

exposure to cells, and up to 210 min exposure period, following adsorption of benzene onto solid-phase silicone polydimethylsiloxane (PDMS) from aqueous solution, and subsequent desorption in cell media: Dulbecco's modified eagle medium (DMEM). Up to 80% benzene was adsorbed to silicone PDMS during the adsorption phase that was conducted under laboratory conditions with temperature control to 22 ± 0.52 ; hence and the desorption in 6-well plates during incubations at 37°C . Finally, the study explored the use of advanced in-vitro digestors, 3-dimensional cell culture approaches and gene expression assays to investigate the interaction between pollutants, metals and the human intestine to help set remediation objectives. Keywords: Chemical risk assessment, Bioavailability, *In vitro* test systems, Epithelial cells

Advancing our Understanding of Contaminants of Emerging Concerns Associated with Plastic and Microplastics: Identification, Analysis, Occurrence and Effects (P)

3.03P.1

The global environmental footprint of indigo denim fibers

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Microfibers are a ubiquitous contaminant group that include natural, semi-synthetic and synthetic textile fibers. While most research has focused on synthetic fibers, natural and semi-synthetic fibers are sufficiently persistent to undergo long-range transport, accumulate in the environment, and potentially cause ecological effects. Because natural fibers may be anthropogenically modified, we describe cellulosic fibers with evidence of anthropogenic modification (e.g., chemical additives, manufactured morphology) as anthropogenic cellulose (AC) fibers. Our goal was to document sources, pathways and sinks of AC fibers. We collected sediment samples from sites across the Canadian Arctic Archipelago, sediment from inland Canadian lakes, sediment and fish from the lower Laurentian Great Lakes, as well as WWTP effluent from plants in Ontario discharging to the Great Lakes. Material composition and surface morphology of fibers enumerated from all samples were analyzed using micro-Raman spectroscopy. An abundant AC fiber present in all samples was indigo denim fibers (6%). Inland lake and Canadian Arctic sediments contained the highest concentrations total fibers of all environmental compartments with 2520 ± 1260 fibers kg dw^{-1} and 1957 ± 1380 fibers kg dw^{-1} , respectively. The high levels in sediments may indicate that this compartment is a sink for AC fibers in the aquatic environment. The most abundant AC fiber in WWTP effluent was indigo denim (37%), as well as other textile fibers (e.g., polyester), suggesting laundering as a likely source of these fibers. To further investigate indigo denim as a source of these fibers, we washed individual pairs of new and used blue jeans, a garment that dominates the indigo denim market. In-lab washing trials showed that all fibers shed from blue jeans consisted of cotton, with most containing indigo dye (84%). New jeans (210 ± 3 fibers g^{-1} of denim) shed significantly more fibers than used jeans (130 ± 14.5 fibers g^{-1} of denim). Our results indicate a single pair of used blue jeans can release $5.6 \times 10^5 \pm 4.1 \times 10^3$ indigo denim fibers in one wash. Chemical composition and surface morphology of indigo denim fibers found in environmental compartments were consistent with those identified from blue jean washing experiments. We conclude that indigo denim garments (e.g., blue jeans), one of the world's most popular textile materials, is a source of AC fibers to the global aquatic environment.

3.03P.2

Biofouling on solid litter: what do they prefer?

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Solid litter affects coastal systems where increasing amounts of synthetic material are being deposited. Litter such as plastic persist in the environment. It can then interact with organisms as a surface to biofouling i.e. a colonization process in which organisms associate with hard substrates. Biofouling can cause fragmentation, degradation and/or sinking of items, besides transport of exotic fauna, increasing the impacts on the ecosystems. This thematic is yet understudied in salt marshes. In the Patos Lagoon Estuary (Brazil) most of salt marshes are contaminated by litter. This work evaluated the association of organisms with solid litter in different zones of a salt marsh in the Patos Lagoon Estuary. The Molhe Oeste salt marsh presents zones that differ flooding rates, which increases from the mudflat (MF) to the low marsh (LM), mid marsh (MM) and high marsh (HM). Solid litter was sampled in 30m-long transects covering all zones. A total of 227 items were analysed, with 13 groups of macroorganisms associated with these items, divided into 6 groups of vagile and 7 groups of fouling/sedentary organisms. There was no difference in biofouling (%) between zones, although

