LUXEMBOURG SOCIETY FOR MICROBIOLOGY

4th LUXEMBOURG MICROBIOLOGY DAY

JULY 1ST 2021, 09:00 - 15:00 [HYBRID EVENT]
University of Luxembourg and Virtual Conference Room

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Luxembourg Society for Microbiology a.s.b.l

Abbreviation: LSfM
Founded in 2016; Registration number F 10800
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Programme

09:00 Welcome to participants & Opening speech by LSfM's President, Dr Jos EVEN

09:15 Keynote presentation by **Dr Manon BOURG: The State Veterinary Laboratory: One lab, two fields of action (LMVE, LU)**

10:00 3 presentations by **post-doc fellows** in microbiology:

- Adaptive genomic and metabolic traits influence microbial islands of life in alpine streams: **Susheel Busi** (Uni.lu)
- Improving metaproteomic identification by focusing on the spectra that matter: **Benoît Kunath** (Uni.lu)
- Functions and structure of electrogenic bacterial communities in bioelectrochemical systems for bioremediation: **Anna Espinoza** (University of Milano-Bicocca, IT)

10:45 Short break

11:00 Science-in-society **roundtable** discussion "**Key research in microbiology meets public health, national policy and society – The example of the national COVID-19 pandemic**"

Introduction & Moderation by **Jean-Paul BERTEMES**, Head of Science in Society, FNR Presentations by **Dr Thomas DENTZER**, Health Directorate, Ministry of Health,
Dr Joël MOSSONG, Sanitary Inspection Services, Ministry of Health,
Dr Henry-Michel CAUCHIE, Luxembourg Institute of Science and Technology (LIST)
Prof Paul WILMES, University of Luxembourg.
Open discussion with all participants

12:30 Lunch break

13:15 Keynote presentation by **Prof Colin MURRELL: Microbial growth on the climate-active gas isoprene (University of East Anglia, UK)**

14:00 3 presentations by **PhD students** in microbiology:

- Substrate-mediated active organization in sessile bacterial colonies: **Ghazaleh Eshaghi** (Uni.lu)
- Generation-scale evolution of the gut resistome under selective antibiotic pressure: **Laura de Nies** (Uni.lu)
- Interaction between the microbiome, epithelial cells and immune cells in the context of allergic airway inflammation: **Lucas Morel** (Lih.lu)

14:30 **LSfM's 5th anniversary** celebration & announcements
Award ceremony for the best oral communications (2 prizes of 250€ each)

15:00 Closing of the Luxembourg Microbiology Day


KEYNOTE
PRESENTATIONS

The State Veterinary Laboratory: One lab, two fields of action

Manon Bourg

Affiliations

Administration of Veterinary Services, Division of the Laboratory of Veterinary Medicine, Dudelange, Luxembourg

Dr Manon Bourg, manon.bourg@asv.etat.lu

Abstract

The Laboratory of Veterinary Medicine (LMVE) is a division of the Administration of Veterinary Services, acting under the responsibility of the Ministry of Agriculture, Viticulture and Rural Development, and the Ministry of Health, and ensuring compliance with laws and regulations. The main activities of the LMVE are 1) the detection of contagious animal diseases subject to mandatory reporting and 2) the detection of pathogenic germs of foodstuffs (food of animal origin or other animal products).

The LMVE has been accredited (ISO 17025) by the Luxembourg Accreditation and Surveillance Office since March 2005 and provides high quality diagnostic services for animals and the products thereof, including necropsy, bacteriology, serology, immunofluorescence, parasitology and molecular diagnostics. The LMVE processes upwards of 10.000 requests with more than 225.000 samples annually.

In addition, the LMVE is the National Reference Laboratory for Listeria monocytogenes, Campylobacter, VTEC, Salmonella and antimicrobial resistance together with the Laboratoire National de Santé (LNS). The LMVE is involved in epidemiological investigations and contributes to research of animal diseases, together with other Luxembourgish and foreign institutions.

