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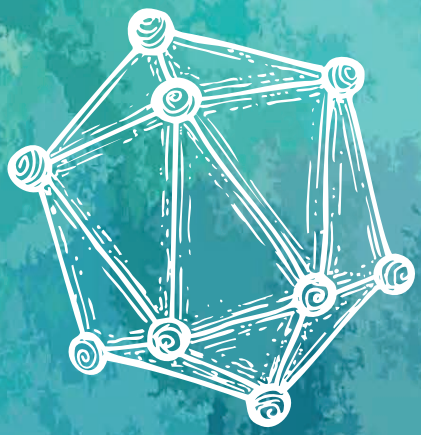

Swiss Society for Microbiology Annual Congress 2026

From 26 to 28 August 2026

2m2c Montreux Music & Convention Center

ABSTRACT BOOK

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NUMBERING LEGEND

K = Keynote talk
M = Main talk
MT = My Thesis in 180 seconds
P = Poster
PF = Posterflash
S = Short talk
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K01 Expansion Microscopy Atlas of Microbial Eukaryotes

Omaya Dudin, PhD

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Summary: Microbial eukaryotes represent the majority of eukaryotic diversity, yet the cellular architecture underlying their remarkable phenotypic range remains poorly characterized. While genomic data has transformed our understanding of protistan evolution, corresponding cell biological information, particularly from high-resolution microscopy, remains sparse, limiting our knowledge of how cellular structures diversify and how this diversity relates to evolutionary transitions. We are developing an Expansion Microscopy Atlas of Microbial Eukaryotes (ExÂME), a systematic effort to characterize cytoskeletal and cellular diversity across hundreds of species using Ultrastructure Expansion Microscopy.

U-ExM overcomes key technical barriers to imaging these organisms: it achieves nanoscale resolution while improving antibody and dye accessibility, addressing permeability constraints that have traditionally limited microscopy of microbial cells. We have optimized high-throughput imaging workflows for non-model organisms collected from natural environments, systematically validated cytoskeletal markers across diverse lineages, and established screening protocols to identify conserved and divergent organizational features.

Critically, we are building this atlas through direct engagement with the research community. Rather than selecting organisms in isolation, we partner with culture collections and research groups to prioritize species based on key evolutionary questions: mitotic diversity, multicellular transitions, and cytoskeletal innovation. This collaborative approach ensures the atlas reflects shared scientific priorities while providing a foundation for mechanistic understanding of how cellular architecture evolves.

With support from the Gordon and Betty Moore Foundation, this project aims to bridge fundamental gaps between genomic and phenotypic data. By making imaging data openly available , we establish a living resource that accelerates discovery and enables the broader adoption of expansion microscopy across microbial research.

K02 Carnivorous Fungi: From Molecules to Ecosystems

Yen-Ping Hsueh

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Abstract not received yet.

K03 Deltaviruses spread through a viral Trojan Horse

Karim Majzoub, PhD

IGMM, Montpellier, France

Hepatitis D-like satellite viruses, known as deltaviruses, have been recently discovered in a wide range of animals. These viruses are thought to expropriate glycoproteins from helper viruses to form infectious particles. During the talk, I will challenge this paradigm and demonstrate that deltaviruses are packaged within helper virus particles, using them as viral Trojan Horses for cell entry. By leveraging orthogonal electron and optical super-resolution microscopy, we visualize deltaviruses enclosed within virions from rhabdo-, herpes-, and arenavirus families. We show that this conserved hitchhiking mechanism ensures concomitant deltavirus-helper virus spread, thereby promoting the dissemination of deltaviruses, broadening their host range, and expanding their tropism. Our findings reveal a previously unrecognized mode of viral transmission, providing a framework to investigate overlooked deltavirus infections outside of the human liver.

K04 Molecular mechanisms of bacterial cell division in filamentous bacteria

Susan Schlimpert

John Innes Centre, Norwich, United Kingdom

Abstract not received yet.

K05 Innovating Against AMR: The Role of Diagnostics

Robin Patel, M.D.

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Antimicrobial resistance (AMR) is a global crisis. Antibiotics are lifesaving, but their lifespan is oftentimes limited. Empiric use is rampant; patients who *need* specific antibiotics may not get them, and conversely, others may receive unneeded antibiotics. Reasons for empiric use are many - an important consideration is that when most antibiotics were introduced, suitable diagnostics supporting their use or non-use were unavailable, so they "had" to be used empirically. Today, we are in a diagnostics revolution; we *can* know exactly what is ailing our patients, including which bacterium is present, if any, and which antibiotic will work against it. We are not all the way in terms of developing what is needed, but we are heading in a good direction. Rather than empiric use, antibiotics should be deployed when there is a bacterial infection that needs treatment, and the "right" antibiotic used. If patients have infections with susceptible bacteria, usual antibiotics can be given; with resistance, antibiotics targeting resistant bacteria should be used. This is good for AMR as it decreases the selective pressure that breeds more resistance. It is good for patients, because it decreases their likelihood of harboring resistant bacteria that may infect them in the future, minimizes microbiome disturbances that may be linked to adverse health effects, and decreases antibiotic toxicity. Novel diagnostics can also inform use of oral instead of injectable antibiotics, often used because of concerns about AMR. Innovation in diagnostics is not just about making more accurate and faster diagnoses; it is also enabling new ways of delivering healthcare, outside of the walls of traditional healthcare facilities. Rapid and point-of-care diagnostics are changing the way people access and use antibiotics, directly impacting AMR. In this lecture, Professor Patel will highlight recent advances in diagnostics and how they may help in the fight against AMR.

M01 Current AST Practices in Swiss Clinical Microbiology Laboratories: A Nationwide Survey

Stefano Mancini

UZH, Zurich, Switzerland

Background: Harmonized antimicrobial susceptibility testing (AST) is essential to ensure comparable reporting, effective antimicrobial resistance surveillance, and optimal patient management across Swiss laboratories and hospitals. The Swiss AST Committee (SAC) conducted a national survey on current AST practices to support future discussions, guideline development, harmonization efforts, and educational activities, enabling anonymous benchmarking.

Methods: An online questionnaire was distributed in early April 2026 to Swiss clinical microbiology laboratories performing bacterial AST.

Results: Responses were received from 41 laboratories, including private laboratories (17/41, 41.5%), cantonal hospital laboratories (13/41, 31.7%), university hospital laboratories (5/41, 12.2%) and others (6/41, 14.6%). Daily AST workload ranged from 10–60 antibiograms in 27/41 laboratories (65.9%), while 5/41 (12.2%) performed more than 100 per working day. VITEK 2 was the most used first-line AST method (27/41, 65.9%), followed by disc diffusion (10/41, 24.4%), whereas other commercial broth microdilution-based systems were used by a minority of laboratories. Overall, DD was used by 37/41 laboratories (90.2%), gradient diffusion by 34/41 (82.9%), and VITEK 2 by 28/41 (68.3%). EUCAST breakpoints were used either exclusively by 17/41 laboratories (41.5%) or in combination with alternative breakpoints when EUCAST criteria are unavailable in the remaining 24/41 (58.5%). The EUCAST "I" category was reported as "susceptible, increased exposure" by 37/41 laboratories (90.2%). EUCAST updates were implemented within 3 months by 21/41 laboratories (51.2%). ESBL confirmation relied predominantly on double-disk synergy testing (30/41, 73.2%). Anaerobic AST was performed by 27/41 laboratories (65.9%), mostly using gradient diffusion (23/27, 85.2%) or DD (14/27, 51.9%). ATU results were mostly managed by repeat testing with an alternative method (20/41, 48.8%). Rapid AST directly from positive blood cultures (RAST) was implemented by 19/41 laboratories (46.3%).

Conclusion: Swiss laboratories show broad adherence to EUCAST recommendations, although relevant variability persists in selected AST practices. These findings support future national harmonization efforts and guideline development.

M02 Taste the Microbiome: The Ecology of Microbial Terroir in Fermented Foods

Lena Flörl

ETHZ, Zurich, Switzerland

Fermented foods are the result of the transformation of a substrate by microorganisms and their interactions with environmental conditions and human practices. Their relatively simple, accessible, and experimentally tractable microbial communities therefore make fermented foods powerful model systems for studying the ecological and biogeographic processes underlying microbiome assembly and function.

These *microbial terroir* processes are Building reflected on the traditional concept of *microbial terroir*, *terroir* also describes called how the biogeographic "taste of patterns placed", microbial communities contribute to location specific characteristics of fermented foods. In winegrowing, this phenomenon has been reported across the globe yet the ecological drivers of these microbial communities remain incompletely understood.

Hence this work investigates how environmental conditions, vineyard management, and host genetics jointly shape microbial community assembly in vineyards and wine fermentation outcomes. A multi-year survey of 95 Swiss vineyards integrating amplicon sequencing, metabolomics, environmental monitoring, and sensory analyses shows that climatic and topographic gradients, together with fruit chemistry, structure microbial communities and influence fermentation dynamics, ultimately contributing to regional differences in wine composition and aroma. Beyond these environmental drivers, vineyard management practices further modulate microbiome composition and function, with cover cropping and fungicide use altering soil and grape associated microbial communities. Finally, quantitative trait locus (QTL) mapping reveals that grapevine genetics also contribute to microbiome assembly, affecting both fermentative yeasts and pathogenic fungi through loci associated with plant immunity and stress response, highlighting host level control over microbial community structure.

Collectively, these findings show that *microbial terroir* emerges from the interaction of environmental conditions, human practices, and host biology. More broadly, they illustrate how fermented foods provide a tractable framework for uncovering general ecological principles of microbial biogeography and community assembly while linking these processes to the sensory characteristics – making it possible to taste the microbiome in action.

M03 Jumping Genes and Pathogenic Schemes: The diverse role that transposons play in plant pathogen evolution

Megan C. McDonald

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Our work over the last several years has sought to understand how transposons drive adaptive evolution in plant pathogenic fungi, namely the necrotrophic wheat pathogens causing spot blotch, tan spot and Septoria nodorum blotch. Fungal genomes are highly variable in their transposon content and contain many novel transposons that have yet to be functionally characterised. We now have very detailed examples of how transposons drive the duplication, pseudogenisation, and horizontal movement of effector genes, all of which can have real world impacts on the incidence and severity of disease in the field. This talk will explore the evolutionary history of the capture and horizontal transfer of a novel Class II DNA transposon, ToxTA, by different giant *Starship* transposons. ToxTA carries the well-characterised necrotrophic effector *ToxA,* whose origins have remained a source of debate since the first horizontal transfer was reported in 2006. Here, we explored the diversity of the two best characterised *Starships,* Sanctuary and Horizon, within the *Bipolaris, Parastagonospora* and *Pyrenophora* genera in order to better define the direction and timing of horizontal gene transfer between these the species carrying the *ToxA* effector. I will also present some preliminary work towards establishing novel experimental systems to study the mechanisms of *Starship* transposon mobilisation and horizontal transfer outside of their native fungal hosts.

S01* Malassezia at the Host Interface in Health and Disease

Ms. Fiona Fischer[1], Prof. Salomé LeibundGut-Landmann[2]

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The human skin mycobiome is dominated by a single fungal genus, *Malassezia.* As a pathobiont, it has important functions in the maintenance of the skin barrier function in a healthy state. However, changes in the skin environment and/or in fungal properties can lead to the disturbance of this symbiosis. *Malassezia* is associated with the exacerbation of several inflammatory skin diseases such as atopic dermatitis (AD), a disease affecting up to 10% of the adult population and characterised by a disrupted epidermal barrier, and a pronounced type 2 immune response. Previous work has revealed the secreted aspartyl protease 1 (SAP1) to be higher expressed in *Malassezia* collected from AD patients compared to healthy volunteers and SAP1-knock-out mutants have shown decreased inflammatory host responses compared to the wildtype strain in a murine model of AD. Another pathogenicity trait of *Malassezia* is the switch from the common yeast form into a filamentous form, which is frequently observed in pityriasis versicolor, a pigmentation defect common in hot and humid climates. In my PhD project, I aim to investigate how *Malassezia* adapts to different skin states, how its morphology and repertoire of secreted factors shifts during the transition from commensalism to pathogenicity and to characterise the subsequent host immune response. By comparing the fungal transcriptome of *Malassezia* grown on AD-like skin to *Malassezia* grown on intact skin, such factors have been identified and will now be validated by generating *Malassezia* mutants and investigating how they affect the host response. Further, I will characterise the hyphal morphology of *Malassezia* and its implications on the fungus-host interaction.

**Student Paper*

S02 Four cases of persistent dermatophytosis caused by terbinafine-resistant Trichophyton indotineae diagnosed in a private laboratory

Mrs. Geraldine Jost[1], Dr. Béatrice Ninet[2], Dr. Emmanuel Ebengo[3], Dr. Janine Fumeaux Roh[4], Dr. Manon Rosselin[1], Dr. Nadia Liassine[1]

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Background:

Trichophyton indotineae is an emerging dermatophyte associated with refractory dermatophytosis and increasingly reported resistance to terbinafine. We present four clinical cases diagnosed in a private laboratory between July 2025 and February 2026 highlighting the diagnostic challenges and resistance patterns of this pathogen.

Methods:

Specimens were examined by direct microscopy and inoculated onto specific media. Identification of isolates was initially performed by morphological identification and/or MALDI-TOF (Bruker). Since May 2025, our diagnostic approach has been reconsidered for patients with a history of persistent dermatophytosis and/or with colonies of *Trichophyton mentagrophytes* from skin samples. The identification was then performed using MALDI-TOF and the MSI library. Sequencing of the ITS region was also added to confirm the identification.

Antifungal susceptibility testing was performed in a microplate using colorimetric detection to determine the minimal inhibitory concentration (MIC) (Sensititre, Thermo Fisher Scientific), according to the EUCAST method, except for the reading of the cell viability by Alamar blue, which changed from blue to pink in the presence of growth.

Results:

Four patients, two from the canton of Geneva and two from the canton of Valais were diagnosed with *T.indotineae*. They all presented with persistent dermatophyte infections including widespread tinea corporis (n=2) and tinea cruris (n=2). One autochthonous case was reported. Treatment failure with terbinafine was observed in three patients. Another patient was unsuccessfully treated with isoconazole and clotrimazole.

Antifungal susceptibility testing revealed high minimum inhibitory concentrations for terbinafine, consistent with clinical failure. Itraconazole was active against all isolates.

Conclusions:

These cases highlight the clinical relevance of terbinafine-resistant *T. indotineae* and the importance of combining culture, MALDI-TOF and sequencing for accurate identification and treatment. Early recognition of *T.indotineae* and resistance patterns supports effective antifungal therapy and help prevent treatment failure and transmission. The autochthonous transmission of *T.indotineae* has been documented in Switzerland.

MT04*/S03*/P008* Exploration of novel mechanisms of manogepix resistance in Candidozyma auris

Ms. Anaïs Hautefeuille[1], Mr. Miguel Suarez Robles[1], Ms. Danielle Brandalise[1], Prof. Dominique Sanglard[1], Dr. Jizhou Li[1], Prof. Frédéric Lamoth[1]

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Context: *Candidozyma auris* is an emerging multidrug-resistant yeast responsible for invasive infections. Manogepix is a novel antifungal agent that inhibits Gwt1, a key enzyme in the glycosylphosphatidylinositol anchor biosynthesis pathway required for proper cell wall protein localization and fungal viability. However, *C. auris* can develop reduced susceptibility to manogepix. *TAC1b* mutations resulting in overexpression of *CDR1* efflux pump and reduced susceptibility to both manogepix and fluconazole have been reported.

Aim: In this study, we plan to explore *TAC1b*-independent genetic mechanisms contributing to reduced manogepix susceptibility in *C. auris*.

Methods: A *TAC1b* deletion strain was subjected to *in vitro* evolution under manogepix exposure. Evolved strains were characterized by antifungal susceptibility testing to both manogepix and fluconazole, whole-genome sequencing, and RT-qPCR of *CDR1*.

Results: Five evolved strains with reduced susceptibility to manogepix were obtained. Two strains harbored mutations in the manogepix target gene *GWT1*, while two other strains carried a mutation in a gene encoding an enzyme involved in phospholipid biosynthesis. All four strains did not show cross-resistance to azoles and did not exhibit *CDR1* overexpression. The last strain carried a mutation in *UBR2* (a ubiquitin-ligase involved in control of *CDR1* expression), which was associated with increased *CDR1* expression and reduced manogepix and fluconazole susceptibility.

Conclusion and perspectives: These findings provide new insights into mechanisms contributing to reduced manogepix susceptibility in *C. auris*. We showed for the first time that *C. auris* can develop mutations in the manogepix target gene *GWT1* and in a lipid biosynthesis gene, under manogepix pressure. The function of these mutations needs to be confirmed by genetic modifications.

**Student Paper*

S04∗ A conditional TE-driven hypermutator state in a major wheat fungal pathogen

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Adaptation to novel environments requires heritable genetic variation, but mechanisms that generate adaptive variants can also compromise genome stability. Transposable elements (TEs) exemplify this trade-off: their mobilization can accelerate adaptation through structural genome variation while simultaneously threatening genome integrity. In most eukaryotes, TE activity is suppressed by epigenetic silencing, particularly DNA methylation (5mC). However, fungi exhibit substantial natural variation in 5mC levels, raising the question of whether loss of 5mC promotes a TE-driven hypermutator state.

In the wheat pathogen Zymoseptoria tritici, 5mC is largely absent due to mutations in the DNA methyltransferase DIM2. Loss of DIM2 occurs predominantly in populations outside the species' center of origin and is associated with increased TE content, suggesting a link between reduced epigenetic TE control and adaptive potential.

To investigate how loss of 5mC influences genome evolution, we combined experimental evolution with long-read sequencing to compare parental and evolved lines differing in DIM2 activity. We quantified mutation and transposition rates across multiple genetic backgrounds and selective conditions. Loss of 5mC was associated with a genotype-dependent hypermutator state characterized by elevated rates of TE mobilization and structural genome variation in populations evolved at optimal conditions. In contrast, populations evolved under heat stress accumulated fewer transposition events, suggesting that directional selection efficiently purges TE-mediated mutations.

Our results indicate that TE insertions accumulate as standing genetic variation under permissive conditions, whereas selective environments increase allele fixation while limiting the persistence of non-adaptive variants. Together, these findings identify loss of epigenetic TE control as a conditional hypermutator state and suggest a mechanism by which fungal pathogens may balance genome stability with evolutionary innovation.

∗Student Paper

M04 Strain transmission and microbiome dynamics in early life

Pascale Vonaesch

University of Lausanne, Lausanne, Switzerland

The human microbiome is a complex community of microorganisms that is established at birth and undergoes a coordinated succession during early life, reaching an adult-like configuration by three to five years of age. This developmental process is shaped by environmental factors, particularly birth mode, diet and lifestyle. Disruptions to the microbiome, commonly termed dysbiosis, have been implicated in the pathogenesis of many diseases, including nutritional disorders. Although extensive research has characterized the human microbiome and its implications in health and disease in populations from Europe, North America, and China, major knowledge gaps remain regarding microbiome development in low- and middle-income countries, particularly in South America, Africa, and Southeast Asia.

In this presentation, I will introduce a prospective mother-child cohort that we established in Vientiane Province, Lao PDR, to investigate how maternal malnutrition influences early-life microbiome development and child health. I will present findings on the effects of maternal and birth factors on the vertical inheritance of bacterial strains and the neonatal seed microbiome. Furthermore, I will discuss microbial developmental trajectories from birth through the first year of life and their modulation by different maternal and dietary/socioeconomic factors. Finally, I will discuss age-associated microbial signatures linked to childhood undernutrition and their implications for the development of microbiota-based strategies to improve child growth and health.

S05 Plasmids and prophages drive horizontal gene transfer in the developing infant gut microbiome

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The first months of life are a critical window for gut microbiome assembly, when bacteria colonise rapidly and compete for limited resources, creating opportunities for horizontal gene transfer (HGT) between co-occurring species. How often and through which vehicles such HGT occurs has not been resolved at a multi-cohort scale.

We tracked recent within-sample between-species (WSBS) gene sharing in 36,500 metagenome-assembled genomes from infants spanning birth to 36 months of age, drawn from 15 published studies in North America, Europe, and Asia. A WSBS event was defined as a sequence cluster sharing 100% nucleotide identity across two or more co-occurring metagenome-assembled genomes from the same metagenome. We identified 10,280 events and fitted hierarchical mixed-effects models accounting for between-study heterogeneity and within-subject longitudinal sampling.

Plasmid and prophage contigs were 2,417-fold and 544-fold more likely to carry WSBS sequences compared to the assembly-level genomic background. This indicates plasmids and prophages were the dominant genomic vehicles for recent inter-species HGT despite comprising fewer than 0.02% of the total contigs in these metagenomes. Two focal pairs anchored the biology: *Escherichia coli* and *Klebsiella pneumoniae* shared 219 genes enriched on plasmids and concentrated at 0–3 months, and *Bifidobacterium longum* and *B. bifidum* shared 248 genes enriched on prophages and transposases, consistent with the commensal *Bifidobacterium* mobilome. A small antimicrobial resistance subset (49 genes; <0.5%) was likewise enriched on plasmids and prophages. Feeding type, but not delivery mode, modulated HGT rate (mixed feeding vs breastfed IRR 1.62, 95% CI 1.29–2.04, $p = 4.2 \times 10^{-5}$).

These findings demonstrate that plasmids and bacteriophages dominate early-life inter-species gene sharing, a process further modulated by early feeding. The mobilome, rather than the static resistance-gene inventory, is a tractable target for early-life AMR surveillance, and the infant gut emerges as a natural system for studying horizontal gene transfer as it happens.

S06* Antagonistic bacterial interactions in the human gut microbiome

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Bacterial interactions in the human gut are essential for community stability and, consequently, host health. In this environment, bacteria encounter selective pressures from both the host's immune defenses and bacterial competitors. Competition is thought to be most intense among strains of the same species, due to shared ecological niches during colonisation. Focusing on the opportunistic pathogen *Escherichia coli*, a wide range of competitive strategies targeting members of the same species have been described, including bacteriocins, bacteriophages, and contact-dependent systems such as the T6SS. While these mechanisms are well studied in simplified systems, their roles in the natural gut environment remain unclear. Here, we investigate antagonistic interactions among 16 different *E. coli* strains isolated from the gut microbiomes of ten healthy adult donors. We focus on three key questions: which inhibitory mechanisms are encoded, under what conditions they are expressed, and whether they influence success during microbiome colonisation. To address this, we combine genomic analyses with pairwise co-culture assays and fecal microbiome batch cultures. We find multiple mechanisms of inhibition encoded by the strains of interest, and their cell-free supernatant showed inhibitory effects against other *E. coli* isolates. Similar inhibitory interactions are also observed in liquid coculture experiments. These findings provide insight into how intraspecific competition may shape colonization dynamics in the gut and may inform the development of targeted strain interventions with probiotics or microbial consortia therapies.

**Student Paper*

S07 Antibiotic stress promotes competitive interactions in gut-derived bacterial communities

Dr. Maxime Batsch[1], Ms. Aline Raub[2], Dr. Tatiana Galperine[1], Prof. Jan Roelof van der Meer[2]

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Exposure to abiotic stress, such as antibiotics, is expected to affect the global composition of a community by counter-selecting susceptible taxa; however, little is known about how community-level susceptibility emerges from interactions among its constituent members. To explore the role of interactions in species-rich communities, we cultivated four different human stool-derived communities, challenged by vancomycin, in bulk liquid cultures or fragmented them into three different water-in-oil microdroplet emulsions, with decreasing numbers of randomly distributed taxa per droplet, down to complete isolation of single founder cells. This fragmentation reduces opportunities for interactions and creates a gradient of interaction density. We then tracked, over one week, the global compositional changes and growth of these fragmented and bulk communities to investigate the impact of interaction density on overall community development under increasing vancomycin concentrations. At all concentrations, communities reached their highest productivity in the isolation regime, which decreased as interaction density increased, suggesting predominantly negative interactions. Conversely, higher interaction density led to the greatest diversity in the absence of vancomycin, indicating the presence of positive dependencies. The addition of vancomycin (up to 500 µg/mL) gradually reversed this trend across all four communities: the number of taxa able to grow was highest in isolation and decreased with increasing interaction density, suggesting that vancomycin promotes competitive interactions. Overall, our results demonstrate how abiotic stress can reverse prevailing ecological relationships within bacterial communities.