total values were higher in the lower zones as items in constantly flooded areas are thus constantly being colonized. The % biofouling was more associated with fragmented, intermediate degraded and malleable items. A contribution index was calculated to evaluate preferences for zones, type of material, fragmentation and colour of items. Most of the groups (8/13) were recorded in the lower zone, which is expected due to the nearly constant water exposure in this area. However, secondary colonizers such as algae and fungi were more present in the higher zone, which can indicate that items did not stay exposed to water long enough or that they lost colonization due to desiccation. Also, this result might indicate that these groups are more resistant to air exposure, which also avoids competition with other groups. Most groups were recorded on plastic (5/13), which might be related to the high fragmentation of these items. Regarding colour, brown items were preferred (4/13). Algae preferred transparent items, which might be related to its sunlight need. Thus, plastic items are more colonized in a salt marsh environment, as well as brown, intermediate degraded and malleable items. The preference for zones remains to be further analysed.

3.03P.3

Low cost and high-throughput: Nile red a tool for microplastic monitoring and hotspot identification.

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Beyond simple identification of either presence or absence of microplastic particles in the environment, quantitative accuracy has been criticised as neither comparable nor reproducible. This is in part due to difficulties faced in the identification of synthetic particles amidst naturally occurring organic and inorganic components. The aim of this study was to test the efficacy of Nile red for microplastic detection by systematically investigating what drives variations in particle pixel brightness (PB). Accordingly, the results showed that PB varied between polymer type and staining procedure, but remained similar among different sized particles. This data informed the use of a lower PB threshold limit of 100 a.u. that improved the detection of 4 out of the 6 polymers tested, and reduced processing time compared to unstained samples. This practical and cost-effective tool will allow for high throughput sampling needed in monitoring and citizen science whereby results need to be disseminated rapidly. Additionally, a large volume of samples may be analysed with sites of particular interest (hotspots of microplastic accumulation) then investigated further with spectroscopic techniques, to chemically confirm the polymer type; allowing laboratories without spectroscopic equipment to outsource a handful of samples for further investigation without incurring a substantial cost.

3.03P.5

Microplastics analysis of Environmental Samples using FTIR and Raman Spectroscopy

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Microplastics are particulates, roughly 20-1000 microns in size, originating from various man-made materials. These enter aquifers where they can join the food chain when consumed by filter feeders. Microplastics are chemically and physically stable, making excretion or digestion very difficult. Sample collection is often done by filtration, and the filter becomes the sample holder for further analysis. FTIR and Raman microscopy are excellent for analyzing these materials non-destructively and without further sample preparation, through either point-by-point identification or through area mapping or imaging. Data collection and analysis using the complementary techniques of Raman and FTIR microscopy is straightforward, providing particle sizes, counts and identities. This is essential for the rapid assessment of the situation and remediation efforts. We will show data collected using the Thermo Scientific DXR3xi Raman and Thermo Scientific iN10 FTIR Microscopes on reference materials and real samples. The balance between simplicity of operation and completeness of the analysis will be discussed.

3.03P.6

Screening of Plastic Additives and Organic Contaminants in Microplastics exposed to the Marine Environment

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Microplastics, defined as plastic particles below 5 mm in size, have attracted great interest and concern in the last years for their prevalence and detection in marine environments, increasing concentration over time and potential for trophic transfer with biomagnification in food chains¹. The presence of plastic debris in the marine