Microbial growth on the climate-active gas isoprene

Colin Murrell

Affiliations

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Professor J. Colin Murrell, j.c.murrell@uea.ac.uk

Abstract

Isoprene (methyl isobutene) is a climate-active volatile organic compound that is released into the atmosphere in similar quantities to that of methane, making it one of the most abundant trace volatiles. Large amounts of isoprene are produced by trees but also substantial amounts are released by microorganisms, including algae in the marine environment. The consequences on climate are complex. Isoprene can indirectly act as a global warming gas but in the marine environment it is also thought to promote aerosol formation, thus promoting cooling through increased cloud formation. We have shown that aerobic isoprene degrading bacteria are widespread in the environment. Rhodococcus AD45 and Variovorax WS11, our model isoprene degraders, oxidize isoprene using a soluble diiron centre monooxygenase is similar to soluble methane monooxygenase and has considerable potential as a biocatalyst for biotransformations and bioremediation. The physiology, biochemistry and molecular biology of isoprene degraders will be described, together with genome analysis, transcriptome analysis and regulatory mechanisms of isoprene degradation by bacteria. The distribution, diversity and activity of isoprene degraders in the environment has also been studied using functional gene probing, DNA-stable isotope probing, metagenomics, metatranscriptomics and metaproteomics. Results indicate that isoprene-degrading bacteria are widespread in soils, leaf surfaces and estuarine sediments and that they are likely to play a major role in the metabolism of isoprene before it escapes to the atmosphere. Focussed metagenomics using DNA-SIP has facilitated capture of the genomes of new isoprene degraders. Concomitant cultivation studies have enabled the isolation and characterisation, at the physiological and molecular level, of novel isoprene degraders, all of which use a common pathway, initiated by isoprene monooxygenase, to degrade isoprene.

ORAL PRESENTATIONS

Adaptive genomic and metabolic traits influence microbial islands of life in alpine streams

Susheel Bhanu Busi[1,*], Valentina Galata[1], Paul Wilmes[1], & Tom J. Battin[2]

Affiliations

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Abstract

Microbial life in streams and rivers is dominated by complex communities organized in multi-trophic biofilms. In glacier-fed streams (GFSs), biofilms colonize various habitats, including streambed sediments and more stable habitats on rocks and boulders. Here we elucidate the cross-domain structure and putative functions of GFS biofilm microbiomes to gain insights into the adaptive strategies of these multi-trophic assemblages to these cold and dynamic ecosystems. We focus on biofilms forming on larger rocks and boulders (i.e. epilithic biofilms) and thus sheltered from physical disturbances.

Using metabarcoding, we first compared epilithic bacterial and eukaryotic biofilm communities to those of the respective stream sediments, finding that the epilithic biofilms harbor distinct communities. Based on subsequent metagenomic assessments, we report for the first time that epilithic biofilms featuring numerous cross-domain interactions are major contributors to microbial lifestyles. Additionally, we identify genomic adaptations tailored towards energy acquisition, including metabolic and biogeochemical pathways related to cold environments. Interestingly, we also find that Polaromonas, an abundant and prevalent bacterial genus identified in epilithic GFS biofilms, is markedly different to currently known species from other biomes, encoding functions associated with cold and stress adaptations. Collectively, our findings highlight the structure and metabolic strategies of epilithic biofilms from GFSs, elucidating previously uncharacterised lifestyles within these pristine ecosystems.

Improving metaproteomic identification by focusing on the spectra that matter

Benoît J. Kunath[1,*], Shengbo Wang[2], Satwant Kaur[2], Oskar Hickl[1], Juan Antonio Vizcaíno[2], Rob Finn[2], Patrick May[1], Paul Wilmes[1]

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Abstract

The human gut microbiome has been extensively studied because of its profound influence on human physiology and involvement in a range of chronic diseases. As microbial proteins perform the majority of cellular functions, their analysis via metaproteomics has emerged as the most relevant approach to characterize the functional expression of a microbial community and elucidate its activity. However, a majority of the mass spectrometry (MS/MS) spectra obtained from shotgun metaproteomics cannot be assigned to a peptide. Indeed, typical studies only achieve an identification rate of ~30% spectra, which substantially limits the analysis of the datasets to their full potential. The low percentage of spectra assigned to peptides is notably due to the limited extent of sequence databases as well as spectra with unknown or unconsidered modifications.

