes with higher DOM lost much less MeHg through the

photodemethylation pathway and a strong seasonal difference due to variation in incoming solar radiation was evident. This model may be appropriate for other aquatic ecosystems by simple standardization techniques depending on water quality characteristics such as DOM photoreactivity (structure), pH, and dissolved ionic species. Overall, this body of work yielded a method for predicting mercury availability to food webs depending on environmental and physicochemical factors. Climate change in temperate and boreal regions of Atlantic Canada is projected to increase rainfall amounts and occurrences and thus lead to browning of freshwaters and further inhibition to the photodemethylation pathway of MeHg reduction.

MOPC27

Polymer inclusion membranes followed by X-ray fluorescence analysis as a new methodology for mercury monitoring in natural waters at low concentration level

G. Elias, University of Girona; E. Margui, University of Girona / Department of Chemistry; S. Díez, IDAEA CSIC Barcelona; C. Fontas, University of Girona / Department of Chemistry

At present, there is a considerable interest in mercury (Hg) monitoring due to its widespread occurrence and high toxicity of most of its compounds. Due to the low concentration levels, the complexity of some natural waters and the poor stability of the metal during sample storage, methodologies overtaking this problems are of main interest. In this context, polymer inclusion membranes (PIMs) can be an interesting tool to help in environmental monitoring. PIMs consist of a polymer, which provides mechanical strength, the carrier, which is the responsible of the extraction process, and sometimes also a plasticizer can be used to provide elasticity. The stability, versatility and easy manufacturing make PIMs as a useful separation technique to be taken into account. In this work, PIMs have been prepared fixing cellulose triacetate as polymer and the ionic liquid trioctylmethylammonium thiosalicylate (TOMATS) as extractant. PIMs were contacted with Hg in natural waters and, once the metal was collected, membranes were mounted in the sample holder of the Energy dispersive X-ray fluorescence (EDXRF) system and were directly analyzed. A good correlation was found between Hg concentration in the natural water (0.5-10 ppb) and the peak area obtained in the analysis of the corresponding loaded PIM, and thus, it can be used as a calibration curve. Optimized conditions of the whole methodology allowed a Hg detection limit of 0.2 µg Hg L^{-1} in water. Moreover, no water matrix effects when testing tap water, river water, sea water and ground water were found. Thus, PIMs can be seen as a global solution for Hg monitoring in all types of natural waters. Additionally, we have investigated for the first time, the possibility of using PIMs as a tool to preserve samples of Hg in environmental waters. PIMs anayzed after 6 months of Hg extraction did not difere from the results obtained the first day of analysis. Hence, PIMs can be viewed as an innovative media to extract low levels of metal from different natural waters and to preserve sample information until the determination of the metal can be performed.

MOPC28

Dissolved organic matter as a modifier of Hg bioavailability to phytoplankton

V. Slaveykova, University of Geneva / Département F.-A. Forel des sciences de lenvironnement et de leau; T. Chonova, I. Worms, University of Geneva / Department FA Forel for Environmental and Aquatic Sciences

Mercury (Hg) is a priority toxin of global concern, which concentrates in biota and biomagnifies in the aquatic food webs. However, mercury interaction with phytoplankton, central for its incorporation in the food webs, and in particular the role of various environmental modifying factors such as dissolved organic matter (DOM) is still to elucidate. The objective of this work is to get new insight in the role of the DOM on Hg bioavailability to phytoplankton. Since trace metal complexation by DOM is expected to reduce its bioavailability, we hypothesized that the reduction of the Hg bioavailability to *Chlamydomonas reinhardtii*, chosen as a model phytoplankton, will be proportional to the fraction of the Hg being complexed by DOM. To get insight into the role of DOM in Hg uptake, *C. reinhardtii* was exposed to two concentrations of Hg in the presence of standard Suwannee River humic acid (SRHA) and in natural water rich in DOM from Onego Lake, Russia. Water was sampled from five sites representing the DOC gradient from River Shuya to open lake. Bioavailability was quantified by determining the adsorbed and intracellular mercury concentrations by Direct mercury analyzer on freeze-dried pellets. Concentrations of Hg in the exposure media were measured with the MERX Automated Total Mercury Analytical System. Chemical speciation of Hg in the absence or presence of DOM was computed with WHAM/model VII. The results showed that adsorbed and intracellular Hg concentrations decreased as compared with exposure in the absence of SRHA only at 0.7nM IHg, when the ratio between the reduced sulfur concentration and IHg is bigger than 100. A significant increase (1.5×) of Hg uptake in *C. reinhardtii* exposed to 70 nM Hg in the presence of 0.5 and 5 mg·L^{-1} DOC was found. In the DOC-rich water from lake Onega, a decrease of the bioavailability with respect to exposure in the absence of DOM was found. However no specific trends in the Hg uptake by *C. reinhardtii* were observed over DOC concentration gradients. The effect of the other factors such as the presence and concentration of different major cations and anions, as well as mercury binding to the Al, Mn and Fe colloids has to be taken into account in addition to the role of DOM. The implications of the obtained results are discussed further with respect to the prediction of the mercury incorporation at the base of the food-webs and the impact in the environmental systems.

Fungicides - an overlooked compound group? Fate, effects, risk assessment and mitigation (PC)

TUPC01

Overview on the risks from fungicides for aquatic organisms

J.P. Zubrod, University of Koblenz-Landau / Institute for Environmental Sciences; J.R. Rohr, University of South Florida / Department of Integrative Biology; G. Arts, Wageningen Environmental Research (Alterra) / Environmental Risk Assessment; C. Bruehl, University of Koblenz-Landau / Institute for Environmental Sciences; M. Bundschuh, Swedish University of Agricultural Sciences / Department of Aquatic Sciences and Assessment; R. Schaefer, University Koblenz Landau / Institute for Environmental Sciences

As fungal pests are a major threat to crop production, the application of fungicides to control fungal infestations is considered indispensable to secure global food supply. The use of fungicides is forecasted to increase due to altered climatic conditions and invasive fungal species. Following their use, fungicides can enter aquatic ecosystems and, given their typically frequent application, ecotoxicologically relevant levels of fungicides can occur in surface water bodies in agricultural catchments throughout the growing season. However, in comparison to herbicides and insecticides, the fate and effects of fungicides have received less attention. To highlight research gaps, we reviewed the current knowledge on fungicide effects for aquatic organism groups (microorganisms, plants, as well as invertebrate and vertebrate animals) with a particular emphasis on the functional and ecosystem level. Related contributions reviewed fungicide exposure and mitigation measures. Within aquatic systems, aquatic fungi appear to be particularly at risk of adverse effects because fungicides are designed to control their terrestrial relatives during crop production. Indeed, structural and functional implications in aquatic fungal communities have been reported in field and laboratory studies. As fungi positively (e.g., conditioning of detritus) and negatively (e.g., via parasitism) interact with other organisms, such effects have been shown to result in indirect fungicide effects on other taxonomic groups. In addition, other taxonomic groups can also be directly affected by fungicides because these substances act on biological processes that are highly conserved (e.g., energy production). Direct effects have been reported for aquatic non-fungal microorganisms, plants, as well as invertebrate and vertebrate animals. We will discuss these effects for several fungicide/mode of action groups that were comprehensively tested in laboratory and semi-field studies. Subsequently, we discuss current risk assessment procedures for fungicides in the light of identified knowledge gaps and provide recommendations for amendments that can be inferred from our findings.

TUPC02

Relative tolerance of aquatic organisms to fungicides

A. Rico, IMDEA Water Institute / Aquatic Ecotoxicology; T. Brock, Alterra, Wageningen University and Research Centre / Environmental Risk Assessment Team; M. Daam, New University of Lisbon

Fungicide compounds are routinely used in intensive agriculture production to treat a wide range of plant pests and microbial pathogens. These compounds may reach aquatic ecosystems by spray-drift, leaching and runoff, posing a potential threat to aquatic organisms. In this study we evaluated the sensitivity and relative tolerance of non-target aquatic organisms as compared to that of the standard test species used in the aquatic risk assessment for fungicides. A toxicity database was created that contained acute and chronic laboratory toxicity data for 182 taxa and 139 fungicidal compounds. Toxicity data was obtained from the US EPA ECOTOX database and complemented with data contained in EFSA draft assessment reports. The data was selected following strict criteria as regards to the endpoints, measured effect and exposure duration proposed by the EFSA Aquatic Guidance Document for the aquatic effect assessment of plant protection products. Sensitivity differences between non-standard and standard test species were assessed following the relative tolerance (Trel) approach i.e., by dividing the toxicity value of a non-standard test species by the toxicity value of the standard test species. Trel values were calculated on the basis of the standard test species used in the acute first tier (*Daphnia magna*, *Oncorhynchus mykiss*) and chronic first tier (*Raphidocelis subcapitata, D. magna, O. mykiss*) effect assessments. Trel values were averaged per taxonomic group and per antimicrobial toxic mode of action of the evaluated compounds. The results of this study reveal that, on average, annelids, mysids and bivalves have a higher acute sensitivity to fungicides than *D. magna*, although such trend was not observed in the chronic sensitivity evaluation. *O. mykiss* was confirmed to be among the most sensitive fish species to fungicides. Regarding the primary producer evaluation, diatoms were found to be more sensitive than *R. subcapitata* in the majority of the cases. Sensitivity differences were generally less than two orders of magnitude in the acute assessment and less than one order of magnitude in the chronic assessment, indicating that the assessment factors applied to toxicity values for standard test species encompas the sensitivity range of non-standard test species under laboratory conditions. A clear Trel pattern related to the antimicrobial toxic mode of action of the evaluated substances was not identified.

TUPC03

Fungicide effects propagate through the detrital food chain in streams

J. Rasmussen, Aarhus University / bioscience; M. Bundschuh, Swedish University of Agricultural Sciences / Department of Aquatic Sciences and Assessment; M. Skov Pristed, Aarhus University / Department of Bioscience

Fungicide use in Europe equals that of herbicides, but the amount of studies addressing ecological effects of fungicides is disproportionately low. Recent studies suggest that particularly freshwater fungi may be susceptible to fungicide exposure leading to changed fungal community structure and reduced fungal biomass. These effects may negatively influence the food quality for higher level consumers, e.g. invertebrate shredders. Fungicides occur rather continously in low concentrations in agricultural streams especially during cropping seasons suggesting that long-term chronic exposure scenarious should be covered in ecotoxicoloigcal research. We conducted a 5 month stream channel experiment using two environmentally reaslistic concentration levels of a quaternary fungicide mixture to investigate long-term effects of chronic fungicide exposure of a leaf decomposer assemblage containing fungal communities and two species of caddisfly shredders: *Chaeopteryx villosa* and *Anabolia nervosa*. Food availability was additionally manipulated ranging from excessive to limited food availability (three treatment levels). Fungal biomass significantly decreased with increasing fungicide concentrations, and the fungal community structure was significantly different in the highest fungicide treatment compared to the lowst fungicide treatment and the untreated control. Fungal species richness was consistently and significantly lowest in the highest fungicdie treatment. Shredder induced leaf decomposition was significantly lower in the treatments containing highest fungicide concentrations and always highest in the untreated control. Emergence success of *C. villosa* significantly decreased with increasing fungicide concentration from >60 % in the untreated controls to < 20 % in the highest fungicide treatment at maximum food availability. Minimum food availiabity further increased fungicide effects. Significant effects occurred at concentrations a factor of 20 to 200 below the $EC50_{(growth)}$ concentrations for chronic algae ecotoxicity tests. Our study highlights that environmentally realistic fungicide exposure may propagate through the detrital food chain in streams at concentrations that are well below the Regulatory Acceptable Concentrations. Hence, supplemental ecotoxicity tests (e.g. based on aquatic fungi) are probably necessary for sufficiently safeguarding stream ecosystems in the risk assessment of fungicides.