environment, including anthropogenized coastal areas, is a consequence of their worldwide use in most human activities (fisheries, domestic, agriculture, industry, etc.) through direct discharges which can be transported from continental areas to the marine environment (rivers, air, etc.)². Furthermore, microplastics can not only cause physical damage to marine organisms⁴ due to absorption or ingestion, but also can provide a potential pathway of exposure to organic chemicals³. Microplastics can contain two types of chemicals: polymeric raw materials (monomers or oligomers) and additives added during plastic production to enhance the performance of plastic products; and other chemicals (mainly hydrophobic organic contaminants) sorbed from the surrounding seawater^{1,2}. The aim of this work is to screen for additives and hydrophobic organic contaminants potentially sorbed to microplastics of low-density polyethylene exposed during 8 weeks in a portuary area in Mallorca (Andratx). Microplastic and spot water samples were weekly sampled and additives and contaminants were extracted by ultrasonic solvent extraction and liquid-liquid extraction, respectively. All samples were analysed by gas chromatography coupled to time-of-flight-mass spectrometry (GC-QTOF). Compounds identification was carried out with the Agilent MassHunter Unknowns Analysis B.10.00 using the SureMass deconvolution algorithm and matching against two high-resolution libraries (one in-house library of 363 compounds and the Agilent pesticides commercial library of 844 compounds) and one low-resolution library (NIST). *Acknowledgements:* The authors acknowledge the financial support provided by Galician Government (Consellería de Economía e Industria, Xunta de Galicia, ref. ED431C2017/36), by the Spanish Government (CTM2017-84763-C3-2-R, CTM2017-84763-C3-3-R, CTM2017-90890-REDT and CTM2017-88332-R) and FEDER funds *References:* [1] Hong, S.H., et al. *Anal. Methods* 9 (2017) 1361-1368. [2] Victor, M.L., et al. *Environ. Pollut.* 236 (2018) 442-453

3.03P.7

Determination of bisphenol A in sheep's urine and faeces

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There are a few published analytical methods for the determination of bisphenols in animal excreta. The objective of our work was therefore to introduce a sensitive and selective analytical methodology for determination of bisphenol A (BPA) and its main metabolite bisphenol A-glucuronide (BPA-GLUC) in sheep's urine and faeces by testing for the presence of both free (aglycone) and total (a sum of free and conjugated) BPA. Conjugated BPA was determined by an enzymatic deconjugation of the glucuronide bond using β -glucuronidase, followed by subtraction of the free BPA from the total BPA. Optimizations of the BPA analysis were made mainly regarding the enzymatic deconjugation, extraction from the matrix, concentration of the extract and chromatographic separation. A clean-up of both urine and faeces samples was performed by solid phase extraction (SPE) using two SPE sorbents, namely Chromabond HR-X and molecularly imprinted polymer (MIP) SPE Bisphenols. Urine samples were applied on the cartridges after dilution, while faeces samples were extracted first by acetonitrile. Final sample extracts were analysed for the presence of BPA by gradient reversed-phase HPLC using water and acetonitrile and/or water and acetonitrile/methanol as a mobile phase, Hypersil Gold C18 (3 μ m) analytical column, and fluorescence detection. The recovery values for determination of BPA in urine and faeces ranged from 52 to 67% and from 41 to 81%, respectively. The repeatability and within-laboratory reproducibility of the measurements, represented by the coefficient of variation (CV) values, ranged from 1.3 to 27.4% and from 8.8 to 32%, respectively. Regarding urine, the estimated limit of detection (LOD) values for determination of free and total BPA were 0.1 and 10 μ g/l, respectively, while the limit of quantification (LOQ) values determined were 0.5 and 100 μ g/l, respectively. With regard to the faeces analysis, LOD and LOQ values were 1 and 2 μ g/kg for both free and total BPA, respectively. The results obtained show that the method could be applied in toxicokinetic and environmental studies of BPA, BPA-GLUC and total BPA in animal urine and faeces and could thus contribute to the risk assessment of environmental contamination.

