Faecalibacterium gut colonization is accelerated by presence of older

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Published in:
The Danish Microbiological Society Annual Congress 2018 - programme & abstracts

Publication date:
2018

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

Programme & Abstracts

ONLINE VERSION
with poster abstracts

The Danish Microbiological Society

Annual Congress 2018

60 YEARS ANNIVERSARY · 1958 - 2018 ·

12 NOVEMBER 2018

EIGTVEDS PAKHUS
COPENHAGEN · DENMARK

The Danish Microbiological Society
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Flash poster presentations

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About DMS

The Danish Microbiological Society (DMS) is a professional association in the fields of human and veterinary medical microbiology, general microbiology, food microbiology, environmental microbiology and biotechnology. DMS has existed since 1958, and is dedicated to the advancement of microbiology, both applied and basic, and promotes microbiological information to the public. These aims are achieved by organizing annual congresses, workshops and symposia - and by taking part in the current microbiological debate.

Furthermore, DMS collaborates with the Danish Pasteur Society on the award of travel grants to students and researchers in microbiology, immunology and related science.

Being a member of DMS, you are part of the advancement of microbiology in Denmark. Additionally, as a member of DMS, you are entitled to discounts at FEMS meetings (Federation of European Microbiological Societies) and for FEMS journals.

About the keynote session:

"Danish microbiology history"
Jan Sørensen, prof. emer.,
University of Copenhagen &
Søren Molin, prof.,
Technical University of Denmark

We illustrate how descriptive studies by early Danish microbiologists were gradually replaced by experimental studies in laboratory cultures and eventually by in situ studies in the field. We further document how molecular approaches, founded by Ole Maaløe and his Copenhagen School in the 1950s, subsequently made most microbiologists think molecular, even in such different disciplines as microbial ecology, medical microbiology and industrial biotechnology.

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Speakers' abstracts

Speakers' abstracts

[O1] When microbial conversations get physical

Gemma Reguera[1]

[1] *Department of Microbiology and Molecular Genetics, Michigan State University*

The prevailing chemical view of cellular life has for long limited our understanding of how microorganisms adaptively respond to environmental signals that are physical in nature. In this talk, I will provide a historical perspective of *electric microbes*, focusing on key experiments by my group that led to the discovery of microbial nanowires, microbe-microbe interactions via electrical signals and the birth of electromicrobiology, a new subfield in microbiology. Unlike chemical signals, electrical signals are subjected less to diffusion constraints, and can propagate through a wide range of media, including biogenic minerals and cells, to enable faster cellular responses and electro-syntrophic interactions. I will also discuss how we expanded these seminal studies into applied projects that exploit the electrical properties of microorganisms and their components to catalyze reactions that would otherwise be thermodynamically limited *in vivo*. Lastly, I will share with the audience new projects in my lab that explore novel adaptive responses of microorganisms to physical signals in an unsuspected environment: the human middle ear. These projects showcase my group's interest in investigating microbial adaptive responses that have only been marginally explored and the unsuspected insights that are to be gained when considering the physical nature of microbial life.

[O2] Evolution and ecology of plague: a disease of today, which changed our history

Nils Christian Stenseth[1]

[1]*Oslo University, Norway*

Plague is a disease caused by the bacterium *Yersinia pestis*. It is first and foremost a wildlife disease which occasionally spills over to the human populations. At present a couple of thousand human cases are reported. In the past plague has cased three big human epidemics, the Justinian plague (from about 541 AD), the Black Death (from about 1330 AD) and the third plague pandemic (from about 1880 AD). During the Black Death about 50% of the European human population was killed. The lecture will provide an overview of the ecology and evolution of the plague illustrating the mutual interaction between ecology and evolution (with partly a focus on the dynamics in the wildlife hosts (rodents) as well as between the wildlife host and humans and within the human population); it will discuss how the bacterium is spread from human to human (mostly by human fleas and lice); it will discuss how the bacterium most likely come to Europe in several waves during the Black Death – using both ecological and genetic data; finally the lecture will discuss how genetic changes in the bacterium changes the behavior of the fleas making it, through evolution, be as effective as possible in spreading the bacterium from one (wildlife) host to another host.

[O3] Valorisation of dairy side streams using bacterial cell factories

Christian Solem[1]

[1]*National Food Institute, Technical University of Denmark*

The dairies generate enormous amounts of whey as a by-product of cheese manufacturing. Much of the whey ends up as whey powder, or is further processed into whey proteins or lactose, however, large under-utilized streams rich in lactose are still generated. The streams often end up being used as animal feed, are used for biogas production or are simply disposed of as waste. By harnessing the chemical machinery of microbes, the waste can be transformed into value added compounds, and this is an attractive solution for generating additional income for the dairies. Here we present some of our past and ongoing efforts to come up with fermentation-based solutions for valorizing low-value dairy streams.

[O4] Yeast cell factories for more sustainable pest control in agriculture/ Engineered Oleaginous Yeast Cell Factories for Fine Chemicals

Irina Borodina[1]

[1]*Technical University of Denmark*

One of the major applications of synthetic biology is development of novel cell factories for sustainable production of bulk and specialty chemicals. The recent advances in CRISPR-based genome editing of yeast made construction of yeast cell factories cheaper and faster. These genetic tools facilitate iterative cycles of metabolic engineering, where the cellular metabolism is systematically re-wired towards higher titer, rate and yield of the target product(s). Oleaginous yeast *Yarrowia lipolytica* recently emerged as a work horse for production of acetyl-CoA and fatty acid derived metabolites. I will present examples of engineering *Y. lipolytica* for production of carotenoid feed additives and insect pheromones for environmentally friendly pest control.

Speakers' abstracts

[O5] Heterologous production of secondary metabolites in filamentous fungi

Uffe Hasbro Mortensen[1]

[1]*National Food Institute, Technical University of Denmark*

We have developed versatile methods that allows for rapid and simple cell factory construction by gene targeting. Currently, we use our methods to reconstruct and elucidate pathways for secondary metabolite production in filamentous fungi like *Aspergillus nidulans*, *A. niger*, and *Trichoderma reesei*. Our tools include stable expression platforms with matching vectors that facilitate construction of the gene targeting substrates containing the new genes. Using our systems it is possible to reconstruct secondary metabolite gene clusters in a stepwise manner, which may facilitate elucidation of the biosynthetic pathway towards formation of that secondary metabolite. Using this strategy, heterologous production of the model polyketide 6-MSA and the block-buster immunosuppressant drug mycophenolic acid, a meroterpenoid, will be presented. It is also possible to introduce entire gene clusters in one or a few gene targeting steps. Using this strategy, we will demonstrate how we functionally transfer the entire gene cluster required for production of geodin in *A. terreus* into *A. nidulans*. Our systems are compatible with CRISPR/Cas9 technology we have recently implemented in fungi. With this technology in place, we can perform marker-free integrations as well we can make deletions and point mutations in a multiplexing setup, hence, setting the stage for efficient metabolic engineering.

[O6] Shades of brown: how ice algae is melting the Greenland ice sheet

Alexandre Anesio[1]

[1]*Aarhus University*

It is now recognised that large expanses of ice in the polar and alpine regions are inhabited by active microbial communities forming one of the biomes of Earth. Microbes on ice are diverse, play an important role in the cycling of nutrients and can also modify the physical environment they live. For instance, microbial processes at the surface of glaciers and ice sheets can lead to the accumulation of labile dissolved and particulate organic carbon and this in turn have consequences to the delivery of nutrients to downstream ecosystems. Furthermore, the accumulation of cells often seen at the surface of the ice results in 'biological darkening' of glacier surfaces. The ice algal community on the Greenland ice sheet (GrIS) is dominated by *Mesotaenium berggrenii* and *Ancylonema nordenkioldii,* and the presence of these algae reduces the albedo of the ice surface, mostly due to a brown-purple purpurogallin-like pigment. Here, I will show that these ice algae are the dominant albedo reducing particulate in the west side of the GrIS, an area known as the dark zone, and demonstrate that light stress is key in stimulating the production of phenolic-based pigments in ice algae. We estimate an albedo reduction of the ice between 12% and 21%, depending on the algal cell abundances. Such darkening increases the amount of incident shortwave radiation available for ice ablation and is a clear contributing element to glacier thinning and wastage.

[O7] Antibiotic resistance genes (ARGs) dissemination to the Antarctic environments by anthropogenic activity

Woo Jun Sul[1]

[1]*Chung-Ang University, South Korea*

Soil is an important environmental reservoir of antibiotic resistance genes (ARGs), which are increasingly recognized as environmental contaminants. Methods to assess the risks associated with the acquisition or transfer of resistance mechanisms are still underdeveloped. Quantification of background levels of antibiotic resistance genes and what alters those is a first step in understanding our environmental resistome. Toward this goal, 62 samples were collected over 3 years from soils near the 30-year old Gondwana Research Station and for 4 years before and during development of the new Jang Bogo Research Station, both at Terra Nova Bay in Antarctica. These sites reflect limited and more extensive human impact, respectively. A qPCR array with 384 primer sets targeting antibiotic resistance genes and mobile genetic elements (MGEs) was used to detect and quantify these genes. A total of 73 ARGs and MGEs encompassing eight major antibiotic resistance gene categories were detected, but most at very low levels. Antarctic soil appeared to be a common reservoir for seven ARGs since they were present in most samples (42%-88%). If the seven widespread genes were removed, there was a correlation between the relative abundance of MGEs and ARGs, more typical of contaminated sites. There was a relationship between ARG content and distance from both research stations, with a significant effect at the Jang Bogo Station especially when excluding the seven widespread genes; however, the relative abundance of ARGs did not increase over the 4 year period. Silt, clay, total organic carbon, and SiO_2 were the top edaphic factors that correlated with ARG abundance. Overall, this study identifies that human activity and certain soil characteristics correlate with antibiotic resistance genes in these oligotrophic Antarctic soils and provides a baseline of ARGs and MGEs for future comparisons. In addition, I will introduce the newly-launched Antarctic ARGs dissemination projects supported by Korea Polar Research Institute.

[O8] Microbial responses to warming and cooling of Arctic soils

Anders Priemé[1]

[1]*University of Copenhagen*

The Arctic is warming. This leads to thawing of permafrost soil and enhanced temperatures of surface soils, which may influence emission of greenhouse gases to the atmosphere. In a laboratory experiment involving an Arctic surface soil experiencing annual thawing and freezing, metatranscriptomic data revealed only minor changes when warming from -10 to -2 °C or cooling from -2 to -10 °C. Following modest transcriptional changes one day after soil thawing, a more pronounced response was observed after 17 days dominated by a large up regulation of genes involved in protein production and an increase in abundance of presumed copiotrophic bacteria. In contrast, the abundance of presumed oligotrophic bacteria decreased probably due to an increase in fast-growing bacterivorous protozoa. Also, transcripts related to cellulose, hemicellulose and chitin degradation increased following thawing and were accompanied by a fourfold increase in CO_2 production. Overall, our experiments revealed dynamic responses of Arctic soil microorganisms to soil thawing, which may have implications for our understanding of soil greenhouse gas emissions in a warming Arctic.

[O9] Azole Resistant A. fumigatus the global perspective

Paul Verweij[1]

[1]Center of Expertise in Mycology, Radboud University Nijmegen Medical Center, The Netherlands

Aspergillus fumigatus is an important cause of fungal diseases in humans including chronic and acute pulmonary aspergillosis. The triazole class represents the main class of drugs for prevention and treatment of aspergillus diseases and are the only class that can be administered orally. The use of voriconazole has significantly improved survival of patients with invasive aspergillosis, including those with central nervous system infection. These benefits are threatened by the emergence of azole-resistance, which was first reported in 1997. Although azole resistance may develop during patient therapy, a second more alarming route is through exposure to azole fungicides in the environment. Resistance mutations that are associated with the environment have been reported worldwide and new mutations continue to emerge. Environmental resistance complicates patient management as most patients with azole-resistant invasive aspergillosis have not been treated with azoles before, and mixed (azole-susceptible and azole-resistant) infections may occur. Furthermore, most patients are culture-negative which requires direct detection of resistance mutations in clinical specimens. Recently studies indicate that azole-resistant invasive aspergillosis has a 20% higher day-42 mortality compared with azole-susceptible infection, when voriconazole was used for primary therapy. Patients that started on voriconazole and switched to appropriate antifungal therapy when resistance was detected also showed a higher mortality than those that immediately received appropriate therapy. Overall, azole resistance causes excess mortality and interventions are urgently needed to retain this class for medical treatment. Understanding how resistance develops in the environment and evaluations of interventions that prevent resistance selection are critical to reduce the resistance burden. This would require prioritization of fungal resistance research preferably by incorporating the problem in current antimicrobial resistance programs.

[O10] Azole Resistant Aspergillus in DK- epidemiology and molecular perspectives

Rasmus Krøger Hare[1]

[1]Statens Serum Institut

Azole resistant *Aspergillus fumigatus* has been detected in Danish clinical samples since 2007, yet, detailed knowledge on the overall epidemiology and burden of azole resistance is still lacking. A laboratory based study covering all Danish clinical isolates received at the national mycology reference laboratory (Statens Serum Institut) from 2010-2017 sought to address this concern. In total, 1511 clinical respiratory *A. fumigatus* isolates was included and susceptibilities, resistance mechanisms and genotypes were evaluated. The incidence of azole resistance was 2.8% (17/613) in 2010-2013 and 4.3% (24/568) in 2014-2017, while the proportion of resistant isolates were 3.5% (26/754) and 6.3% (48/757), respectively. Importantly, around 50% of azole resistance was due to resistance mechanisms originating from the environment (TR_{34}/L98H or TR_{46}/Y121F/T289A). Although the clinical information was lacking, the observed increasing prevalence of azole resistance is a serious concern, further exacerbated by the circumstances of a dominating environmental source. An ongoing environmental study, collecting daily air-samples from multiple crop fields, allow culture-independent detection of azole resistant *A. fumigatus* and may further help estimate the airborne burden of azole-resistant *A. fumigatus* in the Danish environment. Such studies, improved diagnostics and surveillance of aspergillosis in Denmark is required to help reduce the occurrence of azole resistant infections.

[O11] Azole Resistant plant-pathogenic fungi, why should we worry?

Lise Nistrup Jørgensen[1]

[1]*Dept. of Agroecology, Aarhus University*

From nature *Aspergillus fumigatus* is known as a saprophyte widespread in nature. It is typically found in soil and decaying organic matter, such as compost heaps, where it plays an essential role in recycling. *A. fumigatus* is also known as the most important species in human *Aspergillus* infection. Azole drugs are recommended for therapy. In agriculture and industry azole antifungals are similarly recommended for control of a broad range of plant pathogens or as part of material preservation. Evidence indicates that azole resistance in *A. fumigatus* from clinical samples is emerging in several European countries and that the development of resistance may in part be environmentally driven. Selection of resistance is known to take place during long term azole therapy in the clinical setting but is also seen to take place from the environment due to the use of azoles fungicides in agriculture and from anti-fungals applied for material preservation. Two specific resistance genotypes have been found in azole naïve patients and have similarly also been found in the environment. The two mutations have not been found in any of the *A. fumigatus* isolates that have become resistant through patient therapy, indicating that other sources could select for resistance. Azole fungicides used in the farming community may be applied using dipping, drenching, seed treatments or foliar application. The risk for developing of resistance from these different methods is not very well investigated, but is believed to be influenced by specific azoles sensitivity to *A. fumigatus*, the dose applied and the products residual effect. Also the specific environment (e.g. humidity and temperature) in whichit the azole-fungicides are applied and *A.fumigatus* ability to trive in a given environment. is foundn to be importance.