TUPC04

Mitigation of fungicide exposure of stream ecosystems within agricultural catchments

M. Bundschuh, Swedish University of Agricultural Sciences / Department of Aquatic Sciences and Assessment; R. Schulz, University of Koblenz-Landau / Institute for Environmental Sciences

Fungicides are a vital part of the agricultural pest management. As a consequence, fungicides – such as all pesticides – reach surface water bodies mainly through spray drift, run-off and drainage. To mitigate fungicide exposure a range of measures have been put forward. A densely vegetated and wide buffer strip surrounding surface water bodies, for instance, can be efficient to reduce the spray drift of fungicides during application. Also during run-off, buffer strips have been suggested as a potential measure mitigating fungicide exposure by retaining run-off water and providing sites for adsorption as well as degradation. Under field conditions, however, sparse vegetation density and erosion rills undermine the buffer strips' mitigation potential. Once released into aquatic ecosystems, (constructed) wetland and vegetated systems are considered an effective tool for mitigating a downstream transport of pesticides. The efficiency of such systems depends on both the physico-chemical properties of the pesticide of interest as well as system inherent properties. The pesticides affinity to organic carbon (Koc) is one physico-chemical property driving their retention, with more hydrophobic substances being more efficiently retained. Although fungicides are usually rather hydrophilic, their peak concentrations were also shown to be reduced by such vegetated systems. The systems' efficiency in doing so, is modulated by size related properties as well as plant density. Both parameters are increasing the retention time of fungicides and thereby the probability for adsorption and degradation processes to take place. Mitigating the fungicide exposure via spray drift and runoff may thus efficiently be addressed by a combination of measures. Those measures may include the proper management of vegetated buffer strips. This mitigation measure may be supported by the implementation of vegetated systems (such as constructed wetlands) in situations where catchment characteristics suggest a high risk that cannot be controlled by buffer strips or where such buffer strips cannot be realised.

TUPC05

Towards a better exposure assessment of antifungal azoles

N. Creusot, Eawag - Swiss federal Institute of Aquatic Science and Technology / Environmental Chemistry; M. Casado-Martinez, Centre Ecotox; A. Chiaia-Hernandez, Eawag - Swiss federal Institute of Aquatic Science and Technology / Environmental Chemistry; B.J. Ferrari, Centre Ecotox EAWAGEPFL; S. Fischer, Eawag, Swiss Federal Institute of Aquatic Science and Technology / Environmental Toxicology; Q. Fu, Eawag, Swiss Federal Institute of Aquatic Science and Technology / Department of Environmental Chemistry; N. Munz, Eawag / Environmental Chemistry; H. Singer, Eawag, Swiss Federal Institute of Aquatic Science and Technology / Environmental Chemistry; S. Spycher, B. Spycher, Eawag Swiss Federal Institute of Aquatic Science and Technology; C. Stamm, Eawag, Swiss Federal Institute of Aquatic Science and Technology / Environmental Chemistry; A. Tlili, Eawag / Department of Environmental Toxicology; I. Wittmer, Plattform Wasserqualität VSA, c/o Eawag; J. Hollender, Eawag / Environmental Chemistry

Antifungal azoles are a class of contaminants of emerging concern since increasing evidences highlight their potential effect on aquatic organisms at different trophic levels, raising the need to evaluate the associated environmental risk. Although a few of these compounds are routinely investigated, an accurate exposure assessment of most of them is still lacking to evaluate this risk. To address this issue, we first defined a list of 60 antifungal azoles including pesticides and pharmaceuticals based on the use/consumption of these compounds in Switzerland and Germany. We then performed a retrospective suspect screening on a set of data acquired with liquid chromatography-high-resolution mass spectrometry (LC-HRMS) from a large panel of environmental samples to complete previously targeted analyses on azoles. Since antifungal azoles are used both as pharmaceuticals and pesticides these samples included wastewater treatment plant effluents (WWTPs), river surface waters, biota from rivers (fish, gammarids, biofilms), river and lake sediments, soils and groundwater from various sites allowing to encompass different sources of anthropogenic pressures. The results revealed that antifungal azoles are widely distributed in aquatic ecosystems (e.g. from

TUPC06

Is the EFSA effect assessment approach for fungicides sufficiently protective for aquatic ecosystems?

M. Daam, CENSE & New University of Lisbon, Lisboa; T. Brock, Alterra, Wageningen University and Research Centre / Environmental Risk Assessment Team; A. Rico, IMDEA Water Institute / Aquatic Ecotoxicology

In Europe, the EFSA Aquatic Guidance Document describes the procedures for the derivation of regulatory acceptable concentrations (RACs) for pesticides on the basis of tier-1 (standard test species), tier-2 (geomean and SSD) and tier-3 (micro/mesocosms) approaches. The consistency of this tiered approach has previously been evaluated for insecticides. In the present study, results of different tiers are compared for fungicides. To this end, laboratory toxicity data and microcosm/mesocosm data were compiled from open data sources supplemented with data from confidential studies conducted for the Industry. RACs for tiers 1, 2 and 3 were calculated following the guidelines described in the EFSA Guidance Document. This presentation will discuss i) the consistency of the tiered effect assessment approach for fungicides as proposed in the EFSA Aquatic Guidance Document; ii) the predictive value of acute and chronic laboratory toxicity estimates for observed responses in microcosm/mesocosm tests; iii) problems in using the Geometric Mean approach in the acute effect assessment for fungicides with a biocidal mode-of-action; and iv) the taxonomic groups that should be represented in species sensitivity distributions for fungicides with a biocidal mode-of-action.

Developments in the ecological and human health risk assessment of biopesticides: microorganisms, semiochemicals and botanicals (PC)

TUPC07

Ecotoxicological studies performed to assess the potential of a yeastlike fungus, Aureobasidium pullulans, and the response of evaluating authorities

C. Donat, bio-ferm GmbH

In the course of inlcusion of two strains of the species *Auroebasidium pullulans* as active substances to be used in plant protection products in Annex I of the Directive (EU) 91/414, a data package was developed to asses the ecotoxicity of these yeastlike fungi. Back then no methods designed for the evaluation of microorganisms as active substances did exist. Methods were based upon certain parts of OECD methods (Section 2) and some advise was found in the EPA OPPTS Series 855 Microbial Pesticide Test Guidelines published by US EPA. However, in some test systems it seemed appropiate to work with scientists of the university to find adequate solutions. Hence some additional "unconventional" results like the Flying doctors experiments with bees as well as an avoidance test with earthmorms were available. Whereas EFSA identified data gaps, the European Commission decided to include *Aureobasidium pullulans* to Annex I without requiring any additional study. National decisions for the registration of the product varied, some authorities authorized the label without any limitations, whereas others demanded up to 30m distance from surface water. However, it is important to think out of the box by evaluating these products used in agriculture. It is evident that living microorganism are able to proliferate, depending on the local environmental conditions in their micro-niche (influenced by climatic factors as well as microbial population dynamics), it is hardly possible to simulate all these complex conditions in laboratory trials. Therefore, a quantitative assessment does not seem to be a scientific sound, since it is not predictable in which quantities, the microorganisms might or might not be present in the environment at certain time points after application.

TUPC08

Ecological testing and risk assessment considerations for microbial active substances

E.A. McVey, J. Wassenberg, Ctgb

For some types of biological pesticide active substances, the same testing schemes and methodologies as used for chemical active substances suffice. However, for others (for example, microbial active substances), the unique properties of the substances and resulting risk assessment questions result in the need for a different perspective on the appropriate testing guidelines and programs, as well as different considerations for the risk assessment assumptions and methodologies. Comparing and contrasting the risk assessment theories and available testing methods, it is clear that while some areas of the risk assessment can be translated between chemical and biological actives, the majority require unique and thoughtful innovations to address the risk assessment objectives. This is particularly well illustrated in the ecological risk assessment schemes for microbials, where testing should be performed under conditions such that both the (various) test organisms and also the microbial active are in an optimal environment. Unique and unknown mechanisms of action and toxicity may also present, and be dependent upon the exposure conditions. Similarly to chemical actives, exposure estimations with microbials are also highly dependent upon environmental conditions, however, microbial actives may have a much greater potential to increase above initial exposure levels than most chemical actives. Regardless of these obstacles, some logical and objective recommendations can be made, both regarding testing and risk assessment for microbial active substances. Considerations of microbial active substances in groups based on pesticide mechanism of action and/or organism group may allow development of some generic testing recommendations. Knowledge from bothe the biological and chemical testing and risk assessment areas should be comprehensively surveyed and utilities to advise more appropriate and adequate testing for microbial active substances.

TUPC09

Human and environmental Risk assessment for microorganisms - to what extent?

A. Cornelese, J. Süß, GAB Consulting GmbH; A. Jakubowska, GAB Consulting Spain S.L.U.; S. Seehase, R. Hauschild, GAB Consulting GmbH

Biopesticides are an excellent alternative to chemical pesticides, and there is continuously increasing interest with both industry and consumers. The Sustainable Use Directive (SUD; Dir. 2009/128/EC) strongly promotes a targeted use of integrated control of pests and diseases where non-chemical measures are preferred.Plant protection products with a microorganism as active substance could be a solution. Microorganisms have to be approved in Europe in order to be used as active ingredients in biopesticide products. Data requirements for pesticides based on microorganisms are available as separate part of regulation EC 283/2013 and EC 284/2013. The Regulation provides the scope of the data requirements and regulates the approval. Series of guidelines published by OECD, SANTE, or EPPO are available for testing of substances used in plant protection. Many of these guidelines are not adapted for microorganisms. Risk assessment for biopesticides to enter the European market is described in the Uniform Principles, Commission regulation EU 546/2011 Part II. Risk assessment approaches for plant protection products are accompanied by guidance’s and models that are not developed for microorganisms. Thus, no consistent approach is available in the different member states. This leads to uncertainties and non-acceptance of submitted data. We will discuss the current challenges in interpretation of the data requirements and propose solutions for the risk assessment of biopesticides based on microorganisms.