S08* Combined cryo-electron tomography imaging and identification of microbes as a novel tool for environmental microbiology

Mr. Nikolai Petrosian[1], Dr. Ulises Erick Rodriguez Cruz[2], Dr. Charles Ericson[1], Prof. Valeria F. Souza Saldívar[2], Prof. Martin Pilhofer[1]

1. ETH Zurich, Institute of Molecular Biology and Biophysics, Zurich, Switzerland
2. National Autonomous University of Mexico

Cryo-electron tomography (cryoET) is a powerful imaging method successfully used to study archaea and bacteria. However, most archaeal and bacterial species cannot be cultivated in laboratory conditions and the only way to study them is by analyzing complex and diverse environmental samples. Applying cryoET imaging to such samples presents a challenge because cells cannot be identified confidently. Here we present a novel experimental approach to correlate electron microscopy (EM) data with genomic identification of cells. We combine cryoET imaging with laser micro-dissection for single-cell analysis. Single or multiple EM-imaged cells are dissected directly from the EM grid and analyzed via PCR, shotgun sequencing, or 16S rRNA amplicon sequencing. The taxonomy of the dissected cells is then revealed bioinformatically. As a result, we found that for confident species identification, a sufficient number of dissected cells should be five or more. Moreover, from five pooled cells, high quality metagenome-assembled genomes (MAGs) can be obtained. We also determined the electron dose that is compatible with downstream DNA analysis and optimized for the first time laser micro-dissection from the EM grid. With this experimental approach, we provide a new method for species identification and obtaining genomes for assembly in environmental samples combined with cryoET imaging. Our developments allow application of cryoET to environmental samples for detailed study of uncultivated species.

**Student Paper*

M05 A wild-type mouse-adapted SARS-CoV-2 model captures multi-organ long COVID and enables host-focused mechanistic and interventional studies

Stefanie Bader

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Post-acute sequelae of SARS-CoV-2 infection (PASC), or long COVID, comprise heterogeneous, multi-organ manifestations for which mechanism-based therapies remain limited. A key barrier has been the lack of tractable preclinical models that reproduce persistent post-viral pathology in immunocompetent hosts. We established a wild-type mouse-adapted SARS-CoV-2 model using the P21 strain, generated by serial passaging from a human isolate, and followed disease from acute infection to post-viral timepoints after virus clearance.

In C57BL/6 mice, P21 infection caused acute disease followed by a durable PASC-like syndrome affecting lung, brain, heart and gastrointestinal tissues. Post-acute pathology included persistent lung injury with inflammatory infiltrates, haemorrhage and fibrotic remodelling, accompanied by cardiovascular and intestinal abnormalities. Mice also developed neuroinflammatory changes and impaired behavioural and cognitive performance, supporting this model as a platform to dissect multi-system long COVID.

We then used the model to benchmark early antiviral intervention. Treatment with the papain-like protease (PLpro) inhibitor WEHI-P8 improved acute virological and inflammatory outcomes and conferred stronger protection against post-acute lung pathology and brain-associated sequelae than a mouse-equivalent Mpro inhibitor regimen. However, protection was organ-selective, indicating that suppression of acute viral replication can ameliorate, but not uniformly prevent, long-term pathology. To explore mechanisms underlying neurological sequelae, we applied MERSCOPE spatial transcriptomics to post-acute brain tissue. This revealed infection-associated alterations in hypothalamic neuronal populations, including reduced orexin/hypocretin pathway expression and broader dysregulation of neuropeptide-associated genes. Together, these data establish the P21 model as a practical framework for studying multi-organ long COVID, evaluating interventions and identifying host pathways that sustain post-viral disease.

M06 RNA decay and KSHV infection: Deciphering the viral-host arms race to control RNA stability

Prof. Mandy Müller

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Abstract: Rapid and widespread turnover of host mRNA is a cornerstone of infection by the gamma herpesvirus Kaposi's Sarcoma Associated Herpesvirus (KSHV). This process is driven by the viral endoribonuclease SOX, and results in a catastrophic RNA decay event where 70% of total mRNA is lost. This widespread remodeling of the cytoplasmic mRNA landscape causes long range secondary effects, including a sudden loss of mRNA targets for many RNA binding proteins as well as a global hyperadenylation defect in the nucleus and subsequent nuclear export block. While the vast majority of the transcriptome is susceptible to SOX, a subset of host transcripts has been identified as being refractory to this virally-mediated decay event and are efficiently translated into proteins. We showed that these spared transcripts encode a 200nt cis-acting RNA element in their 3'UTR that provides resistance to SOX cleavage. However, while we know how these spared transcripts escape SOX cleavage, we have yet to understand how they escape the nuclear export block associated with SOX-mediated decay. We discovered that these SOX-resistant transcript are hyperadenylated like other transcripts. Intriguingly, these transcripts are still able to be exported out of the nucleus despite carrying these aberrant polyA tails and the longer polyA is maintained in the cytoplasm. Through this work we hope to gain insight into how virally induced alterations to mRNA influences mRNA fate, and to better understand how the dynamics of viral-host interactions alter gene expression.

S09 A primate specific transcription regulator facilitates transcriptase activity of influenza A virus polymerase

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Influenza A viruses (IAV) hijack the host nuclear machinery to transcribe their negative-sense RNA genome, relying on a viral RNA-dependent RNA polymerase (RdRP) that engages host RNA polymerase II (RNA Pol II) and "snatches" capped primers from nascent host transcripts. Despite its centrality, host support for this process remains poorly defined. Here, an ORFeome-wide cDNA screen uncovers a primate-specific transcription regulator as a potent enhancer of primary IAV transcription across diverse viral subtypes, but notably not zoonotic H5N1 and H7N9 strains. Mechanistically, it recruits and positions the viral RdRP at actively transcribing RNA Pol II complexes, promoting efficient cap-snatching and viral mRNA synthesis. These findings reveal a species-specific host dependency that shapes IAV replication and highlight host transcription factors as critical, and previously underappreciated, determinants of viral fitness.

S10* Mechanistically Distinct Therapeutic Strategies Restore Antiviral Immunity in the Presence of Neutralizing Anti-IFNβ Autoantibodies

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Interferons (IFNs) are critical regulators of innate antiviral immunity, and deficiencies in IFN signaling contribute to susceptibility to severe viral disease. In this context, autoantibodies targeting type I IFNs (anti-IFN-I autoAbs) have been identified in patients with severe infections. Their strong association with viral diseases suggests that these autoAbs might exacerbate virus replication by binding secreted IFN-Is and blocking receptor engagement, thereby inhibiting their antiviral activity. However, experimental systems to study the effects of specific anti-IFN-I autoAbs, and their impacts on endogenous IFN deficiencies, remain limited. Here, as part of developing a cell model for studying anti-IFN-I autoAbs in the context of endogenous IFNs, we show that endogenous IFNβ is the main driver of antiviral effects against multiple respiratory viruses in human lung epithelial A549 cells. Neutralization of endogenous IFNβ, but not IFNα, IFNω, or IFNλ1/2/3, enhanced viral replication, and plasma samples containing IFNβ-neutralizing autoAbs similarly promoted virus replication. We next applied this model to develop two mechanistically distinct proof-of-concept therapeutic strategies to counteract the pathogenic effects of anti-IFNβ autoAbs. First, we engineered signaling-inert IFNβ decoy molecules designed to retain autoAb-binding epitopes while abolishing receptor-mediated signaling. Structure-guided mutagenesis identified several IFNβ mutants lacking detectable signaling activity but maintaining strong binding to anti-IFNβ autoAbs. A combination of mutants yielded a decoy molecule restoring endogenous IFNβ-mediated antiviral activity in infected A549 cells by sequestering pathogenic autoAbs and restoring control of viral replication. Second, we evaluated the therapeutic potential of the small-molecule IFN-agonist RO8191, which activates IFNAR2-dependent JAK-STAT signaling independently of IFNs. RO8191-induced antiviral signaling resisted neutralization by anti-IFNβ antibodies and autoAb-positive plasmas and strongly inhibited respiratory virus replication even in the presence of neutralizing anti-IFNβ antibodies. Together, our findings establish a cell model to study anti-IFN-I autoAb effects against endogenous IFNβ and evaluate therapeutic strategies to restore antiviral immunity in autoAb-driven immunodeficiency.

**Student Paper*

M07 One Genome – One Interpretation? Lessons from the Swiss Journey Towards Harmonized NGS Diagnostics

Dr. Tim-Christoph Roloff Handschin

UZH, Zurich, Switzerland

Next generation sequencing (NGS) has become an integral component of modern clinical microbiology, enabling high-resolution pathogen identification, antimicrobial resistance prediction, outbreak investigation and genomic surveillance. However, the increasing implementation of NGS in diagnostic laboratories raises a fundamental challenge: to what extent can genomic data generated by different laboratories be interpreted in a consistent and clinically meaningful manner?

Achieving harmonized NGS-based diagnostics requires standardization across the entire workflow, including laboratory procedures, sequencing technologies, bioinformatics analysis, quality assessment, interpretation, reporting and data exchange. This presentation will discuss the Swiss experience in establishing the foundations for comparable and interoperable genomic diagnostics, highlighting both current achievements and remaining challenges.

The Swiss approach combines technological innovation with the development of shared standards, analytical frameworks and quality assessment strategies. Examples include the implementation of standardized bioinformatics workflows for bacterial whole genome sequencing, national and international proficiency testing initiatives such as INSSIGHT, and efforts to establish interoperable pathogen genomic data infrastructures through initiatives such as the Swiss Pathogen Surveillance Platform (SPSP). These developments illustrate how harmonization can be achieved across institutional and national boundaries while maintaining flexibility in laboratory technologies and analytical approaches.

A particular challenge remains the translation of genomic data into standardized yet context-specific interpretations and reports. Different stakeholders, including clinical microbiologists, clinicians, infection prevention teams and public health authorities, require different levels of information while relying on the same underlying genomic evidence.

Finally, the presentation will discuss the role of international interoperability and the contribution of initiatives such as the PathogenDataNetwork in extending harmonization beyond national borders. The overarching goal is not uniformity of technology, but the generation of reliable, comparable and actionable genomic information—moving towards a future where one genome can support one trusted interpretation across laboratories, healthcare systems and surveillance networks.

M08 Alternative careers in Biotech: Lessons from Entrepreneurship

Giorgia Greter

ETHZ, Zurich, Switzerland

What comes after a PhD in biology? For many early-career scientists, leaving academia can feel uncertain, and understanding how scientific training translates into industry roles is not always straightforward. After completing her PhD at ETH Zurich, Giorgia explored an alternative path through biotech entrepreneurship, cofounding Baxiva AG with support from ETH and the local startup ecosystem. Following her experience as a startup founder, she transitioned into business development at a deeptech startup. In this talk, she will reflect on the opportunities and challenges of moving from academia into entrepreneurship, the transferable skills gained along the way, and the broad range of career possibilities in biotech beyond the bench.

M09 From bench to blackboard

Thomas Fiedler

Gymnasium Muttenz, Muttenz, Switzerland

Leaving academia is often framed as a loss—of talent, of expertise, even of ambition. But is stepping out of the lab really stepping away from science? In this talk, I will argue the opposite: that moving from microbiological research into high-school education can amplify, rather than diminish, scientific impact.

My path was not always straightforward to begin with: an early fascination with nature and microorganisms, years spent exploring languages instead of science, and practical teaching experience gained while supporting my studies all contributed to shaping my professional identity. Alongside my academic training studying infectious diseases abroad and subsequent PhD work in basic science (still involving microorganisms), these experiences led me to reconsider what it means to contribute to science.

Why would someone trained in cutting-edge research trade the bench for the classroom? What happens to scientific expertise when it leaves the laboratory—does it lose its value, or does it reach audiences it never could before? How can a background in microbiological research shape what and how we teach at the high-school level? And, perhaps most importantly, who will inspire the next generation of microbiologists if not those who have experienced research firsthand?

In my talk I will briefly present my own educational journey and outline the transition from research to teaching, including motivation, pedagogical training, and the realities of entering the profession. Further I will touch upon the rewards and challenges of working in education. Finally, I will address the guiding questions outlined above and explore how research training translates into practice: supervising student projects and lab experiments, fostering scientific thinking, and creating platforms for science communication, to which you too are warmly invited to make a contribution!

M10 Leading with Science: Forging a Career as a Molecular Microbiologist in Big Pharma

Dr. Rusudan Okujava

Senior Principal Scientist and Research Team Leader, Infectious Diseases Therapeutic Area (ID TA), pharma Research and Early Development (pRED), Roche Innovation Centre Basel, Basel, Switzerland

Abstract: Dr. Rusudan Okujava is a Senior Principal Scientist and Antibiotics Discovery Research Team Leader at Roche pRED, Basel. Her scientific journey began with an MSc in Immunology from Tbilisi State University, Georgia, followed by a PhD and postdoctoral research in Molecular Microbiology at the Biozentrum, University of Basel. In 2015, she transitioned from academia to the pharmaceutical industry, joining Roche as a Scientist in the Antibiotics discovery group. Since then, she has evolved from a bench scientist into a leader of multidisciplinary teams, driving internal drug discovery projects, managing external academic collaborations, and serving on the Roche Antibiotics Discovery Leadership Team. In addition to her project leadership, she is a dedicated People Leader guiding the performance and career growth of research associates.

In this session tailored for early-career researchers, Dr. Okujava will share her personal trajectory from a local PhD student to a leader in Big Pharma. Her talk will highlight the vital roles of dedication, adaptability, and a collaborative, team-oriented mindset in navigating a successful industry career. Finally, as a mother of two, she will offer an encouraging, real-world perspective on being a "multi-faceted scientist" - demonstrating how to successfully balance a high-impact leadership role in drug discovery with a fulfilling personal life.

M11 The Good, the Bad, and the Ugly of an Academic Career

Assist. Prof. Andrea Vettiger, PhD

Department of Fundamental Microbiology, University of Lausanne, Lausanne, Switzerland

In this career lecture, I will share the highs and lows of life as an assistant professor in academia. I will discuss my career path to an independent faculty position, provide an overview of what the job actually entails, and reflect on lessons I wish I had learned earlier. Finally, I will share my experience of pursuing an academic career as part of a dual-career couple with children, and discuss some of the challenges and opportunities of balancing family life with academia. The talk aims to provide an honest perspective on academic research and practical insights for those considering this career path.

S11 Diversity and distribution of tree-associated fungi in the Caucasus region and their potential risk to European forests

Dr. Luis Ever Vega Cabrera[1], Dr. Carolina Cornejo[1], Ms. Khadija Apbayeva[2], Dr. Dilzara Aghayeva[2], Dr. Simone Prospero[1]

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2. Institute of Botany Public Legal Entity, Ministry of Science and Education of the Republic of Azerbaijan

Forest ecosystems consist of a variety of interacting flora, fauna, microorganisms, and abiotic components that contribute to a healthy environment. Fungal endophytes are fungi capable of living within plant tissues without causing apparent harm. However, they can become pathogenic and threaten forest ecosystems if environmental conditions change, or when they are introduced to novel ecosystems. Global trade is a key driver of biological invasions, and Azerbaijan's transformation into a logistics hub for trade between Asia and Europe increases the risk of introducing invasive species to the country and spreading them between continents. This study presents the results of a monitoring project designed to characterize the community composition of tree-associated endophytic fungi in forest ecosystems in the Caucasus region. Leaves were systematically sampled from trees belonging to four genera (*Quercus*, *Carpinus*, *Fagus*, and *Castanea*) in 12 locations during spring and autumn. The endophyte communities were then assessed using high throughput metabarcoding coupled with long-read sequencing. These surveys have revealed a remarkable fungal biodiversity, as well as the presence of native and non-native fungal pathogens in the Caucasus region. This could threaten local forests and pose a phytosanitary risk to Europe. In this presentation, we will examine the species diversity and geographic distribution of fungal species in forest ecosystems along the southeastern periphery of Europe in the Caucasus region.

S12∗ disentangling between soil properties and fungal plant pathogens to understand tree dieback in urban areas

Ms. Linda Philipona[1]

1. Laboratory of Microbiology, University of Neuchâtel, Neuchâtel, Switzerland

In a context of climate change, trees and soil are key components in cities. They provide ecosystem services, that benefit human well-being, refreshing the surrounding air and thus decreasing heat islands, especially in hot seasons. In urban environments, soils play a crucial role in sustaining tree growth and health. Yet they are often affected by disturbances such as sealing, compaction, and contamination, threatening tree health by restricting root growth, depriving roots of oxygen, and blocking water infiltration. That is why soil is the major problem explaining urban tree health.

Our study aims to investigate the relationships between soil properties, tree health and the potential presence of fungal plant pathogens in the city of Neuchâtel. For this purpose, we selected symptomatic and non-symptomatic cedar and poplar trees, as well as lawns as control sites. At each site, we sampled soil and characterized their physico-chemical (humidity, organic matter content, pH, CHN, cationic exchange capacity, bioavailable phosphorus, presence of heavy metals, granulometry and stability of the aggregates) and biological (earthworms populations and associated bacterial activity via an ecoplate assay) properties. In parallel, we sampled tree tissues and the presence of fungal pathogens was assessed using long read metabarcoding.

Data obtained from the different sites were compared to identify links between soil conditions and the occurrence of fungal pathogens and we compared this to the presence of described symptoms in the associated trees. This approach allows a better understanding of how soil quality may influence tree health in urban environments. In particular, with a focus on fungal tree pathogens that may benefit from adverse environmental conditions to deploy symptoms in their host. Our results will improve understanding of urban tree-soil-fungal interactions and provide insights into the sustainable management of urban green spaces in Neuchâtel.

∗Student Paper

S13∗ Morels co-cultivation with apple trees : a potential for mycosylviculture ?

Ms. Alix Jornot[1], Prof. Saskia Bindschedler[1], Mrs. Melissa Cravero[1], Mr. Robin Jacquat[1]

1. LAMUN, UNINE, Neuchâtel, Switzerland

Prized for their flavour, morels (*Morchella* spp.) have been the focus of cultivation efforts for several decades, particularly in China and the United States. However, commercial production remains challenging due to an incomplete understanding of their life cycle and ecological requirements. Although the trophic status of morels remains debated, current knowledge supports a predominantly saprotrophic lifestyle, with possible endophytic behaviour among certain species or in specific ecological contexts. Several studies have demonstrated the ability of morels to modulate soil characteristics by influencing both the composition of the microbial community and soil nutrients dynamics. It is also well known from morel hunters that certain species thrive with specific tree species. In this context, it is of interest to investigate whether the presence of a plant promotes an interaction with morels and how this affects specific soil characteristics.

In this study, carried out during the 2025–2026 cultivation season, we used an outdoor pot-based co-culture system with two-year-old apple trees co-inoculated with either of the following four *Morchella* strains (three Swiss wild strains and one Chinese cultivar). We then followed the effects of morel co-inoculation on soil physicochemical properties (pH, residual moisture, bioavailable nitrogen, cation exchange capacity, and carbon content) and on soil microbial dynamics (Ecoplate™ physiological profiling and 16S and ITS metabarcoding).

All strains developed conidiophores, asexual reproductive structures thought to play a key role in morel cultivation, although no sexual fruiting bodies emerged during the experiment. Nevertheless, dynamics regarding soil parameters were highlighted, which provided preliminary insights into the interactions among morels, tree roots, and soil microbial communities under cultivation conditions. These results provide a basis for exploring environmental factors associated with fruit tree–morel co-culture systems and contribute to the development of novel mycoforestry approaches.

∗Student Paper

S14 From apple-skin waste to fungal rodlets: discovering class I hydrophobins in Ganoderma adspersum

Dr. Carolina Reyes[1], Mr. Enrico Boschi[2], Dr. Peter Gehrig[3], Ms. Siiri Bienz[4], Dr. Naresh Kumar[4], Dr. Tiago Carvalho[5], Dr. Nico Kummer[6], Mr. Lukas Bürgi[2], Dr. Ashutosh Sinha[2], Prof. Francis W.M.R. Schwarze[5], Dr. Silvia Campioni[2], Dr. Gustav Nyström[2]

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Hydrophobins are amphipathic fungal proteins that self-assemble at interfaces into robust, amyloid-like films and rodlets, enabling surface colonization and offering opportunities for bio-based interfacial materials. Here, we investigate the white-rot basidiomycete *Ganoderma adspersum* as a sustainable source of class I hydrophobins and evaluate food-waste substrates as cultivation inputs. *G. adspersum* was grown in defined, shaken liquid cultures with varied carbon and nitrogen sources, including apple-skin waste as an alternative carbon feedstock. Foam formed at the air–liquid interface was harvested and subjected to trifluoroacetic-acid extraction to enrich for highly stable class I hydrophobins. Protein mixtures were profiled by SDS-PAGE and intact-mass MALDI-TOF, and hydrophobin candidates were identified by LC–MS/MS proteomics. Atomic force microscopy (AFM) revealed abundant rodlet-like nanostructures in foam-derived extracts, and confocal Raman spectroscopy showed β-sheet enrichment consistent with functional amyloid organization. Proteomics identified a previously unreported class I hydrophobin, termed Gad1, as a major component of the foam fraction; additional hydrophobin-like proteins, including a second candidate (Gad2), were also detected, and N-terminal processing was observed in agreement with known hydrophobin maturation pathways. Beyond molecular identification, hydrophobin-enriched extracts exhibited strong interfacial activity: they stabilized oil-in-water emulsions that remained intact for weeks and could be converted into porous, freeze-dried cellulose–protein composite aerogels. Together, these results expand the diversity of known hydrophobins in white-rot fungi and demonstrate that lignocellulosic food waste can support secretion of functional, self-assembling interfacial proteins. This work highlights *G. adspersum* as a promising platform at the interface of fungal ecology and biotechnology, enabling waste valorization while generating biomolecules suitable for sustainable emulsifiers and porous material fabrication.

S15∗ Speckle spectroscopy as a non-invasive tool for mycelium network characterization

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Mycelium is a disordered network composed of micrometric transparent hyphae whose morphology and spatial distribution depend on species-specific growth dynamics and environmental conditions. Such networks constitute complex scattering media, where features across multiple length scales govern light transport and polarization behavior. This work investigates the use of optical scattering techniques to probe species-dependent structural variations in fungal networks. To this end, samples were fabricated using a process based on solid-state surface fermentation, employing three basidiomycete species with distinct hyphal architectures: *Pleurotus ostreatus*, *Trametes versicolor*, and *Ganoderma lucidum*. Under coherent illumination, these filamentous media exhibit multiple light scattering, generating speckle patterns sensitive to the underlying arrangement. Polarized speckle spectroscopy was used to characterize the scattered field through the polarization state distribution, polarization axis, and degree of depolarization. Reproducible species-dependent differences in polarization behavior were observed, consistent with variations in network topology and hyphal arrangement. These results suggest that polarized speckle spectroscopy can serve as a non-invasive tool for differentiating fungal species. Furthermore, this approach could enable monitoring of mycelial network evolution and provide a framework for studying light transport in bio-based scattering media with tunable optical properties.