We developed a workflow to integrate sample-specific metagenomic sequencing information with the recent Unified Human Gastrointestinal Genome[1] (UHGG) catalogue to improve the identification rate in metaproteomics. The workflow utilizes a comprehensive database while retaining the sample specificity and leads to improved MS/MS spectra identification.

Furthermore, metaproteomic samples do not necessarily have associated metagenomic data. Spectral library searching[2] is a powerful technique that could mitigate the need for a sequence database and has the potential to increase the efficiency of the searches. While spectral libraries remain a substantially underutilized resource, here we present spectral clustering and libraries applied for the first time to metaproteomic datasets. We show that our method can extract information from relevant, yet unidentified spectra and allow focusing on those for subsequent identification, notably via de novo analyses. Ultimately, our workflow identifies more spectra and retrieves information even from unidentified spectra, thus improving our metaproteomic analyses.

References

1. Almeida A et al. "A unified catalog of 204,938 reference genomes from the human gut microbiome." Nat Biotechnol 39(1):105-114 (2021).

2. Deutsch, Eric W et al. "Expanding the Use of Spectral Libraries in Proteomics." Journal of proteome research vol. 17,12 (2018)

Functions and structure of electrogenic bacterial communities in bioelectrochemical systems for bioremediation

Anna Espinoza[1], M. Daghio[1], E. Palma[2], F. Aulenta[2], A. Franzetti[1]

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Abstract

Bioelectrochemical Systems (BESs) is an innovative technology that has been recently studied to stimulate the anaerobic degradation of hydrocarbons. Understanding the microbial community structure and genetic potential of anode biofilms is of great interest to interpret the degradation mechanisms that take place in BES.

In this study, the removal of hydrocarbons (phenol, toluene and a mixture of BTEX) was assessed in continuous-flow BES and the composition of the anodic microbial communities in the inoculum and in the anodic biofilm after the treatment was characterized with shotgun metagenome sequencing approach, to obtain taxonomic and functional diversity patterns of the microbial communities.

The system operated with phenol was inoculated twice: first with a municipal activated sludge (inoculum 1) and after 33 days was re-inoculated with a refinery wastewater from a petrochemical plant (inoculum 2). Inoculum 1 was used to inoculate the runs with toluene and BTEX respectively.

The structure of the bacterial community was mainly composed by the genus Geobacter, that was highly enriched on the anodes of the three systems. Geobacter species have been related to BES, due to their capacity to degrade hydrocarbons in absence of oxygen and to use the anode as solid electron acceptor.

To functionally characterize the microbial community, the genes coding for the anaerobic degradation of toluene, ethylbenzene, and phenol were selected as genetic markers for the anaerobic degradation of the pollutants. The genes related with direct extracellular electron transfer (EET) were also analyzed. The inoculum carried the genetic baggage for the degradation of aromatics but lacked the capacity of EET while anodic bacterial communities were able to pursue both processes. The metagenomic approach provided useful insights into the ecology and complex functions within hydrocarbon-degrading electrogenic biofilms.

Substrate-mediated active organization in sessile bacterial colonies

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Abstract

The human gut is a highly dynamic physico-chemical environment, harboured by a highly diverse population of bacteria that actively respond to a multitude of parameters including physical variations, biochemical cues (e.g., hormones) and dietary intake[1]. The aim of the my doctoral research is to develop a bottom-up mechanistic framework of microbial strategies within gut-inspired environment, and analyze their functional ramifications toward maintenance of a healthy gut. Specifically, by varying the substrate properties on which bacteria grows, we decipher how bacteria can selectively sense and switch between growth substrates resembling different consistencies of colonic mucus under fiber-rich and depleted settings. We use a combination of microfabrication and high-resolution imaging techniques to simulate, iterate and optimize gut-relevant cues within custom-built microenvironments. Bacterial species are allowed to grow within these environments and tracked using time-lapse imaging. Our results suggest that changing substrate stiffness, alongside temperature[2], has a significant effect on the morphological transitions during the early stages of colony development. The visualization-based quantitative approach introduces a novel angle, taking us a step closer to the biophysical principles mediating this complex ecosystem, their dynamic emergence, in particular substrate stiffness – a key biophysical determinant of microbial growth. By harness the microscale scale insights, our goal is to develop predictive tools for early diagnosis of inflammatory bowel disease, colon cancer and a host of other gut-induced ailments.