TUPC10

Ecotoxicological testing to support the assessment of Microbials

J. Müther, U. Schöbinger, J. Süß, A. Cornelese, GAB Consulting GmbH; A. Jakubowska, GAB Consulting Spain S.L.U.; R. Hauschild, GAB Consulting GmbH

Biopesticides are an excellent alternative to chemical pesticides, and there is an increasing demand in testing and evaluation of these products. This poster focuses on microbial pesticides based on bacteria, fungi, viruses or protozoans as their active substances. Possible adverse effects to non-target organisms (NTO) are rather limited due to the narrow and specific host range of these microbial pest control agents. However, a complete risk assessment demands testing of NTOs, when exposure and risk cannot be fully neglected. The assessment of microbial biological control agents (mBCA) and microbial biological control products (mBCP) is relatively new and approved testing methods are not yet available in the same extent as they are for chemical pesticides. Not only the toxicity, but also the potential pathogenicity/infectivity needs to be addressed. Currently, the data requirements for mBCAs and mBCPs are issue of Part B of the European regulations 283/2013 (mBCA) and 284/2013 (mBCP). Numerous data requirements listed in these annexes were transformed directly from requirements for chemical pesticides and often cannot be adapted to the biological properties of mBCAs. In order to address the data requirements in a feasible manner, the biological properties of the microorganism have to be taken into account, instead of strictly applying to current test guidelines. It is important to note, that testing is strongly influenced by physico-chemical properties of mBCAs. Microorganisms, *i. a.* with frequently used co-formulants, are not soluble which results in alteration of the test conditions (i.e. turbidity, O_2-demand, spray layers). Furthermore, organic components of the formulated product (i.e. yeast, starch) may lead to increased fungal growth in soil or test media. Additionally, the need to test at high concentration levels, leads to negative effects of particles (i.e. spores or co-formulants like kaolin) on the test organisms which are not related to the active substance and are difficult to interpret. Differences between OECD and OCSPP (formerly OPPTS) guidelines, and requirements of the analytical verification in the test medium are addressed as part of the development of alternative ecotoxicological testing approaches. The findings of our ecotoxicological expertise presented in this poster can be considered as basis for further discussions in proposing different test designs addressing mBCA and mBCP requirements.

TUPC11

Microbiological Quantification Methods for MPCAs - Applicability to a Range of Microorganisms and Different Substrates

M. Zetzmann, F. Kümmich, A. Dabrunz, C. Lang, Eurofins Agroscience Services Ecotox GmbH / Aquatic Ecotoxicology

In the last decade the number of biopesticide registrations in the EU and US have steadily increased. In the EU biopestices are regulated as plant protection products under regulation 1107/2009. Biopesticides cover a wide spectrum of substances including microbial pest control agents (MPCA) defined as products containing microorganisms (e.g. bacteria, fungi, protozoa, viruses). As for chemical plant protection products, regulatory authorities require an analytical verification of the doses applied in ecotoxicological tests also for MPCAs. Guidance can be derived from SANCO/3030/99 rev.4 and OPPTS 885.1400 (1996), but verification procedures need to be adapted on a case by case basis, as each microorganism possesses its own chemical properties and different growth conditions. Just as chemical methods, microbial methods need to be robust, reproducible and specific. Experimental data will be presented with focus on the applicability of microbial quantification methods considering different microorganisms and substrates.

When ecotoxicology meets trophic ecology (PC)

TUPC17

Modelling bioaccumulation of persistent organic pollutants in Arctic food chains

R. Hoondert, Radboud University Nijmegen / Department of Environmental Science; A.M. Ragas, Radboud University / Department of Environmental Science; J.A. Hendriks, Radboud University Nijmegen / Department of Environmental Science

Persistent organic pollutants (POPs) are a group of chemicals with similar physical-chemical characteristics that are resistant to environmental degradation and biodegradation. Not only do these POPs bioaccumulate in the food chain, they are also known to cause adverse effects in fish, wildlife and humans. Although being banned in the previous century, many POPs are still present in high concentrations in Arctic areas, due to a combination of northward marine currents and their semi-volatile nature, high thermal stability and slow degradation turnover rates. As food webs in the Arctic are relatively simple, POP contamination may pose a great risk for animals at higher trophic levels, such as the polar bear (*Ursus maritimus*), hence the growing interest in studying bioaccumulation in the Arctic. Despite the large interest in bioaccumulation in Arctic food chains, the OMEGA model, as well as similar bioaccumulation models, are predominately validated on temperate food chains or relatively straight-forward Arctic food webs. In the present study, we aim to model bioaccumulation of multiple persistent compounds in the Arctic encompassing multiple species, using the OMEGA (Optimal Modelling for EcotoxicoloGical Application) bioaccumulation model. In this study, we aim to validate the model on Arctic areas by using a binning approach to include multiple species, in which species of a similar trophic level were binned.

TUPC18

Distribution and Trophic Magnification of Dechloranes, HBCDs, PCNs, and Other Legacy POPs in the Maritime Antarctic Ecosystem

J. Kim, Korea Polar Research Institute / Division of polar paleoenvironment; Y. Choi, POSTECH Pohang University of Science and Technology; M. Barghi, POSTECH; J. Kim, J. Jung, Korea Polar Research Institute; Y. Chang, POSTECH Pohang University of Science and Technology

This study investigated distribution and trophic magnification of emerging persistent organic pollutants (POPs), including PCNs, HBCDs, Dechloranes, polychlorinated biphenyls (PCBs) and organochlorine pesticides (OCPs) in the maritime ecosystem in King George Island, Antarctica. The samples were collected in the Baton Peninsular in King George Island, Antarctica. from December 2013 to January, and included Antarctic cod, icefish, limpet, amphipods, leopard seal, Gentoo penguin, Chinstrap penguin, kelp gull, and south polar skua. PCNs, HBCDs, Dechloranes, DDTs, HCHs, Pentachlorobenzene (PCBz), Hexachlorobenzene (HCBz), Chlorodanes, PCBs were detected in all samples, and the levels were the detection rates for the legacy POPs were more than 90 %, but those of some new POP compounds were only 50%. The detected POP levels in this study were much lower than the levels in the previsiou study in low and mid latitude region, and even those in the Arctic. The trophic magnification factor (TMF) of each POP compound were calculated based on the ratio of stable isotope nitrogen and the log-transformed POP concentrations. Some of the compounds, OCPs, HBCDs and highly chlorinated PCBs and PCNs, showed significantly positive correlations, suggesting biomagnification of the chemicals. DPs, however, showed insignificant trophic magnification. After the seperated TMF analysis for aquatic and terrestial food web models, TMF values showed different tredns compared to the TMFs in whole sample model. The inclusion of migrant animal, such as south pola skua and kelp gull, also arose an uncertainty to evaluate TMFs. The result of this study presented widespraed contamiation of the Anarctic Environment by the New and Legacy POPs. The levels of most POPs were magnified through trophic levels, while Dechloranes, emerging contaminants, appeared not to enough TMF values. The insufficient detection rate of Dechloranes, compexity of the food web structure, and the overestimation due to migrant animals arose the uncertainties in TMFs, and therefore need to be taken into consieration to interpret the TMF results in this study.

TUPC19

Bioconcentration as the predominant mechanism for fish PCB contamination in alpine lakes.

T. Masset, Universite Savoie Mont Blanc; M. Perga, University of Lausanne / Faculty of Geosciences and Environment; N. Cottin, Universite Savoie Mont Blanc; S. Cachera, CISALB; C. Piot, E. Naffrechoux, Universite Savoie Mont Blanc

Bioconcentration and biomagnification relative contribution to the PCB burden in freshwater fish in alpine lakes ecosystems remain a debated issue. The aim of this study was to identify the relative role of those different processes for two fish species *Coregonus lavaretus* (European whitefish) and *Salvelinus alpinus* (arctic char) in one of the heaviest PCB contaminated alpine ecosystem: lake Bourget (France). The 7 indicator-PCB concentration and lipid content of fish filet were measured in European whitefish (n = 89) and arctic char (n = 55) from 2013 to 2016. Potential explanatory variables for differences in PCB contamination levels in fish were chosen to identify the impact of living and feeding habitat (using $d^{13}C$) and the influence of trophic parameters using $d^{15}N$ and body size. Results showed a decrease of PCB burden in fish after the clean-up of the major input source of PCB in the lake and a steady situation since then. Arctic char was found to be significantly more contaminated than whitefish with a mean concentration of 254±132 ng.g^{-1} w/w and 45±28 ng.g^{-1} w/w respectively. Individual's PCB contaminations in both species were not tied to feeding habitats (p>0.05). Trophic position (characterized with $d^{15}N$) was also not correlated with intra-species concentration variabilities for arctic char and was only slightly positively related to concentration variabilities for whitefish (p=0.04), dismissing the importance of biomagnification as PCB accumulation process. However, fish body size seemed to be a potential explanatory variable for individual's PCB concentration discrepancies in arctic char (p=0.002) and whitefish (p=10^{-4}). This last observation could be explained by fish/water partitioning equilibrium to be reached, where fish would tend to accumulate more PCB through their lifetime, highlighting the effect of the bioconcentration process. A lower clearance rate due to changes in physiological parameters (lower gill/body weight ratio, lower metabolism and/or excretion rate) could also be involved.

TUPC20

The role of diet and age: organohalogen accumulation in an avian top predator

M.E. Løseth, The Norwegian University of Science and Technology / Biology; N. Briels, Norwegian University of Science and Technology / Biology; I. Eulaers, University of Antwerp / Biology; T. Nygård, T.V. Johnsen, J.O. Bustnes, Norwegian Institute for Nature Research NINA; D. Herzke, NILU - Norwegian Institute for Air Research / FRAM Centre Tromso; G. Poma, G. Malarvannan, University of Antwerp / Toxicological Center; A. Covaci, University of Antwerp, Toxicological Center / Toxicological Centre Dep of Pharmaceutical Sciences; B.M. Jenssen, Norwegian University of Science and Technology / Biology; V. Jaspers, Norwegian University of Science & Technology / Biology

Occupying a high trophic level, the white-tailed eagle (WTE; *Haliaeetus albicilla*) can accumulate a wide range of organohalogenated contaminants (OHCs), even at an early age. Their diet consists mainly of fish and seabirds; thus, a long food chain potentially resulting in biomagnification of OHCs. The nestlings can be exposed to high levels of OHCs through maternal transfer from the egg, and later through the diet. As nestlings develop and grow, concentrations of maternally derived compounds are diluted. Few studies are accounting for the biological factors of age and increase in body mass when monitoring OHCs in nestlings. The aim of the present study was to investigate how differences between years, locations and dietary tracers can explain variation in OHC accumulation in plasma of WTE nestlings. Stable isotopes (SI) of nitrogen ($\delta^{15}N$) and carbon ($\delta^{13}C$) were applied as proxies for trophic level and dietary carbon source, respectively. In addition, we included the possible confounding effects of age and body mass on the contaminant variation. Samples were obtained in 2015 and 2016 from 70 WTE nestlings from two archipelagos in Norway, Smøla and Steigen. In total, 14 polychlorinated biphenyls (PCBs), 7 organochlorinated pesticides (OCPs), 5 polybrominated diphenyl ethers (PBDEs) and 8 per- and polyfluoroalkyl substances (PFASs) were quantified in over 50 % of the analyzed plasma samples at each location and year. The WTE is a marine top predator; however due to the topography of the island Smøla WTEs may feed on a mixed terrestrial and marine diet. According to our preliminary analyses, WTEs in Steigen are feeding on a slightly higher trophic level than WTEs in Smøla WTEs, and may consequently accumulate more of the investigated OHCs. Though, in our analyses the SI values were only important in explaining variation in POPs but not PFAS levels. We also observed that age at sampling is an important factor, as legacy POPs are decreasing while PFASs are increasing with age. However, there are differences between years at each location with higher OHC concentrations for Steigen in 2015 and Smøla in 2016, not explained by age or diet. Our analyses also demonstrate large variations within nests at both locations, suggesting that siblings may not always share prey. We hereby emphasize the importance of ecological and biological variables when investigating OHCs in an avian top predator.