∗Student Paper

S16 Frequency-Dependent Growth Responses of Pleurotus spp. to Audible Sound Exposure

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1. Bern University of Applied Sciences, Bern, Switzerland

The influence of physical stimuli on fungal growth has gained increasing attention in recent years, particularly regarding non-chemical approaches to modulate microbial metabolism and biomass production. Among these stimuli, audible sound exposure represents a promising but still insufficiently understood factor affecting fungal physiology. This study investigated the frequency-dependent effects of audible sound on the growth of different *Pleurotus* species cultivated under controlled laboratory conditions.

Mycelial cultures of selected *Pleurotus* spp. were exposed to continuous sound treatments at defined frequencies within the audible range, while control cultures were maintained under identical environmental conditions without sound exposure. Growth responses were evaluated by measuring radial mycelial expansion, biomass formation, and morphological characteristics during incubation.

The results demonstrated that sound exposure influenced fungal growth in a frequency-dependent manner. Certain frequencies promoted significantly enhanced mycelial growth and denser colony morphology, whereas other frequencies resulted in reduced growth rates or no measurable effect compared with controls. The magnitude and direction of the response varied between *Pleurotus* species, suggesting species-specific sensitivity to acoustic stimulation.

These findings indicate that audible sound may represent a novel environmental parameter capable of modulating fungal development. The observed frequency-specific responses provide new insights into fungal mechanosensitivity and may contribute to the development of innovative cultivation strategies for edible mushrooms and fungal biotechnology applications. Further investigations are required to elucidate the underlying physiological and molecular mechanisms involved in fungal responses to acoustic exposure.

M12 Engineering the plant microbiome: large scale application of microbial biostimulants for sustainable agriculture

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Beneficial soil microorganisms have a large potential to reduce dependency on synthetic fertilisers and pesticides and enhance agricultural sustainability. However, despite decades of promising results under controlled conditions, the translation of microbial biostimulants into reliable field-scale solutions remains a major challenge.

Among these microorganisms, arbuscular mycorrhizal fungi (AMF) are of particular interest because they enhance plant nutrient uptake and reduce abiotic and biotic stress. AMF inoculation therefore represents a promising strategy to support plant health and productivity. Yet, most commercial products fail to establish functional symbioses under field conditions, leaving farmers without reliable large-scale solutions.

To address this limitation, we formulated AMF into granules compatible with standard agricultural machinery, enabling large-scale application. We tested this approach in 39 maize and sunflower fields across Switzerland over two growing seasons. The inoculated AMF strain was successfully detected in the roots of treated plants using molecular tools, confirming its establishment under field conditions. We observed that AMF inoculation enhanced plant yield in the field (up to 20% more yield). Based on these results we discuss the economic costs and benefits associated with the application of mycorrhizal granules in the field.

Together, these results highlight both the challenges and opportunities of engineering the plant microbiome as a viable component of sustainable agriculture.

S17 Soil microbial functions and carbon utilization across a large environmental gradient

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Soils harbor immense microbial diversity and are central to global carbon and nutrient cycles. Predicting how ecosystem carbon (C) balances will respond to ongoing and future environmental change is of major socio-economic importance. However, the incorporation of microbially derived compounds into stable soil C pools, and the feedbacks of microbial processes on climate, remain poorly understood. In particular, the drivers of variation in soil microbial diversity and functioning across continental-scale environmental gradients are still unclear. First, we investigated the diversity and network complexity of soil prokaryotic and fungal communities, and their links to soil multifunctionality (SMF)—an integrative index of C-, N-, and P-cycling—along a 3,000 km European latitudinal gradient (37°–62°N). Our results highlight the key role of fungal diversity in sustaining SMF and show that microbial diversity is shaped by both large-scale climatic and biogeographical factors, as well as local soil properties and land cover. These findings provide important insights for maintaining ecosystem functioning. In addition, a common-garden experiment examined short-term climate-change effects on microbial respiration, biomass, and carbon use efficiency (CUE) across biomes. Surface soils from Arctic tundra (Greenland), boreal (Sweden), temperate (Switzerland), Mediterranean (Spain) forests, and desert (southern Spain) ecosystems were incubated under elevated temperature and moisture treatments simulating summer drought followed by rewetting. Warming had little effect on microbial biomass (C and N) and CUE overall, but reduced CUE during drought in all biomes except the Arctic. Rewetting generally increased CUE, though recovery rates varied among biomes. Overall, our results demonstrate that soils from different biomes respond distinctly to drought and rewetting, with important implications for carbon cycling under future climate scenarios.

S18 Laboratory evolution enhances resilience of a symbiont yeast and its honeybee host against agrochemical exposure

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Yeasts are key members of several ecosystems. Yet, the impact of widespread chemical pollutants on yeasts is understudied. Here we report the effect of >1000 chemical pollutants on 14 diverse yeast species spanning the Saccharomycotina subphylum. *Starmerella bombicola*, a symbiont of various bee species, was the most sensitive and was inhibited by both fungicides and non-fungicides. To identify the molecular basis of this ultra-sensitivity, we selected resistant lineages against 9 chemicals using adaptive laboratory evolution. Whole-genome-sequencing uncovered convergent evolution on *YBP1*, a regulator of oxidative stress. Proteomic analysis confirmed the protective role of oxidative stress response pathways, including proteins encoded by horizontally transferred bacterial genes. We find that the evolved *S. bombicola* ameliorates the negative effect of paclobutrazol, a plant hormone regulator, on the bee gut microbiome, sucrose responsiveness and learning. Our findings demonstrate how laboratory evolution can aid mitigating the negative impact of pollutants on pollinators.

S19 Spatial organization of biomass controls intrinsic permeability of porous systems

Prof. Pietro de Anna[1], Dr. Wenqiao Jiao[2], Dr. David Scheidweiler[3], Prof. Alberto Guadagnini[4]

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Biofilms alter the hydraulic properties of porous media, impacting processes from groundwater remediation to industrial filtration. While biomass accumulation is known to reduce permeability, a quantitative link between its spatial organization and system- scale hydraulics remains missing. Here, using microfluidics, time-lapse microscopy, and a novel mechanistic model we demonstrate that biofilm spatial organization is the key control for the resultant permeability decline. With independent experiments, we show that motile Pseudomonas putida sp. and its non-motile mutant grow biofilm attaining identical total biomass, yet cause permeability reductions of 78% and 94%, respectively. This divergence arises because motile cells, escaping nutrient-depleted zones, colonizes the porous system differently in space, confining significant biomass upstream, whereas non-motile cells persistently colonize homogeneously the entire system. Our model, conceptualizing the medium as a series of pores with different size and biomass-modified permeability, accurately predicts these dynamics. We conclude that biomass spatial distribution, not simply its abundance, is the primary control on permeability, offering a new framework to predict and manage clogging in environmental and engineered systems.

S20 Historical nutrient regimes select distinct methane-cycling communities in lake sediments

Dr. Alice Bosco Santos[1], Ms. Eulalie Rose Beyala Bekono[2], Dr. Santona Khatun[3], Dr. Marie-Ève Monchamp[4], Dr. Joana Séneca[5], Dr. Petra Pjevac[5], Prof. Jasmine Berg[2]

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4. Biology Department, McGill University, Montreal, Canada
5. Division of Microbial Ecology, Centre for Microbiology and Environmental Systems Science, University of Vienna, Vienna, Austria

Methane cycling in lake sediments is governed by complex interactions between microbial communities, organic matter composition, and redox conditions. However, the extent to which historical environmental change shapes present-day methane-cycling communities remains poorly understood. Here, we combined sediment geochemistry, stable carbon and sulfur isotopes ($\delta^{13}C_{org}$, $\delta^{13}C_{DIC}$, $\delta^{13}C_{CH4}$, and $\delta^{34}S_{CRS}$), and 16S rRNA gene sequencing to investigate four centuries of methane cycling in a dated sediment core from Lake Joux, Switzerland.

Distinct depositional intervals associated with contrasting nutrient regimes and organic matter sources hosted markedly different methane-cycling microbial communities. Carbon isotope signatures and microbial community composition revealed a transition from methylotrophic methanogenesis in older eutrophic sediments, dominated by Methanomassiliicoccales, to hydrogenotrophic methanogenesis in younger deposits. Elevated sulfide concentrations and enriched $\delta^{34}S$ values in sedimentary sulfides suggest an active coupling between sulfur cycling and methylotrophic methanogenesis in the deeper eutrophic interval. In addition to these long-term shifts in methanogenic pathways, we observed an unexpected abundance of aerobic methane-oxidizing bacteria (MOB) in oxygen-free sediments. The distribution of MOB coincided with isotopic evidence for methane oxidation, including a sharp decrease in methane concentrations and a 13C enrichment of residual methane.

Our results demonstrate that historical nutrient enrichment leaves a long-lasting ecological legacy in freshwater sediments by shaping both the composition and metabolic potential of methane-cycling communities. By integrating microbial ecology with stable isotope geochemistry, we show that past environmental conditions continue to influence methane production centuries after deposition and reveal a potentially important role for aerobic methanotrophs in anoxic sedimentary environments.

M13 Genomic and functional diversification of the bat antiviral innate immunity

Lucie Etienne

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Bats are natural hosts or reservoirs for a diverse range of viruses, including coronaviruses, lyssaviruses, and retroviruses. While many of these viruses are asymptomatic in bats, they can cause severe disease in humans and other mammals. These characteristics may notably result from long-term virus-host coevolution, balancing strong antiviral responses with immune tolerance. Innate immunity, one of the first and ancestral line of defense against pathogens, is a key area of this molecular conflict.

Here, we characterize the functional evolution of bat innate immune responses and investigate how past viral epidemics have shaped bat immune diversification. Through an international partnership (CNRS IRP RAPIDvBAT), we generated novel genomic sequences and primary cell lines from multiple bat individuals and species, and identified key genome-wide adaptations. Through viral infections, functional genetics, and transcriptomics of bat cells, we further uncovered evolutionary and mechanistic insights into candidate genes underpinning bat antiviral immunity.

Our findings reveal functional adaptations across scales, which have shaped bat–virus specificities. These results shed light on how past epidemics have impacted host genomes, possibly influencing modern pathogenesis and molecular determinants of cross-species transmission.

S21* Development of two classes of non-toxic broad-spectrum antivirals

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Viral infections cause millions of deaths annually and continue to pose a significant global pandemic threat. While vaccines are essential for prevention, they provide limited benefit once infection is established. Therefore, there is a critical need for complementary therapeutic strategies, including the development of effective antiviral agents. Many viruses initiate infection by binding to heparan sulfate proteoglycans present on the surface of host cells, which act as attachment factors that facilitate viral entry. Blocking this interaction can inhibit infection at its earliest stage and limit viral spread. In this project, we evaluated two classes of heparan sulfate (HS) mimicking compounds in vitro for cytotoxicity, antiviral activity against multiple viruses, and potential virucidal effects. We also investigated the antiviral activity of these compounds in physiologically relevant tissue models, such as air-liquid interface cultures. The first class consisted of β-cyclodextrins functionalized with sulfated di- and heptasaccharides, while the second was composed of anionic dendrimers bearing carboxylate or sulfonate surface groups. Both compound families demonstrated potent antiviral activity against a broad range of viruses. Low nanomolar activity was observed against strictly HS-dependent viruses, including respiratory syncytial virus and herpes simplex virus type 1, while nanomolar potency was also maintained against viruses that are not strictly dependent on HS, such as human coronavirus OC43, influenza A virus and hantaan virus. In addition, both classes of compounds were effective at limiting and reducing viral spread ex vivo. Moreover, both classes of compounds exhibited virucidal activity, indicating irreversible viral inactivation. Together, these findings highlight the therapeutic potential of HS mimetics as entry inhibitors and virucidal agents, supporting their further development as broad-spectrum antiviral strategies.

**Student Paper*

S22 Rethinking Feline Coronavirus Research: Alternative in vitro systems to study a complex virus

Dr. Kimberly Schmied[1], Dr. Bettina Trüeb[1], Dr. Nadine Ebert[1], Mrs. Nadja Peduto[1], Mr. Hanspeter Stalder[1], Dr. Lisa Thomann[1], Prof. Volker Thiel[1]

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Feline infectious peritonitis is a fatal systemic disease of cats caused by a feline coronavirus (FCoV), specifically feline infectious peritonitis virus (FIPV). Although major progress has recently been made in the treatment of FIP, important aspects of FCoV biology, viral evolution, tissue tropism, and pathogenesis remain poorly understood. Progress in the field is further limited by the lack of physiologically relevant *in vitro* systems and molecular tools that accurately model infection dynamics in the natural host. The emergence of a highly pathogenic FCoV outbreak in Cyprus in 2023 (FCoV-23) further emphasized the need for improved experimental platforms to investigate viral determinants associated with transmission, virulence, and host interactions.

To address these challenges, we are establishing complementary *in vitro* systems for the study of FCoV infection and virus-host interactions. We successfully generated feline intestinal organoids from primary intestinal stem cells, providing a species-specific and physiologically relevant model of the intestinal epithelium, the natural target tissue of the transmissible feline enteric coronavirus (FECV). In addition to maintaining three-dimensional organoid cultures, we established two-dimensional organoid-derived epithelial monolayers to facilitate infection studies. Current work focuses on phenotypic characterization of these cultures and evaluation of their susceptibility to FCoV infection. These organoid-based systems provide an important alternative to *in vivo* studies and enable investigation of early infection dynamics, epithelial responses, and virus-host interactions in a controlled and biologically relevant environment.

In parallel, we are developing reverse genetics clones based on FCoV-23 strains associated with the Cyprus outbreak. As reverse genetics systems for FCoV remain limited, this platform will enable targeted investigation of viral mutations potentially associated with enhanced transmission and potentially altered host interactions.

Together, these approaches establish a versatile experimental framework to study FCoV biology, viral evolution, and mechanisms underlying FCoV-associated disease.

MT10*/S23*/P020* targeting Rhinovirus IRES as new approach for antivirals

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Picornaviridae is a family of viruses that includes different known human pathogens such as Polioviruses, Enteroviruses and Rhinoviruses. Their symptoms range from mild common cold to paralysis and even death. The IRES sequence is a conserved RNA element present in this family that plays a crucial role in the viral life cycle. Targeting the IRES is feasible, as demonstrated by compounds showing activity against EV71 in vitro and in vivo. However, available compounds mostly target stem-loop II, whose relatively simple structure may lead to off-target effects. Additionally, the IRES structure has largely been predicted but has not yet been assessed in infected cells. Therefore, this project begins by refining IRES structural predictions through probing viral RNA in infected cells and characterizing its secondary structure using DMS-MaP. The resulting structure enabled the design of a library of LNA-ASOs (locked nucleic acid antisense oligonucleotides), that either unfold or induce degradation of the target RNA. Among the tested ASOs, four exhibit nanomolar activity against RV8 without detectable toxicity in vitro and in human derived respiratory models. Further optimization of these LNA-ASOs, including viral resistance, synergy evaluation and testing across different enteroviruses (including EV68 and EV71), is currently ongoing.

**Student Paper*

M14 Beyond microbiology: Pragmatic tools for optimizing antibiotic use in lung infections

Prof. Noémie Boillat Blanco

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Respiratory tract infections are among the most common indications for antibiotic prescribing, with diagnostic uncertainty being a key driver of unnecessary use.

This keynote will explore how pragmatic tools can help clinicians navigate uncertainty and optimize antibiotic decisions. From bedside point-of-care diagnostics to artificial intelligence–guided approaches, we will discuss how integrating clinical expertise, diagnostics, and digital innovation can support more targeted care and contribute to tackling antimicrobial resistance.

S24 CRISPRi-seq reveals human body-site-specific pressures on pneumococci

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Background

Pneumococci can disseminate from the bloodstream to various sites in the body. Little is known about how they sustain these different environments.

Methods

We created a CRISPR interference sequencing (CRISPRi-seq) library in a serotype 22F pneumococcal blood isolate, with an anhydrotetracycline-inducible dCas9 system. We cultured the library in sterile cell-free specimens of human cerebrospinal fluid (CSF), pleural fluid (PLF), and synovial fluid (SYF) diluted with low-nutrient medium to conduct genome-wide screens and identify pneumococcal genes that are important for survival in these human environments.

Results

The CRISPRi-seq screens demonstrated differential pneumococcal gene fitness signatures between the human dissemination sites. Those consistent across human donor samples were further validated. The data suggest that in CSF, pneumococcal fitness depended on teichoic acid biosynthesis and lipid metabolism. These findings were confirmed using directed mutants, and by the differential activity of antibiotics specifically targeting these pathways. Phospholipase A_2, present in human CSF, may be responsible. Pneumococci that carry *slaA* can avert phospholipase-mediated damage which was previously identified as meningitis-mediating in population studies. In PLF we observed clumping of pneumococcal cells and CRISPRi-seq footprints of activation of the early competence genes and streptococcin B, possibly mediated by fibrin-formation. While various pneumococcal chain lengths were visible in the other study media, SYF only supported diplococcal growth. Consistent with this phenotype, pneumococcal fitness in SYF relied specifically on expression of the divisome, and relatively less on the elongasome. Clavulanic acid, typically not bactericidal, showed antimicrobial activity in SYF.

Conclusions

Pneumococcal CRISPRi-seq identified body-site specific antimicrobial pressures. This humoral host profile can guide clinical tending towards preferred antibiotics, to fibrinolysis, or repurposing of medicines in disseminated pneumococcal sepsis.

S25 Hidden genome dynamics: structural variation in chronic Burkholderia cepacia complex infections

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Bacteria adapt to diverse niches through single nucleotide variants and structural variations (SVs). SVs remain understudied because reads from standard sequencing techniques are often shorter than the variants themselves, limiting their detection. SVs include insertions, deletions, inversions and translocation and are often mediated through mobile genetic elements. They can affect clinically relevant traits such as antibiotic resistance and virulence. Species of the Burkholderia cepacia complex (Bcc) infect patients with chronic lung diseases, and are an excellent system to study SVs. Bcc can persist for decades within the host, enabling diversification of closely related strains. In addition, they harbour numerous mobile insertion sequence (IS) elements that facilitate intragenomic recombination, and possess three chromosomes whose plasticity remains poorly understood. Here, we aim to characterise SVs within Bcc.

Using publicly available short-read sequenced Bcc genomes (n=1,028), we find a highly variable (i) number of IS-elements per genome (median=7; range=0-34) and (ii) genome sizes (median=7.01MB; range=5.74-9.59MB) suggesting high genome plasticity. To investigate SV in Bcc within the human host, we analysed 38 clinical isolates from 11 patients respiratory samples using long-read whole-genome sequencing (2–4 isolates per patient, sampled 1-1,393 days apart). Using the earliest isolate from each patient as a reference, we identify a median of 2 SV per sample (range: 0–16). Detected SVs include insertions, deletions, and a ~250KB translocation between chromosomes 1 and 2 in isolates from one patient, suggesting dynamic genome rearrangements within the human host.

Future work will link SVs to genomic regions and phenotypic effects to clarify their role in within-host adaptation.

S26* Tailored phage selection for the treatment of chronic airway infections due to MDR Pseudomonas aeruginosa

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Chronic Pseudomonas aeruginosa infections are a leading cause of morbidity and mortality in cystic fibrosis (CF) patients. Treatment is complicated by antibiotic resistance, biofilm-associated persister cells, and rapid adaptation of hypermutator strains in the CF lung. Phage therapy is a promising alternative, yet efficacy depends on selecting phages under conditions that reflect chronic infection phenotypes — which typically involve loss of type IV pili, flagella, and altered lipopolysaccharide (LPS) structure, all of which serve as canonical phage receptors.

We isolated and characterized a phage collection spanning seven genera, selected against clinical isolates and strains with chronic infection-associated phenotypes, including mutants lacking type IV pili (ΔpilA), flagella (ΔfliC), or LPS O-antigen (ΔgalU). Among these, four Pbunavirus phages could infect a triple-deletion strain (ΔfliCΔgalUΔpilA). Resistance mutant selection combined with comparative genomics identified dnpA as a novel gene required for phage receptor expression.

To assess clinical relevance, we tested our phage collection in a polarized Calu-3 airway epithelial cell infection model. Selected phages successfully delayed or prevented disruption of epithelial integrity, demonstrating efficacy in a setting that more closely mimics the infection environment.

We also investigated phage evolution in the context of hypermutator strains. Using a PAO1ΔmutS strain — representative of clinical isolates with mismatch repair defects — phage resistance emerged faster than in wild-type PAO1. Serial co-culture experiments showed that phages evolved in both backgrounds gained enhanced bacterial suppression. Phages evolved in the hypermutator strain accumulated more mutations with greater diversity, establishing this as a powerful platform for directed phage evolution.

Together, this work presents a phage collection tailored to chronic infection receptor profiles, validated in a clinically relevant cell model, and paired with an evolution strategy suited to the dynamic CF lung environment.

**Student Paper*

S27 Performance evaluation of AI-assisted auramine microscopy in routine mycobacterial diagnostics

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Aims: Automation and AI-assisted image analysis are increasingly implemented by microbiology laboratories worldwide to improve efficiency and diagnostic quality and to address the shortage of qualified personnel. Microscopy remains a valuable diagnostic tool in clinical microbiology but is limited by labour-intensive and time-consuming workflows, dependence on highly experienced personnel, and subjective interpretation. In this study, we evaluated the performance of automated AI-assisted auramine fluorescence microscopy for the detection of acid-fast bacilli, including *Mycobacterium tuberculosis* complex and non-tuberculous mycobacteria, in clinical specimens.

Methods: The Metafer System (MetaSystems, Altlussheim, Germany), an automated digital fluorescence microscope that integrates deep neural networks to assist the screening of clinical samples for acid-fast bacilli was used. A customized neural network was trained with 16430 positive and 8628 negative objects before a total of 185 auramine-stained smears, including 132 negative and 53 positive smears, of respiratory (75.1%) and non-respiratory (24.0%) specimens were analysed. Manual screening as the gold standard was compared to AI-assisted interpretation with and without revision by experienced technicians and to culture and PCR results.

Results: AI-assisted interpretation of auramine stains showed high sensitivity (86.8% to 96.2%) and good specificity (70.3% to 93.5%) A cutoff-dependent inverse relationship between sensitivity and specificity was observed, with increasing sensitivity associated with decreasing specificity. Re-evaluation of the digital images classified as positive for acid-fast bacilli by trained technicians increased specificity to 99.0% without affecting sensitivity. Overall concordance with culture and PCR results was high.

Conclusion: AI-assisted auramine microscopy with the Metafer system showed reliable detection of acid-fast bacilli in clinical specimens. Good overall cutoff-dependent sensitivity and specificity were achieved and could be further improved by re-evaluation of digital images by trained professionals, demonstrating the system's suitability for integration into routine mycobacterial diagnostic workflows.

M15 Discovery and modulation of metabolic interactions among plant-associated bacteria

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Metabolic interactions form the basis for ecological networks among host-associated and environmental microbes, shaping community assembly and ultimately influencing host states. Understanding these processes is essential for predicting microbiome structure and for the rational design of communities with desired functions. Here, we combined detailed metabolic characterizations of over 200 representative plant-associated bacterial isolates with computational metabolic models to study the links between metabolism, ecological interactions, and community dynamics. This framework allowed us to accurately predict interaction outcomes for selected microbiota members in planta and provided mechanistic insight into the factors governing community assembly, revealing widespread competition for carbon alongside opportunities for positive interactions mediated by resource exchange. By linking metabolic modeling to emergent population trajectories, we further show how positive interactions can result in a range of ecological states, from competitive exclusion to coexistence characterized by oscillatory population abundances. Together, these results highlight the central role of metabolism in driving microbiome assembly and establish a predictive framework for understanding and engineering microbial community structure and function.