Fungal plant pathogens cause diseases in many agricultural and horticultural crops compromising yield and quality. Yield losses in the range of 10-30% are not uncommon. Effective fungicides have been available for more than 35 years and fungicides are today commonly used for control of plant pathogens in many crops. Azole fungicides constitute the most widely used class of fungicides for control of fungal plant pathogens world wide. Individual azoles are recommended 1-2 times per season using typically 100 to 200 g.active ingredients per ha per season in cereal crops providing control of a range of plant pathogens. Prothioconazole, epoxiconazole, metconazole, propiconazole, tebuconazole and difenoconazole are among the most potent and commonly used azoles in agriculture. In comparison with other groups of fungicides the field performances of azoles have been stable suggesting only moderate risk for development of resistance in fungal plant pathogens. Even though other groups of chemical e.g. strobilurins and SDHIs are available, problems related to resistance are so significant to these groups that azoles are preferred applied alone or in combinations with other agents in order to limit selection of resistance. Azole resistance has appeared in several plant pathogenic fungi and particularly the plant pathogen *Zymoseptoria tritici* has developed significant levels and resistance over the last 10 seasons. Three azole resistance mechanisms have been found to be important, most of which are identical to those described for *A. fumigatus* 1) point mutations in the CYP51, 2) upregulation of target gene production and 3) efflux pumps. The European population of *Z. tritici* is evolving into many molecular types, which have variable sensitivity to specific azoles, depending on the specific CYP51 mutations, efflux and overexpression. Even though the changes in sensitivity have significant impact on the field performance a total loss of azole fungicides would have major impact on the farming communities ability to control attack of plant pathogens.

[O12] Lotus japonicus and rhizobia interactions; from simple to complex associations

Simona Radutoiu[1]

[1]*Aarhus University*

Legume-rhizobia interactions are controlled by protein-carbohydrate recognition events that take place at the epidermal-soil interface. Legumes use LysM proteins to recognize carbohydrates produced by pathogens or symbionts. This suggests that an ancient recognition process has been used in legumes for evolution of elaborated mechanisms for various carbohydrate perceptions.

In *Lotus japonicus* two LysM receptor kinases, NFR1 and NFR5, initiate root nodule symbiosis after perception of Nod-factors secreted by *M. loti*, while EPR3 scrutinizes rhizobial exopolysaccharides controlling the elongation of infection threads. *Lotus* encodes several additional LysM receptors, and we have used reverse genetics coupled with *in planta* functional studies to study their role in *Lotus*. Our studies based on binary interactions identified novel components involved in carbohydrate signaling that contribute to the ability of *Lotus* to distinguish symbiotic and pathogenic microbes.

Recent analyses of bacterial taxa associated with roots of soil-grown *Lotus* wild-type and symbiotic mutant plants identified a previously unsuspected role of the nodulation pathway in the establishment of distinctive bacterial assemblages in root and rhizosphere. However, the role of soil microbiota on legume-*Rhizobium* symbiosis is currently unknown. We have employed specific members of a newly established culture collection to investigate the complex *Lotus*-*Rhizobium*-soil bacteria interactions in tailored microcosms. Our findings from these investigations based on plant and bacterial mutants will be presented.

[O13] Extending the rhizosphere microbiome – bacterial communities associated with hyphae of plant beneficial fungal biofertilizers

Ole Nybroe[1]

[1]*University of Copenhagen*

Fungi from the genus *Penicillium* colonize the rhizosphere and solubilize phosphate (P), thereby increasing nutrient availability to plants. We have isolated beneficial bacteria from *Penicillum* hyphae that promote fungal growth and P solubilization. Exploiting this positive interaction has high potential for development of biofertilizer consortia for increased plant nutrient use efficiency. However, the main drivers for assembly of hyphae-associated bacterial communities, and their functional traits, in soil remain elusive. We developed a novel baiting type microcosm to study colonization of hyphae in soil. The approach was used to investigate the impact of soil type as well as *Penicillium* species on hyphae-associated bacterial communities. 16S rRNA gene targeted analysis showed that hyphae-associated communities had lower diversity and less variation in taxonomic structure than soil communities. Besides the hyphosphere effect, the soil type had a large impact on hyphae-associated communities, whereas the effect of fungal species was visible only for few discriminative taxa and specific enriched OTUs. qPCR analysis revealed increased abundance of genes involved in inorganic P cycling and phosphonate metabolism in several hyphae-associated communities. Taken together, the *Penicillium* hyphosphere represents a unique niche, which may be a hot spot for P turnover, where soil type and fungal species together orchestrate microbiome assemblage and functionality.

[O14] Future Cropping: Microbiomics Support Danish Field Trial with Microbial Fertilizers

Inês Marques Nunes[1]

[1]*Novozymes*

New sustainable solutions to improve crop productivity are needed. Microbial fertilizers is a new technology enabling such solutions.
Rock phosphate is a finite and non-renewable resource which, together with the low mobility in soil and limited plant availability of phosphorus (P), makes a more sustainable and efficient use of P in agriculture, crucial for assuring global food security. The use of microbial inoculants such as *Penicillium* spp. and *Bacillus* spp. has shown a potential to solubilize P (Rodríguez and Fraga, 1999; Wakelin et al., 2004) resulting in an increased plant growth and root development (Hassen and Labuschagne, 2010; Leggett et al., 2015). However, these inoculants do not act in a vacuum and the interaction with the existing microbial communities naturally associated with the root under different soil nutritional status is still poorly understood.
To shed light on these interactions, Novozymes joined forces with the University of Copenhagen under the Future Cropping project to look into the impact of two microbial inoculants on the natural bacterial community of winter wheat, grown in different nutrient stressed environments.
Results show a vertical and temporal stratification of the natural root bacterial communities. Soil nutritional status also affects significantly the bacterial communities with bigger differences between organic and inorganic fertilized soils. However, there is no significant impact on the bacterial communities' structure of applying the tested inoculants under any conditions.
This study thus contributes to a better understanding of bacterial successions in wheat roots across a full growing season and how they are affected by soil nutrient status. Moreover, we show that microbial inoculants have no significant effect on the overall composition of the natural microbial ecosystem.

[O15] Clinical rationality and underlying structuring mental models

Rune Nørager[1]

[1]*IT University & Designpsykologi*

Mental models are internal cognitive representations of the world that affect the way humans reason, behave and make decisions. Our research project aims to understand the genesis and dynamic of mental models related to bacterial biofilm. We hypothesise that the in vitro experience of biofilm among researchers might have generated a shared mental model and whether such a model has led further assumptions related to in vivo contexts. Given that evidence from the scientific community shows that biofilm follows highly distinct pathways when they grow in vivo versus when they grow in vitro, may set the basis for erroneous claims, expectations and decisions related to the treatment of biofilm that is rooted in a mental model that might not adequately support both conditions.

[O16] The clinicians view

Klaus Kirketerp-Møller[1]

[1]*Bispebjerg Hospital*

The lecture will present the view of this specific clinician with substantial expertise in treating chronic wounds. Bacteria are present in all chronic wounds and it is now evident that the bacteria exist in biofilms. When we acknowledge that bacteria are present, we have to decide: Which wounds have to be treated with antibiotics and which should not. Should we take MIC in account and is it appropriate to consider MBIC?

The majority of chronic wounds belong to the subgroups venous leg ulcers, diabetic foot ulcers and pressure ulcers. Despite bacteria residing in biofilms, most of these wounds will heal with proper treatment; the venous leg ulcers must have compression therapy, diabetic foot ulcers must be offloaded and revascularized and pressure ulcers must be off-loaded. This is a paradox. Yet some ulcers do not heal despite adequate treatment and these likely harbor vicious bacteria and requires antibacterial treatment. The concept of biofilm targeted treatment have been introduced, but the concept is not well defined and the role of each element in the treatment must be evaluated.

[O17] The in vivo transcriptome

Blaine Fritz[1]

[1]*University of Copenhagen*

Studying the physiology of bacteria during human infection presents a difficult challenge. *In vitro* lab cultures or *in vivo* animal models do not necessarily reflect the true physiological state of bacteria in human chronic infections. Our research utilizes deep RNA-sequencing of tissue from human chronic infections to explore the gene-expression of pathogens such as *Staphylococcus aureus* and *Pseudomonas aeruginosa* directly within the infection. Our findings as well as those from others suggest that gene-expression during chronic infection is unique to the wound environment as opposed to *in vitro* lab cultures or animal models. In this presentation, I will highlight important physiological processes during chronic bacterial infection and how these contrast what is observed in other systems. I will also describe the power of RNA-sequencing and how modern computational techniques can be applied to further understanding the physiology of bacteria during human infection as well as challenges associated with RNA-sequencing in these types of infection.

[O18] Hepatitis E virus in food - detection methods and infectivity determination

Reimar Johne[1]

[1]*Bundesinstitut für Risikobewertung, Germany*

Hepatitis E is a human disease with increasing importance, which is caused by infection with the hepatitis E virus (HEV). In Europe, the HEV genotype 3 is most prevalent. It is widespread in domestic pigs and wild boars and can be transmitted to humans by consumption of meat or meat products prepared from these animals. However, tools for control of HEV in food are only scarcely available so far.

In order to enable the identification of contaminated food, a detection method for HEV in meat products was developed. The method is somewhat laborious, but ensures an efficient removal of PCR inhibitors present in the food matrix and guarantees sensitive virus genome detection. It was successfully validated with artificially contaminated sausages and in an interlaboratory ring trial.

A major disadvantage of the detection method is its inability to distinguish between infectious virus and those, which has been inactivated during the food production process. Assessing the infectivity of HEV is difficult because the available cell culture methods are mainly inefficient and poorly repeatable. Recently, we optimized a cell culture method, which subsequently allowed the analysis of heat inactivation of HEV. Future applications of the methods may allow the identification of "high risk" foods for HEV transmission as well as the development of methods for efficient inactivation of HEV during food processing.

[O19] Advances in molecular detection of enteropathogenic viruses in food

Anna Charlotte Schultz[1]

[1]*National Food Institute, Technical University of Denmark*

Hepatitis A virus (HAV) and in particular norovirus (NoV) are amongst the leading agents of outbreaks of foodborne illness. Their importance has mainly been established through diagnostics on patient samples combined with epidemiological studies. Reliable detection of viral presence in food samples has been hampered by low concentrations combined with their low infectious doses.
Thus efficient and sensitive methods for detection of viruses in foods are required to improve understanding of transmission routes, to investigate foodborne outbreaks, and to implement preventive measures into food hygiene legislation.
Efforts have been made to develop such tools and the first European standard (ISO 15216-1:2017) for detection and quantification of virus in foods most often involved in virus transmission has been published. Due to difficulties in propagating HAV and NoV in food samples this standard applies realtime RT-qPCR for detection of viral RNA in nucleic acid extracts obtained by separate pre-processing and virus extraction steps for each food matrix. Quality controls are built in along the entire procedure.
Using current methodologies in studies of outbreaks and field screening has led to evidence based determination of foods at risk for virus transmission and to the first EU initiated projects - import control on virus in strawberries and baseline of NoV in oysters. However, the methods do have shortcomings which are constantly targets for further development.
These include low recoveries, inaccurate quantification, information lack of virus viability and strain diversity, all aimed overcome by for example simplified extraction of viral RNA, digital PCR, infectivity or integrity tests, and NGS or metagenomics, respectively.

[O20] Association between exposure to airborne norovirus and gastroenteritis among wastewater workers

Katrine Uhrbrand[1]

[1]*National Research Centre for the Working Environment, Denmark*

An increased incidence of acute gastroenteritis (AGE) has been reported among workers at wastewater treatment plants (WWTPs). The cause is unknown but the symptoms are consistent with AGE caused by norovirus (NoV) infection. The aim of this study was to investigate if exposure to airborne NoVs is the cause of the increased incidence of AGE among WWTP workers.

Personal exposure to airborne enteric viruses was examined monthly among 14 WWTP workers during a one-year period. Individual air sampling was performed from the inhalation zone of the workers throughout a working day. Gastrointestinal symptoms were reported by workers and their stool samples screened for NoV Genogroup (G)I and GII, sapovirus, adenovirus 40/41, astrovirus, rotavirus, enteric bacterial pathogens and protozoa.

NoV genomes were detected in 47% of all personal air samples (n=106). Quantifiable levels of NoV GI and GII were found in 4.7% and 13.2% of the air samples in a geometric mean exposure level of 1423 and 243 genome copies (GC)/m^3 air, respectively. Asymptomatic infection with NoV GI was observed in one of the 14 WWTP workers in two consecutive months (January and February). Both NoV GI and GII were detected in the air sample from this worker in February, but no airborne NoV exposure was observed on the sampling day in January. However, genotyping of NoV in the air sample from February showed the exposure to be from NoV GI.4 and not GI.3, which was found in the stool sample.

The majority of WWTP workers were exposed to airborne NoV on several occasions, but exposure seldom correlated with symptoms and infections. Although NoV infection following airborne exposure to NoV was seen in one worker molecular evidence did not establish a direct association between exposure and infection.

Keynote presentation

[O21] Danish Microbiology History

Jan Sørensen[1] & Søren Molin[2]

[1]*Prof. emer., University of Copenhagen*
[2]*Prof., Technical University of Denmark*

By the 1880s Danish pioneers in microbiology had learned the basics of isolation and cultivation from Pasteur and Koch and aseptic standards and proper strain characterization soon led to better beer and dairy products and to safer diagnostics of disease. By turn of the century, further insights came from the newborn disciplines of biochemistry, genetics and virology. At this time, a new generation of pioneers developed new breeding technology in brewing yeast, disease treatment by new sera and vaccines, and application of plant growth-stimulating inoculants in agriculture. From the Second World War, a third phase of development in Danish microbiology came with the introduction of antibiotics and pesticides – for the good and the bad. First half of the presentation will exemplify important discoveries by Danish pioneers in Microbiology until the 1950s. We hear when Emil Chr. Hansen lost his temper, when Sigurd Orla-Jensen ´invented´ the holes in Emmentaler cheese, and we learn about Hjalmar Jensen who first depicted the denitrification process and about soil microbiologist Hans Laurits Jensen who authored a total of 15 scientific publications in Nature! But first of all, we illustrate how the early, descriptive studies of the microorganisms were soon replaced by experimental studies with laboratory cultures and eventually developed into field studies laying the foundation for modern microbial ecology.

After the Second World War experimental microbiology took a new direction towards addressing more fundamental biological problems. As early as 1946 the famous Lederberg and Tatum experiment demonstrated how genetic information is transfered between bacteria by conjugation, and quantitative microbiology was blooming inspired by a group of physicists who focused their attention on bacteriophages. Thanks to two pioneers – Ole Maaløe and Niels Ole Kjeldgaard – these new developments in biology came to Denmark at an early time and formed an excellent platform for an international scientific environment, which for the next 25 years dominated research in molecular biology. The dominance ended in the mid-1970'ies when gene technologies invaded Danish microbiology research, but the subsequent dispersal could for a long time be traced back to the Maaløe/Kjeldgaard laboratories. During the last 30 years we have witnessed a rapid transfer of molecular microbiology concepts and methods to associated scientific disciplines such as microbial ecology and medical/clinical microbiology. Parallel to this development, Danish biotech industry bloomed after introducing molecular methods for the design of microorganisms which can produce medicine, enzymes for technical applications, and lately also small molecules – all by sustainable fermentation processes.

Industry symposium

Time: 13.00 - 13.25 Room II

Presented by Nordic Biosite and Zymo Research

Standardizing Microbiomics – Removing Bias in Collection, Purification and Analyses

Sarah Hemmasi[1]
[1]*Zymo Research*

The field of microbiomics has developed rapidly in the past several years. However, there are concerns due to poor data reproducibility across labs. To objectively assess the performance of different microbiomics workflows, it is essential to have accessible, well-defined, and accurately characterized mock microbial community standards to serve as reference materials for optimization, validation, and controls for microbiomic workflows. Acknowledging this deficit, the scientists at Zymo Research have created a well-characterized mock microbial community to be used as a reference material for microbiome measurements. Using this microbial standard, we assessed the performance of several of the most cited DNA extraction protocols used in the Microbiomics field and the effect of various library preparation techniques for 16S and shotgun sequencing. Thus, improving all steps involved from sample collection and DNA extraction to sequencing and bioinformatics will harmonize the data generated in this rapidly expanding field of research.

Zymo Research has the goal to provide researchers the best tools for microbiome measurement to ensure standardized microbiomics workflows. The ZymoBIOMICS™ portfolio has been developed to eliminate bias across microbiomics workflows and offers a complete pipeline from start-to-finish for all your microbiome related needs.

Poster sessions

This year the poster sessions are held during two different breaks. If you have a poster at DMS, please be present at your poster during the assigned time according to your poster number.