TUPC21

Fate of PAH, phthalates and their metabolites in an urban river food web

A. Goutte, F. Alliot, EPHE / UMR Metis; H. Budzinski, University of Bordeaux; M. Chevreuil, EPHE / UMR METIS 7619; R. Santos, Hepia, University of Applied Sciences Western Switzerland / Ecology and Engineering of Aquatic systems research group; P. Labadie, UMR CNRS EPOC Universite Bordeaux / UMR 5805 EPOC

Trophic magnification factors have been extensively assessed for persistant organic pollutants, but remain poorly studied for metabolizable pollutants and their metabolites. Polycyclic aromatic hydrocarbons (PAHs) and phthalate plasticizers are continuously released in urban rivers and are rapidely metabolised and excreted by freshwater organisms, thus limiting their bioaccumulative potential. Abiotic and biotic samples, from primary producers to piscivorous fish, were collected in an urban river and analysed for PAHs, phthalates and their metabolites. Stable isotopes of nitrogen were used to determine trophic levels and to calculate trophic magnification factors (TMF) of each compound and its associated metabolite. Our results highlight a trophic dilution (TMF < 1) of all PAHs and phthalates, meaning that predators were less contaminated than their preys. When taking into account the associated metabolites, total body burden of PAHs still declined with increasing trophic levels, confirming the rapid transformation and excretion of these compounds within organisms and a very limited trophic transfer. In contrast, the level of phthalate compound and its associated metabolite(s) increased from prey to predators, suggesting a lower clearance rate of phthalates and a slight bioamplification potential across freshwater food webs. At the light of these results, it appears essential to consider phthlate metabolites, instead of phthalate diesters only, in environmental risk assessment.

Epigenetic and evolutionary toxicology: from mechanisms to risk assessment (PC)

WEPC01

Does pre-exposure to bisphenol A affect the susceptibility of breeding zebrafish upon re-exposure?

H. Littler, University of Exeter / Biosciences College of Life and Environmental Sciences; L.V. Laing, University of Exeter / Biological Sciences; R. Boreham, M. Griffiths, University of Exeter / Biosciences College of Life and Environmental Sciences; M. Trznadel, University of Exeter / Biosciences; J. Fitzgerald, University of Exeter / Biosciences College of Life and Environmental Sciences; G.C. Paull, University of Exeter; R. van Aerle, Centre for Environment Fisheries and Aquaculture Science / Biosciences College of Life and Environmental Sciences; J. Mill, University of Exeter / Exeter Medical School; E.M. Santos, University of Exeter / Biosciences College of Life and Environmental Sciences

Bisphenol A (BPA) is a commercially important chemical used in the production of widely used epoxy resins and polycarbonate plastics and it is ubiquitous in the environment, resulting in widespread exposure of humans and wildlife. BPA was shown to cause reproductive effects via disruption of both the oestrogen and androgen signalling pathways. Recent studies suggest that BPA also affects epigenetic signalling pathways, including alterations in transcription of DNA methylation maintenance enzymes and altered DNA methylation profiles. This study aims to investigate how previous exposure of adult fish to BPA affects their response and the response of their offspring upon re-exposure, and whether there is an epigenetic basis for these effects. Breeding groups of zebrafish (*Danio rerio*) were exposed to 10 and 100µg BPA/L for 5 days, either once (C-10, C-100) or twice (10-10, 100-100) with a 13 day period of depuration in between, and appropriate controls were maintained in parallel. The adult gonads were sampled for transcriptional analysis. Reproduction was quantified over time, and embryos from each treatment group were then exposed to a range of BPA concentrations from 0-72hpf to measure their susceptibility to BPA exposure. There were no effects on reproductive output under our exposure conditions. However, at the transcriptional level, anti-Mullerian hormone (*amh*) was significantly downregulated only in females receiving repeated exposures to BPA (100-100). In addition, embryos originating from adults which received a pre-exposure to BPA (100-100) were significantly more tolerant compared to embryos originating from

naïve adults which received a single exposure to BPA (C-100). This suggests that pre-exposure of adult fish leads to a protective effect on their offspring. We hypothesise that these effects may be due to physiological changes or epigenetic memory between the first and second exposure period, and we will now analyse the promoter DNA methylation of *amh* to investigate this hypothesis.

WEPC02

Zebrafish as a model to assess transgenerational effects of environmental stress via epigenetic inheritance

J. Kamstra, NMBU / BaSam

Although toxicological effects of anthropogenic stressors present in the environment are studied extensively, few studies assess the epigenetic effects of such stressors, and even fewer assess these effects over multiple generations. It is hypothesized that some stressors are able to change the epigenetic state in germ cells causing effects on histones modifications, DNA methylation and miRNA expression, potentially inherited by subsequent generations. Here, the aim was to elucidate the role of epigenetics in transgenerational inheritance of effects of different stressors by measuring all epigenetic layers using the zebrafish model. Transgenerational studies were performed with three different stressors; a DNA methylation inhibitor, 5 azacytidine (5AC); a phthalate metabolite, mono(2-ethylhexyl)phthalate (MEHP) and ionizing radiation. We employed state-of-the-art techniques to assess effects in multiple generations of zebrafish embryos and larvae at all epigenetic layers, but most extensively on DNA methylation. Following early life exposures to 5AC and MEHP, many changes of DNA methylation were found in larvae, and these changes could be linked to gene pathways that are associated to those compounds, such as embryonic development and obesity. Subsequent analysis in two following generations lead to the conclusion that some regions were persistently changed. Concerning ionizing radiation, in F1 embryonic offspring from irradiated parents, many changes of DNA methylation were observed. These changes could be linked to effects that were found in the offspring, such as DNA damage. Follow up analysis in the second and the third generation, revealed persistent effects of DNA methylation in a number of regions. Additionally, miRNA analysis in the offspring revealed a number of differentially expressed miRNA linked to similar pathways as with the DNA methylation dataset. Finally, changes in histone modifications were found at specific loci, but these changes were not observed in the second generation. Our results reveal considerable effects on DNA methylation following exposures during early life in zebrafish to MEHP, 5AC and ionizing radiation, as well as a role for miRNAs and histone post translational modifications for the latter. By linking the DNA methylation data to genes, results indicate a functional role for DNA methylation in zebrafish. Persistent effects in F2 and F3 implies that DNA methylation changes can be inherited by multiple generations.

WEPC03

Can changes in DNA methylation be linked to exposure of plants to radiation over multiple generations?

N. Horemans, Belgean Nuclear Research Centre (SCK-CEN) / Biosphere Impact Studies; E. Saenen, J. van de Walle, SCK-CEN; M. Van Hees, SCK-CEN / Biosphere Impact Studies; S. Gaschak, Chornobyl Center; K. Nanba, Institute of Environmental Radioactivity; R. Nauts, SCK-CEN

In this study the impact on plants of long-term (transgenerational) exposure to radiation coming from nuclear accidents like Fukushima and Chernobyl is investigated and compared with lab experiments in either a chronic, acute, multi-, or transgenerational transgenerational set up. The focus of the study was on changes in DNA methylation could be the basis of transgenerational changes found in field or lab conditions. A field campaign was performed in both Chernobyl (CEZ) and Fukushima affected areas (FEZ) in the course of May 2016. Annual *Brassicacea* plants, *Arabidopsis thaliana* and *Capsella bursa pastoris* in CEZ and FEZ, respectively, were sampled alongside a gradient of enhanced radiation ranging from 0.5 to 50 $\mu Gy.h^{-1}$. Seeds from *Arabidopsis thaliana* were harvested in the CEZ and grown for one clean generation under lab conditions to score for multigenerational effects. In addition further lab experiments were performed on wild type plants of *Arabidopsis thaliana* grown under chronic exposure conditions (at 1 mGy/h) or more acutely (20 to 400 mGy/h) for 14 days in one, two or three generations. Plants were scored for total methylation, photosynthetic capacity and oxidative stress markers as well as germination rate and root growth. In general higher differences are found in plants exposed in a multigenerational setup than in a transgenerational one. The field plants did not show any abnormalities that could be correlated with the exposure gradient although some delay in flowering was observed in plants from medium and high radiation levels. The level of total DNA methylation could not be linked to the radiation gradient present in the field but rather to differences in developmental stage of the collected plants. In lab-exposed plants however global DNA methylation showed a significant increase which was both dose and generation dependent. Significant changes in transcription of methylation regulating genes were also measured in the different generations. Highest differences were present in the S1 generation but seemed to be reduced in the S2 generation. Overall the data hint towards a role of methylation in the response to radiation but its use as marker of exposure or in risk assessment needs further experimental evidence and discussion. This work was supported by European project COMET (7th PCRD EURATOM Contract Number: Fission-2012-3.4.1-604794) (www.comet-radioecology.org)generation.

WEPC04

Evolutionary toxicology: tools to understand impacts of past, present and future environmental contamination

S.E. Crawford, RWTH Aachen University / Institute for Environmental Research, Dept. of Environmental Analysis; J.D. Ouellet, RWTH Aachen University / Department of Ecosystem Analysis; M. Hinderer, Technische Universitat Darmstadt / Institute for Applied Geoscience; A. Schwalb, Technische Universität Braunschweig / Institute for Geosystems and Bioindication; H. Hollert, RWTH Aachen University / Institute for Environmental Research

This research will utilize environmental reconstruction methods along with paleolimnological, paleoecotoxicological, and paleogenomic techniques to understand historical, current and potentially future trends in environmental contamination and associated impacts on lake systems. Long-term exposure to environmental contaminants can cause genetic adaptations in exposed populations of aquatic organisms. The new research fields of evolutionary toxicology and resurrection ecology offer powerful tools for the investigation of changes in sensitivities and adaptive trajectories of populations exposed to contaminants and environmental stressors over decades to centuries. Dormant resting eggs produced by *Daphnia* species (Crustacea: Cladocera) as a result of unfavourable environmental conditions are archived in sediments and can be dated and hatched to produce clonal lineages (i.e., same genotypes) of historical populations. This talk will present an overview of the evolutionary tools available and their current and potential use in toxicological investigations. Additionally, we will present our preliminary research, which examines how genotypes of clonal lineages of *Daphnia* species from single populations, separated through generations of evolution, differ in their response to exposure of stressors. Results will provide insight into the sensitivity and fitness of organisms in response to environmental contaminant exposures and the micro-evolutionary adaptations of genes that evolved in response to changing environments. Furthermore, evolutionary change in aquatic organisms can also result from other environmental stressors, such as temperature. Since increased temperatures are expected to occur in the future as a result of predicted climate scenarios, it is important to examine the fitness of historical and recent clones of *Daphnia* to temperature changes in combination with exposure of contaminants. Toxicological assessments and genomic data obtained from these archives of natural populations will provide unprecedented opportunities to gain insight into long-term and potentially future evolutionary responses of natural populations to environmental changes resulting from environmental stressors, including contamination and climate change.