S28 Glucosinolate transport shapes spatial microbiome assembly along the root axis in two Brassicaceae species

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Plant roots shape distinct microbial communities through the secretion of specialized metabolites into the rhizosphere. Still, the extent to which the spatial organization of these compounds influences belowground microbiome assembly remains unclear. In *Brassicaceae* species such as *Arabidopsis thaliana* and the emerging oilseed crop *Camelina sativa*, glucosinolates (GSLs) constitute a large class of amino acid-derived specialized metabolites well-characterised for their roles in plant defense. However, their distribution within roots and their ecological functions belowground are still poorly understood.

To address this, we combined metabolite profiling with bacterial community analysis across whole roots and defined root compartments in both species, including mutants deficient in the glucosinolate transporters GTR1 and GTR2. We found that both plants display a conserved, transporter-dependent accumulation of long-chain aliphatic glucosinolates at the root tip, indicating an active axial organization of chemical defenses along the root system. Using 16S rRNA amplicon sequencing, we further identified plant species identity as the dominant factor shaping bacterial community composition. Nevertheless, disruption of glucosinolate distribution changed compartment-specific patterns of microbiome assembly in a species-specific manner. In *Arabidopsis*, these effects were primarily observed in the rhizosphere, whereas in *Camelina* both rhizosphere and root-associated microbial communities were affected.

Moreover, our analyses uncovered previously undescribed developmental and local variation in root chemistry and microbial composition across root systems. Together, these findings shows that glucosinolate transport generates chemical gradients along the root axis that contribute to the spatial organization of the root microbiome. This highlights the relevance of spatially structured specialized metabolite allocation as a mechanism by which plants engineer their belowground microbial environment and offering new perspectives for microbiome-informed agricultural strategies.

S29 Microbial community complexity modulates emergent metabolism

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Metabolic activity in bacterial communities is shaped by inter-species interactions. However, the relationship between community complexity and the extracellular metabolome, as well as the predictability of metabolic traits of complex communities from lower-order interaction data, remains poorly characterised. Here, we assembled all 31 possible combinations of five bacterial species and used a combination of targeted and untargeted metabolomics, and a scalable high-throughput pH measurement method to systematically characterise community exometabolomes. We find that higher-order communities display emergent pH changes and metabolite features that are absent from constituent lower-order monocultures and communities, indicating that multispecies assembly can reorganise nutrient use and metabolite production in a manner not wholly captured by simpler communities. We then follow up a subset of communities displaying emergent metabolic features with metaproteomics measurements to uncover the proteome states of each species present in each community. Our results show that community complexity can modulate emergent features of extracellular metabolomes with implications for microbial community function relevant for microbial ecology, natural product discovery and the design of synthetic microbial communities.

S30 Yellow protein co-opted to sustain obligate symbiosis in leaf beetles

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Yellow proteins are best known for their roles in pigmentation, behavior, and development across insects. Here, we uncover their striking evolutionary co-option for a wholly distinct function: sustaining a Paleocene-aged digestive symbiosis in tortoise beetles. We show that a female-specific Yellow forms the gelatinous spheres that encapsulate the bacterium *Stammera* during vertical transmission, allowing it to subsist extracellularly despite its drastically reduced genome (0.24 Mb) and limited metabolic capacity. Yellow expression is highly localized to symbiont-harboring glands in the ovaries, where the protein is assembled into a matrix and secreted during egg-laying. Functional knockdown of *yellow* disrupts sphere integrity and compromises symbiont viability under dry conditions, underscoring the protein's embedding properties and protective role for *Stammera*. These findings reveal a novel function for an ancient gene family and demonstrate how tortoise beetles have repurposed Yellows to overcome the extreme metabolic constraints faced by their symbionts during extracellular transmission.

S31* Modulation of intercellular tunnelling nanotubes of Dictyostelium discoideum by Legionella pneumophila

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Tunnelling nanotubes (TNTs) are thin, actin-dependent protrusions connecting cells and enabling intercellular communication over short to intermediate distances. Unlike classical secretion-based communication, TNTs provide a continuous route for transfer of cellular material, including cargo up to the size of whole organelles. In mammalian systems, TNTs have been implicated in tissue repair, immune regulation, inflammation, tumor progression, neurodegeneration, and host-pathogen interactions. However, despite their emerging biological relevance, the cellular mechanisms controlling TNT formation, cargo selectivity, and directional transport remain elusive. This is partly due to the lack of genetically tractable model system.

This project aims to establish *Dictyostelium discoideum* as a model organism to study TNT-mediated intercellular cargo transfer. *D. discoideum* shares important functional properties with mammalian phagocytes and is widely used as a host model for intracellular pathogens such as *Legionella pneumophila*, the causative agent of Legionnaires' disease. Using genetically encoded fluorescent reporters for mitochondria, endosomes, and lipid droplets, we show that *D. discoideum* forms TNT-like intercellular connections capable of transferring cargo between cells. Moreover, TNT formation is modulated by cellular stress and bacterial infection. Treatment with latrunculin B promotes TNT formation and enhances intercellular transfer of fluorescently labelled cargo, supporting a role for cytoskeletal remodeling in TNT induction. In contrast, infection with wild-type *L. pneumophila* reduces TNT formation, whereas mutants lacking the microtubule-modulating effectors LegG1 and PpgA partially restore TNT formation. These findings suggest that bacterial effectors interfere with TNT-dependent intercellular communication.

Together, these results identify TNTs as intercellular transport structures and establish *D. discoideum* as a powerful model to dissect the molecular mechanisms underlying TNT formation, cargo selection, and infection-dependent modulation. By combining live-cell confocal fluorescence microscopy, genetically encoded cargo reporters, and *L. pneumophila* effector mutants, this project will provide new insight into how cytoskeletal remodeling controls TNT-mediated exchange of organelles and vesicular cargo during stress and infection.

**Student Paper*

M16 Fungi as a Platform for Sustainable Bio-Based Living Materials

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Abstract not received yet.

S32 Fungal-based biosorption: An eco-friendly strategy for heavy metal removal from contaminated water

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Fungal-based adsorbents are gaining widespread attention as an eco-friendly and cost-effective solution for removing heavy metals (HMs), particularly in dilute solutions where synthetic resins are ineffective. The goal of this study was to develop a fungal biosorbent for the removal of HMs from wastewater. For this, *Curvularia lunata*, AD6 strain was isolated from a municipal solid waste and evaluated for its capacity to remove various HMs in non-competitive and competitive systems where the removal efficiency order was Cr > Pb > Cd > Ni > Cu in 2 h. The maximum metal removal capacity were ranged from 3.83 to 6.24 mg/g under optimized conditions. The role of various functional groups and surface chemistry of the fungal biosorbent was determined by FTIR and SEM. Furthermore, the thermodynamic parameters proved that the biosorption process was endothermic, spontaneous and feasible. The adsorption equilibrium isotherms and kinetic modelling studies were significant proven a multi stage physisorption and chemisorption. The biosorbent showed a significant removal of Ni and Cu when applied to the electroplating industrial effluent and metallic leachate in a laboratory scale up study. These findings indicated that the fungal sorbent AD6 possesses potential efficiency for the removal of various HMs from contaminated water.

S33 Synthetic lichen for the conversion of lignocellulosic biomass into fuels and chemicals

Dr. Derek Troiano[1], Mr. Tobias Hofmann[1], Dr. Simone Brethauer[1], Prof. Michael Studer[1]

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Microalgal biomass represents a promising feedstock for the sustainable production of, among other things, biofuels and industrially relevant chemicals, but its use is hindered by high water and energy requirements for growth and harvesting as well as nutrient cost. Growing microalgae in a biofilm may represent one solution to the high water and energy consumption associated with more commonly employed suspended growth approaches. Concerning nutrient cost, which is high due to the requirement for supplemental organic carbon (i.e., for mixotrophic growth) to promote sufficiently rapid microalgal growth, process economics may be improved by replacing typical sources of supplemental organic carbon (e.g., acetate) with cheap and abundant lignocellulosic biomass. While microalgae do not possess the capacity for metabolizing the complex biopolymers available in lignocellulosic biomass (e.g., cellulose), filamentous fungi are excellent at degrading components of lignocellulose into simple molecules. Co-culturing multiple strains of microbes within a single bioprocess, i.e., as a microbial consortium, is emerging as a promising alternative to monocultures for complex biotransformations due to their inherent advantages which include the distribution of metabolic burden by division of labor, the ability to convert complex substrates more efficiently, and their modularity. Here, the model microalga *Chlamydomonas reinhardtii* was co-cultured with the cellulolytic filamentous fungus *Trichoderma reesei* to form a synthetic lichen biofilm utilizing cellulose-based growth media. Ultimately, the engineered symbiosis in cellulose-based media improved microalgal biomass productivity vis-à-vis axenic phototrophic and mixotrophic systems thus representing a promising alternative approach that addresses key bottlenecks in microalgal bioprocesses. The platform was moreover implemented in a robust, continuous production process thereby further enhancing its economic potential.

S34 Effect of Bacillus velezensis K-165 volatiles on Botrytis cinerea and Arabidopsis protection

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The tomato root isolate *Bacillus velezensis* K-165 was shown to promote eggplant growth, but also to protect both eggplant and potato against the soilborne pathogen *Verticillium dahliae*. Moreover, strain K-165 was shown to induce systemic resistance against *V. dahliae* in *Arabidopsis thaliana.*

Our study's aim was to determine the role of volatile organic compounds (VOCs) released by K-165 in the direct inhibition of the pathogen *Botrytis cinerea* as well as in the induction of plant defenses. We show that the volatiles released by K-165 were dependent on its cultivation medium.

The VOCs released by K-165 when grown on PDA (but not on LB) inhibited the mycelium growth of *B. cinerea*, while *A. thaliana* plants exposed to VOCs released by K-165 grown on LB (but not on PDA) were more resistant to *B. cinerea*, suggesting that these VOCs induced plant defenses. Camalexin involvement in the VOCs-mediated plant defense stimulation was confirmed by an upregulation of genes involved in camalexin synthesis, an increase of the leaf camalexin concentration as well as by the loss of the VOCs' protective effects on *pad3* mutant and *cyp79B2/B3* double mutant plants infected with *B. cinerea*.

The VOCs released by K-165 cultivated on PDA and LB were identified by GC-MS. Five ketones were identified only when K-165 grew on PDA, while 2,5-dimethylpyrazine was released only when the strain grew on LB. The ketones were tested for effects on *B. cinerea* mycelial growth and shown to inhibit it when applied at high concentrations. 2,5-dimethylpyrazine was tested as plant defense inducer. Interestingly, exposure of *A. thaliana* wild-type (but not *pad3* and *cyp79B2/B3* double mutants) to 2,5-dimethylpyrazine showed a tendency for higher resistance against *B. cinerea*, suggesting that this VOC might be involved in K-165's induction of plant defenses.

S35 Population control by the T3SS sustains a bacterial–fungal endosymbiosis

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Endosymbiotic interactions, in which one cell resides within another, are key drivers of biological complexity, enabling organisms to expand their metabolic capabilities. However, the establishment of stable cell-in-cell interactions remains poorly understood. Characterizing crucial endosymbiotic factors can pave the way towards engineering novel endosymbioses to transfer desirable traits. An emerging model system for understanding mutualistic endosymbiosis is the filamentous fungus *Rhizopus microsporus*, which depends on its bacterial endosymbiont *Mycetohabitans rhizoxinica* for sporulation. This system provides an opportunity to characterize the mechanistic foundation of endosymbiosis, as both partners are genetically tractable and can be cultured axenically. Here, we established a workflow to compete bacterial mutants against wildtype bacteria *in fungo*, comparing their phenotypes and fitness. Our findings show that the density of bacterial endosymbionts within *R. microsporus* is strictly regulated, with consistent population thresholds being reached in mature mycelium. Disruption of the type-three secretion system (T3SS) leads to increased bacterial proliferation and disrupted vertical transmission, which is rescued in the presence of wildtype bacteria. This suggests that a functional T3SS is essential for transmitting signals that regulate endosymbiont densities and vertical transmission within the fungal host, but not receiving them. Together, these findings highlight the importance of controlling endosymbiont population dynamics to maintain symbiotic homeostasis.

M17 From dividing to dormant: the microbial activity spectrum and its ecological consequences

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Microbial activity has traditionally been treated as a binary state (active or dormant), oversimplifying a biological continuity that remains technically difficult to quantify. This simplification limits our understanding of key ecological properties of environmental microbial communities, including how activity switching among cells and taxa contributes to system resilience. This talk will explore the microbial activity spectrum and its ecological consequences. I will show how readily available, single-cell phenotypes can be used to differentiate active cells and quantify community activity intensity. Then, I will apply these data to examine soil microbiome stability and responses to environmental perturbations. I advocate that routine, quantitative accounting of total cells, active cells, and activity intensity should become standard practice in microbiome studies to strengthen ecological interpretation. Together, this work provides insights into the capacity of environmental microbiome to resist and recover from disturbances associated with climate change.

S36*/P155* Sublethal pesticide exposure and the honey bee gut microbiota: interactions and impacts on bee health

Ms. Jamille Viray[1], Dr. Andrew Quinn[2], Dr. Richard Jacoby[3], Mr. Lenard Kreis[3], Prof. Philipp Engel[1], Prof. Michael Zimmermann[3], Ms. Thi Huong Giang Nguyen[2]

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Bees are essential pollinators whose health is increasingly threatened by agricultural intensification and widespread agrochemical exposure. While pesticide risk assessment largely focuses on acute toxicity, chronic sublethal exposure can also impair immune competence, pathogen resistance, and foraging behavior. Because these same functions are partly supported by the specialized honey bee gut microbiota, pesticide-induced perturbation of this community may contribute to sublethal effects on the host. Conversely, the gut microbiota may modulate pesticide toxicity through biotransformation or by altering their persistence and bioavailability.

Here, we use the honey bee gut microbiota to investigate reciprocal pesticide–microbiome interactions. Building on a high-throughput *in vitro* screen of agrochemical effects on bee-associated bacterial isolates, we exposed gnotobiotic honey bees colonized with a defined synthetic community to selected agriculturally relevant pesticides at sublethal concentrations corresponding to 1% of the reported bee LD50. Microbial community dynamics were followed across exposure and recovery using qPCR and full-length 16S rRNA amplicon sequencing.

Overall, the synthetic gut community was largely resilient at these concentrations, although selected pesticides caused subtle strain-specific shifts in community assembly. Follow-up experiments comparing mono-colonization with *Gilliamella apicola*, a conserved honey bee gut bacterium, to full-community colonization at higher pesticide doses further suggest that community context can buffer pesticide sensitivity of vulnerable strains. In parallel, targeted and untargeted LC-MS analyses revealed pesticide-dependent differences in gut metabolite profiles and pesticide persistence, providing a basis to explore microbiota-mediated pesticide biotransformation and broader metabolic changes *in vivo*. Finally, pathogen challenge experiments with *Serratia marcescens* showed that some pesticide treatments increased infection frequency, suggesting that sublethal exposure may weaken microbiota-mediated colonization resistance. This provides an initial readout for exploring how pesticide-induced microbiota perturbations may affect broader bee health outcomes. Together, our results support the gut microbiota as a key factor in modulating the sublethal impacts of agrochemical exposure on bees.

**Student Paper*

S37* Understanding soil microbiomes' drivers using function-based global ecosystems classifications

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We present a prototype framework for the research of soil microbiomes using an externally defined functional ecosystem classification system. This ecosystem typology is the IUCN Global Ecosystem Typology, a hierarchical system of 34 terrestrial Ecosystem Functional Groups (EFGs) defined by their main ecological drivers. By linking EFG definitions to soil samples from MicrobeAtlas database via their geographic coordinates. We matched 5,635 samples to nearly all the terrestrial EFGs. To validate the classification's applicability, we examined within- and between-community compositional dissimilarities. The EFGs showed higher discriminative power than categories based purely on pH (PERMANOVA mean adjusted R^2: 0.36 vs. 0.27). Alpha diversity estimates revealed that understudied EFGs —such as tropical dry forests— may represent unrecognized biodiversity hotspots. We hypothesized that each EFG must contain foundational taxa that strongly influence community structure and function. For every EFG with sufficient samples, we built a co-occurrence network to predict these foundational taxa. We identified them through topological features that achieved >85% accuracy in identifying such taxa within simulated networks. By including environmental metadata in the networks, we could also identify drivers capable of having pivotal influence over these ecosystems. Overall, the Global Ecosystem Typology provides ecologically meaningful ecosystem descriptions that allow for strong comparisons of soil microbiomes, establishes a framework for future research, and aids in recognizing prokaryotic taxa that have a disproportionate impact on their environment.

**Student Paper*

S38 Stickland metabolism operates as a distributed community property governed by hierarchical thermodynamic constraints

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Amino acid fermentation requires balancing oxidative and reductive half-reactions, yet how this balance is achieved at community scale remains unresolved. Using genome-resolved metatranscriptomics of controlled anaerobic enrichment communities, we show that Stickland metabolism is partitioned across community members rather than confined to individual genomes.

A three-tiered hierarchy structures this partitioning: methanogenesis sets the dominant thermodynamic boundary, suppressing glycine reductase community-wide, and reshaping the entire electron economy; feed stoichiometry enforces obligate coupling between oxidative and reductive Stickland arms, with neither operating independently when cognate substrates are absent; and amino acid concentration gates a biosynthetic growth program only when both upstream constraints are satisfied. Among MAGs encoding both Stickland arms, co-activation was observed in fewer than half, with the majority deploying a single arm.

These patterns demonstrate that redox balance in amino acid fermentation is an emergent property of the community ensemble, maintained through distributed functional specialization rather than intra-genomic coupling.

S39 rethinking chain-length distribution dynamics in diatom populations through cell size and cellular organization

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Diatom blooms play an important role in the global biological pump through fixation of inorganic carbon. During bloom formation many species undergo a progressive mean size reduction as they proliferate. Yet much of our understanding of these dynamics derives from bulk population measurements, which obscure the single-cell behavior that ultimately drives them. Deciphering the mechanisms linking cell division, cell size, and bloom formation therefore requires a closer look at individual cells. To better understand bloom formation and modulation of division rates at the single-cell level, we used a multi-scale approach, observing cell size, cell cycle progression, and the impact of variations within these on population growth in chain-forming diatoms. High resolution, time-lapse observations of cells propagating in time and space along a lengthening chain within a growing population allow us to elucidate the relationship between division rate and cell size, and identify potential morphological indicators of division rates in growing chains. We further explore the relationship between morphological parameters, cell division rates, and population growth across different phases of the life cycle using cryo-expansion microscopy. Our experiments were done using *Chaetoceros decipiens*, a chain- and bloom-forming diatom frequently found in marine environmental samples. We describe growth dynamics and test predictions for geometric parameters which affect population-level growth, and potentially bloom formation. This work allows us to begin creating a link between single-cell behavior and emergent shape and growth dynamics, gaining a better understanding of the role that diatom cell dynamics play in the formation of ecologically successful blooms.

M18 Dissecting Zoonotic Virus Infection Through Advanced Cellular Imaging

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Abstract not received yet.

S40* Viral inhibition through structure targeting the frameshifting element of SARS-CoV-2

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The SARS-CoV-2 programmed −1 ribosomal frameshifting element is essential for viral replication and represents a promising antiviral target. However, all inhibitors validated to date were tested against a minimal 84-nucleotide RNA fragment derived from in vitro studies. Recent cellular probing experiments reveal that the frameshifting element adopts alternative conformations in cells, including a longer-range interaction spanning 1.4 kb. These cellular conformations are proposed to be the predominant forms in infected cells, with the minimal structure appearing only transiently near the translating ribosome. Targeting these biologically relevant structures offers two advantages: inhibiting the most abundant conformations and potentially blocking the conformational switch required for frameshifting. Yet no screening efforts have focused on these cellular structures, leaving unexplored the possibility of identifying more potent inhibitors.

My PhD research addresses this gap through an integrated experimental and computational approach. I conducted high-throughput screening of an RNA-targeting compound library using a dual-luciferase reporter assay with a 527-nucleotide construct including the 5' region of the long-range interaction. Hits were counterscreened against positive and negative controls to eliminate false positives. In parallel, I performed virtual screening of the same library using RNA-ligand docking. Validated hits were then tested in SARS-CoV-2-infected cells to assess antiviral efficacy. Eleven molecules successfully impeded viral replication, and one molecule was identified by both experimental and computational approaches. Computational analysis revealed potential binding pockets that will now be used to perform virtual screening of additional compound libraries and guide the design of optimized analogs. This strategy aims to discover inhibitors with improved potency against cellular RNA conformations and establish a framework for developing broad-spectrum antivirals targeting structurally related frameshifting elements across RNA viruses.

**Student Paper*

S41*/P151* Hemagglutinin-associated stability of H5N1 influenza A virus in exhaled respiratory droplets

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The stability of influenza A virus (IAV) in exhaled respiratory droplets is a key determinant of airborne transmission, as virions are rapidly exposed to inactivating conditions such as low pH, and high salinity. Different strains of IAV vary in their ability to transmit via aerosols and the viral hemagglutinin protein (HA) plays a central role in transmissibility. To assess whether HA contributes to viral stability during the airborne phase, we characterized a pair of biosafe replication-incompetent H5N1 reporter viruses. Both viruses are based on the A/Vietnam/1203/2004 isolate but differ in their HA protein: While one (Viet/04-Renilla) carries the HA of the non-airborne transmissible A/Vietnam/1203/2004 isolate, the other one (Viet/04-Renilla-Tx) carries HA of A/Texas/37/2024, which belongs to the currently circulating H5N1 2.3.4.4b clade associated with airborne transmission in the ferret model. Our data indicate that Viet/04-Renilla-Tx displays increased thermal stability compared with Viet/04-Renilla. Under high-salinity conditions in bulk solutions, no discernible difference in stability was observed between the two viruses. However, in drying 0.5 and 5uL droplets, which mimic efflorescence and salt supersaturation during aerosol evaporation, Viet/04-Renilla-Tx exhibited increased stability relative to Viet/04-Renilla. In a next step, controlled aerosol chamber experiments are planned to assess viral stability in aerosols. Together, our findings indicate that Viet/04-Renilla carrying HA from A/Texas/37/2024 exhibits greater resistance to inactivating conditions during the airborne phase, which may contribute to its increased transmissibility.

**Student Paper*

S42 Tracking West Nile Virus in Southern Switzerland

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West Nile virus (WNV) circulation has increased across Europe, becoming a growing public and veterinary health concern. This mosquito-borne zoonosis is maintained in an enzootic cycle between local birds and mosquitoes, while humans and equids act as dead-end hosts. Although most infections are asymptomatic, severe neuroinvasive diseases may occur.

Given its proximity to Northern Italy, where WNV has been circulating since 2008, the Canton of Ticino established an arbovirus surveillance system in 2010, which has since been progressively expanded. Between 2020 and 2025, entomological monitoring increased from 5 to 13 sampling sites in natural areas with permanent water bodies, focusing on *Culex pipiens*, the primary vector. Female mosquitoes were collected biweekly from June to October using gravid traps equipped with honey-baited FTA cards, which preserve viral RNA released by feeding mosquitoes. Samples were tested using pan-flavivirus RT-PCR and RT-qPCR assays for WNV and Usutu virus (USUV).

Findings indicate an epidemiological shift: USUV was detected in 2020, while 2021 yielded no positives. In 2022, WNV lineage 2 was identified for the first time in Switzerland in both FTA cards and mosquito pools, persisting into 2023 alongside USUV. In 2024, only sporadic USUV detections occurred, while WNV was absent. Both viruses reappeared in the following season.

After the first detection of WNV in native mosquitoes, a bird-ringing pilot study in 2023 found no PCR-positive swabs but identified 15 ELISA-positive birds, indicating prior exposure. To strengthen integrated surveillance, a two-year pilot study launched in 2025 is assessing WNV seroprevalence in blood donors from Italian-speaking Switzerland through collaboration among specialized institutions and cantonal authorities.