Presentation time	Categories	Poster number
12.45 - 13.45	Bioconversion Polar Microbiology Resistant fungi Other	Even numbers
15.15 - 16.15	Rhizosphere microbiology The microbiology of chronic wounds Food and environmental viruses Other	Uneven numbers

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1. Statens Serum Institut, EPI nyt (Stor stigning i legionellatilfælde), 8. november 2017. www.ssi.dk

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P02	Microscale H2 dynamics at Nickel-Molybdenum cathode surfaces during electromethanogenesis	Bioconversion	Karen Maegaard
P03	From soil to plant protection: Exploiting secondary metabolites of Bacillus subtilis	Rhizosphere microbiology	Heiko Thomas Kiesewalter
P04	Producing PHBs from renewable sources: Using excess electricity to produce bioplastics from CO2	Bioconversion	Daniel Jensen
P05	Developing a CRISPR/Cas9-assisted Recombineering system for natural soil Pseudomonads	Rhizosphere microbiology	Morten Lindqvist Hansen
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P07	Mining the rhizosphere of Christmas trees (Abies nordmanniana) for plant growth promoting bacteria	Rhizosphere microbiology	Adriana Garcia
P08	Optimising acetate production from H2 and CO2 in a tricle-bed bioreactor	Bioconversion	Morten Kok Lund
P09	Isolating novel strains for in situ alkaline organic phosphorus mineralization	Rhizosphere microbiology	Sabrina Pittroff
P10	Construction of various microbial consortia for biodegradation based on novel dilution to extinction cultures	Bioconversion	Dingrong Kang
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Poster abstracts

[P1] Attachment of Bacillus subtilis cells to the hyphae of Aspergillus niger depends on the biofilm fibre-protein

Bodil Kjeldgaard[1], Anne Richter [2], Stevanus A. Listian[2], Valliyammai Ramaswamhi[3], Ákos T. Kovács [2]
[1]Technical University of Denmark, Kgs. Lyngby, Denmark
[2]Technical University of Denmark
[3]Friedrich Schiller University Jena

Bacillus subtilis is a well-studied rod-shaped soil bacterium capable of forming multicellular communities within a matrix. Biofilm formation depends on the genes in the *epsA-O* operon, which encode the machinery responsible for the production of exopolysaccharides (EPS). Alongside EPS, the matrix is composed of amyloid fibres of the secreted protein TasA, which is essential for structural integrity of the extracellular matrix. Hydrophobicity of the biofilms is ensured by a small protein, BslA in *B. subtilis*. Previous studies demonstrated that the genes involved in biofilm formation also facilitate root colonization of *Arabidopsis thaliana*, EPS and TasA deficient mutants are unable to produce adequate biofilm to colonize roots successfully. In addition to plant colonization, *B. subtilis* also specifically attaches to the hyphae of the black mould fungus, *Aspergillus niger*.
Here, we examined whether any of the biofilm matrix components is required for hyphal colonization of *A. niger*. Interestingly, the *tasA* mutant exhibited reduced attachment to *A. niger* hyphae which is otherwise observed for the wild-type (WT) strain. However, attachment is recovered when the *tasA* mutant is co-inoculated with WT suggesting that the mutant utilizes the secreted matrix protein from the WT. In addition, we show that mutation of the *epsA-O* operon abolished robust biofilm formation on the fungal hyphae, but single cell attachment was still occurring suggesting that the production of exopolysaccharides is not responsible for the direct cell-hypha adhesion. Furthermore, a strain with impaired *bslA* gene showed no impaired hyphae attachment.
Our results suggest that TasA is strictly required for the attachment of *B. subtilis* cells to *A. niger* hyphae.

[P2] Microscale H2 dynamics at Nickel-molybdenum cathode surfaces during electromethanogenesis

Karen Maegaard[1], Frauke Kracke[2], Jörg S. Deutzmann[2], Alfred M. Spormann[2], Niels Peter Revsbech[3]
[1]Section for Microbiology, Department of Bioscience, Aarhus University, Aarhus, Denmark
[2]Stanford University
[3]Aarhus University

Microbial electrosynthesis of CH_4 is a CO_2 capture and energy storage technology. In integrated bio-electrochemical systems, electrons become available at a cathode surface where H_2 is formed or the electrons are transferred directly to the microorganisms. However, the spatial distribution of H_2 in such systems has not previously been determined. In this study, we determined the microscale H_2 dynamics during electrosynthesis of CH_4 by *Methanococcus maripaludis* and compared the gradients with abiotic controls. An electrochemical H-cell setup with a 10-cm^2 Nickel-Molybdenum cathode was modified to allow for determination of microscale H_2 profiles with a H_2 microsensor at 50 μm resolution. The reactors were operated at constant current of -1 mA. The abiotic reactors produced H_2 at rates of 16.4±6.8 (SD, n=5) μmol h^{-1} and no CH_4 production was detected. In the biotic reactors, CH_4 was produced at rates of 5.7±3.4 (SD, n=14) μmol h^{-1} while the remaining measurable H_2 production rates were 0.3±0.4 (SD, n=14) μmol h^{-1}. In the abiotic reactors, the H_2 flux away from the cathode was 0.12±0.00 (SE, n=11) nmol cm^{-2} s^{-1} while the H_2 flux away from the cathode in the biotic reactors was 0.07±0.01 (SE, n=9) nmol cm^{-2} s^{-1}. The higher H_2 flux away from the cathode in the abiotic reactors indicates that the microbial transformations are located within <<50 μm of the cathode surface. These findings have important implications for the design of integrated bio-electrochemical systems for the sustainable production of chemicals from gases.

[P3] From soil to plant protection: exploiting secondary metabolites of bacillus subtilis

Heiko Thomas Kiesewalter, Jonas Greve Lauritsen[2], Regina Åris Schürmann[2], Akos Kovacs[3]
[1]Bacterial Interactions and Evolution, Dtu Bioengineering, Kgs. Lyngby, Denmark
[2]Technical University of Denmark
[3]Technical University of Denmark, Department of Bioengineering, Kgs. Lyngby, Denmark, Denmark

Bacillus subtilis is well-known for its ability to develop biofilm under laboratory conditions, a characteristic that is necessary for bacterial attachment in the rhizosphere, therefore important for plant root colonization. The extracellular matrix of *B. subtilis* is mainly composed of exopolysaccharides, fiber-protein, and hydrophobin. To understand the abundance, the biofilm formation ability and biocontrol properties of *B. subtilis* isolates, a comparative strain analysis of diverse soil ecosystems was performed. In Germany, the isolation focused on diverse ecosystems, whereas in Denmark, we aimed at isolating strains from soil of grasslands with a non-agricultural background as well as in close proximity of different mushrooms. Several soil isolates from different ecosystems were phenotypically characterized and genetically confirmed as *B. subtilis* strains. Importantly, the abundance of *B. subtilis* depended on the soil ecosystem used for sampling. The isolated *B. subtilis* strains showed robust biofilm development both *in vitro* and on the roots of *Arabidopsis thaliana*. In addition, secondary metabolite production was tested against various phytopathogenic fungi and bacteria. Targeted gene knockouts of the isolates highlighted that biocontrol properties of the isolates depend on 4'-phosphopantetheinyl transferase, required for production of surfactin, plipastatin and bacillaene. We propose that *B. subtilis* isolates from soil ecosystems

are promising targets to use them as biocontrol against plant pathogens with plant root colonization ability.

The project is connected to the Center for Microbial Secondary Metabolites that is supported by the Danish National Research Foundation (DNRF137).

[P4] Producing PHBS from renewable sources: Using excess electricity to produce bioplastics from CO2

Daniel Jensen[1]
[1]*Aarhus University, Aarhus C, Denmark*

Carbon dioxide (CO_2) emissions into the atmosphere and pollution from non-biodegradable plastics are major problems of today. Bioplastics derived from CO_2 could provide a green alternative to petrochemical-derived plastics as it can be produced by sustainable sources and simultaneously counteract CO_2 emissions. Using *Clostridium ljungdahlii* and *Cupriavidus necator* as biological catalysts, CO_2 can be refined to more usable high-value molecules. In this study, the synthesis of PHB bioplastics using acetate derived from bioelectrosynthesis is elucidated. Results show that PHB was produced at high carbon-efficiencies. This method of using biological catalysts to convert CO_2 and surplus electricity into bioplastics could pave the way for cheap and effective carbon utilization, moving us a step closer to becoming petrol-independent.

[P5] Developing a CRISPR/CAS9-assisted recombineering system for natural soil pseudomonads

Morten Lindqvist Hansen, Lars Jelsbak[2]
[1]*Dtu Bioengineering, Kongens Lyngby, Denmark*
[2]*Department of Systems Biology, Technical University of Denmark, Dep of Biotechnology and Biomedicine, Kongens Lyngby, Denmark*

Fluorescent Pseudomonads are producers of a variety of secondary metabolites, but little is known of how these molecules actually influence the diversity and functionality within natural microbial communities. One approach to study secondary metabolites and their function involves genetic engineering of specific biosynthetic gene clusters. Currently, methods are available for performing genetic engineering in Pseudomonads, but they are laborious and time-consuming.

The scope of this project is to develop/adapt genome editing tools based on recombination-mediated genetic engineering (recombineering) and CRISPR-Cas9 based counter-selection systems for *Pseudomonas protegens* and other members of the Pseudomonas fluorescens group. This will enable fast and streamlined construction of mutants across multiple strains of pseudomonas sampled from natural soil samples. This will further allow us to interrogate

the function of secondary metabolites and to experimentally test the potential relationships between Pseudomonas populations and their secondary metabolites, as well as the microbial diversity.
The strategy and results from the recombineering system will be presented. The current work focus on elucidating the function of an uncharacterized secondary metabolite gene cluster discovered in a *Pseudomonas protegens*.

[P6] Electron uptake from solid surfaces by two methanosarcina

Mon Oo Yee[1], Oona Snoeyenbos-West[2], Bo Thamdrup[2], Lars Ottosen[3], Amelia-Elena Rotaru[2]
[1]University of Southern Denmark, Odense M, Denmark
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Members of *Methanosarcinales,* one of the most environmentally and biotechnologically relevant methanogenic groups, were previously shown to retrieve electrons from an extracellular partner microorganism performing direct interspecies electron transfer (DIET) and were proposed to be electroactive. Nevertheless, their electroactivity has never been examined. In this study, we tested two methanogens*, Methanosarcin barkeri* and *Methanosarcina horonobensis* regarding their ability to accept electrons directly from insoluble electron donors like electrodes, via conductive particles and from other cells. Both methanogens were able to carry out DIET in respective co-cultures with *Geobacter metallireducens*. Electrically conductive particles (granular activated carbon) accelerated electron transfer in all co-cultures and favored methanogenesis interspecies electron transfer. However, only *M. barkeri* produced 2.2 times more methane with electricity as the sole electron source supplied via a cathode at – 400 mV (vs. SHE). This suggested that the electron uptake mechanisms could differ between methanogens depending on the surfaces and the partners involved. Nevertheless, the nature and growth conditions of *M. horonobensis* make it a more attractive candidate for genetic studies of methanogens participating in DIET. Lastly, a strict hydrogenotrophic methanogen, *Methanobacterium formicicum*, did not produce any methane at the same electrochemical conditions, indicating that the route of electron transfer in this study was not likely to be H_2-mediated.

[P7] Mining the rhizosphere of Christmas trees (Abies nordmanniana) for plant growth promoting bacteria

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Abies nordmanniana is a major Christmas tree species in Europe, but prolonged growth hamper their production, an early root development is important for plant development. The tree rhizosphere represents a diverse source of plant growth promoting bacteria that may influence root development however; the *A. nordmanniana* rhizosphere community remains unknown. This study aimed to characterize bacterial communities associated with roots of *A. nordmanniana* at nursery stage, and isolate rhizosphere bacteria able to regulate plant growth. Composition of the bacterial communities from bulk soil and rhizosphere of *A. nordmanniana* at different sampling sites was compared by 16S rRNA gene sequencing. There were clear differences in community composition between rhizosphere and bulk soil, and significant effect of sampling site on both rhizosphere and bulk soil communities. *Proteobacteria, Actinobacteria*, *Acidobacteria* and *Bacteriodetes* dominated in the rhizosphere. The same phyla dominated in bulk soil, but the relative abundance of *Acidobacteria* was higher while that of *Proteobacteria* was lower than in rhizosphere. Among bacterial strains isolated from the rhizosphere we identified strains able to improve seed germination, increase root development and branching when seeds were bioprimed with bacterial suspensions, 22 bacterial isolates were tested, seed germination percentage improved up to 40% when the seeds were bioprimed with strain 040, furthermore a bigger development of secondary roots and root hair was also observed. These isolates shown capability to produce auxins in pure culture. These results suggest that plant growth promoting bacteria is associated with the rhizosphere of *A. normanniana* and could enhance growth.

[P8] Optimising acetate production from H2 and CO2 in a tricle-bed

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With the prospect of a future exhaustion or abandonment of fossil fuels, CO_2 from waste and industrial off-gas will become an important resource for production of a variety of different organic compounds such as acetate. Homoacetogenic bacteria catalyze the production of acetate by gas-fermentation of CO_2/H_2, but the production of acetate is often limited by methanogens competing for CO_2 and H_2 as substrates for methane production. Since acetate is industrially more valuable than methane, finding ways to favor production of acetate over methane, could provide an industrially relevant biotechnology. This study seeks to investigate

how different media pH-values will alter the production-ratio of acetate/methane, as well as the microbial community structure, in a mixed cultured batch system enriched with CO_2 and H_2. The pH-condition in the batch experiment resulting in the most promising acetate production and methanogenesis inhibition, will be applied to a small-scale bioreactor, simulating a trickle-bed reactor design.

[P9] Isolating novel strains for in situ alkaline organic phosphorus mineralization

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Phytate constitutes the majority of identified organic P in many soil types, is inaccessable to plants, and is accumulating in soil with increasing application of P fertilizer[1,2]. While phytate is quite resistant to chemical hydrolysis in the soil, enzyme hydrolysis via a specialized phosphatse; phytase, is known to breakdown available phytate and release P. Therefore, re-incorporating phytase-producing biofertilizers into soil presents a viable and environmentally responsible way of utilizing P from the phytate pool in agricultural soils. By creating a microcosm that enriches for phytate degraders present *in situ (ie.* within the soil), we aim to harvest robust bacterial biofertilizer candidates capable of digesting oprganic phosphorus within complex and competitive soil environments. A microcosm using a calcium phytate baiting system was designed to perform *in situ* (ie. in soil) bacterial selection. Bacteria were tested on calcium phytate precipitate plates, and then further tested for enzymatic potential comparing their activity towards both sodium and calcium phytate substrates under buffered (pH7.2, pH5.5) phosphorus starvation condiditons. Bacterial isolates recruted by the microcosm were functionally competent in solubilizing and/or degrading CaPhy in the absence of acid production. Enzymatic extracts of selected strains showed preference towards insoluble calcium phytate substrate (pH7) as compared to Na-phy (pH5.5), indicating the use of a beta-propellar phytase; a phytase that funtions optimally at neutral pH and necessitaties calcium-bound phytate.

[P10] Construction of various microbial consortia for biodegradation based on novel dilution to extinction cultures

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Microbial degradation has been extensively applied from environmental remediation to industrial application. Microbial consortia as multi-members coexistence are supposed to divide labor and increase functionality during degradation. Their application presents an enormous potential for enhancing the efficiency and yield of industrial processes, particularly when dealing with recalcitrant substances that are resistant to decomposition by conventional methods. Co-occurrence networks of an efficient keratinolytic microbial consortium were investigated to reveal the microbial interactions during keratin degradation. Subsequently, dilutions series with 24 replicates of the microbial consortium were performed and characterized. 10^{-9} dilution was selected to construct a library of simplified microbial consortia based on a comprehensive comparison. Functional microbial consortia were obtained from the library and the taxonomic diversity was proved by 16s rRNA gene amplicon sequencing. A novel dilution to extinction cultures approach with four steps was summarized, which is able to select various functional microbial consortia for biodegradation. It provides an effective way to settle the potential issues of microbial consortia in practical applications.