WEPC05

Chemical and physical stressors shape the population genetic structure of aquatic invertebrate populations

P. Inostroza, University of Gothenburg / Effect Directed Analysis; I. Vera-Escalona, Dalhousie University / Biology; A. Wicht, Eberhard Karls Universität Tübingen / Chemistry; M. Krauss, W. Brack, Helmholtz Centre for Environmental Research UFZ / Effect-Directed Analysis; H. Norf, Helmholtz Centre for Environmental Research GmbH - UFZ / River Ecology Aquatic Ecosystems Analysis and Management

Organisms are rarely exposed to only one single stressor in the environment, but rather to multiple human-derived threats working simultaneously. Environmental pollution can modify population genetic structure via ecological bottlenecking, adaptation of species to contaminants, altered gene flow, or increasing mutation rate. Organic micropollutants such as pesticides, biocides, pharmaceuticals, personal-care products, or industrial chemicals are ubiquitous in the aquatic environment and their effects are considered a relatively new and emerging anthropogenic pressure over evolutionary processes, especially potential effects of pollutants on genetic population structure may be more disruptive regarding ecosystem functioning than individual-level effects. Despite the bunch investigations on genetic variation in wildlife, our understanding about the individual stressor effects on genetic variation is still limited. Recently, there has been an increased interest to integrate environmental chemistry and evolutionary toxicology approaches into the assessment of direct and indirect effects of anthropogenic pressures on genetic variation. To address these challenges the genetic structure of a shredder invertebrate, *Gammarus pulex*, was examined using evolutionary toxicology and body burden of organic micropollutant approaches. Exposure to chemical pollution alone and in combination with the presence of weirs resulted in a depression of allelic richness in native *G. pulex* populations. Our results suggest that the input of a mutagenic effluent from a WWTP resulted in a strong increase in private alleles over the affected populations. In addition, the presence of weirs disrupted the migration across the river and thus the gene flow between *G. pulex* upstream and downstream. This study provides strong evidence that the assessment of genetic variation including private alleles together with the contamination of mutagenic and nonmutagenic chemical pollution offers new insights into the regulation of genetic population structure and highlights the relevance of emerging anthropogenic pressures at the genetic level.

WEPC06

Histone methylation as exposure biomarker of environmental chemicals

N. Chatterjee, University of Seoul / Environmental Engineering; J. Jeong, University of Seoul; J. Choi, University of Seoul / School of Environmental Engineering

Epigenetics, phenotypic characters without modification of gene sequence, possess reversibility as well as the heritable transgenerational transfer of epigenetic marks which argues for it inclusion in environmental health risk assessment. The aim of the present study was to identify chemicals which can cause histone modification with the relevance of in vitro and in vivo model systems. To this end, will employ three model systems - EZH2 knock out cell line (CRSPR-CAS technique) and a transgenic *Caenorhabditis elegans* strain (NL2507) pkIs1582 [let-858::GFP + rol-6(su1006)], which shows demethylation of H3K9me3 and H3K27me3 marks with GFP expression in germline. We used various group of environmentally relevant chemicals, such as, heavy metals, endocrine disruptors, nanomaterials, biocides etc. A number of chemicals exhibited altered histone methylation – among them some chemicals showed species specificity while some of the chemicals exhibited conservation of histone methylation changes in both in vitro and in vivo systems. Taken together, our study showed histone methylation as a sensitive epigenetic biomarker for chemical screening and in turn risk assessment.Acknowledgement: This study was supported by Basic Science Research Program (2016R1D1A1B03931553) through the National Research Foundation of Korea (NRF) funded by the Ministry of Science, ICT and Future Planning

What's your take on communication? Don't Panic! Reports on how to accurately communicating science and risk (PC)

WEPC07

Dangerous misconceptions - Consumers need help!

U. Klaschka, University of Applied Sciences

Previous surveys revealed that average consumers and even more illiterate persons are struggling with risk communication instruments for harmful substances in commodities. The majority of consumers do not understand risk communication instruments as intended by legislators or do not use them at all. In contrast, the present survey focused on ‘best-case’ consumers who are interested in the topic, have a good education or a good self-reported knowledge in chemistry. These ‘best-case’ consumers use preferentially hazard pictograms in accordance with the Regulation on Classification, Labelling and Packaging (86%), reports in the media (80%) and information printed on the products (77%) to learn about harmful substances in consumer products. Surprisingly, smartphone applications (< 10%) and information by authorities (14%) were not indicated as frequently used information sources. Most respondents considered information published by consumer and environmental organizations (75%) and the hazard pictograms (74%) as trustworthy. Interestingly, the respondents considered legislators (94%), consumers (75%), manufacturers (71%) and environmental and consumer organizations (61%) as responsible for risk reduction. It is alarming that many of these ‘best-case’ participants assumed that food (up to 62%), products with an environmental label (36%), natural personal care products (36%), homeopathic products (30%), natural pharmaceuticals (26%) and products without hazard pictograms (11%) would not contain substances harmful for health or the environment. Nearly one out of ten respondents did not know that consumer products can contain harmful substances. These results show that motivation and knowledge in chemistry help, but are not sufficient. Consumers need support to understand risk communication instruments, they need support to understand which products might contain harmful substances, they need support to determine the impact of harmful substances in commodities and they need support for suitable risk reduction behavior.

WEPC08

The European Union Observatory for Nanomaterials (EUON): A new platform for communicating information on the safety of nanomaterials

A. Sumrein, European Chemicals Agency (ECHA); J. Holmqvist, T. Aitasalo, European Chemicals Agency ECHA; C. Constantin, ECHA-European Chemicals Agency / Evaluation Directorate; L. Walin, European Chemicals Agency ECHA; L. Deydier Stephan, European Chemicals Agency / Evaluation Directorate; A. Elwan, V. Amenta, European Chemicals Agency ECHA

The REACH and CLP regulations are two key regulations addressing the manufacture and use of chemicals and the impact of these chemicals on human health and the environment. Although these regulations do not contain explicit requirements for nanomaterials, the regulations nevertheless address all chemical substances, including nanomaterials. In addition, over the past years, significant research has been conducted on the safety of nanomaterials. In spite of this, there is a perception that there is insufficient information available in the public regarding the safety of nanomaterials. As a result, the European Commission entrusted ECHA with the creation, management, and maintenance of the European Union Observatory for Nanomaterials (EUON) [1] via a delegation agreement in December 2016[2]. The aim of the Observatory is “to give objective and reliable information on markets and safety aspects of nanomaterials in the EU market”. The presentatoin will provide an overview of the activities of the EUON, including the background, the current content of the Observatory, and planned future developments. \n

WEPC09

Roadmap for the unknown

M. Luitwieler, M.H. Wagelmans, Bioclear earth

The main environmental themes have been addressed in the last decades. Think about acidification, nutrients and bulk industrial chemicals for which environmental guideline values have been derived within legal frameworks. changes are ongoing in the scale level at which environmental problems are regarded as well as the scale level of industrial production. in the past large volumes of bulk chemicals were produced, now and in the future lower volumes of more specialised compounds are and will be produced. that means that more and more compounds will enter the environment in low volumes. also time-to-market of new developments and technologies decrease which leaves less time for a thorough risk assessment. last but not least, techniques for measuring compounds are improving. More and more compounds can be measured in increasingly low concentration while the risks of these compounds in low concentrations are not known or just being studied. For the Province of Groningen en the Water Company Groningen these were the resons to develop a policy for emerging contaminants in the environment with the main question: What to do when emerging contaminants are found in soil or water. Bioclear earth has developed a roadmap for policy and/or actions by the province, municipalities, water company and water boards for emerging contaminants in the soil-water system. Our framework was: compounds and organisms that enter the environment or can be spread by human actions and that potentially have a negatove effect on humans, nature or agriculture and for which no guideline values have been derived within the Soil Law, Water law (Water framework directive) and Drinking water decree. The first step in the roadmap is to determine if a compound or organism in the environment can cause an actual or large risk. If no risks are present, no further investigation is needed. In all other cases, the roadmap needs to be followed. In the presentation we will describe the process that has been followed to come to this roadmap as well as the background information. In the roadmap we describe the role of different stakeholders, including communication, enfrocement, measurements to further prevent contamination or spread. In the presentation these roles will be further highlighted. Additionally we will organise a workshop regarding to licence to discharge in January for province, municipalities, water boards, water company and RWS. The results of this workshop will also be highlighted.

WEPC10

EVOKED: enhancing the value of climate data - translating risk and uncertainty utilizing a Living Labs approach

A.M. Oen, Norwegian Geotechnical Inst. / Environmental Technology; B. Kalsnes, Norwegian Geotechnical Institute / Natural Hazards; L. Van Well, M. Zetterlund, Swedish Geotechnical Institute; G. Ellen, R. van der Brugge, DELTARES; J. Koerth, B. Vollstedt, Christian Albrechts University of Kiel

The impacts of climate change are broad and although much focus has been on disaster risk reduction and coastal management, climate change will also have consequences for environmental management where the transport of contaminants, organism acclimation and vulnerable communities will be important to consider for future human and ecological risk assessments. In this context it is useful for environmental scientists to be familiar with the concept of climate sercvices which are defined as the transformation of climate-related data into products (trends, assessments, best practices) in relation to climate that may be of use for the society at large. Since the climate service sector is relatively new, there is a need to engage knowledge providers, users, and translators to identify improvements to climate services through co-design, co-development and co-evaluation. The EVOKED project aims to address this challenge by re-framing the risk and uncertainty associated with climate data into knowledge products more understandable and useful for end-users concerned with risk mitigation and adaptation. The project team will engage end-users in a Living Labs approach to ensure a user-contribution innovation methodology at established case study sites in Norway, Sweden, Germany and the Netherlands. The Living Lab methodology has emerged in recent years as a form of experimental and potentially inclusive mode of urban planning. Although the scope and character can vary depending on the issue at hand, the institutional level and the scope of the problem, the general idea is to involve a range of committed stakeholders in a real-life “laboratory” setting to test and develop alternative solutions for complex challenges, such as climate adaptation. The first activity for the Living Labs at each case study site will be a co-design process to encourage stakeholders to share their perceptions of risk and uncertainty. Since there are many different definitions and interpretations of risk, understanding the local perceptions of risk is a prerequisite for communicating risk. Thus, EVOKED supports the development of the field of climate services to improve our capacity to manage climate-related risks.

WEPC11

Communicating monetary values of environmental impacts - case studies related to ISO DIS 14008

B. Steen, Chalmers University of Technology; K. Hallberg, AkzoNobel; P. Hanarp, Volvo Group; J. Lindberg, IVL Swedish Environmental Research Institute; E. Riise, Essity; M. Romare, IVL Swedish Envirionmental Research Institute; T.