References

(1) Martens et al., Interactions of commensal and pathogenic microorganisms with the intestinal mucosal barrier, Nature Reviews Microbiology 16, 457-470. 2018
(2) Dhar, Thai, Ghoshal, Giomi & Sengupta, Trade-offs in phenotypic noise synchronize emergent topology to actively enhance transport in microbial environments, arXiv:2105.00465, 2021

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Generation-scale evolution of the gut resistome under selective antibiotic pressure

Laura de Nies[1,*], Susheel Bhanu Busi[1], Mina Tsenkova[2], Rashi Halder[1], Elisabeth Letellier[2], Paul Wilmes[1]

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Abstract

Prior to the advent of antibiotics, bacterial infections were the leading cause of disease and mortality in humans. However, many bacterial taxa have evolved antimicrobial resistance (AMR) to several classes of antibiotics, and multidrug-resistant bacteria have now emerged, preventing comprehensive treatment thus representing an ever-growing healthcare challenge worldwide. Nevertheless, the mechanisms and time-scales shaping this resistome remain elusive. Therefore, using a cocktail of antibiotics administered to a murine model along with a detailed, longitudinal sampling strategy, we identify the mechanisms by which gut commensals acquire AMR after a single course of antibiotic treatment.

While most of the resident bacterial populations were depleted due to the treatment, Akkermansia muciniphila and members of the Lachnospiraceae family developed resistance and remained recalcitrant. We further identified specific genes conferring resistance against the antibiotic cocktail in the corresponding metagenome-assembled genomes (MAGs) and traced their origins within each genome. Additionally, we identified AMR genes encoded on mobile genetic elements (MGEs). While we found MGEs, including plasmids and phages, contributing to the spread of AMR, we observed that integrons represented the primary factors mediating AMR in the antibiotic-treated mice. Consequently, our findings suggest that antibiotic treatment alone may act as the selective pressure driving AMR acquisition and incidence in gut commensals on a generational timescale.

Interaction between the microbiome, epithelial cells and immune cells in the context of allergic airway inflammation

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Abstract

Allergic asthma is a chronic inflammatory disease triggered by inhaled allergens and other environmental factors. In asthmatic patients, chronicity is characterized by airway inflammation and epithelial alterations. Several studies have shown the role of microbiota in the regulation of immune function and the development of asthma. Other studies have explored the link between immune cells and nerve cells in the case of asthma and it turns out that Neurturin (NRTN), which is a member of the glial cell line-derived neurotrophic factor family, has beneficial effects on the inflammatory response. In addition, the absence of the receptor of NRTN, RET, leads to alter microbial communities in the gut [1]. The objective of the project is to study the interactions between epithelial cells, immune cells and the microbiome in the context of allergic airway inflammation. Our team has described that NRTN Knock-Out (KO) mice have more severe asthmatic phenotype than Wild-Type mice [2, 3] and we have shown that receptors of NRTN (GFRα2 and RET) are expressed on immune cells and lung epithelial cells. As changes in gut microbiota have a link with asthma development and as there is a communication between lung and gut microbiomes, we compared the microbiome composition of NRTN KO and WT mice by 16S sequencing first in the gut at different timepoints and lastly in the lungs. Mice where at steady state or in asthma condition obtained by nasal instillation of House dust mite (HDM) for two weeks. We found a few significant differences when looking at the taxa frequencies of the gut microbiome.

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