Ticino presents all conditions for sustained WNV circulation: competent vectors, reservoir birds, and increasingly warm summers. Continuous integrated surveillance matching entomological, animal and human epidemiological data (One Health approach) remains essential to evaluate risk and protect public and animal health.

M19 Modeling host-microbe interactions in immunocompetent engineered human gut tissues

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As a primary port of pathogen entry, the intestinal mucosa has developed a sophisticated set of defenses to ward off infections. Investigating the human-specific intestinal barrier responses to pathogens is limited by the lack of models integrating the necessary architectural complexity with an autologous immune compartment. Here, we developed engineered barrier models of the human small intestine and colon combining a multilineage epithelium, mucus layer, accessible microbial compartment, and autologous tissue-resident immune cells. These tissues recapitulate morphological and functional properties that are region-specific and compare to those of the native organ. We use the model to characterize the human epithelial response to Salmonella Typhimurium at the single-cell level, uncovering the distinct gene regulatory networks that underpin luminal versus intracellular pathogen presence. Invaded epithelial cells initiate an IL-18 signaling axis that triggers cytotoxic T-cell activation, resulting in epithelial shedding and barrier disruption. Overall, this work allows for the modular integration of epithelial, microbial, and immune compartments providing a versatile system for studying human intestinal physiology and pathologies.

S43 Staphylococcus aureus localization in infected human bones

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Staphylococcus aureus is the leading cause of difficult-to-treat bone and joint infections (BJIs), with failure rates of 20-30% despite combined surgical and antibiotic therapy. Proposed underlying mechanisms include *S. aureus* biofilm formation, intracellular localization, and invasion of the bone canalicular network, yet patient-derived evidence remains scarce and qualitative.

To address this issue, we imaged bone biopsies from 10 infected human patients, sampled at multiple sites and/or surgeries, using decalcification, vibratome sectioning, and immunohistochemistry. The labelled biopsy sections were automatically imaged using confocal microscopy combined with AI-based bacterial detection (neural network).

Analysis of >26,000 *S. aureus* cells in human bone biopsies revealed heterogeneous infection patterns across bone and surrounding tissues. Bacteria were detected both intra- and extracellularly. Intracellular *S. aureus* was predominantly found in phagocytes within bone marrow and bone-associated soft tissues, whereas infection of bone-resident cells (osteoblasts, osteocytes, osteoclasts) was rare. Occasional bacteria were observed within bone marrow adipocytes. Extracellular *S. aureus* appeared mainly as single cells or small clusters in soft tissues, while large aggregates were confined to specific microanatomical niches, such as bone cracks in one patient. Only a limited number of bacteria were detected within the canalicular network as demonstrated before.

Our dataset suggests limited infection of the bone matrix. Thus, bone penetration of antibiotics and immune cells might be less critical than previously assumed. Our data inspire laboratory models mimicking relevant aspects of bone infections as a basis for developing more effective BJI therapies.

S44 Distinct Pseudomonas aeruginosa subpopulations inflict tissue damage during infection

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Pseudomonas aeruginosa is a Gram-negative bacterium and opportunistic pathogen that can infect the airways of susceptible individuals and is particularly problematic due to rising antibiotic resistance. Distinct bacterial subpopulations of *P. aeruginosa* arise upon surface sensing or in biofilm conditions, due to the existence of different microenvironments or to intrinsic heterogeneity of the bacterial population, which can influence virulence or antibiotic resistance profiles. The prevalence of this phenotypic heterogeneity *in vivo* and its relevance for infection are uncharacterized. In this project, we developed tools and approaches to characterize the physiology of *P. aeruginosa* subpopulations that arise under infection-mimicking conditions.

Specifically, we screened >70 fluorescent transcriptional reporters that provide single-cell information on the physiology of *P. aeruginosa* cells in *in-vitro* infection models, including Synthetic Cystic Fibrosis sputum Medium (SCFM) and A549 alveolar epithelial cells. Around 25% of tested reporters display heterogeneous expression in at least one condition. We next validated that at least 5 of these reporters are also heterogeneously expressed in a transwell model of the airway epithelium, including pathways that respond to oxygen levels, metabolite concentrations and others. We developed approaches based on fluorescence-associated cell sorting (FACS) coupled with high sensitivity proteomics to characterize the physiological state of these bacterial subpopulations and implemented state-of-the-art image analysis pipelines to assess their virulence potential. Lastly, we generated *P. aeruginosa* strains locked in ON or OFF states of expression of the pathways under study to study the impact of these subpopulations on the host tissue. These approaches point to a role of previously unrecognized pathways in *P. aeruginosa* pathogenesis, and further highlight the importance of quorum sensing and the type III secretion system for the pathogenic process. These findings hint towards novel antivirulence approaches that may contribute to driving *P. aeruginosa* cells towards avirulent states.

S45∗/P153∗ A genome editing pipeline in human lung cells enables functional dissection of epithelial barrier protection against P. aeruginosa

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The opportunistic pathogen *Pseudomonas aeruginosa* (*PA*) is a leading cause of life-threatening respiratory infections. During airway colonisation, the bacterium uses diverse strategies, including surface adherence, virulence induction, and tissue dissemination, to adapt to the respiratory niche. However, studying these complex dynamics in murine models is limited by the lack of high temporal and spatial resolution, while many cell models fail to faithfully recapitulate human airway physiology. To provide a more translatable and physiologically relevant understanding of the interaction between *PA* and the airway epithelium, we adapted a Transwell-based air-liquid interface (ALI) model from primary human bronchial epithelial stem cells. This microtissue faithfully recapitulates the native airway epithelium, showing proper epithelial barrier function and mucociliary differentiation.

In order to define the host–pathogen interactions that facilitate or restrict *PA* invasion, we established a highly efficient, virus- and plasmid-free CRISPR-Cas9 genome editing pipeline. This platform allows us to systematically deconstruct the epithelial defence layers, such as mucus production and ciliary function, to determine their individual roles in bacterial infection. As a functional validation, we targeted FOXJ1, a master regulator of ciliogenesis, which led to marked changes in airway epithelial morphology and loss of ciliation. By enabling the precise genetic manipulation of the human airways, this approach allows for a detailed dissection of host-pathogen dynamics with a level of precision difficult to achieve *in vivo*.

∗Student Paper

S46 A novel human in vitro bladder model to study patient-specific responses to uropathogenic E. coli

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Urinary tract infections (UTIs) are highly prevalent, with huge heterogeneity in the course of infection, ranging from quick clearance to chronic disease. The heterogeneous response to UTI has been investigated in mouse models, but little is known about the heterogeneity in humans. To understand UTI and the tissue immune response comprehensively, there is a need for human, patient-derived models. We obtained patient-derived bladder biopsies and blood samples to generate a patient-specific microtissue model to study patient-specific responses to UTI.

Bladder biopsy and blood samples are received from UTI patients and non-UTI controls. PBMCs were isolated from the blood samples and frozen for later integration. Urothelial cells were isolated from biopsies and expanded as urothelial organoids following published protocols. After expansion, the urothelial organoids were dissociated into single cells and seeded on Transwell membranes to generate stratified urothelial microtissues. The microtissues were infected with uropathogenic *E. coli* (UPEC) strain UTI89 and the infection process was followed with microscopy.

Urothelial cells could be successfully isolated from biopsies and expanded as organoids. Biobanking of organoids and PBMCs is ongoing. Organoid-derived urothelial microtissues have been generated from three patient cell lines, containing all three urothelial cell types, as confirmed with immunohistochemistry. Barrier function has been confirmed with transepithelial electrical resistance measurements. UPEC infections have shown that bacteria attach to the tissue and invade. Integration of monocyte-derived macrophages to study their response to UPEC infection is ongoing.

Next, patient-derived microtissues will be generated from additional organoid lines, to study patient-specific infection dynamics. Patient-derived PBMCs will be integrated to study the immune response to UPEC infections. With the unique access to urothelial tissue and donor-matched immune cells, this model offers an exceptional opportunity to study patient-specific responses to UTI, elucidating heterogeneity in tissue responses to infection and antibiotic treatment.

M20 Being Lazy is not always bad: Cyclic di-AMP signaling in cyanobacteria

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Because of their photosynthesis-dependent lifestyle, cyanobacteria evolved sophisticated regulatory mechanisms to adapt to oscillating day-night metabolic changes. How they coordinate the metabolic switch between autotrophic and glycogen-catabolic metabolism in light and darkness is poorly understood. Recently, we discovered that the second messenger c-di-AMP is implicated in diurnal regulation[1]. The carbon-sensor protein SbtB[2,3] was identified as a major c-di-AMP receptor[1,4]. Adding to its cellular functions catalog, for first time, we show also that c-di-AMP influences on nitrogen metabolism, implying new cellular roles for c-di-AMP[2]. Additionally, we found that the cyanobacterial pilus biogenesis and natural competence[5] are regulated by c-di-AMP/c-di-GMP and showed that the ComFB signaling protein is a novel c-di-NMP-receptor protein[6], widespread in bacterial phyla[6], and required for DNA uptake[5].

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S47* Mycomembrane proteins are inserted at zones of polar growth and linked to the cell wall

Ms. Dieuwertje de Bruin[1], Dr. Elizabeth Hart[2], Dr. Victoria Marando[2], Ms. Marina Arnau Alemany[1], Dr. Andrea Vettiger[1], Prof. Thomas Bernhardt[2]

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The Mycobacteriales are a unique order of bacteria that includes several important human pathogens such as *Mycobacterium tuberculosis*. They possess a complex cell envelope architecture that includes a two-layered cell wall composed of peptidoglycan (PG) and arabinogalactan, and an atypical outer membrane consisting of mycolic acids called the mycomembrane (MM). This layer mediates a strong permeability barrier that contributes to intrinsic resistance to many antibiotics in these organisms. At the same time, the MM contains a high number of mycolate outer membrane proteins (MOMPs), a subset of which are thought to form pores for nutrient uptake. However, unlike porins in the Gram-negative outer membrane, the mechanisms governing the secretion and assemby into the MM remain largely unknown.

Using *Corynebacterium glutamicum* as a model organism, we combined fluorescence microscopy, biochemistry, and genetics to show that MOMP incorporation is coupled to PG synthesis and occurs specifically at the cell poles. We further demonstrate that, once inserted into the mycomembrane, MOMPs become non-diffusive and remain stably associated with the underlying cell wall. Furthermore, we show that MOMP assembly depends on hetero-oligomerization and a unique post-translational modification, O-mycoloylation, and we identify the responsible proteins for this process. Finally, we present a high-confidence AlphaFold model of the hetero-pentameric channel formed by the MOMPs PorA and PorH.

Overall, our findings reveal surprising mechanistic parallels to outer membrane porin assembly in Gram-negative bacteria, despite fundamentally distinct envelope architectures and the absence of a BAM-like assembly machinery.

**Student Paper*

S48 One does not simply kill Brucella: Intracellular antibiotic tolerance, population heterogeneity, and an RND Efflux Pump

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Brucella abortus and *Brucella melitensis*, risk group 3 pathogens requiring biosafety level 3 (BSL-3) containment for laboratory manipulation, cause Brucellosis, a neglected zoonosis that remains a major public health threat and imposes substantial burdens on human health and animal husbandry in low- and middle-income countries. Human brucellosis requires prolonged combination therapy with doxycycline plus either rifampicin or streptomycin, as recommended by the WHO since 1986. Despite this intensive regimen, relapse rates remain high (5-15%), and the underlying mechanisms remain poorly understood.

We have previously demonstrated that a small subpopulation of phagocytosed *Brucella abortus* can survive antibiotic treatment and retain full virulence in a macrophage infection model, potentially serving as a reservoir for relapses. To better characterize this surviving subpopulation, we developed a BSL-2-compatible infection model based on *Brucella microti*, which recapitulates the previously observed "persister" phenotype—displaying biphasic killing in broth and intracellular survival after antibiotic treatment. Using *Brucella microti* as a proxy, we show that phagocytosed bacteria retained in the endocytic network survive antibiotic exposure displaying high antibiotic tolerance. Notably, the stage of intracellular trafficking at treatment initiation does not affect survival rates, indicating the existence of tolerant bacteria before treatment start. Using transposon sequencing analysis of *Brucella abortus*, we show that a conserved efflux pump of the RND superfamily is essential for treatment survival of the non-replicating subpopulation.

Building on these findings, we are developing a high-throughput screening platform to identify drugs capable of targeting this tolerant intracellular subpopulation. Given that only a subset of wild-type bacteria transition to the replication-permissive, ER-associated niche—while the majority remains in a growth-arrested state—we hypothesize that this physiological heterogeneity represents a classical bet-hedging strategy employed by *Brucella* to ensure survival in adverse conditions and a better understanding of this strategy can help adopting more efficacious treatment schemes.

S49* Bacterial L-form cellular fusion creates interspecies mosaic genomes

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Exposure to cell wall-targeting agents, such as β-lactams, can induce bacteria into a wall-deficient L-form state adapted to envelope stress. Without a peptidoglycan layer, L-forms exhibit altered cellular behaviors, including multinucleation and cell-cell fusion. These properties suggest that L-forms may provide a unique route for horizontal gene transfer beyond classical mechanisms such as transformation, transduction, and conjugation. However, the evolutionary consequences of L-form cellular fusion remain poorly understood. Here, we demonstrate cross-species cellular fusion between *Listeria monocytogenes* and *Listeria innocua* L-forms, sharing approximately 87% average nucleotide identity. Using distinct fluorescent and antibiotic resistance markers in each parental strain, we observed fused L-form cells co-expressing phenotypes from both parental strains. However, the hybrid state was unstable without continued dual selection, highlighting the highly plastic genomic organization of L-forms. Under prolonged exposure of fused L-form cells to dual selective pressure, we observed revertant cells, which regained a cell wall and returned to a monoploid-like state while maintaining phenotypes from both parental strains. Whole-genome long-read sequencing of these revertants cells revealed enlarged mosaic chromosomes containing contributions from both parental genomes, with genome sizes increased by 4.3–18.1%, as well as novel coding sequence variants. Altogether, these findings demonstrate that L-form fusion can transcend species barriers, serving as a mechanism for extensive genomic recombination and bacterial evolution.

**Student Paper*

S50 MtvS1 and MtvS2 Interact With RNA Polymerase to Regulate the Francisella Type V-A CRISPR-Cas System

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CRISPR-Cas systems endow bacteria and archaea with adaptive immunity against mobile genetic elements, playing a fundamental role in shaping microbial communities. Many organisms harbor more than one CRISPR-Cas system, and little is known about whether and how they are differentially regulated, in many instances due to the impossibility of studying CRISPR immunity in native hosts. Here we studied the regulation of endogenous type II-B and type V-A CRISPR-Cas systems in opportunistic human pathogen *Francisella novicida* U112. Fluorescence microscopy and transcriptomics experiments revealed that while the type II-B system is constitutively expressed, the type V-A CRISPR-Cas system is differentially expressed at stationary phase and high cell density. Using mass spectrometry and genetics, we identified MtvS1 and MtvS2 as factors required for the differential expression of the type V-A CRISPR-Cas locus. In addition, MtvS1 and MtvS2 modulate transcription of many genes in stationary phase, some of which are required for *Francisella* virulence. Pull-down experiments revealed both *Francisella* MtvS proteins bind to RNA polymerase. MtvS1 is predicted to interact with the β′ subunit of the RNA polymerase, and MtvS2 with multiple RNA polymerase subunits as well as MtvS1. We propose that MtvS1 and MtvS2 constitute noncanonical alternative sigma factors involved in the regulation of the expression of CRISPR loci and other genes in *Francisella*. Last, we show that *E. coli* MtvS1 homolog YgfB is also required for expression of the type I-E CRISPR-Cas system in *E. coli*. Intriguingly, MtvS1 rescued the phenotype of a *ygfB* mutant, suggesting functional redundancy and that MtvS1 homologs may have a broader role in gene expression in bacteria.

M21 Fungal Infections in times of Climate Change

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Climate change is having a growing impact on fungal diseases, affecting both the behavior of fungal pathogens and the vulnerability of human populations. Rising temperatures, changing rainfall patterns, and an increase in extreme weather events create ideal conditions for fungi to spread and survive in new environments. This has led to a growing incidence of fungal infections—caused by *Candida auris*, *Histoplasma*, *Sporothrix*, and *Talaromyces marneffei*—some of which are becoming increasingly resistant to treatment [1], and also to the mergence of new fungal pathogens [2]. Natural disasters linked to climate change, such as floods, hurricanes, and wildfires, can lead to increased exposure to fungi, particularly through trauma and environmental contamination [3]. At the same time, human susceptibility to fungal infections is rising due to weakened immunity, malnutrition, urbanization, and climate-related migration. Social factors such as poverty and limited access to healthcare further increase the risk, especially in low- and middle-income countries [4].

The spread of fungal diseases is now a global issue that affects countries across all income levels. However, poorer regions are often hit hardest because of weaker health systems and limited diagnostic resources. Addressing these challenges requires global cooperation, increased investment in healthcare infrastructure, better access to antifungal treatments, and more research on fungal pathogens [1]. Wealthier nations have a responsibility to support global efforts, as fungal diseases can emerge anywhere and spread rapidly. Understanding how climate change influences fungal diseases is essential for improving prevention, early detection, and effective treatment. By working together, countries can strengthen their ability to manage the growing threat of fungal infections in a changing climate.

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S51 Candidalysin orchestrates homeostatic Th17 immunity in response to C. albicans oral colonization

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Microorganisms that colonize mucosal surfaces continuously interact with the immune system while avoiding excessive inflammation or tissue damage. In the oral cavity, these interactions occur across a highly structured stratified epithelium that physically separates microbes from immune cells, raising the question of how commensal organisms are sensed by the host. Here, we show that the common oral fungus *Candida albicans* uses the pore-forming peptide candidalysin to engage antigen-presenting cells (APCs) and promote efficient Th17 induction. When *C. albicans* is put in direct contact with APCs, candidalysin enhances antigen uptake and presentation without altering the production of APC-derived polarizing cytokines. Using a mouse model of persistent oral colonization, we identified Langerhans cells as the key APC subset mediating Th17 induction through the extension of dendrites across the stratified epithelium. Candidalysin enables fungal access to these extensions by facilitating traversal of the outer keratinized epithelial barrier. Together, our findings suggest that candidalysin, originally characterized as a virulence factor, has likely evolved primarily as a commensalism factor that promotes mutually beneficial host-fungal interactions without damaging the host.

S52 Sticky situation: uncovering the molecular and mechanical basis of Candida adhesion

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Fungal pathogens pose a major and growing challenge to human health, particularly in vulnerable patients and healthcare-associated settings. Our work examines the earliest stages of fungal colonization from a molecular and biophysical perspective, focusing on how cell-surface proteins mediate adhesion to host-associated surfaces under mechanical forces prevalent in the human body. Because these forces can strongly influence molecular interactions, understanding adhesion in this context is critical for clarifying pathogen colonization.

To investigate this, our study integrates yeast-surface display, spinning-disk adhesion assays, single-molecule atomic force microscopy, and high-throughput deep mutational scanning. Together, these approaches enable the investigation of adhesion across multiple scales, from population-level behavior to molecular-scale binding events, providing a comprehensive view of adhesion in pathogenic *Candida* species. This workflow is readily adaptable for investigating adhesion in other fungal pathogens, and a broader understanding of these early host-pathogen interactions may help inform future strategies to limit infection and transmission.

S53* Exploring Mitogen-Activated Protein Kinases (MAPK) pathways in Candida auris

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Candida auris is an emerging pathogenic yeast first identified in 2009 that has rapidly spread worldwide and caused numerous healthcare-associated outbreaks. Due to its multidrug resistance and high pathogenic potential, the World Health Organization (WHO) and the Centers for Disease Control and Prevention (CDC) have classified *C. auris* as an urgent public health threat. Its remarkable stress tolerance, including optimal growth at 37°C (and possibility to grow at 42°C) and high salinity resistance, suggests a strong adaptive capacity that may have contributed to its emergence.

Mitogen-Activated Protein Kinase (MAPK) pathways regulate essential cellular processes such as stress adaptation, proliferation, and differentiation. In *C. auris*, deletion of the Stress-Activated Protein Kinase (SAPK) *HOG1* increases sensitivity to osmotic stress while restoring susceptibility to amphotericin B and caspofungin, indicating a major role for MAPK signaling in stress response and antifungal resistance.

Morphogenetic transitions, including pseudohyphal growth, aggregation, adhesion, and biofilm formation, are key virulence traits in *Candida* species, yet their regulation in *C. auris* remains not totally understood. We hypothesize that MAPK pathways are central regulators of these pathogenic phenotypes and directly contribute to environmental adaptation and virulence.

To address this question, we will construct reporter genes specific to major morphogenetic transitions and analyze their expression under different environmental conditions. MAPK deletion mutants will be generated to assess their impact on stress adaptation, morphogenesis, adhesion, and biofilm formation. We expect that disruption of specific MAPK pathways will impair virulence-associated phenotypes while increasing antifungal susceptibility. These results should identify MAPK signaling as a critical regulator of pathogenicity in *C. auris* and reveal promising targets for novel antifungal strategies.

**Student Paper*

S54 Deciphering the Tac1a / Tac1b network of azole resistance in Candidozyma auris

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Context: *Candidozyma auris* is a concerning pathogenic yeast because of its ability to develop antifungal resistance. Resistance to fluconazole (one of azoles) is a hallmark in *C. auris* and the efflux pump Cdr1 plays a key role in this process. The gain-of-function mutations in the transcription factor Tac1b are known to overexpress *CDR1*. *TAC1b* is located in tandem with *TAC1a*, whose function is not yet elucidated.

Objective: We aimed at investigating the respective roles of Tac1a and Tac1b in fluconazole resistance.

MethodS: The functions of *TAC1a/*b were studied by hyperactivation using a tagging strategy. RNA-sequencing and ChEC-sequencing were performed to identify potential targets of Tac1a/b. *In vitro* microevolution experiments were conducted to assess the evolution of *TAC1a*.

Results: Hyperactivation of Tac1a resulted in markedly decreased fluconazole susceptibility similar to Tac1b hyperactivation. Tac1a and Tac1b shared a large common set of downstream targets, including *CDR1*, and use the same binding motif. Other common targets included the efflux pump Cdr4_1, with no role in fluconazole resistance, and the transcription factors Zcf13 and Mrr1b, that demonstrated some ability to impact fluconazole susceptibility but independently from Cdr1. Tac1a was able to bind to the *TAC1b* promoter (and not inversely), suggesting that Tac1a is located upstream of Tac1b in this regulatory pathway. Finally, our attempts to induce *TAC1a* mutations were unsuccessful, showing a different evolutionary fact from *TAC1b*.

Conclusions and perspectives: Tac1a/b have redundant functions but with distinct roles in the evolutionary process of azole resistance with Tac1a being the ancestral conserved gene and Tac1b having evolved to trigger azole resistance.

M22 Ecological diversity of microbial immunity

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Bacteria and archaea have evolved numerous defense pathways to fend off infections by viruses and other genetic elements. However, the current census of prokaryotic immune diversity is heavily biased towards cultivated lineages, focuses on molecular mechanisms or evolutionary conservation, with little information on ecological distribution. Here, we screened 3.9 million genomes, including >2 million recovered from over 10,000 metagenomic samples, and identified 17 million defense systems. We then propose a framework to organise this diversity in defense system families using unsupervised iterative clustering at scale. With this approach, we identify 300,000 defense system families across microbiomes, find that environments shape immunity independently of taxonomy, identify distinct immune repertoires within ecosystems, and track immune dynamics in the human gut. This works contributes to a new picture of prokaryotic immunity by providing an ecological lens at global scale.