Rydberg, IVL Swedish Environmental Research Institute; A. Wikström, Chalmers University of Technology
Monetary valuation of environmental impacts from human activities is a way of understanding and communicating its societal significance. However, monetary values are easy to accept without knowing the many ways they can be determined and the many perspectives they may represent. ISO TC 207/SC1 has set up a working group to develop a framework standard on monetary valuation of environmental impacts and related aspects (=emissions and use of resources) to increase transparency and its use in management. The standard contains requirements and recommendation on how to document and report information (metadata) about what a monetary value represents and how it is developed. As a part of the Swedish contribution to the work, three case studies were made to test the standard approach and to find out which metadata that had the greatest influence on decisions made. The cases represented three choices of ways to produce energyware, one between natural gas and waste and heat from waste incineration, one between different vehicle propulsion techniques, and one between different ways of sludge treatment and energy recovery. We have used the EPS 2015dx method to value emissions and resources and a national Swedish database used for cost benefit studies. The results indicate that the most important metadata to report is the system boundaries of the impact valuation i.e. which impacts on which environmental goods and services that is included in the valuation. The system boundaries of impacts may vary in time, and object that is valued. The object may be chosen anywhere in a cause- effect chain. System boundaries also exist for the population whose values are assessed, and for the emissions and resources used. Other metadata that has a large influence on the choice is discounting rate and other assumptions relating to future conditions. The cases, where the alternatives mainly differ due to more or less use of fossil fuel is rather insensitive to how the valuation was made with respect to the ranking of alternatives. In cases where there is a trade-off between use of fossil resources and scarce metals, the choice is very sensitive to the temporal system boundary of the impacts and affected population. A database format is proposed for documenting monetary values and related metadata.

WEPC12

Full STEAM Ahead: Merging Science and Communications to Investigate Environmental Questions

G.K. Bielmyer-Fraser, Jacksonville University / Chemistry; A. Kent-Willette, Jacksonville University / Communications; M. Simmons, Jacksonville University / Biology and marine sciences
This project involved a case study and best practices surrounding successful STEAM interdisciplinary research. The collaboration was fostered through events at the campus CTL developing into a two year research agenda. The study focused on graduate and undergraduate interdisciplinary research in the fields of Environmental Science and Communications. Specifically, the influence of changing land use along the lower St. Johns River, Fl. was investigated, and the project and resulting data were publicized using modern communication tools such as social media, in tandem with more typical scientific means such as presentations at meetings and journal publications. The presenters will discuss how the collaboration lead to grant applications and ultimately secured funding, successfully incorporated service learning and research opportunities for students, pursued and communicated meaningful research and managed teaching across very different disciplines.

WEPC13

Let's go visual, a picture is worth a thousand words: How to explain Emerging Contaminants using animations

N. Ospina-Alvarez, S. Schneider, University of Potsdam / Institute of Earth and Environmental Sciences
The development of new technologies has enhanced the use of several elements in information and communication technologies, semiconductors, electronic displays and ‘green energy’ related technologies. Platinum, indium, thallium are good examples of those kind of elements, that during long time were laboratory curiosities but that now have an important place as raw materials in high-tec products (optics, electronics, medicine). However, many of these elements are toxic (*e.g.* thallium), during the mining processes of its extraction can be transferred to the water, increasing its availability in the environment. Basic research about Technology-Critical Elements (TCE) and Emerging Contaminants (EC) is needed, but also is part of the research process to transfer this knowledge to a general public. Within this framework, the University of Potsdam and GeoEd (http://geoeducation.de/) started a pilot project to develop teaching and learning material related to emerging contaminants in the environment. In this presentation, we will show how a complex topic, can be easily included in modern science classes, going from a general concept (TCEs and EC) to a particular study case (phytoextraction of thallium from soils using mustard plants). All the material produced implements the Open Educational resources (OER) concept, which aims towards free access, documents with open license and media useful for teaching, learning, as well as for research purposes. The OER concept allows to new initiatives and projects, produce educational material accessible without time-wise or spatial barriers. **Acknowledgment:** This project is supported by an Outreach Grant of AXA Research Fund (Paris, France) and the Research Focus of Earth Sciences (RFES), University of Potsdam (Germany). **Key-words:** Emerging Contaminants, Technology-Critical Elements, raw materials, science animations, outreach

WEPC14

Improving transparency, consistency and efficiency of ecotoxicological teaching: development of an open online textbook Environmental Toxicology

C. van Gestel, Vrije Universiteit Amsterdam / Ecological Science; N. van Straalen, Vrije Universiteit Amsterdam / Department of Ecological Science; T. Hamers, VU University Amsterdam, Institute for Environmental Studies (IVM) / Department of Environment and Health; S. Moes, Vrije Universiteit Amsterdam / UBVU; M. Kraak, University of Amsterdam / IBED-FAME; J. Parsons, University of Amsterdam / IBED-ELD; S. Droge, University of Amsterdam/IBED Institute / IBED; J. Hermens, Utrecht University / Institute for Risk Assessment Sciences; M.G. Vijver, CML Leiden University / Conservation Biology; N. van den Brink, Wageningen University / Dept of Toxicology; A.M. Ragas, Radbound University / Department of Environmental Science; A. Löhr, F. van Belleghem, Open University
Although several textbooks are available, teaching environmental toxicology in general seems to suffer from a lack of a well-elaborated, up-to-date and consistent textbook that covers all aspects of the field. As a consequence, every university is developing its own training materials in addition to a textbook, but only little of this material is available online. And if materials are online, they are not consistent, lack novelty or do not cover the entire field. A Dutch consortium therefore took the initiative to develop an open online textbook on Environmental Toxicology that should cover the field in its full width, including aspects of environmental chemistry, ecotoxicology, toxicology and risk assessment. The initiative is sponsored by the Netherlands Ministry of Education. The project aims at developing an online open access book on Environmental Toxicology that is useful for training at BSc, MSc and higher levels. The book will be designed in a modular way, each module having a clear training goal/attainment level and flagged with a number of keywords. The book will also contain tools for self-study and training, like exercises and questions. With the book, we aim at improving quality, continuity and transparency of the education in environmental toxicology. We also want to make sure that fundamental insights on fate and effects of chemicals gained in the past are combined with recent approaches of effect assessment and molecular analysis of mechanisms causing toxicity. To guarantee quality of the book and associated training materials, we aim at having 1-2 authors for each module and also 1-2 reviewers from outside the team of authors. In addition, an advisory board will be involved in supervising the project, as well as educational advisors, while the project team will serve as an editorial board. The project team, consisting of environmental toxicologists and chemists from six Dutch universities, does not possess all expertise to cover the width of the field. We therefore solicit contributions from as many colleagues as possible from within the SETAC community. With that, we hope we can produce a book that is written and supported by SETAC, that is covering the entire field, and is useful for training within e.g. the SETAC Europe Certified Risk Assessor (CRA) programme. The publication as an open online book will allow continuous updating of the book, providing a possible role of SETAC in sustaining the book.

WEPC15

Policy learning through professional forums in the field of environmental toxicology: What role for the Society of Environmental Toxicology and Chemicals (SETAC)?

M. Mondou, McGill University - Macdonald Campus / Dept Natural Resource Sciences; G. Hickey, McGill University - Macdonald Campus / Natural Resource Sciences; S. Maguire, McGill University; D. Crump, Environment and Climate Change Canada / National Wildlife Research Centre; M. Hecker, University of Saskatchewan / School of the Environment & Sustainability and Toxicology Centre; N. Basu, McGill University / Faculty of Agricultural and Environmental Sciences
Leveraging more than a hundred years of experience and knowledge (Gallo 2008), modern toxicology has crystalized in a set of highly codified and standardized practices that are used by industry and regulatory agencies internationally to assess the risk of chemicals to the environment. Toxicity testing methods using whole animal studies have long provided the general framework of instrumental beliefs concerning the most appropriate way to pursue the goals of environmental toxicology. While such *in vivo* methods are useful for determining the acceptable levels of single chemicals in the environment, they have a number of limitations that are broadly accepted within the profession. Most significantly, conventional *in vivo* testing methods are very expensive and time-consuming, relying on large numbers of animals to deliver confidence in the results. Such recognized shortcomings have led to sub-optimal policy outcomes that are increasingly being acknowledged by all stakeholders. For example, the general public and regulatory agencies are being forced to overlook the potential effects of most chemicals in the environment due to a lack of data. On the other hand, the chemical industry is being frustrated by slower access to potentially lucrative markets. Beyond human welfare concerns, the welfare of test animals is also a major consideration, particularly for groups mobilized around the issue of animal rights. Alternative testing methods are being increasingly available, such as using *in silico* computational models and *in*

vitro cell- or genomics-based testing strategies (Waters and Fostel 2004). For more than a decade, these alternatives have been discussed and debated in a range of high profile forums (National Research Council 2007) as offering potential answers to the various challenges facing chemical risk assessment. However, the accepted regulatory approaches to determining the risk of chemicals in environmental toxicology have remained, for the most part, unaffected. This poster explores the role of SETAC in policy learning using primary survey data collected from participants in previous SETAC forums. We will summarize the instrumental and core policy beliefs concerning alternative testing methods of respondents and assess their self-reported policy learning experiences at SETAC. We will then consider the significance of SETAC as a professional forum through which policy actors learn and adapt to emerging challenges in regulatory science.

WEPC16
SETAC Science and Risk Communication Interest Group
T. Seiler, RWTH Aachen University / Ecosystem Analysis

Thinking green and circularly about microparticles, nanomaterials and composite materials: approaches for recovery, recycling and reuse (PC)

WEPC17
Biochar-mortar composites for construction materials
S. Oh, T. Seo, University of Ulsan / Department of Civil and Environmental Engineering; Y. Seo, University of Ulsan / Civil and Environmental Engineering
Feasibility of biochar for construction material was examined through synthesis of biochar-mortar composites and evaluation of their construction and environmental properties according to mixing ratios. Characterization of biochar-mortar composites showed that 3-5% biochar inclusion did not significantly change their flowability, compressive strength, and thermal conductivity. As biochar content increased in biochar- mortar composites, benzene concentration in air was accordingly reduced, suggesting that biochar may be favorable to remove toxic contaminants causing sick building syndrome. Toxicity characteristics leaching procedure (TCLP) and Micotox® bioassay tests showed that biochar-mortar composite were not toxic. Our results suggest that biochar-mortar composites may be promising environmental-friendly materials for building and infrastructure construction area.