3.03P.8

Degradation products from novel biodegradable polymers in marine settings

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There is increasing concern about plastic in the ocean due to its longevity, its potential toxicity and its ability to harm wildlife. The oceans are where much of the waste plastic we create accumulates but are not an ideal environment for degradation because of lower temperatures and fewer microbes than in an ideal environment like compost. It can take decades for large plastic pieces to degrade in the ocean but during this time they are not inert. As they break down they can release chemical additives many of which are toxic. Biodegradable plastics could be a solution since they are designed to break down into carbon dioxide and water, however information on this degradation process is scant as is knowledge of the released chemical by-products and their impact. Four different types of biodegradable plastics (polycaprolactone, polypentadecalactone, polylactic acid,

and polyvalerolactone) were produced at the University of Hull. These plastics were made from monomers derived from plant material rather than hydrocarbons and are expected to degrade far better in the natural environment. The four plastics, a commercially produced biodegradable plastic (polylactic acid straw) and a conventionally produced plastic (polystyrene coffee cup lid) were aged in seawater for one month to discover if and how they degraded in a marine environment. Analysis by GC-MS showed some chemicals including the monomers of polycaprolactone and polyvalerolactone were released to the water from aging plastics. Biodegradable polymers released more chemicals than commercially produced plastics (polystyrene released the fewest), which could indicate faster degradation. Chemicals released included hydrocarbons, ketones and esters that would be expected to result from the degradation of the plastic molecules (such as stearic acid and triphenylmethane, a possible environmental contaminant). Some chemicals used in the creation of the polymers such as benzyl alcohol were also found. Other analysis by Fourier transform infrared (FTIR) spectroscopy suggested some breaking of bonds in the weathered samples compared to the unweathered samples but that the physical structures of the plastics did not change much over the weathering period. Further experiments involving a more long-term degradation and an analysis of plant based plasticizers and toxicity tests could provide more information on whether biodegradable plastics are better for the oceans than conventional plastics.

3.03P.9

Investigation of microplastic particles in rural karst groundwater systems and their links with on-site domestic wastewater effluent

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Groundwater from karst aquifers, through springs and wells, is a major source of drinking water for one quarter of the world's population. These aquifers are exceptionally vulnerable to pollution as a result of predominantly rapid recharge of water from the surface and strong aquifer heterogeneity. Groundwater pollution is a complex problem that can be associated with a variety of sources, but human wastewater effluent and diffuse agricultural sources are generally considered among the most significant threats to groundwater quality worldwide. This is particularly true in rural and suburban areas where the primary wastewater treatment options for communities without access to centralized wastewater treatment facilities are on-site domestic wastewater treatment systems (DWTSS). The domestic wastewater is primarily discharged from toilets, washing machines, showers and dishwashers, thus, a wide range of contaminants eventually reach the environment even after on-site wastewater treatment processes. Microplastic particles, as contaminants of emerging concern, are found with other solid materials in the wastewater effluent principally due to household washing and cleaning processes. While microplastics occurrences and related ecological concerns have been well studied in marine and other aquatic environments in the past, investigations in groundwater systems are extremely rare. Toxicological and health concerns with microplastics presence in groundwater used or potentially used for human water supply are a result of ability of microplastic particles to absorb persistent organic pollutants (POPs), while the wider ecological and environmental concerns are related to springs – points at which water flows from an aquifer to the land surface affecting surface water quality and ultimately contributing to microplastic pollution in the oceans. In this study, a number of karst springs in the west of Ireland have been sampled over time for quantification and identification of microplastic particles using Fourier-transform infrared spectroscopy (FTIR) and for the analysis of fluorescent whitening compounds (FWCs; well-known indicators of human contamination since their origin is mostly from laundry detergents). Our results show a significant correlation between microplastic particle counts and detected FWCs signals at different springs, which helps to understand the contribution of household-derived contaminants to this environmental problem.

3.03P.10

First record of the occurrence and composition of microplastics in sediments in Eyjafjörður, Iceland.

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There is a growing concern regarding plastic pollution, its distribution and effects on the ecosystem, both within the scientific community and among the public. Scientists have measured microplastic (plastic fragments less than 5 mm in size) in almost every corner of the world, including in the marine environment, both in the sediment and the water column. The accumulation of the microplastic in the food web is threatening the ecosystems health but microplastic has been reported in fish and other marine organisms as well as in birds, mammals and lately in human stool in number of publications. The sewage is an important source of microplastic pollution in the coastal sea, depending on the sewage treatment, more or less of the microplastic in it ends up in the sea. This study is the one of first records of the occurrence and composition of microplastics in coastal sediments in