S55 Social isolation accelerates divergent evolution in the gut microbiota

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The gut microbiota—a key regulator of host health—is shaped by the host's social environment. Social interactions can drive the transmission of gut bacteria between hosts, altering the taxonomic composition of the gut microbiota. However, relatively little is known about the effects of the host's social environment on the evolution of gut symbionts. We show that social isolation accelerates the genomic divergence among gut bacterial populations in two social animals: wild-derived house mice (*Mus musculus domesticus*) and common eastern bumblebees (*Bombus impatiens*). Using genome-resolved metagenomics, we quantified genomic dissimilarity between populations of gut bacterial species in single- and group-housed animals. We found that after prolonged social isolation, bacteria in single-housed animals diverged not only from conspecifics in group-housed animals but also from those in other single-housed animals. These results support our hypothesis that disrupting the social transmission of gut bacteria changes the strength of gene flow and random genetic drift acting on bacterial populations, accelerating their divergent evolution. More broadly, they demonstrate that the host's social environment shapes not only the ecology but also the evolution of gut symbionts.

S56* A minimal bacterial community provides insights into the microbial ecology of marine chitin degradation

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Bacterial metabolism drives global biogeochemical processes, including carbon cycling in marine ecosystems. Here, bacteria degrade organic matter and direct carbon either to remineralisation, or sinking and sequestration in the seafloor. Polysaccharides such as chitin are major components of marine organic carbon, whose extracellular degradation by specialised degraders attracts metabolically diverse non-degrading bacteria to the particle environment. This co-occurrence enables both metabolic and non-metabolic interactions between bacteria, which can influence the degradation process and thus the fate of carbon in the ocean. However, these interactions are often studied using pairwise strain combinations, which cannot capture the taxonomic and functional diversity present in particle-associated communities. To study interactions between chitin-associated marine bacteria in a community context, we developed a defined minimal community consisting of 4 stably coexisting natural isolates (2 degraders and 2 non-degraders). We investigated interactions between community members using transcriptomics, supported by time-resolved metabolomics, growth, and composition measurements. With this approach, we could show the community reproduces dynamics known to be important in particle-associated communities, including competition for chitin degradation products, biotin crossfeeding that supports an auxotroph, and type VI secretion system deployment that may play a role in nutrient acquisition. Further, we could pinpoint metabolites that are externalised by other community members during chitin degradation and crossfed by non-degraders. Looking ahead, we hope that the minimal community presented here will be used more broadly to study marine particle-associated communities involved in polysaccharide degradation and carbon cycling. Possible applications include exploring general ecological principles of such communities, as well as investigating their responses to environmental perturbations and measuring process rates for the parametrization of community- or ecosystem-level models.

**Student Paper*

S57* full-length 16S sequencing reveals strong effects of host phylogeny on gut microbes in wild micromammals

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The gut microbiome plays key roles in host metabolism, immunity, and pathogen resistance. Different animal species harbor distinct microbial communities in their gut; a pattern known as host specificity. While this seems common in some mammals, its prevalence remains poorly understood – especially in wild animals. Here we probe host-gut microbiome specificity using a model of small rodents and insectivores (micromammals) in their natural habitat at a local scale. We characterize the gut microbial diversity of wild mice, voles, and shrews by capturing these animals alive and sampling fresh fecal content as a proxy for the intestinal microbiome. We use full-length 16S rRNA sequencing and multivariate methods to quantify host specificity. To our knowledge, this is the first instance of this long-reads (1.5kb) approach used for describing bacterial communities in wild micromammals, instead of classical partial (0.2-0.5kb) 16S rRNA sequencing techniques. The full-length approach shows strong signals of host specificity – even between closely related sympatric hosts. We will further compare these results against partial 16S data (sequencing ongoing) to disentangle whether observed differences reflect primer/PCR biases or gains in taxonomic resolution. By combining ecological sampling with explicit methodological comparisons, we demonstrate how full-length 16S sequencing enhances the characterization of complex, previously undescribed microbiomes in wild systems. Our results provide both biological insight into host specificity and a framework for improving microbiome studies in non-model organisms.

**Student Paper*

S58 Resource-dependent interactions drive community assembly in a defined gut bacterial community

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Ecological interactions sustained by the transformation and exchange of resources underpin the assembly, stability, and function of microbial communities. Yet how species-level resource utilization gives rise to these interactions, and ultimately shapes collective community behavior, remains a major challenge.

To address this, we assembled a 26-member human gut bacterial community, spanning seven phyla, and systematically manipulated the availability of dietary polysaccharides. By comparing monoculture and community liquid culture experiments to spatially structured microfluidic experiments, we show how species-level metabolic traits translate into ecological interactions and community-level outcomes across diverse resource environments.

We find that while resource environments strongly influence community composition, individual metabolic performance alone poorly predicts community outcomes. Instead, ecological interactions, including the influence of a small number of keystone species, substantially reshape community structure. Spatial structuring further reorganizes these communities along resource gradients, revealing interaction regimes not captured in liquid culture conditions. Together, these results show how ecological interactions, rather than individual metabolic capabilities alone, play a central role in translating resource availability into community organization.

M23 Molecular mechanisms of viral-bacterial co-infections

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Deepening our knowledge of host-pathogen interactions is essential to reduce the threat posed by emerging infections, antibiotic resistance and the increasing size of endemic areas of tropical diseases. Despite advances in understanding the clinical impact that co-occurring pathogens have on the affected patient, the molecular basis remains understudied, warranting a systems biology approach to decipher co-infections. To disentangle the host-pathogen-pathogen interface, we are employing unbiased, high-throughput screening approaches, followed by mechanistic characterization. Thereby, we highlight the clinical relevance of co-infections in two settings highly prone to pathogen co-occurrence: the intestinal system and the lung.

Firstly, we conducted a systematic, pairwise screening of enteric bacterial and viral pathogens, generating a large-scale dataset encompassing host processes that mediate interactions during co-infections in different host models. Using this, we deciphered two distinct molecular mechanisms of how secondary bacterial infection is influenced by viral pre-infection: 1) Murine-Adenovirus-3 reduces host cell innate immune signaling and causes antagonistic interactions with co-infecting bacteria by dampening ASC-dependent inflammasome activation. 2) A biologically distinct mechanism is triggered by murine-Adenovirus-2, which changes in the regulation of phagocytosis and thereby causes pathogen-specific, increased uptake of *Yersinia* bacteria in macrophages.

Secondly, we conducted a genome-wide CRISPRi screen in *Streptococcus pneumoniae* during single infection and co-infection with Influenza-A-virus. We identified and validated bacterial genes and cellular processes conditionally essential for two principal aspects of pathogenicity. We were further able to disrupt attachment to host cells using small molecule drugs that target these conditionally essential processes at sublethal concentrations. Finally, unbiased metabolomics during single and co-infection allowed us to link nucleoside scavenging and metabolism to *Streptococcus* pathogenicity.

Taken together, this work combines systems biology approaches with mechanistic, hypothesis-driven research to significantly expand and deepen our understanding of viral and bacterial pathogenicity during co-infection.

M24 Early metabolic reprogramming licenses Streptococcus pneumoniae for Influenza-driven superinfection

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Bacterial pneumonia remains a major cause of morbidity and mortality following influenza A virus (IAV) infection. However, the adaptive mechanisms that enable pathogen expansion in the post-viral lung remain poorly defined. Here, we characterize bacterial transcriptional reprogramming *in vivo* and identify alcohol dehydrogenases (AdhA and AdhE*)*, that support NAD^+ regeneration during mixed-acid fermentation, as key determinants of bacterial fitness specifically in the IAV-primed lung. Genetic deletion of these results in a pronounced fitness defect during superinfection but not in primary bacterial pneumonia. Consistently, pharmacological inhibition of alcohol dehydrogenases limits bacterial expansion and dissemination following IAV infection. Mechanistically, we show that IAV infection profoundly remodels the lung environment, inducing hypoxia and increasing the availability of alternative carbon sources, which together impose a metabolic dependency on Adh for bacterial expansion. Our findings place metabolic adaptation as a central driver of pneumococcal outgrowth following viral infection and reveal exploitable vulnerabilities for therapeutic intervention.

S59 A respiratory tract commensal bacterium with direct antiviral action against Influenza A virus

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Lower respiratory tract infections are the fifth leading cause of death worldwide. With the rising threat of antiviral and antibiotic resistance, probiotics represent a promising alternative therapeutic approach. We previously demonstrated that the probiotic application of the natural commensal *Ligilactobacillus murinus* (Lm) protects against *Streptococcus pneumoniae* superinfection in the context of prior Influenza A virus (IAV) infection. Building on this work, we now use human precision-cut lung slices to test whether *Lm* can prevent or clear IAV infection and investigate how this protection works in human lung tissue.

M25 Wartime Wound Infections in Ukraine: Evolution of Antimicrobial Resistance among ESKAPE Pathogens

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Background: Armed conflicts are major drivers of antimicrobial resistance (AMR). Investigating wartime microbiota alterations is crucial for optimizing healthcare strategies and mitigating resistant wound infections.

Objectives: To evaluate wound microbiota changes and AMR distribution among Ukrainian military and civilian patients across different conflict stages, assessing the role of ESKAPE pathogens.

Methods: Wound specimens from infected civilian and military patients (2024–2025) were analyzed. Clinical isolates were identified and tested for antimicrobial susceptibility according to EUCAST guidelines.

Results: In 2024, (non-combat regions), the most frequently isolated pathogens among militaries evacuated after primary treatment were *A. baumannii*, *E. faecalis*, and *K. pneumoniae*, whereas *S. aureus*, *K. pneumoniae*, and *E. faecalis* predominated among civilian patients. Conversely, in 2025 (active regional hostilities), military patients arrived directly from the frontline, shifting their pathogen profile to *S. aureus* and *E. faecalis*, while *S. aureus*, *E. faecalis*, and *S. epidermidis* dominated in civilians. Multidrug-resistant (MDR) *S. aureus* rates in military patients surged from 0% to 40%, aligning with civilians (~40%). Military MDR *E. faecalis* rates remained substantially higher than civilian rates (2024: 100% vs 33%; 2025: 75% vs 29%). MDR *A. baumannii* persisted at critical levels in both cohorts (military: 94% in 2024, 100% in 2025; civilians: 100% in 2024, 50% in 2025).

Conclusions: Active hostilities and altered evacuation logistics shifted military wound pathogens from healthcare-associated *A. baumannii* (2024) to acute-phase *S. aureus* and *E. faecalis* (2025). This transition was marked by a severe escalation of MDR *S. aureus* in soldiers. High military MDR *E. faecalis* and persistent MDR *A. baumannii* underscore the urgent need for adaptive antimicrobial protocols during active conflicts.

MT03*/S63*/P006* Pyoverdines as antimicrobial agents against human opportunistic pathogens

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The emergence and spread of pathogens that are resistant against antibiotics has sparked interest in the development of novel or alternative treatments, including bacteriophages, antimicrobial peptides and new antibiotics. One way to discover new antibacterials is to explore the plethora of secondary metabolites bacteria produce themselves to interact with their environment and other microbes. One group of such potential antimicrobial agents are siderophores. Siderophores are a chemically diverse group of iron-chelating agents secreted by microorganisms to scavenge iron, which is essential for bacterial growth. Certain siderophores are highly species specific and can therefore lock iron away from competitors with incompatible uptake systems.

In our previous work, we showed that environmental *Pseudomonas spp.* produce chemically different variants of pyoverdine – a group of siderophores with strong iron affinity and high specificity. Some of these pyoverdines strongly inhibited the growth of human pathogens *in vitro*, increased survival of wax moth larvae upon infection and showed low potential for resistance development.

In our current work, we examine the clinical potential of pyoverdines as novel antimicrobial agents. We found that pyoverdine 3G07 (our top candidate) effectively inhibited the growth of multiple antibiotic-resistant clinical isolates, including key ESKAPE pathogens like *Acinetobacter baumannii*, *Staphylococcus aureus* and *Klebsiella pneumoniae*. Furthermore, we observed no toxicity following injection of 3G07 into zebrafish larvae, indicating its suitability for *in vivo* applications. In addition, net growth modelling based on time-kill data revealed that pyoverdine acts bacteriostatically. In a next step, we aim to test whether pyoverdine synergies with bactericidal antibiotics or antimicrobial peptides as we hypothesize that a combination therapy may be most potent.

Taken together, our work provides first key insights into the therapeutic potential of pyoverdines and aims to contribute to the global efforts in developing new innovative antimicrobial treatment.

**Student Paper*

S60 Rising spread of Carbapenemase producing enterobacterales in Switzerland over the last five years report from the NARA reference center

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The global spread of carbapenemase producing Enterobacterales (CPE) constitutes a major public health threat and is classified by the WHO as à high-priority concern. The Swiss National Reference Center for Emerging Antibiotic Resistance (NARA), established in 2017, monitors carbapenem-resistant Gram-negative bacteria. This study provides an updated nationwide overview of CPE epidemiology and resistance mechanisms in Switzerland over the past five years.

After deduplication, a total of 2895 CPE isolates were received from all cantons and analyzed. A significant increase over the years was observed: 2021 (n=318), 2022 (n= 454), 2023 (n=596), 2024 (n=752), 2025 (n=775). The samples include specimen from both screening and infected patients. Four main species predominated: *Escherichia coli* n= (1190), *Klebsiella pneumoniae* (n=1112), *Enterobacter cloacae complex* (n=222) and *Citrobacter freundii complex* (n=177).

All major Carbapenemases were detected as well as less prevalent class A IMI and class D OXA-23. A large majority of OXA-48-like producers (n=1404) and NDM producers (n=854) were identified, while KPC, VIM and IMP enzymes were less frequently identified (n = 283, 126 and 3 isolates, respectively). Few isolates co-produced two carbapenemases such as OXA-48-like/NDM, OXA-48-like/VIM, OXA-48-like/KPC, NDM/KPC, NDM/IMP, NDM/VIM and KPC/VIM. High genetic diversity was observed,such as NDM (8 variants detected); OXA-48 (9 variants); KPC (7 variants); VIM (7 variants) and IMP (3 variants). Since 2023, metallo-β-lactamase producers showed susceptibility rates of ~70% to cefiderocol and ~95% to aztreonam-avibactam. In parallel, KPC producers remained highly susceptible (~99%) to imipenem-relebactam, meropenem-vaborbactam, and ceftazidime-avibactam.

In conclusion, this nationwide analysis demonstrates a sustained rise in CPE in Switzerland over the past five years. The diversity of species and resistance mechanisms. The emergence of co-producers and variable treatment susceptibility underscores the urgent need for enhanced surveillance, rapid diagnostics, and coordinated infection control strategies to limit dissemination and protect public health.

S61 Hidden plasmid transmission by temperate phages drives antimicrobial resistance spread in phage-resistant staphylococci

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Coagulase-negative staphylococci (CoNS), particularly*Staphylococcus epidermidis*, are ubiquitous skin commensals that have emerged as primary opportunistic pathogens responsible for intractable, device-associated nosocomial infections. These bacteria frequently harbour small, non-conjugative antimicrobial resistance plasmids, yet how phage-mediated transduction contributes to their spread remains unclear.

Here, we show that the *S. epidermidis* temperate phage PG93, a member of a newly defined species within the genus *Rockfellervirus*, transfers plasmids well beyond its plaque-defined host range. Across a 179-strain panel, PG93 formed plaques on only 15 strains (5 staphylococcal species). In contrast, quantitative transduction assays utilising the 3.8 kb chloramphenicol-resistance plasmid pUR2865-2 across a 114-strain recipient subset demonstrated successful plasmid delivery into 54 distinct strains encompassing 10 staphylococcal species. Crucially, 42 of these transductant-yielding recipients were non-permissive to lytic infection, exhibiting either complete plaque resistance or lysis-from-without. PG93 also mediated plasmid transfer into these non-permissive hosts during both liquid and solid surface cocultures, proving that productive lytic replication is not a prerequisite for successful horizontal gene transfer. Furthermore, genomic analyses revealed that PG93 lysogenizes at a highly conserved chromosomal integration site across divergent staphylococcal species. While prophage integration uniformly conferred superinfection immunity against subsequent PG93 lytic attacks, it impacted secondary plasmid acquisition in a strictly strain-dependent manner, highlighting that phage-mediated immunity does not intrinsically abolish plasmid uptake.

Our findings demonstrate that staphylococcal phages act as highly promiscuous vehicles for horizontal gene transfer, driving antimicrobial resistance dissemination across species barriers and significantly expanding the evolutionary potential of microbial communities.

S62 Development of the Rapid ESBL-Carba Combo PN test, a combined biochemical assay for detection of ESBL and carbapenemase in Enterobacterales

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Background: Rapid detection of extended-spectrum β-lactamases (ESBLs) and carbapenemases in Enterobacterales is essential for early optimization of antimicrobial therapy and infection control. Current rapid phenotypic assays generally detect these resistance mechanisms separately, increasing workload, reagent consumption, price, and turnaround time.

Methods: A chromogenic biochemical assay was developed, the ESBL-Carba Combo PN test, enabling simultaneous detection of ESBL and carbapenemase activity using a single chromogenic substrate (B-Lac; D-Tek) and a low-cost Tris-based lysis buffer. The assay was evaluated using 160 well-characterized clinical Enterobacterales isolates, including ESBL producers, carbapenemase-producing Enterobacterales (Ambler classes A, B, and D), co-producers, and negative controls. Whole genome sequencing (WGS) served as the reference standard. Results were interpreted after 20 minutes and one hour of incubation.

Results: For ESBL detection (n=67), sensitivity was 93.8% after 20 minutes and 97.8% after 1 hour, with 100% specificity at both time points. Carbapenemase detection (n=92) achieved 100% sensitivity and specificity at both readings. The combined assay (n=85) showed 100% sensitivity and specificity for detection of ESBL and/or carbapenemase activity. Carbapenemase class discrimination was excellent for Ambler class A and B enzymes, reaching 100% sensitivity and specificity. For class D carbapenemases, sensitivity reached 91.3% at 20 minutes and 95.5% after 1 hour, with 100% specificity.

Conclusions: The ESBL-Carba Combo PN test provides a rapid, inexpensive (<1 CHF/sample), and simple workflow for simultaneous functional detection of the main β-lactam resistance mechanisms in Enterobacterales. This assay may support earlier targeted antimicrobial therapy and strengthen antimicrobial stewardship in routine clinical microbiology laboratories.

M26 Role of mobile genetic elements in spreading phage resistance in bacterial populations

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Bacteria have evolved a remarkable diversity of defence mechanisms to counter mobile genetic elements (MGEs) such as phages and plasmids. In recent years, hundreds of these systems have been identified, with a striking observation: defence genes are far more densely encoded on MGEs than on bacterial chromosomes. This suggests that MGEs themselves play a central role in spreading immunity within bacterial populations. Yet, the factors driving the selection and dissemination of these defensive MGEs remain poorly understood.

To address this gap, we focus on plasmids, well-known vectors of horizontal gene transfer—particularly for antibiotic resistance genes—but whose role in disseminating defence systems is still unclear. Through large-scale bioinformatic analyses, we demonstrate that plasmids frequently encode defence systems. Moreover, we identify plasmid mobility and host range as key traits positively associated with the presence of these systems. Strikingly, defence systems and antibiotic resistance genes often co-occur, particularly in specific plasmid families.

Our findings highlight plasmids as major vectors for the spread of defence systems, offering new insights into the ecology and evolution of prokaryotic immunity and plasmid dissemination within bacterial communities. These results also carry critical implications for phage therapy, where understanding the spread of resistance is essential for designing effective treatments.

S64 The Integrase encoded in a widespread ICE colocalizes with the type IV secretion system to mediate efficient DNA transfer.

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Mobile genetic elements drive horizontal gene transfer, a mechanism that enables the exchange of genetic material between bacteria. This process is crucial for microbial evolution, as these DNA elements often carry adaptive functions like antibiotic resistance, metabolic pathways, and defense systems. Integrative and Conjugative Elements (ICE) are a class of mobile genetic elements which combine phage-like and plasmid properties. In our work we use the model element ICE*clc* in *Pseudomonas putida* UWC1, which is widespread in Proteobacteria. ICEs rely on an integrase to excise from the host chromosome and re-integrate in the new host. Previous studies have shown that ICE encoded recombinases are characterized by three domains, two DNA binding domains and a catalytic one. In this study, we discovered that ICE*clc* encodes an integrase significantly larger than previously studied ones, containing an extended C-terminus made of an unstructured linker and a globular part. In order to understand its function we produced several integrase variants, where we deleted either the globular part, the extended linker or the entire C-terminus. Our data show that the integrase C-terminus is not required for ICE excision, however its deletion causes a significant decrease in the conjugation efficiency of ICE*clc*. We further used fluorescence microscopy to study integrase's cellular localization both for the wild-type protein and the deletion mutants. Surprisingly, we observed multiple foci forming at the cell membrane for the wild-type, while mutants were misslocalized. In addition, we observed significant colocalization with a core component of the conjugative machinery. Since, this integrase does not possess any transmembrane domain we hypotesize an interaction with one or more membrane proteins encoded by ICE*clc*. Overall, we show that ICE*clc* encodes a unique integrase necessary not only for excision but also for efficient conjugation, likely mediating ICE recruitment to the cell membrane via its specialized C-terminal domain.

S65* Synergy or antagonism? Decoding the rules behind 2,000 phage-antibiotic combinations

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The rapid rise of antibiotic resistance requires alternative strategies to restore antibacterial efficacy and limit resistance evolution. Here, we are exploring at a large scale how combining antibiotics with phages could improve treatment and help fight antimicrobial resistance. To do so, we probed interactions between a broad collection of *Escherichia coli* phages and a diverse panel of antibiotics spanning major modes of action, aiming to identify general principles governing synergy, antagonism, and resistance emergence.

The resulting dataset captures infection dynamics and final treatment outcomes across more than two thousand pairs. Most phage-antibiotic combinations were neutral when considered globally, yet nearly all exhibited strong synergistic or antagonistic effects at specific concentrations. Early infection dynamics were frequently shaped by antagonistic combinations, particularly with antibiotics targeting transcription and protein synthesis. In contrast, antibiotics affecting cell wall integrity and DNA metabolism showed a higher propensity for synergy with phages.

At later time points, synergistic outcomes predominated, revealing effects that are not apparent from early growth observation alone. A major driver of late synergy was the prevention of resistance emergence, most often phage resistance suppressed by sublethal antibiotic exposure. Importantly, resistance-promoting antagonisms were comparatively rare. The outcomes of combinations were strongly antibiotic-dependent and could not be explained by phage taxonomy, underscoring substantial biological heterogeneity.

Together, these results provide a framework for the development of phage-antibiotic combination therapies and aim to guide their translation toward more predictive and robust antimicrobial interventions.

**Student Paper*

S66* Unraveling the functional dark matter in phage genomes with HIDEN-SEQ

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Phages and their bacterial hosts are locked in a continuous battle that drives coexistence and evolution. Molecular innovations arising from this conflict have laid the foundation of modern molecular biology and continue to drive advances in biotechnology. More recently, the global rise of multidrug-resistant bacterial pathogens has renewed interest in phages as therapeutic agents. However, the vast amount of genomic "dark matter" composed of genes of unknown function in phage genomes remains a major obstacle for the molecular understanding of phage-host interactions. Here we present HIDEN-SEQ, a transposon-insertion sequencing method for phages that allows systematic disruption of phage genes by combining transposon mutagenesis and CRISPR-Cas13a/anti-CRISPR selection. In diverse phages, we demonstrate high-resolution mapping of phage gene essentiality and provide experimental access to large fractions of genes with unknown function. Across multiple bacterial hosts and growth conditions, HIDEN-SEQ uncovers extensive sets of conditionally essential phage genes, including previously uncharacterized anti-defense factors that we could link to specific antiviral systems of the respective hosts. Together, these results expand the known repertoire of phage counter-defenses and reveal that successful infection relies on diverse and largely uncharacterized set of accessory phage genes. More broadly, HIDEN-SEQ provides a scalable functional genomics platform for phages that will accelerate discoveries in phage biology by uncovering functions of viral dark matter with direct relevance for microbial ecology, biotechnology and phage therapy.