WEPC18
Complex Formation Trends of Ligand Binding toward In(III) and Ge(IV)
D. Wondrousch, G. Schuurmann, Helmholtz centre for environmental research - UFZ / Department of Ecological Chemistry
In recent years, the demand for strategic elements such as Indium and Germanium has increased due to strong global economic growth, especially in the realm of semiconductors. Dwindling resources and growing demand necessitate new recycling strategies and the reassessment of existing repositories in the light of newly developed technologies. In this context, QSAR methods can be utilized in the development of chelating ligands designed for high affinity toward strategic elements. Through this, novel ligand concepts can be rapidly assessed and synthesis can be prioritized toward promising ligands, resulting in a shortened development cycle and reduced research costs. In our quantum chemical study, we analyze a systematic set of chelators with respect to their complex formation energies toward selected In^{3+} and Ge^{4+} complexes. Following a first principles approach, both Density Functional Theory and higher levels of theory have been used for the calculations, also addressing bulk solvation effects. The study focuses on both affinity and selectivity. General trends in binding affinity to selected ions are discussed as related to the electronic structure of the compounds. Chelator selectivity toward In^{3+} and Ge^{4+} is investigated in comparison to Fe^{2+}, Fe^{3+}, Cu^{2+} and Zn^{2+}. The importance of both properties arises from expected high concentrations of these interfering ions relative to the strategic elements of interest. Financial support from the Krüger Research School “Biohydrometallurgical Center for Strategic Elements” BHMZ (Nr. 02110205) is gratefully acknowledged.

WEPC19
Cellulose Nanofibers as building blocks for innovative materials for remediation
A. Fiorati, INSTM local unit @ Politecnico di Milano / Department of Chemistry, Materials and Chemical Engineering G. Natta and INSTM Local Unit; A. Graziano, L. Melone, Politecnico di Milano / Department of Chemistry, Materials and Chemical Engineering G. Natta; I. Corsi, University of Siena / Physical, Earth and Environmental Sciences; G. Grassi, University of Siena / Department of Physical Earth and Environmental Sciences; N. Pastori, Politecnico di Milano / Department of Chemistry, Materials and Chemical Engineering G. Natta; C. Punta, Politecnico di Milano
From the point of view of circular economy cellulose is one of the most interesting biopolymer since it derives from renewable sources which, thanks to its peculiar structural properties, it is widely used in the design of simple and advanced materials for different applications. Recently, cellulose nanofibers (CNF) were proven to be versatile building blocks for many preparations. The regioselective oxidation of the C6 primary hydroxyls of the anhydroglucose units, catalysed by TEMPO in the presence of the NaBr/NaClO oxidant system, leads to the formation of TEMPO-oxidized CNF (TOCNF), confering to the cellulose many interesting properties.[1,2] We recently report the synthesis of sponge-like nano-structured materials by cross-linking TOCNF and branched polyethyleneimine (bPEI).[3] These materials were fully characterized from a chemical, structural and mechanical point of view. Quantitative information on their inner microstructure were collected by Micro-Computed Tomography (μ-CT) analysis.[4] In addition the material can be easily modified in order to introduce additional chemical or structural properties. As an example, by functionalizing the bPEI with pNO_2-phenyl-urea units it is possible to obtain a material that can be employed for the heterogeneous and selective sensing of fluoride anions.[5] Furthermore, the addition of citric acid (CA) as co-crosslinker enforce the mechanical and structural performances.[4] Another application of bPEI-TOCNF sponges is the controlled release of active principles. They have been used it for adsorption and release of two model drugs, amoxicillin and ibuprofen. The material showed very good performances in adsorbing both model drugs (~200 mg g^{-1}) from methanol solution. Interestingly, the presence of CA led to slower kinetic release in aqueous environments if compared with materials obtained without CA.[4] The ongoing NanoBonD project is focused on the use of these sponge-like materias for soil and water remediation. We have demonstrated that these materials exhibit superb performances in removing contaminants both from fresh and from sea water. In particular we are able to remove heavy metals (Zn^{2+}, Cd^{2+}, Pb^{2+}, Cr^{3+} and Cu^{2+}) and organic contaminants (*e.g.* perfluorooctanoic acid).[3,4] The processed waters have also been evaluated from an ecotoxicological point of view by testing their performances towards exposure to marine and freshwater species such as algae and bivalves, hence the definition of eco-friendly materials.

WEPC20
Zn-Al layered double hydroxides: a promising eco-friendly engineered nanomaterial
R. Martins, Department of Biology, University of Aveiro / Department of Biology and CESAM; J. Figueiredo, University of Aveiro / Biology; A.M. Soares, University of Aveiro / department of Biology & CESAM; J. Tedim, University of Aveiro / Department of Materials and Ceramic Engineering CICECO; S. Loureiro, Universidade de Aveiro / Biology
Layered double hydroxides (LDH), also known as anionic nanoclays, are a class of inorganic engineered nanomaterials with a plate-like structure featuring a lateral size of 20–40 nm. LDH are characterized by positively charged metal hydroxides (e.g. Zn^{2+}, Al^{3+}), stabilized by anions (e.g. NO_3^-) and water molecules between layers. LDH have remarkable physico-chemical properties, namely anionic intercalation and exchange capacity, controlled release capacity, high specific surface area and stability. The safe-by-design structure and composition as well as the properties of Zn-Al LDH have led their use in several industrial and material engineering greener applications, as well as in medicine and pharmaceutics for a safe controlled release of drugs. Despite LDH have been regarded as having low toxicity in humans/mammals, little is known regarding the effects in other organisms (Avelelas et al. 2017; Martins et al. 2017). Martins et al. (2017) showed no acute effects on marine clams (till 100 mg/L), but significant biochemical effects, even at low exposure concentrations. The present study aimed to assess the hazard of Zn-Al LDH in several marine species representing different trophic levels. Exposure tests were carried out with 15 species, including bacterium (*Vibrio fischeri*), cyanobacterium (*Arthrospira maxima*),microalgae(*Isochrysis galbana*, *Nannochloropsis gaditana*, *Phaeodactylum tricornutum*, *Tetraselmis chuii* and *Thalassiosira pseudonana*), rotifer (*Brachionus plicatilis*), bivalves (*Cerastoderma edule* and *Mytilus* galloprovincialis), polychaete (*Hediste diversicolor*), crustaceans (*Acartia tonsa*, *Artemia salina* and *Palaemon varians*) and sea urchin embryos (*Paracentrotus lividus*). Acute and short-term chronic exposure tests followed standard or well described guidelines, with appropriate adaptions in some cases. Exposure concentrations ranged between 0.01 and 100 mg/L depending on the species tested. The nanomaterial exhibited no toxic effects in most of the tested species, even at the highest exposure concentration. However, adverse sub-lethal effects, such as changes in the enzymatic activity or the physiological endpoints were noted in bivalves and bryozoans. The predicted no-effect concentration (PNEC) of Zn-Al LDH for seawater was set at 0.2 µg/L, based on the lowest NOEC available (Martins et al. 2017). The results suggest that Zn-Al LDH is a promising engineered nanomaterial featuring a very low environmental hazard.

WEPC21
Studying microfibre release from textiles towards improved clothing design
R. Johansson, Helly Hansen; S. Kubowicz, SINTEF Materials and Chemistry; I. Yousef, S.W. Haugen, Helly Hansen; A. Booth, SINTEF Ocean / Environmental Technology
Clothing manufactured from synthetic fabrics such as polyester and nylon can release hundreds of thousands of microfibres to waste water systems when washed in domestic washing machines. Fleece fabrics have been of particular focus, however, fleece clothing can be made from a wide variety of different fabric constructions, which may exhibit different microfibre release characteristics. Mechanical and chemical finishing of the yarn and fabric will influence the size and

volume of the microfibers released. In the current study, we assess the release of microfibres from different fleece fabrics with the aim of identifying production methods and fabric properties that release the fewest microfibres during domestic washing machines. A filter unit consisting of a 300 µm filter and a 100 µm filter in series was connected to the effluent pipe of a domestic washing machine. Each fleece test fabric (140cm x 90cm) was prepared by overlocking the edges to prevent loose fibres being released. The fabric samples were washed on a standard synthetic clothing program (1 hr, 40°C). Weights inside the washing machine assured same mass for each material assessed and a consistent water flow into the machine. Effluent water was collected in a clean container and a sub-sample (1 L) passed through a 20 µm filter to collect any microfibres that pass through the filter unit. Each test fabric was first washed to study release in new clothes, and then washed a second time to determine release related to structural shedding. To improve the accuracy of the results, the two washing procedures were repeated in triplicate. A rinse cycle was run between each test wash to ensure removal of any remaining microfibres from the system. The hoses connecting the filters contained residual microfibres after washing; these were collected after each wash by cleaning the hoses manually. The pre-weighed filters were allowed to dry before the mass of fibres was determined. The number of fibres was then estimated based on microscopy counting of a pre-weighed sub-sample of the microfibres. Preliminary results show that ~80-90% of fibres in the effluent water are trapped by the 300 µm filter. Furthermore, the fibre release declines with successive washings. A detailed study of the underlying mechanisms is currently ongoing and the results will be used as a basis for ranking.

WEPC22

Exploring a Potential Nanofertilizer: Effects of Silica Nanoparticles on Alfalfa (Medicago sativa)

F. Schwab, Adolphe Merkle Institute / Materials Science; M. Maceroni, Adolphe Merkle Institute / BioNanoMaterials; A. Petri-Fink, B. Rothen-Rutishauser, Adolphe Merkle Institute / BioNanomaterials Group

Nano-agrochemicals promise higher efficiency than conventional pesticides, but much has to be learned about the gain of efficiency compared to conventional products, and the risk of directly applying such new types of yield enhancers on agricultural soil. Due to the relatively low acute toxicity and high natural abundance of silica nanoparticles (SiO_2-NPs), they are highly attractive for benign-by-design strategies in agriculture. Here, we present initial results of experiments that are in the process of being conducted on a laboratory scale to compare the effects of SiO_2-NPs and conventional fertilizer and pesticide ingredients, and combinations thereof, on the agricultural legume alfalfa (lucerene, *Medicago sativa*). The SiO_2-NPs used for the experiments were ~60 nm in primary particle diameter. As reference substance for conventional pesticides, the broadband fungicide tebuconazole was tested. Seed germination and infection tests, and a plant growth test were conducted. The Si was quantified by inductively coupled plasma – optical emission spectroscopy (ICP-OES). Beneficial effects of SiO_2-NPs were found for the fungal infection and germination rates in alfalfa, while the growth rates in the seedlings transferred to and grown in soil remained largely unaffected. The results confirm the moderate protective effects of silica nanoparticles on plants that have been reported previously, likely linked to the release of orthosilicic acid ($Si(OH)_4$) acting as a phytostimulative micronutrient. The use of silica in nanoagrochemicals promises to reduce the organic pesticide burden of agricultural soil and crops. *Acknowledgement* - The authors thank the Swiss National Science Foundation (http://p3.snf.ch/Project-168187) and the Adolphe Merkle Foundation for the support and funding of the study. We thank Laura Rodriguez-Lorenzo, Dimitri Vanhecke, and Sandor Balog for helpful discussions regarding ICP-OES analytics, electron microscopy, and dynamic light scattering, respectively.