[P11] The conformation of fibronectin determines success of bacterial attachment

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Staphylococcus epidermidis is responsible for implant-associated infections, due to its ability of forming biofilms. Attachment to the implant occurs through interactions with adsorbed host proteins. One receptor for *S. epidermidis* is extracellular matrix binding protein (Embp), which binds to fibronectin (Fn). Embp, like other bacterial adhesins, bind to host proteins soluble in bodily fluids as well as adsorbed to the surface of implants. So how can interaction with these proteins promote attachment to the surface? We hypothesized that Embp only mediates attachment to immobilized Fn, and this is due to the availability of binding domains in that conformation of Fn.
To investigate these, using immunofluorescence assay it was observed that *S. epidermidis* interacted with adsorbed but not soluble Fn. Soluble Fn is in globular, while adsorbed Fn can either remain globular, or change conformation to form fibrils. To study the bacterial

interaction with Fn in these two conformations, we produced surfaces coated with (poly)methyl acrylate (PMA) and (poly)ethyl acrylate (PEA), which adsorb Fn in different conformations. Atomic force microscopy confirmed that Fn adsorbed to PMA remained globular, while Fn adsorbed to PEA fibrillated. We then quantified Embp-mediated bacterial attachment to the two surfaces, using *Staphylococcus carnosus* expressing recombinant Embp. Fibrillar Fn promoted bacterial attachment while globular Fn did not. This result supports hypothesis that adsorption-induced conformational changes dictate if a host protein promotes or prevents bacterial attachment to an implant surface. Our results add a new layer to the considerations made in materials design for implant materials that prevent biofilm infections.

[P12] Searching for Novel Photosynthetic Bacteria in the High Arctic

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Photosynthetic bacteria are among the earliest inhabitants of our planet Earth. They have been demonstrating an amazing capability to adapt to various extreme environments, including the High Arctic, by harvesting sunlight as a supplementary energy source when thriving in those harsh conditions. In this study, we asked two questions: (1) *Are there any novel diversity of photosynthetic bacteria awaiting discovery in the High Arctic ecosystems?* (2) *How the unique light conditions in Arctic have shaped the genomic features of those phototrophs?*

During the years of 2017 and 2018, we collected water samples from of three lakes in Kangerlussuaq and Nuuk, snow/ice/soil/stream samples from the VRS and ZERO research stations in Greenland, and permafrost soil samples from Svalbard. Bacteria were isolated using 1/5 R2A media under 10 °C or RT for up to eight weeks and then were screened for bacteriochlorophylls signal with an infrared imaging system. A collection of >500 photosynthetic strains was established in the lab. Our results show that a highly diverse community of photosynthetic bacteria present in Arctic ecosystems, making up 10-30% of total culturable bacteria.

We found 11 genera in Proteobacteria where phototrophs were first described, including Cereibacter, Loktanella, Methylobacterium, Methylovirgula, Tabrizicola and Tardiphaga (Alphaproteobacteria), and Chitinimonas, Ideonella, Iodobacter, Massilia and Undibacterium (Betaproteobacteria). We sequenced representative species from each genus using both Illumina Nextseq and Nanopore MinIon platforms in house, revealing a diverse organization of their photosynthesis gene clusters and the strong evidence of HGT of these gene clusters.

[P13] SMARTDIAGNOS: Sample concentration integrated solid-phase PCR for next-generation of pathogen

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With an emergency of pathogens, that cause sepsis and foodborne illness worldwide. We, SMARTDIAGNOS, are bringing the next generation of technology for rapid, accurate and sensitive detection and identification of multiple pathogens not only for sepsis detection but also for foodborne pathogens detection. In the project, a number of innovative technologies such as pathogen concentration, direct PCR, solid-phase PCR, supercritical angle fluorescence microlens array will be developed and combined into a streamlined point-of-care and a LAB devices to archive ultrasensitive, rapid, and unlimited multiplexing pathogen detection in blood. So far, the immune-magnetic beads could efficiently concentrate bacterial pathogen at a low concentration of 10^1 to 10^2 CFU/mL from the human pathogen spiked blood sample within 40 min. Integrated direct solid-phase PCR and supercritical angle fluorescence (SAF) microlens array embedded into a lab-on-chip allows for highly sensitive and multiplexed pathogen detection and their antimicrobial resistance genes within 3 hours with a detection limit of 10 CFU/mL of the blood sample. Fast, sensitive, and accurate detection of the pathogen causing sepsis and their AMR genes will improve patient outcome, shorten intensive care stay and thus reduce mortality and health care costs.
Reference: http://www.smartdiagnos.eu

[P14] Detection of airborne bacterial communities in environmental samples collected from a municipal wastewater treatment plant in Denmark

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Exposure to high concentrations of microbial particles in outdoor environments e.g. at wastewater treatment plants (WWTPs) can potentially be health risk for workers. However, up to now, little is known about the airborne microbial communities at WWTP in Denmark. In this study, airborne bacteria were captured by commercial vacuum cleaners (Kärcher) outdoor near the aeration tanks at a municipal WWTP (Roskilde) and they were analyzed using 16S rRNA gene sequencing to investigate their diversity at the plant. The bacterial communities in all the collected air samples were similar and the most abundant bacterial phyla identified were Actinobacteria, Bacteroidetes, Chlroflexi, Cyanobacteria, and Proteobacteria. There was similarity between bacterial species found in the air and in the wastewater from the aeration tank. However, Cyanobacteria were much more abundant in

air samples than in the aeration tanks indicating that Cyanobacteria are more likely to be aerosolized than the other bacterial inhabitants of the aeration tank. The results provide a better understanding of bacterial communities in outdoor WWTP, which might be used for an evaluation of potential hazards for workers related to airborne particles.

[P15] High-resolution in situ transcriptomics of Pseudomonas aeruginosa unveils genotype independent patho-phenotypes in cystic fibrosis lungs

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Life-long bacterial infections in cystic fibrosis (CF) airways constitute an excellent model both for persistent infections and for microbial adaptive evolution in complex dynamic environments. Using high-resolution transcriptomics applied on CF sputum, we profile transcriptional phenotypes of *Pseudomonas aeruginosa* populations in patho-physiological conditions. Here we show that the soft-core genome of genetically distinct populations, while maintaining transcriptional flexibility, shares a common expression program tied to the lungs environment. We identify genetically independent traits defining *P. aeruginosa* physiology in vivo, documenting the connection between several previously identified mutations in CF isolates and some of the convergent phenotypes known to develop in later stages of the infection. In addition, our data highlight to what extent this organism can exploit its extensive repertoire of physiological pathways to acclimate to a new niche and suggest how alternative nutrients produced in the lungs may be utilized in unexpected metabolic contexts.

[P16] Shifts in microbiome structure during summer stratification in temperate lakes

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Temperate lakes often feature a seasonal stratification of the water column, resulting in a hypoxic or anoxic hypolimnion. This heavily impacts the microbiome, however, we only have started to understand how this perturbation effects the ecological connections between different microbes and their abilities for the biogeochemical cycling of elements.

Here we present the spatiotemporal analysis of the microbiomes of two meso-/oligotrophic dimictic lakes experiencing summer stratification. By employing multivariate statistics and co-occurrence network analyses of 16S rRNA gene amplicon data, we observed the clustering of bacterial communities correlated to oxygen concentrations throughout stratification. Co-occurrence patterns among abundant and ubiquitous freshwater Actinobacteria and Proteobacteria suggested microbial niche partitioning potentially accompanied by differential specialization in carbon substrate metabolisms. The hypolimnion showed diverse communities with the potential for a wide variety of respiration processes. Hypoxic/anoxic conditions distinctly defined a module of OTUs co-occurring in the hypolimnion during summer stratification, containing anaerobic Chloroflexi and Chlorobi, but also methanotrophs. We further detected a module including OTUs of the candidate division OP3, Planctomycetes and Nitrosomonadaceae, occurring at low temperatures in the hypolimnion, predominantly in the oxygen gradient before complete anoxic conditions developed.

[P17] PRO-DIAG: improved diagnosis of chronic prosthetic joint infection

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Prosthetic joint infection (PJI) is a rare but serious complication. Surgical revision and a prolonged period of antibiotic therapy have a serious impact on life quality, and costs of hospitalization and social needs are significant. Early postoperative and acute hematogenous infections are often straightforward to diagnose. However, chronic infections remain a diagnostic challenge, and a combination of specimen types and a prolonged incubation period are required. The PRO-DIAG project aims at the latter group of patients and evaluate whether 16S rRNA gene-based diagnosis can compete with culture-based diagnosis in speed and accuracy. The project is a cooperation between Aalborg University and Aalborg University Hospital and is expected to run from January 2018 to June 2019. It is a prospective randomized clinical study aiming at recruiting 40 patients (N-20170084). The inclusion criterion is a suspected long-term chronic PJI infection. An alpha-defensin test is performed on joint fluid directly in the operation theater. Tissue samples a.m. Kamme, joint fluid, and sonication fluid from prosthetic components are incubated for 14 days. Joint fluid and sonication fluid are submitted to 16S rRNA-based sequencing including Sanger sequencing, Miseq sequencing, and Nanopore sequencing. The performance of the methods will be evaluated at the end of the project.

[P18] Transposon mutagenesis in Bifidobacterium longum subsp. longum and characterization of a gene involved in carbohydrate assmilation

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Bifidobacteria are a major component of the gut microbiota in humans; however, molecular mechanisms of their colonization have been poorly understood. To address this issue, the application of transposon mutagenesis into these organisms is effective. In this study, we developed a transposon mutagenesis system for *Bifidobacterium longum* subsp. *longum* 105-A (JCM 31944), using an endogenous IS*3* family insertion sequence IS*Blo11*. To construct this system, we overexpressed the IS*Blo11* transposase under the control of xylose-inducible or constitutive bifidobacterial promoters in *B. longum* subsp. *longum* cells. Next, an artificial transposon plasmid with IS*Blo11* terminal inverted repeats was introduced into the strains expressing transposase, resulting in the insertion of the plasmid into the genome with an efficiency of 10^3 CFU/μg DNA. The plasmid was inserted into various genomic locations, but with a preference for noncoding regions. Characterization of a transposon insertion mutant revealed that a putative α-glucosidase gene contributes to the assimilation of palatinose and trehalose. Collectively, our transposon mutagenesis system will accelerate functional genomic analyses in *B. longum* subsp. *longum*.

[P19] Evolutionary highways to persistent bacterial infection

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Persistent infections require bacteria to evolve from their naïve colonization state by optimizing fitness in the host. This optimization involves coordinated adaptation of multiple traits, obscuring evolutionary trends and complicating infection management. Accordingly, we screen 8 infection-relevant phenotypes of 443 longitudinal *Pseudomonas aeruginosa* isolates from 39 young cystic fibrosis patients over 10 years. Using statistical modeling, we map evolutionary trajectories and identify trait correlations accounting for patient-specific influences. By integrating previous genetic analyses of 474 isolates, we provide a window into early adaptation to the host, finding: 1) a 2-3 year timeline of rapid adaptation after

colonization, 2) variant "naïve" and "adapted" states reflecting discordance between phenotypic and genetic adaptation, 3) adaptive trajectories leading to persistent infection via 3 distinct evolutionary modes, and 4) new associations between phenotypes and pathoadaptive mutations. Ultimately, we effectively deconvolute complex trait adaptation, offering a framework for evolutionary studies and precision medicine in clinical microbiology.

[P20] Benchmarking and Standardizing workflows for Comparative Metatranscriptomics

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Metatranscriptomic analysis has been used widely for investigation and quantification of microbial activity and its response to external stimuli. By assessing the genes expressed by the microbial community, this can provide an understanding of the interactions between different functional guilds and their environment. Metatranscriptomics typically utilize short sequence reads, which can either be directly aligned to external reference databases, or first assembled into contigs for alignment. However, no independent and direct comparison between these two alternative approaches has been performed. Here we compared workflows representing both alternatives, using simulated datasets. We evaluate their accuracy of ability in precision and recall using the Md5nr and eggNOG hierarchical databases. We show that assembly-based method provided the best F-scores and lowest False Discovery Rates (FDR) resulting in more precise identification and quantification of functional genes. At a low confidence threshold assembly-based identification resulted in a maximum FDR of 3.5% (<1% using an optimized threshold) whereas direct alignment of reads resulted in FDRs up to 15% (>3% even at very strict confidence). Using segmented cross validation, we also show that the assembly-based method is significantly more robust with customized or environment specific databases. We also compared these methods using real metatranscriptome samples to investigate responses to warming and cooling of microbial communities in permafrost soil. By virtue of the comparative analysis we also present an open source metatranscriptomic analysis workflow written in python.

[P21] Breaking bad of bacterial viruses during experimental evolution

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Prophages have tremendous impact on bacterial populations, but we do not understand the factors that cause phages to cooperate with or to kill their bacterial hosts. Here we conducted experimental evolution of a stable lysogenic *Bacillus subtilis* strains under various selection regimes. We discovered that selection for dormant spores repeatedly leads to an evolutionary activation of a previously silent SPβ prophage. The evolved SPβ variant can easily spread in the population, because it kills the ancestral lysogens and transduces the residual subpopulation of survivors. Genomic data suggests that the prophage activation involves multiple mutations within the SPβ and genetic rearrangements between prophage elements residing in *B. subtilis* genome. Although the phage awakening scenario was highly reproducible in independently evolving ecosystems, we observed diversity at the level of lytic activities and genetic architecture. To better understand how initially stable SPβ assembled into lytic hybrid phage, we performed *de novo* genome sequencing of selected evolved lysogens using PacBio platform. Based on the obtained results, we propose that phage reshuffling can be a common evolutionary scenario in natural populations of Bacilli, with crucial consequences for ecology of the species.

Reference:
Martin M.*, Dragoš A.*, Hölscher T.*, Maróti G., Bálint B., Westermann M., Kovács AT. 2017. De novo evolved interference competition promotes the spread of biofilm defectors. Nature Communications | DOI: 10.1038/ncomms15127
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[P22] The response of the microbial community in the OMZ off Peru to changes in dissolved O2

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The anthropogenic increase in atmospheric carbon dioxide already had severe consequences, including the melting of the polar caps, ocean acidification, and a decrease of dissolved oxygen (O_2). The latter is most obvious in the tropics, where oxygen minimum zone (OMZ) waters were shown to expand and intensify. Still, the response of microbes to this progressive deoxygenation is not understood, yet. We explored microbial sensing, detoxification of reactive oxygen species, and formation of microbial consortia in the OMZ off Peru, which is the largest and one of the most intense OMZs, using a metagenomic approach.
Our study revealed increased expression of the key functional marker for ROS scavenging, *KatG*., in the OMZ core. As only traces of oxygen were possibly left there, we interpret an extreme sensitivity of the respective organisms to O_2 or other oxygen-containing molecules. More importantly, we identified a dependency of glycosyltransferase activity as an indicator for formation of microbial consortia on O_2 levels. Microbial consortia on marine snow are of critical importance for oxygen consumption, export and consumption of organic material in OMZ waters. Our results suggest however that microbial consortia in OMZs need O_2 to

develop. Given the ongoing deoxygenation of OMZ waters, a loss in marine snow-bound microbial consortia may occur, which in turn may have decrease nutrient remineralization and respiration in OMZs and could thus be regarded as a feedback mitigating OMZ expansion.

[P23] Quorum sensing affects prophage induction and biofilm formation in V. anguillarum

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Wild fish and shellfish populations cannot regenerate quickly enough to make up for the rate at which they are fished, thus leaving aquaculture as the only option for meeting the future demand for seafood. The industry is however challenged by pathogenic strains of *Vibrios* increasing fish mortalities. While the continuous emergence of antibiotic resistance is cause for concern, it also emphasizes the need for an alternative. The potential of phages for biological control is being re-examined, but in order to improve the application of phage therapy further analysis of phage-host interactions is required.
Quorum sensing (QS) affects expression of virulence factors, including biofilm formation, and within *Vibrios* there are examples of both QS-activated and -repressed biofilm formation. By in-frame deletion of genes encoding the QS master regulator (Δ*vanT*) and response regulator (Δ*vanO*) in *V. anguillarum*, we have constructed otherwise isogenic density-independent mutant strains locked in states of low- and high-cell densities, respectively. The biofilm phenotypes of these mutants show that QS strongly represses biofilm formation in *V. anguillarum*. Remarkably, we find that induction of the H20-like prophage of *V. anguillarum* is also QS regulated. The master regulator VanT has affinity to a motif upstream of the CI repressor encoded by the H20-like prophage and reduces induction of this prophage at high cell densities. Prophage-free strains harbouring the QS-mutations mentioned above indicate that the prophage is stimulating biofilm formation at low cell densities.