**Student Paper*

S67 Functional Characterization of Borg Proteins via Heterologous Expression in Methanosarcina acetivorans C2A

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Borgs are giant linear extrachromosomal DNA elements associated with methane-oxidizing *Candidatus* Methanoperedens archaea. They lack cultivated hosts and have few protein functional annotations, precluding their classification as plasmids, viruses, or other known entities. Strikingly, *in silico* structure prediction revealed putative capsid proteins, alongside megabase-scale linear genomes with inverted terminal repeats and broad metabolic gene repertoire, raising the possibility that Borgs represent a new class of giant archaeal virus. However, this hypothesis remains untested, and viruses infecting methane-oxidizing archaea have never been isolated or experimentally characterized. Elucidating what Borgs are and what impact they have on methane-oxidation is substantially hindered by the inability to isolate *Methanoperedens* as a pure culture. To address this, the present study aims to functionally characterize Borg-encoded genes through heterologous expression in *Methanosarcina acetivorans* C2A, the phylogenetically closest cultivable relative of the predicted Borg host. Six genes were selected based on their putative roles in distinct cellular processes: the S-layer protein (Green Borg) involved in surface remodeling, the *mcr* cluster (Lilac Borg), and carbon monoxide dehydrogenase (CODH, Black Borg) involved in core energy metabolism, Hfq-like RNA chaperone (Black Borg) potentially involved in post-transcriptional regulation, and a multiheme cytochrome nanowire cluster (Brown Borg) potentially mediating extracellular electron transfer. The pC2A-based shuttle vector pEM1225 was optimized for tunable expression. The resulting *M. acetivorans* strains will be characterized for growth physiology, metabolism and morphology via electron and fluorescence microscopy. Target proteins will be purified for biochemical characterization of enzyme activities, including methyl-coenzyme M reductase activity (MCR), CO oxidation (CODH), heme-dependent electron transfer kinetics (nanowire), and RNA binding-affinity (Hfq-like protein). Together, this work aims to establish the first experimental platform for Borg protein characterization, providing first insights into the nature and functional significance of these enigmatic elements in archaeal methane cycling.

M27 Circular viroid-like RNA infectious elements infect fungi altering their biological properties

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Infectious circular viroid-like RNAs in fungi represent an emerging paradigm at the interface between RNA genetics, host immunity, and ecological fitness. In the last years, their presence has been highlighted by high-throughput meta-transcriptomic approaches, but the fundamental rules governing how these elements enter fungal cells, establish replication, and spread between hosts remain insufficiently defined. Moreover, while infection is expected to reshape fungal physiology, the specific biological consequences for fungal growth, immune responses (e.g., RNA interference), and—where relevant—virulence or biocontrol traits are still largely unclear. Here, we provide experimental evidence from two complementary fungal systems. First, in the plant pathogen *Fusarium graminearum*, we established the first reverse-genetics system for an ambivirus, Fusarium graminearum ambivirus 1 (FgAV1), using infectious cDNA clones. Successful replication and infection require a head-to-tail dimeric genome construct, and unexpectedly, a reverse-oriented dimer is also replication-competent. Targeted mutagenesis demonstrates that all ambivirus core elements are non-redundant: ORF-A catalytic core and ORF-B translation are each indispensable for replication and horizontal transmission, while embedded ribozyme motifs are absolutely essential, as their inactivation through point mutations abolishes accumulation of authentic viral RNAs in both polarities. FgAV1 infection triggers a canonical fungal RNA interference (RNAi) response, with strong accumulation of viral small RNAs peaking at ~21 nt, consistent with antiviral targeting of replicating circular RNA species. Phenotypically, infection suppresses fungal growth kinetics and significantly reduces virulence on wheat. In the biocontrol fungus *Trichoderma spirale*, we characterized the transmissible viroid-like RNA TsvlRNA1. Bioinformatic and molecular analyses identify TsvlRNA1 as a circular RNA harboring a hammerhead ribozyme whose catalytic activity is required for autonomous replication. Infectious clones reveal that replication initiates from a dimeric transgene via self-processing, and, surprisingly, horizontal transmission occurs between *Trichoderma* species through hyphal routes, while vertical transmission via conidia was demonstrated only in *T. spirale*. RNAi signatures are evident as 21–23 nt viroid-derived small RNAs, and TsvlRNA1 modulates antagonistic capacity against *Rhizoctonia solani* in a strain/genotype-dependent manner, with effects ranging from enhanced to reduced biocontrol. Together, these studies establish that circular viroid-like infectious elements in fungi are replication-competent, ribozyme-dependent (and ambivirus protein-factor-dependent), trigger host RNAi, and can reshape host biological properties through horizontal spread, including interspecies transmission for the genus *Trichoderma*, with potential implications for fungal ecology and biocontrol deployment.

MT07/S68/P014 Mapping the temperate phage landscape and integration hotspots across Staphylococcaceae

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Temperate bacteriophages are ubiquitous in staphylococci, yet their diversity and integration patterns remain under-characterized beyond *Staphylococcus aureus*. This study establishes a genomic–experimental framework to map the prophage landscape, integration hotspots, and mobilization potential within non-aureus members of the Staphylococcaceae family.

Using an in-silico pipeline to analyze 114 bacterial genomes (41 coagulase-negative-staphylococci), we identified intact prophages in 68% of strains, and 45% enclosed phage-like elements (incomplete or questionable phages). Of these lysogens, 47, 42 and 14% carried one, two or three intact prophages, respectively. Notably, 37% and 9% of lysogens carried antimicrobial resistance (AMR) and virulence genes, respectively, while 10% harbored AMR-carrying plasmids.

Experimental induction yielded novel phages from *S. epidermidis* (ILRIcamel_95, ILRIcamel_96) and *Mammaliicoccus sciuri* (EG97). These phages integrate into specific sites: tRNA-Ser-Asn, the FAD-coding gene (*merA*), or a VOC-family protein gene, respectively. These attachment sites were present in 72%, 6% and 1% of our Staphylococcaceae lysogenic collection, respectively, with 16%, 10%, 63% truncated by integrase-leading elements, identifying those loci as integration hotspots for phage-related elements. While ILRIcamel_95 and ILRIcamel_96 exhibit *S. epidermidis*-specific infection range in-vitro, EG97 shows sporadic cross-species activity. Furthermore, in-silico prediction of integration sites was performed on *S. simulans* lysogens (n=11). These phages were predicted to integrate into *sufB*, RNA chaperone or tRNA-Ser-Glu. These attachment sites were present in 61%, 10% and 35% of our Staphylococcaceae lysogenic collection, with 20%, 64%, 40% truncated by integrase-leading elements. These prophages could not be recovered on the tested host panel, and alternative in vitro approaches are underway to assess their ability to excise and induce lysis independently of detectable propagation in indicator strains.

This integrated genomic–experimental framework provides a scalable strategy to uncover hidden temperate phage diversity and integration hotspots within Staphylococcaceae, providing insights into bacterial evolution and new opportunities to recover and study experimentally elusive functional temperate phages.

S69* Monitoring of single-cell bacterial lysis by phages within integrated optical traps

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Regarding their high bacterial strain specificity, rapid and accurate selection of therapeutic bacteriophages is critical in clinical protocols of personalized phage therapy. In this work, we report the use of photonic crystal cavities as on-chip optical nanotweezers for phage susceptibility testing (PST) at the single-bacterium level. On a silicon photonic chip, resonant photonic crystal cavities enable the trapping of a single *Escherichia coli* cell while simultaneously sensing stress-induced modifications of the bacterium. This detection is achieved by monitoring the transmitted optical power through the photonic chip, which carries information about the properties of the trapped cell. *E. coli* cells were exposed to T4 *Myoviridae* and T1 *Tunaviridae* phages prior to injection into the trapping device. We report the direct observation of bacterial lysis by phages inside the optical cavity. Morphological changes induced by phage infection are detected through variations in the transmitted optical signal and corroborated by real-time microscope imaging. The lytic event results in a sudden reduction of the refractive index contrast between the bacterium and the surrounding medium, recorded as an abrupt drop in transmission. Lysis detection occurs within 40 ± 5 minutes, significantly faster than conventional culture-based phagograms, paving the way for ultrafast single-cell PST. Interestingly, we observe different drops in transmission depending on the phages used to lyse the bacterial cell, correlated with the estimated burst size of the phages. We also correlate the facility at which an *E. coli* bacterium is trapped with the O-antigen it carries. This demonstrates that our setup can obtain information even at the proteic level. This method offers a rapid and precise approach for ultrafast PST at the single-bacterium level, addressing a major bottleneck in phage therapy implementation and paving the way for real-time bacteriophage selection in clinical settings

**Student Paper*

S70 Novel Mycobacteriophages as a Promising Adjunct in the Fight Against Drug-Resistant TB and NTM Infections.

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Mycobacterium smegmatis was used as a non-pathogenic model host for bacteriophage isolation. Environmental sampling and soil-extract processing enabled the recovery of sixteen novel mycobacteriophages. Genomes were assembled from Illumina sequencing reads, and phylogenetic relationships were inferred using VICTOR. Lytic activity on M. smegmatis was evaluated using diluted drop tests (DDT) and turbidity assays. Phage-resistant M. smegmatis mutants were analyzed to investigate potential resistance mechanisms. Host range profiles were determined on clinical isolates from the *Mycobacterium tuberculosis* (MTB) complex and *Mycobacterium abscessus*. In parallel, nanomotion-based phage susceptibility testing (PST) was explored as an ultra-rapid phenotypic approach to assess phage activity against mycobacteria.

Phylogenetic analysis revealed that seven newly isolated phages belong to cluster A, the most represented cluster of mycobacteriophages. Phage 8008 was assigned to cluster K, while phages 8009 and 8011 grouped within cluster C. Fred313, Muddy, and Fionnbharth from the University of Pittsburgh were included as reference phages. All sixteen phages demonstrated strong lytic activity against *M. smegmatis*. In addition, the attenuated MTB strain H37Ra was evaluated as a preliminary tuberculosis model, and 11 out of 16 tested phages showed lytic activity against this reference strain. Preliminary experiments also suggested potential synergistic effects between phages and standard anti-TB or anti-NTM antibiotics. These findings support ongoing evaluation on a broader panel of MDR/XDR clinical isolates.

This work highlights the therapeutic potential of bacteriophages against mycobacterial infections in the context of increasing antimicrobial resistance. The identification of multiple phages active against MTB H37Ra further supports their potential application against tuberculosis. Future work will focus on direct phage isolation using MTB H37Ra instead of *M. smegmatis* and on evaluating phage activity against Lausanne University Hospital clinical MTB isolates. In parallel, ultra-rapid nanomotion-based PST is being developed to rapidly assess phage efficacy against drug-resistant MTB isolates.

S71* Diverse wastewater staphylococcal phages reveal a novel family, intron-spliced terminase genes, and phylogeny-taxonomy associated infection patterns

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Bacteriophages are increasingly explored as alternatives to antibiotics against opportunistic pathogens such as *Staphylococcus epidermidis*. However, the genetic diversity, host specificity, and functional features of staphylococcal phages remain insufficiently characterized, limiting the rational selection of phages for therapeutic applications. Here, we present a comprehensive characterization of 86 staphylococcal bacteriophages isolated from a single wastewater treatment plant in Zürich. Whole-genome sequencing and comparative genomics revealed substantial diversity, including members of the *Herelleviridae* (65 myovirus-like phages), *Rockefellervirus* (4 siphovirus-like phages), a *Picoviridae* phage (podovirus-like), and a novel family, *Edelweissviridae* (16 siphovirus-like phages with long non-contractile tails). Introns truncating the large terminase subunit gene (*terL*) were identified in 60% of genomes across *Herelleviridae* and *Edelweissviridae*. Functional analyses demonstrated intron splicing, where excision of a homing endonuclease-encoding intron restores a full-length *terL* protein. Host range assays against 40 clinically relevant *S. epidermidis* isolates revealed higher infectivity toward veterinary strains, particularly for phages of the genus *Sepunavirus* and the new subfamily *Aletschvirinae*. Additionally, wall teichoic acid glycosylation influenced phage susceptibility in a family-dependent manner, consistent with distinct predicted receptor-binding proteins. Altogether, our findings expand the known diversity of staphylococcal bacteriophages and provide new insights into their genomic features, host interactions, and therapeutic potential.

**Student Paper*

M28 Gut microbiota drug biotransformation as a tool to unravel the mechanisms of metabolic microbiota-host interactions

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The variability of the human gut microbiome—the collective community of microorganisms inhabiting the intestine—far exceeds that of the human genome and has been linked to numerous aspects of human health and disease. Although differences in microbiome composition are frequently associated with altered metabolism, our understanding of metabolic interactions between the microbiota and the host remains largely descriptive. This is due to several key limitations. First, most sequencing-based human microbiome studies rely on correlations between microbiome composition and host phenotypes, are constrained by incomplete microbial genome annotations, and are not designed to identify functional traits that emerge at the community level. Second, many metabolites can originate from both microbial and human metabolism, making it conceptually and experimentally challenging to disentangle the respective contributions of the microbiota and the host.

To overcome these limitations, our laboratory employs systematic bottom-up approaches to mechanistically investigate metabolic microbiota–host interactions by harnessing the capacity of gut microorganisms to biotransform, or chemically modify, drug molecules. Because of their broad chemical diversity and exogenous origin, therapeutic drugs provide particularly well-suited molecular probes for studying microbiota–host interactions under controlled in vitro and in vivo conditions. We combine high-throughput microbial cultivation, genetic screening, metabolomics, genomic analyses, gnotobiotic mouse models, and computational modelling to connect interpersonal variation in microbiome composition with differences in the metabolic functions of individual gut microbial communities and, ultimately, with molecular host phenotypes.

The resulting mechanistic insights and resources are essential for defining fundamental principles of the microbiota–host relationship. At the same time, our findings have direct medical relevance by revealing actionable microbiome-based mechanisms that contribute to interpersonal variability in drug response, which remains a major challenge in clinical practice.

S72 From correlations to interactions: predicting and engineering butyrate production in microbial communities

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Oral bacterial overgrowth in the small intestine is associated with dysbiosis, nutrient malabsorption, inflammation, and growth defects in undernourished children. These microbiome alterations include reduced abundance of key Clostridia butyrate producers and lower intestinal butyrate levels. However, current corrective nutritional interventions often fail to restore microbial balance, highlighting the need for rationally designed probiotic communities that could enhance beneficial metabolite production. Here, we aimed to develop a predictive framework to engineer synthetic microbial communities with high butyrate output by integrating ecological co-variation, genome-scale metabolic modeling, and experimental interaction assays.

Using species-level abundance profiles from approximately 90,000 gut metagenomes, we first identified positively and negatively associated bacterial taxa using SparCC correlation analysis. From these results, we selected a defined set of 21 strains, including butyrate-producing bacteria and four *Streptococcus salivarius* strains, to explore potential cooperative-like and competitive-like interactions. In parallel, whole-genome sequences are used to reconstruct genome-scale metabolic models with CarveMe, and pairwise competition and complementarity indices were computed with PhyloMint to estimate metabolic niche overlap and syntrophic potential. These predictions were then experimentally tested and compared using spent-medium assays, where growth responses were quantified to determine inhibitory or cross-feeding effects between strain pairs.

Preliminary analyses suggest that correlation trends from metagenomics analyses are partially consistent with metabolic model prediciton, although associations remain modest. Early spent-medium assays also indicate biologically relevant interactions, including negative interactions between *Coprococcus comes* and *S. salivarius*.

In the next steps, optimal and non-optimal five-strain synthetic communities will be assembled, ranked by butyrate production using GC-MS, and compared against computational and experimental predictions.

By comparing co-variation analyses, metabolic models, and in vitro assays, this framework aims to clarify how different computational and experimental approaches could be combined to guide a more efficient design of specific metabolite-producing communities for future testing in stool-derived and in vivo models.

S73 Hitchin' a ride: Studying how bacteriophage-mediated transduction shapes bacterial evolution in complex intestinal communities

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Horizontal gene transfer is a major driver of bacterial evolution in the gut microbiome, yet the role of bacteriophage-mediated transduction, the transfer of bacterial DNA by a phage to a new host, remains poorly understood. Key parameters such as the frequency, host range, and genetic impact of transduction events in complex intestinal ecosystems are largely unknown. Here, we utilize transductomics—a cutting-edge sequencing-based approach that detects DNA mobilized by phages and virus-like particles (VLPs)—to characterize transduction-driven genetic exchange in murine gut communities under homeostatic and disease-associated conditions. We specifically investigated the impact of newly introduced phages and VLPs, reflecting therapeutic intervention-mediated microbiota perturbations.

We assessed the transduction-potential of therapeutic phages targeting the inflammatory bowel disease-associated pathobiont *Enterococcus faecalis*. Among tested lytic phages, two exhibited signatures of generalized transduction, while others promoted prophage induction, suggesting diverse mechanisms of DNA mobilization. Mobilized DNA included genes encoding antibiotic resistance, stress, and metabolic adaptation, highlighting the potential for phage-mediated dissemination of fitness-enhancing traits.

To probe transduction in a polymicrobial community context, we introduced gut-derived VLPs into fecal outgrowth communities (FOCs). *In vitro* propagation of FOCs reduced bacterial diversity and enriched facultative anaerobes, recapitulating features of dysbiosis. While the input *in vivo* communities showed higher frequencies of unique transduction events, *in vitro* growth amplified the abundance of specific transduction events overall. VLP exposure had minimal impact on the abundance of specialized, prophage-associated transduction or other distinctive patterns indicative of generalized transduction; however, the associated mobilized DNA contained diverse functional determinants including whole CRISPR-arrays, toxins, and multidrug-resistance genes.

Together, these findings demonstrate that phage-mediated transduction actively reshapes microbial genomes and genetic networks in the gut. This work provides a framework to assess the evolutionary consequences of phage therapy and VLP-containing fecal transplants and highlights the need to consider transduction in the design of microbiome-targeted therapies.

S74* Exploring Phyllosphere Bacterial Community Dynamics in a Continuous Culture System

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Rational engineering of microbial communities remains an outstanding challenge, limited by our incomplete understanding of the metabolic interactions that govern community assembly, stability, and function. This project aims to develop a metabolic framework for predicting and manipulating synthetic microbial communities, using bacteria isolated from the *Arabidopsis thaliana* phyllosphere as a tractable model system.

Using a platform of 64 independent mini-bioreactors, we inoculate large, diverse SynComs (>100 strains) under chemostat-like conditions that more faithfully recapitulate phyllosphere dynamics than batch systems while still allowing for precise control over these communities' metabolic environment. We characterize the growth of these SynComs with metagenomic and metabolomic methods, allowing us to identify the metabolic factors driving community dynamics and to benchmark these dynamics against *in planta* growth. We then aim to systematically test community response to environmental perturbations including nutrient stress, temperature shifts, and pathogen invasion to investigate community stability and define the tipping points that drive transitions to alternative stable states.

Characterizing the assembly and dynamics of these communities provides necessary context for their eventual manipulation. By integrating global information about community structure with strain-specific metabolic data, we ultimately aim to develop a framework for the targeted engineering of complex microbial communities.

**Student Paper*

S75*/P156* Individual bacterial taxa drive colonisation resistance to methicillin-resistant Staphylococcus aureus in human nasal microbiome samples

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Nasal carriage of the pathobiont *Staphylococcus aureus* represents a major risk factor for infections, yet the ecological factors that determine *S. aureus* colonisation success within healthy nasal microbiomes remain poorly understood. Our aim is to identify and characterise *S. aureus*-antagonistic interactions in the nasal microbiome and quantify the contribution of pairwise interactions to community-level outcomes.

Using an in vitro microcosm approach to cultivate nasal microbiome samples from healthy donors, we find that samples differ markedly in their ability to inhibit growth of incoming methicillin-resistant *S. aureus* (MRSA), and that these differences are associated with community composition. By assembling nasal model communities reflecting our samples and applying targeted drop-in and drop-out designs, we show that individual taxa can drive colonisation resistance to MRSA. Together, our results establish causal connections between microbiome composition and clinically relevant community-level outcomes, thereby bridging the gap between associative, sequencing-based microbiome studies and mechanistic work on pairwise bacterial interactions.

Ongoing work investigates the molecular mechanisms underlying these inhibitory interactions, with the aim of informing both our understanding of nasal microbiome ecology and the development of novel therapeutic strategies.

**Student Paper*

PF01*/P001* Development and implementation of an evolutionary algorithm for engineering microbiomes via environmental modulation

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The availability and composition of environmental resources – particularly carbon – has a profound effect on the structure and function of microbial communities. This connection presents a promising avenue for engineering communities with specific functions through targeted environmental manipulations.

However, as even slight alterations in resource availability and composition can yield drastically different ecological outcomes, such an approach to community engineering would require testing community responses in a combinatorially intractable space of environmental compositions. To address this challenge, here we develop and experimentally implement a search algorithm to identify combinations of carbon sources that produce bacterial communities with desired properties.

This algorithm navigates combinatorial spaces of carbon sources through an iterative process inspired by evolutionary theory. Communities are assembled in candidate environments, their resulting functions are compared to a desired target, and new candidate environments are generated by recombining carbon sources from the top scoring environments. Thus, with every cycle, the search space is reduced until carbon source compositions that induce the desired function are identified. Utilizing simulated growth data, we have confirmed the efficacy of this approach, assessed the navigability of the search space, and identified optimal parameters for execution *in vitro*. We now begin *in vitro* implementation, selecting for precise abundances of individual community members in a soil-derived microbial community.

Beyond assessing the capability of evolutionary optimization to induce desired properties in microbial consortia, this work will generate a large-scale mapping of environmental compositions to community function, enabling opportunities to examine metabolic mechanisms underlying community assembly patterns.

**Student Paper*

PF02/ P003 Impact of OprF porin deletion on the efficacy of imipenem therapy in a murine sepsis model caused by Pseudomonas aeruginosa producing NDM-1 carbapenemase

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Background: Carbapenem-resistance in *Pseudomonas aeruginosa* infection is driven by multiple mechanisms, including carbapenemase production (particularly NDM-1) and altered outer membrane permeability (OprD defect). OprF, the major outer membrane porin, plays a central role in membrane integrity and possibly virulence. This study evaluated whether OprF disruption could restore carbapenem efficacy in NDM-1-producing strains during experimental sepsis.

Methods: Isogenic PAO1 and PA14 *oprF*-deficient mutants, including NDM-1-producing derivatives, were analyzed *in vitro* by motility and growth competition experiments, and a murine peritoneal sepsis model. Groups of CD-1 female mice were infected intraperitoneally (ip) with the minimum lethal dose (MLDs) previously determined for each strain. Treatment lasted 24 hours and started two hours post-inoculation and animals were randomly grouped into: i) controls, infected, untreated; ii) suboptimal treatment of imipenem (30mg/kg/q4h/ip/24h). Survival was monitored over 24 hours. Bacterial loads in spleen (log_{10}CFU/g) and blood (log_{10} CFU/mL), and bloodstream-infection (BSI) rates were assessed. Comparisons were performed using Mann-Whitney U and Fisher's exact tests.

Results: Although *oprF* disruption reduced motility and fitness, *in vivo* virulence remained preserved, with similar MLDs among strains. In mice infected with NDM-1-producing PAO1 strain, imipenem treatment failed to improve survival or BSI rates despite slight reductions in spleen and blood bacterial concentration. In contrast, disruption of *oprF* in the NDM-1-producing strain is associated with increased expression of OprD, restored imipenem efficacy, resulting in significant reductions in bacterial concentrations in spleen and blood, complete clearance of BSI, and significantly improved survival, reaching outcomes comparable to those observed with the imipenem-susceptible PAO1 strain.