LCA and beyond - integrating sustainability and/or other dimensions to improve decision support (PC)

WEPC23

Environmental Footprint for pasta production - the PEF pasta pilot

L. Ruini, Barilla G.e.R. Fratelli Societa per Azioni; L. Laurenza, UN.A.F.P.A.; L. Marchelli, Barilla G. & R. Fratelli; P. Borla, Life Cycle Engineering

UN.A.F.P.A, representing all the European pasta manufacturers, is the main proponent of the EU pilot on PEF for pasta production. Furthermore, three Italian companies of pasta producers (Barilla, Pasta Zara and Garofalo) decided to be directly part of the Technical Secretariat. The proponents of the pilot for pasta together represent about the 30% of the total production of pasta in the European Union. The PEF pilot, while encouraging the development of sustainable production all over the supply chain from farm to fork, enhances fair competition across EU on sustainability through instruments aimed at: Setting up and validate the developments process of product group-specific rules (PEFCR), including the development of performance benchmarks; Testing different compliance and verification systems, to set up and validate proportionate, effective and efficient compliance and verification systems; Testing different business-to-business and business-to-consumer communication vehicles for PEF information in collaboration with stakeholders The secretariat sees the PEF pilot as a big opportunity for the pasta sector since there are some pasta producers that already measure and communicate the environmental impacts through voluntary certification schemes. A methodology promoted by the European Commission can encourage other producers to communicate the environmental footprint of their pasta, making PEF a tool able to increase competitiveness with important benefits for sustainable agriculture and food production. This approach would be good also for consumers. Giving people reliable and comparable information about the environmental impacts and credentials of products and organizations will enable them to choose the most resource efficient and environmentally-friendly products. During the PEF pilot, an average impact value, representative of the category of dried pasta has been obtained to allow the environmental performance comparison among different products in the same category. This benchmark impact highlighted the process hotspots as cereals cultivation, pasta production and cooking phase. All adopted rules and hypotheses in the PEFCR document have been established with the maximum clarity in order to increase the suitability and robustness of the LCA implemented in the PEF method for pasta sector. The main difficulties noticed during the pasta pilot were about the hotspots management, when the producers do not directly manage those processes.

WEPC25

Life Cycle Assessment of applying Algal Oil in salmon aquaculture; challenges for methodology and tool development

H. Bosch, DSM Nutritional Products; A. Wojciechowski, Evonik Technology & Infrastructure; M. Binder, Evonik Nutrition & Care GmbH; F. Ziegler, RISE Research Institutes of Sweden

Evonik and DSM founded the joint venture Veramaris®, introducing a new Algal Oil based omega-3 fatty acid source for aquaculture. This intracellular oil is produced in a biotechnological manufacturing process using non-marine resources. The rationale for this development is that the capacity to generate omega-3 fatty acids through fish is not sufficient to fulfill the dietary requirements of a growing population, and that many fish species used as feed in aquaculture are either fully utilized or overfished, leaving little room for expansion. Algal Oil reduces the dependency of salmon aquaculture on marine fatty acid production and fish stocks, by replacing marine ingredients with algal oil and crop-based ingredients. To illustrate the environmental impacts and potential tradeoffs of this new product, a Life Cycle Assessment (LCA) was performed. Indicators developed for application of LCA to fisheries were used in an LCA to assess the marine ecosystem impact of replacing fish meal and fish oil by Algal Oil in salmon feed. The analysis had to be performed in a separate calculation outside the LCA software, because the software does not include the required data and methods. The study demonstrated that the use of Algal Oil as a source of omega-3 fatty acids leads to a considerable reduction of impact on marine ecosystems of farmed salmon. This improvement is accompanied by an increase in impacts associated with agriculture. Current LCA methodologies do not allow weighting of these opposing effects. However, to make informed choices between the available options this would be required. Because the availability of natural marine resources is limited, the strong growth expected in salmon aquaculture requires innovative feed solutions decoupled from limited fish stocks to meet future requirements for omega-3 fatty acids. Veramaris® Algal Oil in combination with vegetable crops enables growth of salmon aquaculture that is independent of limited fish stocks. To support the choices discussed in this presentation, and similar choices, implementation of fishery impact assessment methods in LCA tools and development of weighting methodology is essential. Just as for other biotic impacts, there are also challenges of non-linearity and temporal and spatial variability connected to fishery-specific impacts, which are of a more local nature than global-scale life cycle impacts.

WEPC26

Balancing Environmental and Health Impacts of Food Production and Consumption

C. Walker, Institute of Environmental Engineering, ETH Zurich; S. Hellweg, ETH Zurich / Institute of Environmental Engineering

An individual's food choices can affect not only the magnitude of their food related environmental footprint, but can also have a direct effect on their personal health. People consuming lower amounts of meat tend to have lower environmental impacts, but it has also been found that they also tend to have lower risk of certain disease. On the other hand, people with low vegetable or fruit consumption may also have relatively lower environmental impacts, while having increased risk of disease. This study investigates the daily eating patterns of a European population sample to identify and compare each individual's environmental impacts due to their food production as well as the health impacts that can be expected due to their food consumption patterns. The Global Burden of Disease has identified dietary risk factors that have been proven to contribute to increased disease risk such as low fruit, vegetable, nut and seed, or omega-3 intake and high red meat or processed meat intake. The relationship between the environmental impacts from producing foods classified in the dietary risk factor categories are compared to the health impacts associated with consuming these foods. From this investigation, we can estimate the magnitude of the health benefits associated with additional food production, as is shown in the example of whole grain consumption. Results show that for individuals under-consuming whole grains (less than 125 grams daily), for every 1 micro disability adjusted life year (µDALY) increase in production impacts, there is a health benefit of 141 µDALYs. Similar results were found for all dietary risk categories in which under-consumption of a particular food group was considered a dietary risk. In cases where overconsumption poses a health risk, as is the case in certain meats, sodium, and sugar sweetened beverages, a 1 µDALY increase in production impacts is associated with increases in health impacts to varying degrees, ranging from 1.2 µDALYs for red meat up to 36.8 µDALYs for sugar sweetened beverages. This study found that for most of the dietary risk categories, health impacts due to consumption far outweighed the environmental impacts (measured in terms of µDALYs), however this study did not include environmental impacts as they are related to other impact categories such as ecosystem damage and resource depletion, which must also be considered to fully capture food production impacts.

WEPC27

What not to waste? Improving decision support for Food Loss and Waste (FLW) mitigation by considering food security and environmental sustainability

F. Sessa, Quantis; M. Ruth, World Business Council for Sustainably Development (WBCSD); D. Pollard, Nestlé; K. Cooper, A. Cairns, World Business Council for Sustainably Development (WBCSD); X. Bengoa, S. Humbert, M. Vargas Gonzalez, A. Ernstoff, Quantis

LCA-based methods provide evidence of the largescale environmental impacts of food production. Nearly a third of food produced is lost or wasted, meaning production impacts occur with little to no societal service. The primary focus of response e.g. the UN SDG 12.3 has been on avoiding food loss and waste (FLW) quantity—for example halving food waste by 2030. Decision-makers, thus may prioritize FLW mitigation based on quantity, economical gain, and ease of implementation. To support multi-criteria sustainability decision-making we develop a framework and perform a global screening to prioritise FLW mitigation efforts based on two material issues: environmental impacts and nutrition security. LCA-based methods quantify environmental impacts related to FLW streams, and nutrition and global burden of disease data are used to quantify nutrition security. A global screening of FAO data on food production, supply, and FLW for various food categories (e.g. grains) is performed in 15 countries**.** Results demonstrate vastly different environmental impacts and nutrition security potential associated with various FLW streams. The results suggest that there is sufficient production of most nutrients globally, suggesting that in most cases food systems do not need to grow, but need to be optimized to reduce FLW and offer appropriate regional supply.

WEPC28

ARIADNA Project. Analysing the sustainability of implementing a mandatory Deposit-Refund System in Spain

A. Bala, UNESCO Chair in Life Cycle and Climate Change (ESCI-UPF) / UNESCO Chair in Life Cycle and Climate Change. Escola Superior de Comerç Internacional ESCI; P. Fullana, B. Díaz, Universitat Pompeu Fabra UPF / UNESCO Chair in Life Cycle and Climate Change Escola Superior de Comerç Internacional ESCI; R. Colomé, Universitat Pompeu Fabra UPF / Escola Superior de Comerç Internacional; J. Ribas, Universitat Pompeu Fabra UPF; S. Ayuso, Universitat Pompeu Fabra UPF / MANGO Chair in Corporative Social Responsability; I. Mu�oz, 2.-0 LCA consultants; B.P. Weidema, Aalborg University / Department of Planning

There is currently an ongoing debate in some Spainish regions on whether the implementation of a mandatory deposit-refund system (DRS hereafter) would be appropriated. A DRS can be simply defined as a system in which consumers pay a certain amount in concept of "packaging deposit" added to the price of a product and receive the refund back when they return the used packaging. This is a system in place in some European Countries such as Germany, Finland or Denmark whereas others such as France or the UK have refused its implementation. From a regulatory point of view, the Spanish Law 22/2011 (which transposes the correspondent European Directive 2008/98/CE), clearly states that the implementation of a DRS should be based on the analysis of its technical and economic feasibility, a set of environmental, social and human health impacts and must ensure the proper functioning of the internal market. The study presented here follows this regulatory framework, analysing the sustainability of implementing this new system, combining environmental, economic and social studies in order to get an holistic picture of its feasibility. In particular, the following methodologies were used: (1) Life Cycle Assessment (LCA) for the environmental study; (2) Classic Cost Accounting for the economic study and (3) Social Footprint and Quantification of the Integrated Social Value for the social study. All of them applied using the same data, system boundaries and modelling restrictions. The study clearly concludes that the introduction of a DRS in Spain, under the studied conditions, is not advisable. This is because: a) according to several environmental indicators, although it could achive an improvement in the overall recycling rate, it carries a higher environmental impact, mainly due to the duplication of collecting systems and the increase demand in transportation; b) represents a 4.6-fold increase in the economic cost to society (11 times if we only consider the management of DRS containers); and c) from a stakeholders' point of view, it will have a significant increase both in space and time needs, and it will cause a higher Social Footprint.

Keyword Index

AUTHOR INDEX

A

B

C

D

E

F

G

H

I

J

K

L

M

N

O

Ø

P

Q

R

T

U

V

W

X

Y

Z

SETAC Europe Office

Avenue de la Toison d´Or 67
B-1060 Brussels, Belgium
T +32 2 772 72 81
F +32 2 770 53 86
setaceu@setac.org

SETAC North America Office

229 South Baylen Street, 2nd Floor
Pensacola, FL 32502, USA
T +1 850 469 1500
F +1 850 469 9778
setac@setac.org

setac.org

The Society of Environmental Toxicology and Chemistry (SETAC) is a not-for-profit, global professional organisation comprised of some 6000 individual members and institutions dedicated to the study, analysis and solution of environmental problems, the management and regulation of natural resources, research and development, and environmental education. SETAC Europe is one of the five Geographic Units of the global Society, established to promote and undertake activities of SETAC in Europe, and to support activities of SETAC in the Middle East and Africa. The Society is dedicated to the use of multidisciplinary approaches to examine the impacts of stressors, chemicals and technology on the environment. We also provide an open forum for scientists and institutions engaged in the study of environmental problems, management and regulation of natural resources, education, research and development, and manufacturing. SETAC Europe is incorporated in Belgium as a not-for-profit organisation. The Society is governed according to its articles of association and by-laws. SETAC Europe maintains its administrative office in Brussels, Belgium.

SETAC®

Environmental Quality Through Science®

Pictures: Andrea Cenni