[P24] Isolation and Whole Genome Sequencing of Novel Aerobic Anoxygenic Phototrophic (AAP) Bacteria from the Phyllosphere of Wheat Plants in

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Aerobic anoxygenic photoheterotrophic (AAP) bacteria are ubiquitous in aquatic environments, ranging from freshwater lakes, oceans and permafrost, to hot springs and hydrothermal vents. They absorb light through Bacteriochlorophyll α (BChl-α), which they use in photosynthetic reactions to produce energy without fixating carbon or producing oxygen. Recently, their presence in the phyllosphere was discovered. In this study we investigate their presence in the phyllosphere of winter wheat (*Triticum aestivum* L.) in Denmark (Roskilde area). We collected swab samples from wheat leaves and grew them in R2A agar plates at room temperature. Bacterial colonies capable of harvesting light were detected directly on the plates using an infrared CCD camera. The more than 100 isolates were placed into 25 groups based on their emission spectra using MALDI-TOF and representatives of each group were sequenced on the Illumina NextSeq platform and draft genomes were assembled. Based on phylogenetic analyses of the 16S region of the sequenced strains, their average nucleotide identity, and pairwise comparisons of their proteomes, 10 strains were selected for further sequencing on the Oxford Nanopore minION platform in order to successfully close their genomes. The presence of the photosynthetic cluster of AAP bacteria, found predominantly in their genome and not on plasmids, was verified using homology searches. We proceed with bioinformatics analyses to compare the photosynthetic gene clusters, explore possible horizontal gene transfer of their genes, study the phylogenetic relations of the identified strains and characterize novel species.

[P25] A myriad of new bacteriophage genera and spcies with potential use in phage therapy against bacterial plant pathogens

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It is estimated that at least 10% of global food production is lost due to plant pathogens[1]. Consequently, farmers are turning to pesticides and chemical sprays (e.g. copper) for the control of pathogens. However, some of these treatments carry negative effects to both human health and the environment. These harmful effects are pushing stricter regulations on the use of pesticides and chemical sprays. This is especially true for antibiotics that are banned from use in agriculture in many countries, because of the risk of antibiotic resistance development. This leaves few options to combat bacterial plant pathogens and new methods to combat plant diseases are therefore necessary. One such promising alternative is phage

therapy. Here we present the isolation and sequencing of over 150 different phages belonging to 13 new bacteriophage genera and more than 40 new species, acting against 9 different species of bacterial plant pathogens. We then test two cocktails in two independent phage therapy trials, where phages are used to effectively prevent soft rot infections in potato tubers. Phage treatment lowered soft rot disease symptoms by 64% and 75% in tubers infected with *P. atrosepticum and D. solani* respectively, indicating that phage therapy can be used to control bacterial plant pathogens and prevent or delay onset of disease in storage produce.

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[P26] Hi-C data allow linkage of plasmids and their host genomes in a wastewater activated sludge community

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Plasmids play a crucial role in bacterial evolution and the dissemination of antibiotic resistance genes in specific lineages such as in Enterobacteriaceae, a family containing numerous pathogens. It is thus important to develop new techniques that usher a better understanding of antibiotic resistance transmission routes, from the environmental resistome to clinical pathogens. Since wastewater treatment plants constantly mix environmental and gut bacteria together with residual pharmaceutical compounds, they are highly relevant systems to study plasmid-mediated dissemination of antibiotic resistance. However, studying plasmids in complex environments such as wastewater treatment plants (WWTP) remains a challenge.
In this study, we investigated the plasmid content of activated sludge from Odense WWTP. We reconstructed plasmid sequences and linked those to their host genomes by combining shotgun metagenomic sequencing with an *in-situ* Hi-C crosslinking technique that reveals information on the spatial proximity of DNA molecules within cells. Shotgun metagenomic short reads were assembled in contigs. Hi-C cross-linked library information was then used to cluster contig into genomic bins and identify plasmid-host linkage. By using specific chromosome or plasmid markers genes, we identified the contigs' taxonomical origin and molecule type. Furthermore, antibiotic resistance genes were investigated for association with mobile genetic elements and presence on plasmid molecules. Overall, the assembly followed by Hi-C clustering recovered hundreds of genomic bins originating from various

Archaeal or Bacterial taxa, including an almost complete Archaeal genome, and several large contigs of plasmid origin.

[P27] Detection of hepatitis A virus by direct extraction of viral RNA from dates implicated in a disease outbreak in Denmark

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Outbreaks of viral jaundice have been linked to consumption of hepatitis A virus (HAV) contaminated berries, pomegranate, tomatoes and dates. During winter 2017-2018 a hepatitis outbreak was identified by the RT-qPCR detection of HAV RNA in stool samples from 19 of 31 cases showing symptoms of hepatitis from all parts of Denmark and in a Norwegian case. Sanger sequencing and phylogenetic analysis grouped all the detected strains to HAV genotype 3A. Epidemiological investigations through patient interviews and a case-control study pointed towards different batches of dates from the same producer as the vehicle of infection.
10 boxes of dates from 8 suspected batches were collected from the patients' homes and analyzed using a quality controlled direct lysis method, including simultaneous virus elution from dates and RNA extraction of virus followed by further nucleic acids purification. Extracts were tested for presence of HAV and norovirus (NoV) RNA using RT-qPCR.
RNA from NoV GII (<LOQ), or from both HAV (ca. 1×10^3 genome copies/g) and NoV GII (<LOQ), was detected in 2 of 8 batches of dates, where 1 or 2 of 23×25g samples tested positive, respectively. The achieved average extraction efficiency of spiked model virus murine norovirus was 20±13%. RT-qPCR inhibition during detection was 26±39% for NoV GI, -3±28% for NoV GII and -25±49% for HAV.
The HAV genome detected in the dates matched by sequence 100 % to the HAV genotype 3A detected in the stool sample collected from both the Danish case from whose household the specific date sample was obtained – and the Norwegian case that had eaten dates from a different batch. This confirmed, to our knowledge, for the first time a sequence link between a HAV outbreak and consumption of contaminated dates.

[P28] Growth potential of pathogens in reverse osmosis filtrated whey intended for water re-use in cheese production

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Large volumes of water are needed during the production of cheese and constitutes a high cost for the industry. Whey is a by-product of cheese production and cleaned by reverse osmosis (RO) filtration with the aim to extract valuable substances. The dairies are interested in using the RO water for cleaning and cooling purposes. This will require an ability to store the RO water for up to two days without risk of pathogen growth. To determine if the RO water is safe to store, the industry needs an easily tested indicator. For this study, RO water samples were achieved monthly over a period of 3 months. Growth experiments at different temperatures and parallel testing of biochemical parameters were performed on samples from four different dairies. A five strain cocktail for each obligate pathogens *Salmonella, Listeria monocytogenes* and shigatoxin producing *E. coli* (STEC) and a three strain cocktail of the opportunistic pathogen, *Klebsiella* were used to inoculate sterile filtered RO water, double RO filtrated water, and Biological filtered RO water at a starting concentration of approximately 10^2 cfu/ml. Samples were incubated 10°C, 20°C and 30°C for seven days. Cell counts were established by plating on Tryptic Soy Agar. Combase was used to establish estimates for lag phase, growth rate and maximum cell density. A response variable: time to four cell divisions was constructed for each bacteria at each temperature and used in a Machine Learning (ML) model including the biochemically parameters pH, chloride, nitrate, total phosphate, nitrogen, urea, lactose and chemical oxygen demand.A preliminary output from the ML suggested total phosphate to be a relevant indicator to qualify if RO water is safe to store with regard to potential growth of pathogens.

[P29] A high-throughput screening to reveal interactions between slaughterhouse isolates and the potential pathogen Listeria monocytogenes in dual-species biofilms

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Bacterial exposure in food processing environments is a critical aspect of the quality and safety of the products. The sanitization processes may be inadequate in eradicating resilient multispecies biofilms. Various interspecies interactions that occur in the multispecies biofilms may enhance the tolerance towards the sanitizers used in the food industry. The enhanced tolerance is of particular concern in regards to potential pathogenic or spoilage bacteria that may be incorporated - and protected - in a multispecies biofilm.
Here, we present a novel high-throughput method to screen a large selection of isolates collected from a meat processing environment for their capability of adjusting the prevalence of the foodborne pathogen *L. monocytogenes* in dual-species biofilms. Furthermore, the tolerance towards disinfectants commonly used in the food industry were investigated in mono-, dual- and multispecies biofilms.
We demonstrate that a slaughterhouse isolate enhanced the tolerance of *L. monocytogenes* to the commonly used disinfectant hydrogen peroxide. These results indicate that the residing biofilm-forming, non-pathogenic bacteria may facilitate and protect food pathogens and thereby contribute to their persistence in food-processing environments even after sanitization processes.

[P30] Short-term co-culturing as an accelerator of co-adaption for improving bacterial consortia performance

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The characterisation and improvement of biotechnological relevant bacteria is usually based on single strain isolation and cultivation methods, yet they differ from the complex and co-evolved communities, shaped by interspecies interactions and dependencies, they normal live in. Recent short-term co-cultivation (1-2 weeks) led to the development of genetic variants, which interacted differently compared to their ancestors, as their productivity was enhanced. Thus, they benefitted from the changes induced by the short-term co-adaption.

The present research is designed to examine the possibility of applying short-term co-culturing to accelerate co-adaption, and thus improve bacterial performance. We aim at generating geno/pheno-type variants which display enhanced efficiency during co-

cultivation, compared to ancestral variants. We will through short-term experimental evolution, boost the co-adaption for a co-culture of lactic acid bacteria, and select for a relevant trait. We will investigate the enhanced performance and mechanisms behind co-adaption, by considering genetic modifications and spatial organization.

The results will provide an in-depth understanding of the interactions and mechanism behind co-adaptation that shapes co-cultures, hence it will be a major step forward in designing methods that can promote bacterial performance.

[P31] Spoilage in plant-based meat-alternatives is primarily due to lactic acid- bacteria

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Between 2010 and 2017, the fraction of Danish consumers that ate "meat-free" at least once per week increased from 17% to 28%, corresponding to Danes eating meat-free on average every 9th day. Meat-alternative products look like, and are prepared like, tradition meat and are in demand from consumers due to convenience.
We investigated the bacterial spoilage in 6 commercially available meat-alternatives (2 plant-based mince, 2 plant-based cold cuts and a plant-based paté) using a combination of culture-dependant and molecular methods. The meat-alternatives had a longer shelf life (3-4 weeks) compared to just a few days typical for meat containing products. The bacterial counts in the plant-based products were low ($<10^3$ cfu/g) and the counts in cold cuts and paté an increased less than one log-unit by the label expiry date. The counts in the plant based minced meat increased by 3-4 log-units, primarily due to the growth of lactic acid bacteria (evidenced by lactic acid plate counts and 16S rRNA gene amplicon sequencing), specifically *Lactobacillus*, *Lactococcus*, *Brochothrix* and *Leuconostoc*. The pH was stable in all but one of the plant-based mince products where there was a considerable drop in the pH corresponding to the growth of *Leuconostoc*.
Amplicon sequencing libraries prepared with universal primers targeting the V3-V4 region of the 16S bacterial rRNA gene, contained 60-75% mitochondrial and chloroplast rRNA gene sequences. To accommodate this >50 000 reads were sequenced per sample.a

[P32] Microbial Biodiversity of Sedimented Dust from Six Pig Farms

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Farmers and other farm workers are known to be affected by asthma and other airway maladies due to exposure to bioaerosols during their working day. However, although the association between bacterial endotoxin and dust have been studied, there has been little research on the microbial diversity present in these bioaerosols.
In this study, electrostatic dust collectors (EDCs) were used to collect settling dust from 6 pig farms. Extracted dust was analysed using matrix assisted laser desorption/ionisation time of flight mass spectroscopy (MALDI-TOF MS) for culturable bacteria and fungi while Illumina's Miseq platform was used for amplicon sequencing of the bacterial 16S rRNA V4 region and the fungal ITS region.
Constrained analysis of the microbes present in these bioaerosols was explored using constrained redundancy analysis (RDA) plotting which revealed that samples taken from the same farm resembled each other more than samples taken from different farms. In addition, the stable type from which samples were taken from was not observed to have as great of an effect on the bacterial β-diversity compared to the farm they were taken from. However, the same trend in data was not observed for fungal species indicating that the fungal β-diversity in these bioaerosols is not as greatly affect by farm location or age of pigs.

[P33] Abundance of cell-cell communication networks governs adaptation to distinct life

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The biofilms of the ubiquitous Gram-positive bacterium, *Bacillus subtilis* display complex population heterogeneity. The response regulator aspartyl- phosphate (Rap) phosphatases and their cognate phosphatase-regulator (Phr) peptides have principal impact on differentiation of *B. subtilis*. We assessed the role that distinctive selective conditions play on shaping the regulatory network via cell-cell communication by using an iterative experimental competition simultaneously containing single and double rap-phr mutants (in total 78 strains next to wild type). The strain abundances were tracked using DNA barcoding followed by high throughput sequencing that enabled us to analyze the impact of individual Rap phosphatases in the adaptability of *B. subtilis*. Using distinct time periods and life style conditions, we observed that all conditions presented different selection pressure, which resulted certain sets of strains being favored during the competition. We exposed that adaptation favoring certain

combination of rap-phr deletions is transient, and selection benefits distinct strains on a longer time span with additional mutations. To verify the observed trends, the detected fitness differences were specifically verified. Selected ancestor Rap mutant strains were compared to the wild-type, or isolated evolved Rap mutant strains were assayed in competition to its cognate ancestor Rap mutant to reveal fitness differences at the start of the experiment or after experimental evolution. We conclude that the Rap/ Phr-mediated cell-cell communication in *B. subtilis* allows this bacterium to efficiently modulate its genetic regulatory network in order to efficiently adapt to new ecological niches.

[P34] Exposure Characteristics of Airborne Bacteria during a Haze Pollution Event at Oinling Mountain, China

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Bioaerosols are major constituents in atmosphere and significantly influence the climate and affect human health. Recently, many studies have focused on airborne microorganisms in urban environments, but research on other environments, such as the mountains, remains very limited. In this study, airborne microbial samples from Qinling Mountain were collected during a haze pollution event that occurred on January 2017. Plate count method was used to quantify the culturable airborne bacteria. The bacterial community profiles were studied by high-throughput sequencing technology from which diversity and abundances in the samples were determined. Results showed that the concentration of airborne culturable bacteria in Qinling Mountain was higher than that of the surrounding city, and most of this was associated with coarse particles. The coarser particulate matter (PMs) had higher bacterial diversity and abundance than fine PMs. The community structures of airbrone bacteria in PMs were similar. Source tracking analysis indicated that a small portion of the airborne bacteria could have been from the surrounding environment, while most come from the neighbor cities or other areas that aggregated by long-range air masses transport. The results of this work provide an important reference for mountain environmental science and improved our understanding of the differences between urban and field environments.

Keywords: Air pollution; Bioaerosols; Fine particles; Bacterial diversity; Sources.

[P35] Unlocking the historical and biological diversity with Podaxis (Fungi; Basidiomycota) from herbarium collections

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Perhaps the greatest concentrations of fungal biodiversity on earth are to be found in herbaria. A single herbarium may contain over 1 million fungal specimens and tens of thousands of species collected over centuries. Using the fungal genus *Podaxis* as a model, we are unlocking some of this biological and historical diversity in contemporary experiments. *Podaxis* is a genus found in both deserts and termite mounds, but despite the frequent reporting of *Podaxis* fruiting bodies in these environments, very little is known about the genus. It was first described from India by Linnaeus before he described a second species from South Africa. The description of this second species came with an as of yet untested claim that recommends its use in the treatment of cancerous ulcers. However, very little is known about the diversity of the genus or its capacity for the production of secondary compounds. We have developed a protocol for the germination of *Podaxis* spores from herbarium specimens, which allows us to tap into the wealth of biological material available from herbaria. We have already successfully germinated spores from the specimens deposited by Linnaeus to describe the genus and its first two species, the oldest of which is almost 250 years old. We are now able to incorporate these and other herbarium specimens into modern genomics and metabolomics experimentation to further investigate the diversity of *Podaxis* and its potential for the production of novel chemical compounds.

[P36] Can interaction specificity in the fungus-farming termite symbiosis be explained by nutritional requirements of the fungal crop?

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Fungus-growing termites are associated with genus-specific fungal symbionts, which they acquire via horizontal transmission. Although this mode of transmission typically leads to looser associations between symbionts, the symbiosis displays a high degree of interaction specificity between termite hosts and fungal cultivars. This selection of specific symbionts may be explained by the provisioning of specific, optimal cultivar growth substrates by the termite farmers. By performing single-factor growth assays (varying carbon sources), and a two-factor geometric framework experiment (varying carbohydrate and protein availability), we tested whether differences in growth performance of *Termitomyces* cultivars from nests of three termite species are correlated with the interaction specificity of their hosts. We found

quantitative differences between *Termitomyces* strains in carbon-source use, showing that growth is correlated with termite host genus rather than species, while growth on different ratios and concentrations of protein and carbohydrate was correlated with termite host species. Our findings provide support that specificity between termite hosts and fungi is not only visible from phylogenetic reconstructions, but also reflected in fungal physiology. Differences between *Termitomyces* of different hosts could facilitate fungal selection during the colonization of the substrate collected by termites in incipient nests. This could potentially contribute to maintaining termite-fungus interaction specificity if physiological differences cause variable growth during competition between multiple fungal strains at the onset of fungus-garden formation.

[P37] Nitrogen fixation in the upwelling ecosystem off Cape Verde

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We investigated di-nitrogen (N_2) fixation at the eastern boundary upwelling ecosystem located at the Cape Verde frontal region, North-West Africa. This region is rich in nutrients due to intense coastal upwelling along the continental shelf, as well as trace metals as a result of dust deposition from the Saharan desert.
We measured N_2 fixation in July/August 2017 from the surface down to 4000 m depth. Detectable N_2 fixation rates were mainly found in the euphotic zone with rates >0.50 nmol N L^{-1} d^{-1} at several sites from the coast to the open ocean. The highest rates were measured in surface waters south of the frontal zone off the coast of Cape Blanc (~5 nmol N L^{-1} d^{-1}) and at the Cape Verde Islands (~2.5 nmol N L^{-1} d^{-1}).
To identify groups of diazotrophs future work will include analysis of diazotroph community composition based on phylogenetic examination of the functional gene *nifH*. Additionally, the active part of the diazotroph community will be established by analysis of *nifH* mRNA.
The importance of aggregates as loci for N_2 fixation will be investigated by analysing *nifH* sequences derived from size-fractionated samples as well as *in situ* sampled individual marine aggregates. To directly measure aggregate-associated N_2 fixation, aggregates collected *in situ* will be analysed by nanoSIMS.
The results obtained indicate that N_2 fixation is an important source of bioavailable N sustaining productivity in this upwelling ecosystem.

[P38] Exploring interactions between Blastocystis sp., other intestinal parasites and the gut microbiomes of wild Chimpanzees in Senegal

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The old friends hypothesis links the loss of many immunoregulatory microbes, such as intestinal protozoa (e.g., *Blastocystis*) or helminths (e.g., *Strongyloides*), with the rise in immune disorders in high income countries. This may be hard to study in humans, but studying simians closely related to humans may provide insight into the ancestral human relationship with their gut microbiota (GM). Chimpanzees may be particularly promising, as they harbour a GM that is very similar to humans, are thought to in similar ways coevolve and have dependencies with their GM, and because many intestinal parasites colonize both species. We obtained eighty-two faecal samples from chimpanzees (*Pan troglodytes verus*) in Senegal, and examined associations between bacterial and archaeal members (16S rRNA amplicon seq.) and the intestinal parasites *Blastocystis* (n=33/82), *Strongyloides* (n=10/79) and *Giardia* (n=1/82). We found a positive association between *Blastocystis* presence and *Methanobrevibacter* and *Prevotella* abundances, consistent with previous work in humans. In contrast, we found a negative association between *Blastocystis* and *Faecalibacterium*. Several α diversity indices were lower with *Blastocystis* infection, while *Enterobacteriaceae* abundance increased. *Strongyloides* presence was significantly associated with an *Alloprevotella* increase. Both parasites significantly affected β diversity, as did diet and group of origin. *Blastocystis*, unlike *Strongyloides*, seems to be associated with a GM indicator of generally poor health. Our results suggest that studies on the wild chimpanzee GM, less disturbed than human GM, may reveal insight into the role of intestinal protozoa and helminths on the GM and hosts, ape or human.

[P39] Linking of plasmids to bacterial genomes in complex samples using Hi-C

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Genome-centric metagenomics enables genome studies of unculturable microbes in complex samples. Hereby, representing a powerful tool for investigating the vast diversity of organisms at individual genome level. However, using conventional differential coverage binning or tetra nucleotide signatures some genomes remain impossible to differentiate, and important information such as plasmid and genome association is missing. Hi-C seems like a promising technique for obtaining this information as it captures *in-vivo* proximity interaction in read-pairs. Inter-chromosomal junctions (e.g. genome-plasmid interactions) provide a mean for grouping mobile elements to the associated genome. Together with intra-chromosomal interactions this information can also be useful to differentiate more closely related species and achieving higher genome resolution in complex samples.

This project describes a general protocol for generating Hi-C data, and demonstrates the potential of this technique in combination with Nanopore assembled metagenomes. This is done by validating the technique in mock communities and further testing on full-scale complex samples that provide insight in plasmid association and sharing together with genome assemblies.

[P40] DnaA Rejuvenation Sequences are vital for accumulation of DnaA-ATP and initiation of replication

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In *Escherichia coli* and most other bacteria, chromosome duplication is regulated by accumulation of the initiator protein DnaA on its active form (bound to ATP). This fact suggests a coupling between the cell cycle and energy metabolism in such a way that under growth conditions where the cell has ample energy supply, chromosome duplication is promoted. Our objective is to establish and characterize the connection between the energy charge of the cell and chromosome replication in bacteria.
To test the role of the energy content in the cell in controlling DnaA activity, the ATP/ADP ratio has been lowered by two different ways. On one hand we have induced the expression of an ATP synthetase that is decoupled and whose activity is to deplete ATP (Koebmann *et al*., J Bacteriol, 2002). On the other hand we have tested the effect of natural energy depletion on late-phase cultures. We have studied the effect of this low energy charge situation on mutants affecting initiation of replication (*DARS1* and *DARS2*, involved in DnaAATP

rejuvenation). The effect on cell cycle properties and initiation of replication has been analyzed by flow cytometry.
We can conclude that lowered cellular ATP/ADP ratio has no influence on initiation frequency in the presence of any of the mechanisms reactivating DnaA. Nevertheless, when the cellular energy is reduced at least one of the DARS regions is required to ensure the accumulation of active DnaAATP. We suggest accumulation of DnaAATP as the absolute limiting factor for initiation of replication.
Fellowship support for B. M-C from Fundación Alfonso Martín Escudero (Spain) is gratefully acknowledged.

[P41] Swarming behavior in bacteria associated with cable bacteria filaments is closely linked to electric current

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Cable bacteria are long, filamentous bacteria which can transfer electrons over centimetre distances by coupling the half reaction of sulfide oxidation in anoxic sediment zones with oxygen reduction at the sediment surface. In freshwater sediment enrichments we observed diverse motile bacteria swarming around segments of cable bacteria in the anoxic zone. The swarming was transient, occurring only when the cable bacterium extended all the way to oxygen, several millimetres away. Cell tracking of the swarming bacteria showed that they spend most of their time within 50 µm of the cable filament and were highly diverse in their cell morphology. Amplicon sequencing of sediment enrichments selecting for motile bacteria show that chemoorganotrophic bacteria are differentially more abundant when cable bacteria are present, relative to the cable free controls. Swarming cells increase their swimming speed near the cable bacterium, and the detection frequency by fluorescence in situ hybridization is increased, indicating a higher ribosome content. This indicates that the interaction with the cable bacteria is metabolically positive to the swarming cells. The swarming ceases within one minute in cable parts that were disconnected from the oxygen exposed parts by cutting with a dissection laser microscope. Preliminary Raman microscopy of swarming cells suggests the redox state of their cytochrome c is more oxidized near the cable bacteria. These results strongly suggest some type of electron exchange whereby the swarming bacteria take advantage of long distance electron transfer by cable bacteria.

[P42] High-throughput phage isolation - from sample to sequence

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The use of bacteriophages (phages) to treat bacterial infections (phage therapy) is a promising alternative and/or supplement to conventional antibiotics. Phages are the viral antagonists of bacteria, they proliferate by infecting their bacterial host and taking over their replication machinery to produce progeny which is then released by cell-lysis, killing the host in the process. However, efficient phage therapy requires a collection of multiple virulent phages, either targeting a wide range of pathogenic hosts or having highly specific target sites.
A novel, high-throughput screening method based on incubation in microtiter well-plates allows for a faster and more efficient screening of a high number of samples requiring only small sample volumes (<1 ml). This enables the identification of hundreds of phages within a week. The workflow entails sample enrichment, a purification step and a final spot-test which facilitates the fast identification of positive wells for phage isolation, DNA-extraction, library building and sequencing. Observed plaques can be directly harvested for isolation, characterisation and production of phages of interest.
Here we present screenings of 188 individual wastewater samples applying two different enterobacterial hosts (*Escherichia coli* and *Salmonella enterica*). Within one week, 32 novel *S. enterica* phages and 109 novel *E. coli* phages were identified, including one potential novel *S. enterica* and two potential novel *E. coli* phage genera. The presented screening method is highly effective and can become a valuable tool in the process of constructing efficient phage cocktails suitable for treating bacterial infections.

[P43] Uncovering the hidden diversity of the Asgard archaea

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The representation of all living organisms in the tree of life constitutes a cornerstone in biology. However, concurrently with new methods developing, major changes to the tree of life are being proposed. Instead of the conventional three-domain topology, recent studies support the notion that eukaryotes evolved from within the archaeal domain – more specifically from within the Asgard archaea superphylum.
In this study, we aimed at specifically populating the Asgard archaea clade in the tree of life by generating high-quality, near full-length small subunit ribosomal RNA sequences with a primer-based approach.

By designing 12 low specificity primer-sets targeting novel lineages in the Asgard phylum, we were able to generate thousands of new, near full-length Asgard archaeal sequences (>1200 base pairs in length). A total of 17,664 near full-length sequences were retrieved from which approximately 80% belonged to archaea (representing about 20% of the total number of archaeal sequences currently present in the SILVA database). Aligning all of the new sequences against the latest SILVA 132 database revealed plenty of potential novel diversity with an average identity score of 92.8%.
The previously unexplored Asgard archaeal diversity uncovered in the present study represents an exciting avenue for studying this still poorly studied branch in the tree of life and its predicted evolutionary connections to the eukaryotic domain of life.

[P44] Metabolite profiling of social spiders along a climate

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Social spiders of the genus *Stegodyphus* live in colonies of hundreds of individuals. They live in close association with microorganisms, including bacterial symbionts, which together with the spider outline the social spider holobiont. Social spiders are highly inbred and have extremely low genetic variation across populations. Yet, social spiders are ecologically successful and widely distributed over multiple climate zones in nature.
We hypothesize that the social spider holobiont copes with environmental challenges by producing low molecular weight metabolites, for example known thermal stress protectants such as trehalose or proline. If so, the spiders' metabolite profiles should reflect their environment, e.g. by containing higher amounts of cold protectants in spider populations collected from colder sites. Furthermore, due to the low genetic variation in the host, such metabolites may also originate from the bacterial symbionts.
Social spiders were sampled in triplicates from three populations along a climate gradient in Namibia. Whole animals were extracted with methanol and water, after lyophilization extracts were analyzed by GC-MS and ^{1}H-NMR.
Metabolite profiles were highly reproducible within each population, while showing differences between the three populations. A possible link between metabolite profiles and temperature tolerance is currently tested in controlled lab experiments, and we are working on identifying key metabolites and their (potentially bacterial) origin.

[P45] Online surveillance of microbial communities in activated sludge

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Sanitation plays a key role in the interaction between humans and the environment. The activated sludge process is an important step in wastewater treatment, where nutrients are metabolised by Bacteria in sludge flocs. This allows separation of sludge and water, whereby cleaned water can be let out into the environment without causing eutrophication. Different functional groups must co-exist to yield a high treatment efficiency. Certain Bacteria are known to affect the treatment efficiency negatively if allowed to increase in abundance. The standard method of identification is by DNA sequencing, which requires expensive equipment and weeks or months of sample-batching and sequencing. This approach is good for studying the communities in activated sludge, but not feasible for surveillance.
In this project, we developed an easy-to-use workflow to enable microbial surveillance on-site at a full-scale wastewater treatment plant with the Nanopore MinION DNA sequencer. The workflow was performed in 6 hours and resulted in a community abundance profile. Further comparisons between the developed workflow and the standard Illumina workflow indicates that there is a difference in the two platforms, but the variation within the Nanopore platform is not larger than in the Illumina MiSeq. The developed workflow is suitable for regular microbial surveillance, where samples are sequenced on-site and data processed on a remote server. It is very feasible that DNA sequencing can be implemented at larger wastewater treatment plants in a near future, where it will give operators key insights to maximize treatment efficiency.

[P46] The Impact of Variation in Diet on the Gut Microbiota of Omnivorous African Viverrids

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The gut microbiota affects many aspects of host ecology and evolution through different interactions and processes performed by bacteria within the gut ecosystem. The link between diet and microbiota composition has been established in many hosts, but has primarily been studied in species with well-known diets. However, omnivores species may be particularly interesting in this respect, because their diets vary greatly depending on environmental factors such as the availability of different food items. When exploring this link, extracting information about the diet and gut microbiota from the same host and sample, could allow for determining the role of temporary shifts in diet on microbiota composition. We obtained 91 faecal samples from omnivorous species of the family Viverridae (*Civettictis civetta* and *Genetta* sp.) from three locations in southern Africa from September to December 2017. We characterised the gut microbiota using MiSeq amplicon sequencing of the V4 region of the 16S RNA gene. Five universal primer sets for metabarcoding of vertebrate, invertebrate, and plant DNA were used, to determine the diet of the individuals that deposited the faeces. Analyses are still in process, but the diet results this far, have shown that the species are omnivores as predicted. Furthermore, preliminary insights suggest differences in the gut microbiota between viverrid species and changes in microbiome richness over time for all species. This suggests that the viverrids utilize different diets and that dietary changes temporally have an impact on gut microbiota composition.

[P47] Detection of unrecognized vancomycin-resistant enterocci in a hospital

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Vancomycin-resistant enterococci (VRE) has become increasingly more prevalent nosocomial pathogens and represents an increasingly growing health concern worldwide. VRE may be difficult to detect with conventional culture methods. We applied in-house real-time PCR assays for vanA and vanB genes to ensure detection of VRE in patient samples.
Over a course of one month, all urine culture plates with growth of *E. faecium* or *E. faecalis* at the Department of Clinical Microbiology, Odense University Hospital, were screened regardless of the number of colony forming units reported. A loop-needle was stroked through the bacteria growing on the plate and prepared for PCR. Samples from five to ten culture plates were pooled. When a pool of samples was positive for vanA or vanB gene, the plates from the pool were tested individually and bacteria from the original culture plate were seeded at selective VRE plates.

357 patient samples were screened, 15 of them were positive for vanA gene and none for vanB gene.
The finding raised concern that VRE may have escaped detection for a longer period of time. The laboratory stores all pathogenic bacteria isolated from blood. The screening procedure was applied on 319 stored blood isolates of *E. faecium* from Jan 2016 to April 2018 and 141 *E. faecalis* from Oct 2016 to May 2018. Only enterococci previously reported as VRE were found positive for vanA or vanB.
This study shows that conventional culture methods may underestimate the prevalence of VRE and that PCR of pooled samples is a simple, effective, and inexpensive method to monitor the occurrence of VRE in a hospital. No evidence of previous misclassification of VRE as susceptible to vancomycin was found.

[P48] A modified iChip for isolation of antimicrobial drug-producing bacteria from social spider

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Antibiotic resistance in pathogens is an ever-increasing problem for humans and an extraordinary effort is needed to meet the requests for novel antimicrobials. Social spiders (*Stegodyphus dumicola*) are widespread in Southern Africa. They are highly inbred and show extremely low genetic variation. Despite their low genetic diversity they seem rarely affected by pathogens.
We therefore hypothesized that a highly efficient microbial defence system might be at play, and thus these social spiders may represent an untapped source for novel antimicrobials.
The goal of our study was to apply *in situ* cultivation to isolate a large diversity of microbes from spider nests for antimicrobial testing. We modified a previously described isolation chip (iChip) to fit the arid environment of Northern Namibia. The modified iChip is equipped with a water reservoir to prevent the dehydration of the agar layer inside the iChip, and it is miniaturized for application inside spider nests.
After two weeks of *in situ* cultivation, most iChip wells showed growth of microcolonies, of which on average 80% could be further propagated in the lab on standard agar plates. Several hundred isolates were retrieved, and the diversity of isolates was much broader compared to direct isolation on nutrient-rich agar. Antimicrobial testing and genome sequencing of isolates

is currently ongoing. Our modified iChip design offers a simple solution for *in situ* cultivation in environments that are not water-saturated.

[P49] Evolution, Transmission and Function of Social Spider Symbionts

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The social spiders *Stegodyphus dumicola* live in Sothern Africa with habitats covering a wide range of environmental conditions. These highly inbred spiders have extremely low species-wide genetic diversity, leading us to hypothesize that specific symbiotic bacteria contribute to environmental adaptation for their host. Using 16S rRNA gene amplicon sequencing we characterized the microbiome of multiple spider populations in South Africa. The spiders have no obligate core symbionts but share several recurring dominant amplicon sequence variants (ASVs) across populations. The microbiome of each individual spider tends to be dominated by one or a few of these dominant ASVs, in a pattern that is highly nest-dependent, but with no clear population-dependency. All of the dominant ASVs are related to known pathogens or endosymbionts, and phylogenetic analyses revealed that most of them share an evolutionary history with other arachnid-associated bacteria. Using fluorescence in situ hybridization (FISH) on whole body sections of *S. dumicola,* we localized cells representing the most prevalent and abundant ASVs, i.e. *Mycoplasma*- and *Borrelia*-like symbionts throughout the midgut of adult spiders, while eggs seem to be sterile. Preliminary studies demonstrate an effect of antibiotics treatments on the spiders' temperature tolerance. Our data suggest that the symbionts of social spiders are evolutionarily old, transmitted horizontally within the spider colonies, and functionally diverse.

[P50] Faecalibacterium gut colonization is accelerated by presence of older

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Faecalibacterium prausnitzii is a highly abundant human gut microbe in healthy individuals, but is reduced in individuals with gastrointestinal inflammatory diseases. It has thus been suggested to constitute a marker of a healthy gut, and is associated with anti-inflammatory properties. However, factors affecting the colonization of *F. prausnitzii* in the human gut during early life are very poorly understood. By analysis of 16S rRNA amplicon sequencing data from three separate infant study populations we determined the colonisation dynamics of *Faecalibacterium* and factors affecting its establishment in the gut. We found that particularly the presence of older siblings was consistently associated with *Faecalibacterium* gut colonisation during late infancy, and conclude that acquisition of *Faecalibacterium* is very likely to be accelerated through transfer between siblings.

[P51] An experimental setup for colonization and enrichment of marine bacteria on plastic pellets

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Since the introduction of mass production in the 1940s, plastic production has been growing continuously resulting in about 8 million metric tons of plastic waste annually entering the oceans. We are only in the very beginning of understanding the impact of plastic on the marine environment, including its transport, degradation, and potential risk. Microorganisms could influence the fate of plastic debris by colonization or degradation. As a basis to investigate bacteria-plastic interactions, our purpose was to develop a simple experimental setup to estimate bacterial colonization on plastic pellets and utilize it to identify plastic colonizers by enrichment. Neither addition of nutrients nor incubation longer than 24 h had an effect on pellet colonization of a bacterial isolate. Testing the setup with six strains and three common thermoplastics showed that only for one strain, the plastic type influenced the level of colonization. After incubation of natural seawater with plastic pellets for two weeks, bacterial communities on plastic were distinct from the initial seawater diversity demonstrating a bacterial enrichment on the pellets. This included species known for plastic colonization and hydrocarbon degradation. In the future, the setup will allow estimation of

the impact of bacterial colonization on plastic transport in the aquatic environment as well as isolation and identification of possible plastic-degrading microorganisms.

[P52] The identification and study of adaptive intergenic mutations in bacterial pathogens

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Bacterial pathogens become subjected to different selective pressures inside their hosts, which drives their adaptation through acquisition of nucleotide-level changes. Previous studies have largely focused on the adaptive changes within coding regions. Therefore, little is known about the role of intergenic mutations in the development of adaptive processes. Specifically, there has been no attempt to investigate this in a systematic and robust manner.

We have developed a bioinformatic pipeline that enables a systematic analysis of intergenic regions from large genomic datasets of bacterial pathogens. We focus on identifying enriched intergenic mutations that occur in different lineages of a defined phylogeny. We have combined the bioinformatic tool with functional genomics to study the effects of enriched intergenic mutations in *Salmonella enterica* serovar Enteritidis isolates. We identified a potentially adaptive mutation within a promoter of *folA*, a gene coding for a dihydrofolate reductase, which is a target for trimethoprim antibiotic. After generating reporter fusion constructs in S. Enteritidis, we were able to demonstrate that the mutation results in more than 2-fold increase in expression of the *folA* gene in standard media.

Our results show the strength of combining bioinformatic and functional genomic approaches in order to gain new important insights into the significance of intergenic mutations for bacterial adaptive processes.

[P53] Investigating the biofilm properties of Borrelia ssp.

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Background
The disease Lyme borreliosis is caused by vector-borne spirochetes from the *Borrelia* genus. The vast majority of borreliosis cases can be treated with antibiotics.
Approximately 10% of infected patients transit to a chronic stage, chronic Lyme disease (CLD), where the symptoms manifest themselves after months or years after the initial infection. This raises an important question - how can the symptoms of borreliosis persist for months or years despite vigorous antibiotic treatment and the patient's own immune response? Several features ascribed to CLD have characteristics found in many chronic biofilm infections, yet there is a lack of observations of biofilm formation for the three most common borreliosis-causing pathogens: *Borrelia burgdorferi*, *Borrelia afzelii* and *Borrelia garinii*.
Aim
We aim to determine if any *Borrelia ssp.* can form a biofilm *in vitro* and *in vivo*.
Method
This study aims to elucidate the biofilm forming ability of *Borrelia* spp. by growing unattached biofilm aggregates in a semi-solid system which mimics an *in vivo* environment. Environmental specimens of ticks were collected during the spring and summer of 2018 and were sectioned, prepared and stained with PNA-FISH for microscopy for determination of any biofilm phenotype within ticks.
Results
Results from our semi-solid experiments indicate the ability of *Borrelia* spp. to form biofilm-like aggregates resembling what can be found in other biofilm forming species. We were able to identify spirochetes within ticks but cannot with certainty determine any biofilm phenotype at this point.
Conclusion
The ability of *Borrelia* spp. to form biofilm could potentially explain several aspects of the persistent chronic Lyme disease.

[P54] Direct Mobilome method reveals plasmid-encoded genes from bacterial community in Roskilde Fjord

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Circular Mobile genetic elements (MGEs) such as plasmids, small insertion sequences (IS-elements) and bacteriophages allows dissemination of advantageous genetic traits between microbes and are the major players in the rapid evolution of the bacteria. The pool containing these types of DNA vectors are referred to as Mobilomes, and they all allow genetic plasticity. Here, we attempt to build sequencing libraries, which are highly enriched in circular MGEs within a bacterial community. Through construction and analysis of these libraries, we enter the mysterious world of cryptic molecular parasites, their mechanisms, and how these MGEs ensure the flow of traits between species and drive the adaptation in microbial communities.

In this preliminary study, we generated a Mobilome from Roskilde fjord in order to describe the plasmid-encoded genes present in the bacterial community in shallow coastal waters, and we compare the results with other environments. We show that there are plasmids potentially carrying bacterial light-harvesting gene cassettes. Additionally, we discuss the limitations there are in Mobilomics due to the lack of un-bias strategies and absence of high-quality bioinformatics tools to extract and analyze plasmids from sequencing data.

[P55] Biodegradation of pesticide residue in sand filter columns treating membrane residual water

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Background: Drinking water resources, such as groundwater, are threatened by pollution. One concern is the pesticide metabolite 2,6-dichlorobenzamide (BAM) frequently found in groundwater in concentrations exceeding the EU legal limit of 0.1 µg/L. Studies have therefore attempted to add BAM-degrading bacteria to sand-filters at drinking water treatment facilities. This biotechnology has shown promise in purifying BAM polluted water. However, the degradation potential was lost over time due to a decrease of the degrader population.
Aim: to overcome the constraints leading to loss of degraders from inoculated biofilters. Our approach to this was threefold: 1) development of a novel inoculation strategy, 2) lowering the flowrate to reduce washout of cells, and 3) increasing the concentration of nutrients in a smaller inlet water stream. The two latter were achieved via modifications of the inlet water by applying membrane treatment which, besides producing an ultra-pure water fraction, produced a residual water stream with all nutrients including BAM concentrated in a 10x reduced volume. This was done to alleviate starvation of degrader bacteria in the otherwise oligotrophic biofilters and to enable a decreased flowrate.
Results: we achieved 100% BAM removal over a period of 40 days in sand-filter columns inoculated with the BAM-degrader *Aminobacter* sp. MSH1. Molecular targeting of the degrader strain showed that the population of degrader bacteria persisted throughout the sand-filter column and over the entire timespan of the experiment.

[P56] Finally, Bulk Typing of Bacterial Species down to Strain Level using ON-rep-seq

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We have developed a new method for fast and cost-effective bacterial species level identification and strain level differentiation using Repetitive Extragenic Palindromic based amplicon sequencing on MinION platform. The method utilizes an optimized version of rep-PCR followed by dual-stage rep-PCR-2 during which sample specific barcodes are incorporated. DNA enrichment together with barcoding takes less than 5h and ensures highly repetitive and evenly distributed reads per sample. Our results demonstrate that sequencing of the rep-PCR genomic fingerprint profile with Oxford Nanopore Technology generates highly reproducible read length counts (LCp) profiles. We have developed a pipeline that by correcting the random error of individual reads within each peak generates a set (~10 reads per sample; 300bp - 3Kb) of high quality (>99%) consensus reads. The information from high quality reads is used to retrieve species level identification. Furthermore, we have developed an algorithm that compares integrals of the peaks profiles allowing for strain level discrimination.

[P57] Bacterial dispersers along preferential flow paths of a clay till depth profile

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Active dispersal is considered essential for soil bacterial colonization and survival. However, very little is known about the dispersal potential of bacterial communities and the role of active dispersal in the heterogeneous soil environment. Here we applied a recently developed expansion of the porous surface model that allows the capture and identification of bacterial communities dispersing under controlled hydration conditions on a soil-like surface. The method was used to assess the dispersal potential of five bacterial communities derived from contrasting compartments along a fractured clay till depth profile; i.e. plow layer soil, preferential flow paths (biopores and the tectonic fractures below) and matrix sediments, down to 350 cm below the surface. Using 16S rRNA gene amplicon sequencing,

the dispersing communities were found to be less diverse than corresponding total communities. The dominant dispersers in most compartments belonged to the genus *Pseudomonas,* and in some cases to *Rahnella.* As an exception the dispersing community in the matrix at 350 cm below the surface was dominated by *Pantoea.* An increased proportion of amplicon sequence variants representing dispersing communities as compared to non-dispersing ones, were shared between the hydrologically connected compartments (plow layer, biopores and tectonic fractures). This suggests that active dispersal is important for colonizing these compartments. These results highlight the importance of including soil profile heterogeneity when assessing the role of active dispersal and contribute to discerning the importance of active dispersal in the soil environment.

[P58] Danish wastewater harbors multiple mobilized colistin resistance (mcr) genes: a preliminary study on the environmental mcr reservoir

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Despite its neuro- and nephrotoxicity, colistin constitutes a last-resort antibiotic against multidrug-resistant Gram-negative pathogens as safer drugs gradually become obsolete. Thus, the fast-paced emergence and mobility of colistin resistance (*mcr*) genes pose a severe public health concern. To date, research on the prevalence of *mcr* genes has primarily focused on clinical and veterinary isolates, while the study of *mcr* genes in environmental reservoirs has been greatly overlooked. Incidentally, the recent emergence of *mcr-1* and other *mcr* genes calls for epidemiological studies of the dynamics of plasmid-borne *mcr*. In this context, wastewater treatment plants (WWTP) are of high research interest since they comprise horizontal gene transfer hot spots for bacteria, and hence play a critical role in the dissemination of plasmid-borne antibiotic resistance. In this study, we investigated into the activated sludge samples from the Odense municipality WWTP. We confirmed the presence of colistin resistant *Enterobacteriaceae* and the responsible *mcr* genes variants were identified by multiplex PCR targeting 5 different *mcr* genes (*mcr*-1; *mcr*-2; *mcr*-3; *mcr*-4; *mcr*-5). Plasmid DNA extracted from *mcr* PCR positive samples were sequenced and assembled thus revealing the array of plasmids vectors harbouring *mcr* genes in an urban WWTP and their host association.

[P59] Microbiology of whey water after UF- and RO- filtration

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Membrane filtration technology such as Ultrafiltration (UF) and Reverse Osmosis (RO) is applied in the dairy industry for up-concentration of whey protein and lactose, creating at the same time permeate water for reuse. Permeate water for reuse in direct or indirect product contact should not negatively affect safety and quality. In a whey stream line with UF- and two consecutive RO-treatments, followed by UV, we investigated the microbial level and diversity of the permeate water after each filtration step to assess treatment efficacy and microbiological quality of the permeate water. Heterotrophic Plate Count was enumerated using non-selective PCA agar. A representative number of colonies was isolated (n=100) and identified by 16S rRNA sequencing. UF permeate relatively high levels (3 $\log_{10}$CFU/mL) were reduced to 32 (CFU/mL) after RO and 14 (CFU/mL) after ROP. The microbial diversity was higher after UF than after RO and ROP treatment with the composition changing from a mixture of *Bacillus spp*., lactic acid bacteria and gram- to a microbiota dominated by gram+ bacteria (*Bacillus spp.)*. No *E. coli*, total coliforms or Enterococci were detected in 100mL ROP. Therefore, ROP reclaimed water was considered of drinking quality from a microbiological point of view, suitable for reuse in direct or indirect product contact. Upon storage ROP permeate could support bacterial growth, thus monitoring and control of storage and distribute conditions is essential as variations in treatment efficacy were seen and regrowth of survivors and contaminants may potentially jeopardize the microbiological water quality.

[P60] Turnover of soil bacteria rRNA at different temperatures

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Background: Ribosomal RNA (rRNA) is used widely to investigate the potentially active microbiome in environmental samples. Many microbiomes in soil, shallow-water sediments, surface water, poikilothermic animals, or on plants are subjected to pronounced seasonal variation in temperature. However, little is known about the turnover of intracellular microbial rRNA at environmentally relevant temperatures.

Results: We analyzed the turnover at four different temperatures of RNA isolated from a soil microbiome amended with ^{14}C-labelled uridine. We found that the half-life of recently produced rRNA increased from 5.6 days at 20 ˚C to 25 days at 5 ˚C, and 226 days at -4 °C, while no degradation was detected at -18 °C during a one-year period.

Conclusions: The strong temperature dependency of rRNA turnover and its long half-life at low temperature may lead to misinterpretation of microbiome data based on rRNA isolated from environmental samples.

[P61] Preferential flow paths shape the structure of clay till bacterial communities

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Preferential flow paths in subsurface soils serve as transport routes for water, dissolved organic matter and oxygen. Little is known about bacterial communities in flow paths or in subsoils below about four metres. We compared communities from preferential flow paths (biopores, fractures and sand lenses) with those in adjacent matrix sediments of clayey till from the plough layer to a depth of six metres. 16S rRNA gene-targeted community analysis showed bacterial communities of greater abundance and diversity in flow paths than in matrix sediments at all depths. Deep fracture communities contained a higher relative abundance of aerobic taxa and plant material decomposers such as *Planctomyces, Nitrospirae* and *Acidobacteria* than adjacent matrix sediments. Similarly, analyses of the relative abundances of bacterial *amoA*, *nirK* and *dsrB* genes indicated transition from aerobic to anaerobic nitrogen and sulphur cycling at greater depth in preferential flow paths than in matrix sediments. Interestingly, preferential flow paths contained more indicator OTUs from the plough layer community than the matrix sediments. This study indicates that the availability of oxygen and organic material and downward transport of bacteria shape bacterial communities in preferential flow paths, and suggests that their lifestyles differ from those of bacteria in matrix communities.

[P62] Time lapse confocal microscopy imaging of fibrin formation in growing Staphylococcus aureus biofilms

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Staphylococcus aureus biofilms are a leading cause of implant associated infections, which are notoriously difficult to treat using antibiotics. Fibrin is a key component of the extracellular matrix of *S. aureus* biofilms *in vivo*. *S. aureus* secretes two coagulases, coagulase (Coa) and von Willebrand factor binding protein (vWbp), which trigger fibrin formation by binding to prothrombin and hijacking the host coagulation cascade. We are studying fibrin formation in wild type *S. aureus* ATCC 29213, as well as the mutants *S. aureus* ATCC 29213 ΔvWbp and *S. aureus* ATCC 29213 ΔCoa, that lack vWbp and Coa respectively, in order to study the role of each coagulase without the interference of the other. Preliminary time lapse confocal microscopy images of the first 2 hours of growing *S. aureus* ATCC 29213 ΔvWbp and *S. aureus* ATCC 29213 ΔCoa biofilms indicate that Coa is responsible for fibrin formation at the surface of cells, whereas vWbp does not cause fibrin formation in any particular location. We will continue to use time lapse imaging of these bacteria to address fundamental questions regarding the formation of fibrin in *S. aureus* biofilms.

[P63] Better alone: Methanosarcina from the Baltic sea corrodes iron

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Microbial corrosion is an economical and environmental threat in different environments. Over the course of three years we investigated corrosion by a community from coastal Baltic Sea sediments. We established sediment slurries from the methanogenic zone and provided them with iron as sole electron donor. Black crust developed on the metal surface within the course of couple of days and scanning electron microscopy showed the presence of three morphotypes, sarcina-like cocci, vibrio-like cells and spore-forming like cells. High-throughput sequencing showed that *Methanosarcina*, *Clostridium* and *Desulfovibrio* dominated the community after the fourth transfer. The closest relative to the Baltic *Methanosarcina* was *Methanosarcina lacustris*. To determine if the enriched *Methanosarcina* was capable of iron corrosion without a bacterial partner, we eradicated the bacteria with a cocktail of antibiotics. After 16 days, we observed that indeed this Baltic *Methanosarcina* was capable of corrosion independently, as shown by higher Fe^{2+} production per day (62.1 ± 3.1 µM/day) compared to

cell-free controls (11.9 ± 6.2 µM/day), and ca. 60% higher than the mixed community (38.9 ± 3.8 mM/day). The Baltic *Methanosarcina* without bacteria showed more than eight times higher methane-production rates (70.6 ± 9.7 µM/day) than expected from H_2 alone (8.4 ± 4 µM/day). This indicated they could retrieve electrons from iron either directly or via an enzymatic-mediated mechanism. Baltic *Methanosarcina* generated also 40% more methane and were more corrosive releasing 60% more Fe^{2+} than the mixed community suggesting their access to iron and overall growth is inhibited by fast growing acetogens.

[P64] Degradation and sorption of organic micropollutants (OMPs) during laboratory study simulating managed aquifer recharge (MAR)

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Due to the growing problem of drinking water deficits in many places around the world, sustainable methods ensuring safe and reliable source of groundwater – and, hence, drinking water – are of significant importance. Technology based on managed aquifer recharge (MAR) is a promising approach for areas struggling with (temporary) groundwater deficiency. MAR is an efficient and economic tool for sustaining groundwater reservoirs (aquifers) being primary source of drinking water. Within ACWAPUR project (acwapur.eu) we provide a deeper insight into processes during MAR, to understand and prevent leaching of pathogens, inorganic nutrients, organic pollutants and their degradation products to aquifers.
With a set of laboratory columns we simulated operation of a MAR barrier. The columns operated for 4 months and were fed with synthetic wastewater (~secondary effluent). Our focus was on removal of organic micropollutants (OMPs), being chemicals classified as emerging organic contaminants. Although such chemicals are often present at very low concentrations (ng-µg/L), their presence in groundwater and subsequently in drinking water is adverse. We analyzed fate of 10 OMPs, with varying characteristics and applications, including antibiotics, painkiller, psychiatric drug, UV-filter, anti-corrosives and pesticides. The removal of the OMPs by (bio)degradation and sorption was measured by SPE-LC-MS/MS. Inoculation of the barrier material in columns, and addition of compost, both supported removal of the OMPs. Monitoring of O_2 levels and N-species provided insight to redox conditions and biochemical processes. Microbial community structure will be analyzed for both water and solid samples (16S rDNA sequencing).

[P65] Regulation of Initiation of DNA Replication in Staphylococcus aureus

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The increasing challenge of dealing with clinical infections caused by *Staphylococcus aureus* strains resistant to a wide range of conventional antibiotics, calls for the urgent need to develop novel approaches, such as identifying new classes of antimicrobial drugs. Gaining a much needed insight into how regulation of initiation of DNA replication in *S. aureus* is regulated will provide the basis for novel screening techniques aiming to find antimicrobial drugs inhibiting DNA replication, and identify the specific mode-of-action of these compounds.
In this study, we have optimized a flow cytometry-based protocol for investigating the dynamics of DNA replication initiation in *S. aureus*. Using this tool, we have aimed to investigate several mechanisms possibly involved in maintaining replication initiation control in *S. aureus*. These include:
Identifying potential DnaA binding sites affecting the control of initiation of DNA replication in *S. aureus*. We have used bioinformatic predictions in combination with ChlP-Seq analyses to find such chromosomal regions, followed by deletion analyses of these sites to assess their role in regulation of replication initiation.
Determining if and how immediate re-initiation is prevented in *S.aureus* by using a temperature sensitive *dnaA*^{TS} mutant strain.
Examining the role of a YabA homologue in maintaining replication initiation control in *S. aureus* by constructing mutant strains either overproducing or lacking a homologue to YabA. YabA acts as a negative modulator of initiation of DNA replication in *B. subtilis.*

[P66] Detection of deazapurine modifications in phage DNA using nanopore sequencing

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Bacteria, continually exposed to bacteriophages, have developed several defense mechanisms e.g.
restriction-modification or CRISPR-Cas systems. On the other hand, bacteriophages have developed several strategies to evade these mechanisms.
Escherichia coli virulent siphophage CAjan, belonging to Seuratvirus genus and the closely related
genus nonagvirus, contains a homolog of the tRNA-deazapurine modification synthesis pathway.
The evidence is piling up, that CAjan and other similar bacteriophages use these deazapurine modifications in their DNA to evade bacterial restriction-modification systems. In order to investigate this novel DNA modification pathway in detail, we have used several methods including; direct phage plaque sequencing, CRISPR-Cas editing of phage genome and nanopore sequencing of viral DNA. Through generation of specific mutants within the deazapurine modification synthesis pathway followed by nanopore sequencing, we were able to 1) obtain a restriction-sensitive phenotype in the CAjan bacteriophage and 2) detect the modified bases using nanopore sequencing, thus providing new insights on the use of alternative bases by bacteriophages.

[P67] Cable bacteria: from single filament taxonomy to clonal enrichments in autoclaved sediment

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Cable bacteria are multicellular, centimeter-long filamentous bacteria that conduct electrons from sulfide in deeper sediment layers to oxygen at the sediment surface. This recently discovered life form, based on long-distance electron transfer, is functionally and morphologically conspicuous but has so far evaded isolation into pure culture. Whole genome amplification (WGA) using individually picked cable bacteria filaments resulted in almost complete 16S rRNA, which formed the basis for proposing two novel candidate genera, the marine *Ca.* Electrothrix and the freshwater *Ca.* Electronema. However, cable bacteria genomes obtained by WGA are incomplete and fragmented, resulting in erroneous genome-to-genome comparisons and uncertain taxonomic assignments. The alternative to WGA, assembly of cable bacteria genomes from metagenomes, is hampered by high cable bacteria (micro)diversity in natural sediments. Here we present a method to establish clonal cable bacteria enrichments, facilitating assembly of high-quality cable bacteria genomes for taxonomy and omics analyses.
Autoclaved freshwater sediment was inoculated with single cable bacteria filaments using glass hooks. Microsensor measurements confirmed the development of active cable bacteria.

After repeated single-filament transfers, an almost complete genome of a new cable bacterium species could be reconstructed from a mini-metagenome. Comparing average nucleotide identities between transfers verified the clonal nature of the cable bacterium enrichment. Similar approaches with single-cell transfers into natural medium may aid the genomic characterization and taxonomic delineation of other hard-to-culture microbes.

[P68] Presence of extended-spectrum cephalosporin (ESC) resistance Escherichia coli in two Danish poultry slaughterhouses

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The prevalence of extended-spectrum cephalosporin (ESC) producing *Escherichia coli* was investigated from two Danish broiler slaughterhouses. Samples were collected as skin samples and fecal samples. 149 ESC *E. coli* isolates from 2017-18 were whole genome sequenced. Among isolates from 2018 two isolates were saved from ten samples. Presence of ESC resistance genes and multi-locus sequence typing (ST) are presented here.
The $bla_{\text{CMY-2}}$ was detected in 118 isolates (79%), $bla_{\text{CTX-M1}}$ in six isolates (4%), $bla_{\text{TEM-52}}$ in three isolates (2%), one isolate of $bla_{CTX\text{-}M14}$ and one of $bla_{SHV\text{-}1}$. In 20 (13%) isolates upregulated *ampC* gene was detected. The $bla_{\text{CMY-2}}$ was in 76% detected ST2040, 13% in ST429, and the rest in different ST-types. ST429 containing $bla_{\text{CMY-2}}$ has been detected in broiler breeding system in Denmark. For upregulated *ampC* (n = 20), these were found in ST23 and ST4663 and $bla_{\text{CTX-M1}}$ (n = 6) was found in ST115, ST580, ST616 and ST1616. All three $bla_{TEM\text{-}52}$ isolates were ST115.
In 33 matching isolates from skin and fecal samples ESC genes and ST-types were compared. Among these, 33% (n=20) from slaughterhouse A and 15% (n=13) from slaughterhouse B revealed different STs or ESC genes. This discrepancy indicates two possibilities; either more clones exist per sample or a possible cross contamination has occurred during slaughter.

[P69] q2-CSCS: A QIIME2 plugin for integrating chemical information from LC-MS/MS experiments into PCoA analysis

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Metabolomics has become an important technique in microbiology that can elucidate many molecular aspects of both isolates and in situ microbial behaviour. Tandem mass spectrometry (MS) has the potential to substantially improve metabolomics experiments by augmenting chemical structural information through spectra of fragmented ions. These fragmentation spectra can be represented as mass spectral molecular networks identifying similarities in chemical structures. The chemical structural and compositional similarity (CSCS), a similarity between the full set of signals in sample pairs has previously been derived. Mass spectral molecular networking is facilitated by the online processing platform Global Natural Products Social Molecular Networking (GNPS) and has become a widely used tool for chemical identification and analog searching because it represents a much richer set of information about ions than retention time and m/z values alone. This information has been used both for in depth investigations of natural products and large-scale metabolomics projects such as the American Gut Project and the Global Foodomics project. This poster will present the implementation of q2-CSCS, a complete and fast implementation of the CSCS distance that can be downloaded here (https://github.com/madeleineernst/q2-cscs). This plugin, along with the rest of the QIIME2 and GNPS infrastructure can facilitate the analysis of LC-MS/MS data in a robust and user-friendly manner. We demonstrate constructed examples of LC-MS/MS data that highlight situations in which chemical information is particularly helpful and we show improvement in data analysis of a real-world dataset.

[P70] GC-bias in high-throughput sequencing impacts genomics and metagenomics

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High-throughput sequencing (HTS) is now a cornerstone of many fields of biology. However, coverage biases (systematically uneven coverage) in HTS impact applications such as genome sequencing and shotgun metagenomics, among others. Here, coverage biases related to GC-content were investigated in various HTS workflows (Illumina, Nanopore and Pacbio platforms and different library preparation protocols) for shotgun metagenome sequencing datasets and for single genome sequencing experiments of a range of bacteria with contrasting average GC-contents. Generally, AT-rich and GC-rich 500 bp genome regions were seen to receive much less coverage relative to the optimal coverage seen in genome windows with moderate GC-contents (ca. 45% to 65% GC). The manifestation of GC-biases in HTS datasets was shown to differ considerably between platforms and between library preparation methods, particularly in the metagenome datasets we analysed. On Illumina's MiSeq platform, libraries prepared with the Nextera XT kit showed very strong GC-biases, with regions outside of 50-60% GC content suffering from severe under-coverage. We also show that GC-content correlates very tightly with coverage biases in some workflows and it may be possible to account for coverage biases during data processing, while in other workflows the correlation between GC-content and relative coverage is likely too weak to correct during data processing. Our results indicate that abundances of organisms with non-optimal genome GC-contents are severely underestimated in metagenome studies.

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Med kunden i fokus

Hos Ninolab har vi fokus på høj kvalitet og sætter altid dine behov i centrum. Vi hjælper dig med at skræddersy en løsning hvad enten det gælder enkelte produkter eller hele systemer. Vores kvalitetsmål er at kunne levere et korrekt tilpasset produkt, med de rigtige funktioner, til aftalt tid og ikke mindst til den rigtige pris. De produkter Ninolab har valgt at forhandle, er alle nøje udvalgt blandt leverandører fra hele verden.

Tryghed når du vælger Ninolab

For dig som kunde er det en tryghed at vide, at når du vælger Ninolab som leverandør vil du altid blive tilbudt de bedste produkter. En god support i kraft af veluddannet personale er også en del af denne tryghed og hos Ninolab er vi meget bevidste om at vores ansvar strækker sig længere end til leveringen af selve produktet. Vores produktspecialister er til rådighed ved spørgsmål om applikationer og support generelt og vores serviceafdeling tilbyder alle former for serviceydelser på dine produkter.

Produkter fra verdens bedste leverandører

Vi følger konstant og systematisk udviklingen på vort område. Vi udvælger og skaber kontakter med de producenter der er kendte for deres seriøsitet og ansvarlighed i fremstillingen af laboratorieprodukter i højeste kvalitet. Det er producenter der satser på forskning og udvikling og tillægger deres produktion strenge kvalitetskrav. Ninolab forhandler på nuværende tidspunkt produkter fra omkring 30 udvalgte producenter fra hele verden.

Kvalificerede og engagerede medarbejdere

Det er os der taget ansvaret. Vi har specialister indenfor alle områder og samlet giver det den styrke der kræves for at tage et aktivt ansvar for avanceret laboratorieudstyr. Vores produktspecialister er til rådighed ved spørgsmål om applikationer og support generelt og vores serviceafdeling tilbyder alle former for serviceydelser på dine produkter.

Kontakt os hvis du vil vide mere om hvordan gode produkter bliver endnu bedre med den rette leverandør!

Ser du Netflix? Har du set The Rain, så er alt filmet laboratorieudstyr leveret af A/S Ninolab!

Din leverandør af basisudstyr til laboratorier

Vores kerneværdier:

Vores engagement for at levere topkvalitet

Kunde fokus:

Betjener vores kunder og opfylder deres behov

Engagement:

Vi giver aldrig op i vores ambition om at overgå vores kunders forventning

Pålidelighed:

Vi skal levere hvad vi lover og altid tage ansvar for vores handlinger

Fleksibilitet:

Vi tilpasser os kundernes behov og ønsker

Åbenhed:

Vi kommunikerer åbent og ærligt

Innovation:

Ved at fokusere på en innovativ tankegang skal vi give den bedste løsning til vores kunder

Stabilitet:

Vi har en sund finansiel platform til et langsigtet samarbejde

1% Eliteleverandør til det offentlige 2018 UdbudsVagten

ISO 9001:2015 CERTIFICATION Intertek ISO 14001:2015 CERTIFICATION Intertek

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