Conclusions:

1. Loss of OprF porin restores imipenem efficacy against NDM-1-producing *P. aeruginosa*, improving bacterial clearance and survival.
2. Outer membrane permeability alterations may increase susceptibility to antibiotics and enhance therapeutic responses in MBL-producing *P. aeruginosa*.
3. These results highlight the interaction of two specific outer membrane porins, OprF and OprD, in carbapenem resistance.

PF03∗/P005∗ Host specificity depends on spatial scale: insights from micromammal gut microbiota

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Gut microbial communities (microbiota) and their animal hosts are deeply interconnected. Hosts play a role in shaping microbiota composition, and in turn, gut microbiota contribute to host fitness. This tight interplay is reflected in patterns of host specificity, whereby microbiota composition clusters by host species: composition is more similar among individuals of the same species than among individuals from different species.

Quantifying host specificity precisely is therefore a powerful tool for assessing how tightly interdependent a given host species and its gut microbiota are. Surprisingly, the degree of host specificity measured varies substantially across studies, even within the same groups of animals (e.g. rodents). In this study, we hypothesize that this discrepancy originates from the spatial scale at which specificity is measured. Studies focusing on small spatial scales may underestimate within-species variability because sampled individuals are more likely to be closely related. This, in turn, inflates the measured degree of host specificity, making it appear higher than in studies conducted at broader spatial scales.

Using micromammals as a model, we measure the effect of spatial scale on host specificity by sampling feces across multiple wild host species at both local and regional levels. Our preliminary results show that sampling design is an important determinant of the degree of host specificity observed. We confirm that sampling at small spatial scales does indeed inflate host specificity measures. In addition, we test the extent to which host diet (assessed by plant metabarcoding) mediates the host specificity effect we observe.

This study sheds light on potential causes of discrepant observations of host specificity. It therefore helps bridge the gap between host specificity studies and contributes to our understanding of host–microbiota interactions.

∗Student Paper

PF04*/P007* Establishing genetic modification in primary-like human airway epithelial cultures

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Influenza A viruses cause a respiratory disease in humans that poses a major burden on human health. With the long-term goal of developing antiviral strategies, influenza virus-host interactions are mostly studied on a cellular and molecular level using cancer-derived cell lines. While these cell lines are well suitable for genetic modifications and thus allow for mechanistic studies, they do not recapitulate the complexity of the human airway epithelium. This limitation can be overcome by using primary human airway cultures, a physiologically relevant model of airway epithelia. However, genetic modification of these cultures has thus far been difficult. In this study, we aim to establish a protocol to genetically modify human airway epithelial cultures to assess and understand virus-host interactions, using the immortalized primary-like cell line BCi-NS1.1. We show that these cells can differentiate into the different cell types that are found in human airway epithelia, that they can be infected with several influenza A virus strains and are also amenable to genetic modification via lentiviral transduction. Ongoing work aims at generating and characterizing airway cultures deficient of the type I interferon receptor to validate our protocol. Our approach should thereby enable the study of influenza A virus-host interactions in a physiologically relevant model.

**Student Paper*

PF05∗/P009∗ Chimerophore antibiotics: combining host defense peptides for multimodal antimicrobial activity

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Ribosomally synthesized host defense peptides (HDPs) represent a promising yet clinically underexploited reservoir of antimicrobial substances. Non-membranolytic HDPs offer high specificity and low host-cell toxicity by targeting intracellular machinery rather than disrupting bacterial membranes. However, their translation is often hindered by limited potency and low activity in physiological fluids. To overcome these barriers, we employed a semi-rational peptide chimerization strategy, fusing the pharmacophores of non-lytic HDPs into diverse chimeric architectures to trigger intramolecular synergy and enforce multimodal MOAs within a single molecule.

While previous efforts to explore chimeric HDPs have been constrained by the high costs and low throughput of chemical synthesis, here we circumvent these bottlenecks by ribosomally synthesizing of a combinatorial library of 99,235 DNA-encoded *chimerophores* (chimeric pharmacophores). Using the high-throughput self-screening platform (Me^X) in *Escherichia coli* for intracellular activity assessment, we identified over 30,300 active variants, representing a significant expansion of the known functional space of chimeric HDPs. We isolated potentiated broad-spectrum candidates that remain active in serum, maintain low host-cell toxicity, use multiple membrane-transport modalities and simultaneously act on multiple intracellular targets. By integrating these orthogonal pathways, our lead candidate cp9 successfully suppressed the evolutionary escape of *Pseudomonas aeruginosa*. This work establishes a scalable, high-throughput platform for the systematic design of next-generation antibiotics capable of eradicating multidrug-resistant pathogens through enforced multimodality.

∗Student Paper

PF06∗/P011∗ Decoding Type IV Pilus Recognition of Pseudomonas aeruginosa Phages

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Pseudomonas aeruginosa is a leading pathogen responsible for healthcare-associated infections and presents a significant clinical challenge due to its widespread multidrug resistance. Bacteriophages, viruses that selectively infect and kill bacteria, offer a complementary approach to traditional antibiotic therapies. Type IV pili (T4P) are important virulence factors of *P. aeruginosa* that mediate motility, adhesion, biofilm formation, and host colonization. They also serve as receptors for a diverse range of bacteriophages, making them important drivers of phage-host specificity and co-evolution. Understanding the molecular basis of pilus-mediated viral recognition is therefore essential for the development and selection of effective phages for therapy.

In this study, we isolated and characterized a collection of novel T4P-targeting phages infecting *P. aeruginosa*. Phage-pilus interactions were analysed using integrated genomic analyses, phenotypic assays, structural biology, and electron microscopy. Our results identified distinct receptor-binding modules shared across genetically diverse phages, suggesting that T4P recognition relies on a limited set of conserved yet adaptable molecular strategies. To investigate their role in receptor recognition, we first analysed the host range of phages encoding homologous but distinct pilus-binding modules across a diverse collection of clinical *P. aeruginosa* isolates expressing different T4P variants. We subsequently exchanged homologous receptor-binding modules between phages with divergent host ranges using genetic engineering. Our findings proved the role of these modules in pilus binding and directly linked them to phage host range.

Together, these findings do thus not only broaden our knowledge of phage-pilus interactions but also provide a foundation for the rational application of phage therapy to treat multidrug-resistant *P. aeruginosa* strains.

∗Student Paper

PF07/P013 Integrated metagenomic and metabolomic features Predict opportunistic pathogen expansion in allogeneic hematopoietic cell transplant recipients

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Allogeneic hematopoietic cell transplantation (allo-HCT) is a potentially curative therapy for blood cancers. However, receipt of conditioning chemotherapy and prophylactic antibiotics during allo-HCT disrupts the gut microbiota, whose composition is associated with survival, infection risk, and transplant-related complications. Opportunistic bacterial and fungal pathogens can expand in the gut, leading to invasive infections, yet the ecological and pharmacological drivers of these expansions remain poorly understood.

We analyzed 1,279 samples from 156 allo-HCT patients, integrating bacterial composition, pathways, and gene abundances (shotgun sequencing), fungal composition (ITS1 sequencing), fecal metabolomics, and medication records. We developed a dual framework to bridge mechanistic discovery and patient risk stratification: (1) to identify pre-expansion features associated with the subsequent expansion of five key pathogens (*Enterococcus, Klebsiella, Escherichia coli, Candida albicans,* and *Candida parapsilosis*) and (2) to build robust predictive models for these events.

Using generalized linear models and feature selection approaches, we identified 2-62 bacterial pathways, genes, metabolites, and medications associated with subsequent expansion for each of the studied species. In parallel, machine-learning models trained on pre-expansion samples predicted pathogen-specific expansion, explaining 39% and 63% of the variance of *C. albicans* and *Enterococcus* relative abundance, respectively. A classification model predicted *Klebsiella* expansion with 98.1% specificity and 100% sensitivity. Notably, a methyltransferase gene emerged as a strong predictor of *Klebsiella* expansion, and alone outperformed *Klebsiella* pre-expansion abundance.

To ensure robustness, we expanded our dataset with an additional 702 samples for independent *in silico* validation. This comprehensive pipeline serves as a foundation for future *in vitro* and *in vivo* experiments to functionally validate the identified variables and establish causal links between specific microbial features and pathogen expansion.

Overall, our preliminary results show that future opportunistic pathogen expansion after allo-HCT is partially encoded in pre-expansion microbial pathways, fecal metabolites, and medication history, enabling both accurate clinical prediction and biological insight.

MT01*/P002 Spatiotemporal interactions between biocontrol bacteria and pathogenic oomycete

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Biocontrol bacteria employ a sophisticated, multi-layered strategy to suppress pathogenic fungi. However, relying solely on antifungal metabolites rarely achieves complete hyphal inhibition and often allows hyphae to breach weak points in antagonistic activity, leading to wider infection. Moreover, certain antifungal metabolites may pose potential risks to humans, animals, and the environment, which is why the EFSA (European Food Safety Authority) considers Pseudomonas fluorescens to be an effective biocontrol agent that is "not presumed safe by default."

When genes associated with the production of these antifungal metabolites are knocked out, the reduced growth burden enhances the response rate to chemical signals released by pathogens and improves motility. The bacteria actively track and colonize hyphal surfaces using flagella and pili. This targeted bacterial motility often outpaces the spread of diffusible metabolites, thereby effectively restricting hyphal expansion.

**Student Paper*

MT02∗/P004 exploring the ecological and evolutionary roles of persisting bacteriophages in traditional food fermentation

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Traditional fermented foods are simple microbial ecosystems shaped by long-term human influence. Multiple microorganisms interact, adapt, and evolve under dynamic environmental conditions. While the diversity and functional roles of bacteria in these systems are well understood and form the basis of current knowledge on fermentation processes, the ecological and evolutionary contributions of bacteriophages (phages) remain largely underexplored. Recent research shows that phages and their bacterial hosts can stably coexist in fermented foods without negatively affecting fermentation outcomes, challenging the traditional perception of phages as purely disruptive agents. This project aims to advance a comprehensive understanding of the diversity, ecological roles, and evolutionary dynamics of phages in traditional food fermentation systems. In my PhD, I will analyze large-scale genomic and metagenomic datasets to construct an extensive catalogue of phages persisting in these environments. These data will underpin the development of predictive models of phage–host interactions and their ecological functions within microbial communities, while also guiding targeted phage isolation strategies to establish a collection of persistent phages for experimental characterization. By integrating computational and experimental approaches, this work seeks to uncover how phages influence microbial community structure and ecosystem functioning. Ultimately, it aims to reshape current perspectives on phages in food fermentation and reveal beneficial phenotypic impacts that could be leveraged to improve fermentation processes and product quality.

∗Student Paper

MT03*/S63*/P006* Pyoverdines as antimicrobial agents against human opportunistic pathogens

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The emergence and spread of pathogens that are resistant against antibiotics has sparked interest in the development of novel or alternative treatments, including bacteriophages, antimicrobial peptides and new antibiotics. One way to discover new antibacterials is to explore the plethora of secondary metabolites bacteria produce themselves to interact with their environment and other microbes. One group of such potential antimicrobial agents are siderophores. Siderophores are a chemically diverse group of iron-chelating agents secreted by microorganisms to scavenge iron, which is essential for bacterial growth. Certain siderophores are highly species specific and can therefore lock iron away from competitors with incompatible uptake systems.

In our previous work, we showed that environmental *Pseudomonas spp.* produce chemically different variants of pyoverdine – a group of siderophores with strong iron affinity and high specificity. Some of these pyoverdines strongly inhibited the growth of human pathogens *in vitro*, increased survival of wax moth larvae upon infection and showed low potential for resistance development.

In our current work, we examine the clinical potential of pyoverdines as novel antimicrobial agents. We found that pyoverdine 3G07 (our top candidate) effectively inhibited the growth of multiple antibiotic-resistant clinical isolates, including key ESKAPE pathogens like *Acinetobacter baumannii*, *Staphylococcus aureus* and *Klebsiella pneumoniae*. Furthermore, we observed no toxicity following injection of 3G07 into zebrafish larvae, indicating its suitability for *in vivo* applications. In addition, net growth modelling based on time-kill data revealed that pyoverdine acts bacteriostatically. In a next step, we aim to test whether pyoverdine synergies with bactericidal antibiotics or antimicrobial peptides as we hypothesize that a combination therapy may be most potent.

Taken together, our work provides first key insights into the therapeutic potential of pyoverdines and aims to contribute to the global efforts in developing new innovative antimicrobial treatment.

**Student Paper*

MT04*/S03*/P008* Exploration of novel mechanisms of manogepix resistance in Candidozyma auris

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Context: *Candidozyma auris* is an emerging multidrug-resistant yeast responsible for invasive infections. Manogepix is a novel antifungal agent that inhibits Gwt1, a key enzyme in the glycosylphosphatidylinositol anchor biosynthesis pathway required for proper cell wall protein localization and fungal viability. However, *C. auris* can develop reduced susceptibility to manogepix. *TAC1b* mutations resulting in overexpression of *CDR1* efflux pump and reduced susceptibility to both manogepix and fluconazole have been reported.

Aim: In this study, we plan to explore *TAC1b*-independent genetic mechanisms contributing to reduced manogepix susceptibility in *C. auris*.

Methods: A *TAC1b* deletion strain was subjected to *in vitro* evolution under manogepix exposure. Evolved strains were characterized by antifungal susceptibility testing to both manogepix and fluconazole, whole-genome sequencing, and RT-qPCR of *CDR1*.

Results: Five evolved strains with reduced susceptibility to manogepix were obtained. Two strains harbored mutations in the manogepix target gene *GWT1*, while two other strains carried a mutation in a gene encoding an enzyme involved in phospholipid biosynthesis. All four strains did not show cross-resistance to azoles and did not exhibit *CDR1* overexpression. The last strain carried a mutation in *UBR2* (a ubiquitin-ligase involved in control of *CDR1* expression), which was associated with increased *CDR1* expression and reduced manogepix and fluconazole susceptibility.

Conclusion and perspectives: These findings provide new insights into mechanisms contributing to reduced manogepix susceptibility in *C. auris*. We showed for the first time that *C. auris* can develop mutations in the manogepix target gene *GWT1* and in a lipid biosynthesis gene, under manogepix pressure. The function of these mutations needs to be confirmed by genetic modifications.

**Student Paper*

MT05*/P010* Whole-genome enrichment of Mycobacterium tuberculosis from clinical samples: a sensitivity assessment

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Tuberculosis (TB), caused by *Mycobacterium tuberculosis* (MTb), remains a major global health concern, with more than 10 million active cases annually. The rise in drug-resistant strains necessitates their rapid detection for appropriate treatment selection. Existing commercial tests for second-line drug resistance in multidrug-resistant (MDR) or extensively drug-resistant (XDR) TB are largely unavailable or have limited sensitivity. Culture-based diagnostics, regarded as the gold standard, are very slow due to MTb's 20-hour doubling time. Whole-genome enrichment directly from clinical material offers a promising approach to rapidly enabling resistance detection while providing valuable information on lineage and transmission dynamics. We aimed to assess whether target enrichment, also referred to as sequence capture, could be implemented as a culture-free genome sequencing method for routine clinical diagnostics of TB. Using serially diluted sputum spiked with MTb (H37Rv and MDR strains) and clinical samples from the Swiss National Reference Centre for Mycobacteria (NZM), we compared amplicon-based Deeplex® Myc-TB (GenoScreen) and two whole-genome target enrichment approaches, QIAseq® xHYB *Mycobacterium tuberculosis* Panel (Qiagen) and Illumina DNA Prep with Enrichment (IDPE) with custom pan-genome MTb baits, to gold standard diagnostic tests performed at the NZM, including sequencing from culture and phenotypic drug susceptibility testing (pDST). Sensitivity calculations based on genome coverage and mean read depth were derived from read mapping against the H37Rv reference genome using snippy. Lineage determination and drug susceptibility testing used TBProfiler and tbtAMR, and core genome multilocus sequence typing used MBioSEQ Ridom Typer. Preliminary data show that for Deeplex® Myc-TB, only 50% of clinical samples report results with quality score +, and their predicted drug resistances from the easy-to-use web application match pDST results. For QIAseq® xHYB and IDPE, higher bacterial loads produced results with better correlations across the bioinformatic tools. We show promising results for whole-genome target enrichment of MTb, with procedure optimisation needed.

**Student Paper*

MT06*/P012* Lineage and microenvironment effects drive heterogeneous quorum sensing activation in Pseudomonas aeruginosa microcolonies

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Quorum sensing (QS) is a bacterial cell-cell communication process in which bacteria produce, detect, and respond to extracellular signalling molecules called autoinducers. QS enables bacteria to sense population density and coordinate collective behaviours such as swarming and biofilm formation. Although QS has traditionally been considered a population-wide and homogeneous response, recent studies have revealed substantial heterogeneity in QS gene expression even among clonal cells. We hypothesized that this heterogeneity originates from stochastic activation events at the onset of QS and is amplified in spatially structured environments through limited autoinducer diffusion and local cell-cell interactions.

To investigate QS heterogeneity, we grew single-copy fluorescent reporter strains of *Pseudomonas aeruginosa* on agarose pads and monitored microcolony formation and QS activation using time-lapse fluorescence microscopy at single-cell resolution. Deep learning-based cell segmentation and tracking tools were used to analyse temporal, spatial, and lineage effects on QS activation and propagation in the two homoserine lactone-based QS systems, Las and Rhl.

We observed strong heterogeneity in the expression of the autoinducer synthesis genes *lasI* and *rhlI*, whereas the cognate receptor genes *lasR* and *rhlR* and the effector genes *lasB* and *rhlAB* were expressed more uniformly across cells. This pattern suggests stochastic activation of autoinducer production in a subset of cells, followed by limited local propagation that generates hotspots of QS activity within colonies. Random forest regression models trained on the single-cell dataset predicted *lasI* and *rhlI* expression levels with an accuracy of 76% and 81%, respectively, and revealed that lineage effects explained ~95% of the observed variation, while spatial proximity accounted for only ~3%. Together, these findings indicate that QS signal production is primarily driven by a few highly expressing lineages, whereas induction of neighbouring cells through signal diffusion is relatively rare.

**Student Paper*

MT07/S68/P014 Mapping the temperate phage landscape and integration hotspots across Staphylococcaceae

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Temperate bacteriophages are ubiquitous in staphylococci, yet their diversity and integration patterns remain under-characterized beyond *Staphylococcus aureus*. This study establishes a genomic–experimental framework to map the prophage landscape, integration hotspots, and mobilization potential within non-aureus members of the Staphylococcaceae family.

Using an in-silico pipeline to analyze 114 bacterial genomes (41 coagulase-negative-staphylococci), we identified intact prophages in 68% of strains, and 45% enclosed phage-like elements (incomplete or questionable phages). Of these lysogens, 47, 42 and 14% carried one, two or three intact prophages, respectively. Notably, 37% and 9% of lysogens carried antimicrobial resistance (AMR) and virulence genes, respectively, while 10% harbored AMR-carrying plasmids.

Experimental induction yielded novel phages from *S. epidermidis* (ILRIcamel_95, ILRIcamel_96) and *Mammaliicoccus sciuri* (EG97). These phages integrate into specific sites: tRNA-Ser-Asn, the FAD-coding gene (*merA*), or a VOC-family protein gene, respectively. These attachment sites were present in 72%, 6% and 1% of our Staphylococcaceae lysogenic collection, respectively, with 16%, 10%, 63% truncated by integrase-leading elements, identifying those loci as integration hotspots for phage-related elements. While ILRIcamel_95 and ILRIcamel_96 exhibit *S. epidermidis*-specific infection range in-vitro, EG97 shows sporadic cross-species activity. Furthermore, in-silico prediction of integration sites was performed on *S. simulans* lysogens (n=11). These phages were predicted to integrate into *sufB*, RNA chaperone or tRNA-Ser-Glu. These attachment sites were present in 61%, 10% and 35% of our Staphylococcaceae lysogenic collection, with 20%, 64%, 40% truncated by integrase-leading elements. These prophages could not be recovered on the tested host panel, and alternative in vitro approaches are underway to assess their ability to excise and induce lysis independently of detectable propagation in indicator strains.

This integrated genomic–experimental framework provides a scalable strategy to uncover hidden temperate phage diversity and integration hotspots within Staphylococcaceae, providing insights into bacterial evolution and new opportunities to recover and study experimentally elusive functional temperate phages.

MT08*/P016* In situ characterisation of corynephage infection in mycomembrane-bearing minicells.

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The increase of multidrug-resistant (MDR) pathogens within the Corynebacteriales order, e.g. *Mycobacterium tuberculosis*, demands the development of alternative antimicrobial strategies. Corynebacteriales harbour a mycolate-rich outer membrane, the mycomembrane, which forms a low-permeability barrier contributing to their MDR. *Corynebacterium glutamicum*, a BSL1, genetically tractable model organism, used across the food, pharma, and cosmetics industries, enables the investigation of this complex cell envelope. Here, I use a non-contractile tailed corynephage to study how they overcome the mycomembrane. This forms a powerful system to elucidate the molecular details underlying bacteriophage-host interactions and mycomembrane remodelling mechanisms. To visualise this process at high-resolution, I perform *in situ* cryo-electron tomography (cryo-ET) on *C. glutamicum* minicells. These minicells are generated through the deletion of plasmid segregation genes, producing mutant miniature cells. During my master's project I have established a method to obtain near-homogeneous minicell populations and confirmed their morphology and cell envelope integrity by widefield fluorescence microscopy. Their small size and electron transparency make them highly suitable for cryo-ET, without requiring sample thinning by cryo-focused ion beam (cryo-FIB) milling. This increases the observable infectious events and improves the achievable resolution. I have therefore established these minicells as a robust proxy for cryo-ET analysis of corynephage infection, aiming to visualise infection stages from attachment to lysis at nanometer resolution. My first goal is to characterise the conformational changes within the corynephage baseplate which enable tail tube opening and DNA injection into the host. This work will provide the first mechanistic insights into corynephage infection in Corynebacteriales, advancing our understanding of mycomembrane biology and enabling industrial and antimicrobial applications. The minicell model will further support high-resolution studies of bacterial division, mycolate cell envelope remodelling, and mycobacterial biology, laying the groundwork for potential phage-based antimicrobials strategies.

**Student Paper*

MT09∗/P018∗ Effects of combinations of antibiotics on antibiotic resistance selection in aquatic environments

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The emergence of antimicrobial resistance (AMR) is a major global health threat. It is increasingly recognized that AMR selection is not limited to clinical settings but also occurs at low antibiotic concentrations in environmental systems. However, the effects of environmentally relevant antibiotic mixtures on AMR selection remain poorly understood.

This study investigates whether combinations of antibiotics lead to non-additive selection of AMR in wastewater microbial communities. In controlled microcosm experiments, wastewater inocula were grown in artificial wastewater medium. Communities were exposed to single antibiotics and mixtures of antibiotics representing four classes: macrolides, quinolones, tetracyclines, and dihydrofolate reductase inhibitors. Antimicrobial resistance genes (ARGs) enrichment was quantified using quantitative PCR, while shifts in community composition were assessed through 16S rRNA gene sequencing.

We will present initial data and analysis on comparisons between single antibiotic and mixture exposures to evaluate how interactions of antibiotics influence resistance selection and microbial community dynamics. These results will contribute to improving environmental risk assessment frameworks and support the development of water quality guidelines that account for AMR under realistic mixture exposure scenarios within a One Health context.

∗Student Paper

MT10*/S23*/P020* targeting Rhinovirus IRES as new approach for antivirals

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Picornaviridae is a family of viruses that includes different known human pathogens such as Polioviruses, Enteroviruses and Rhinoviruses. Their symptoms range from mild common cold to paralysis and even death. The IRES sequence is a conserved RNA element present in this family that plays a crucial role in the viral life cycle. Targeting the IRES is feasible, as demonstrated by compounds showing activity against EV71 in vitro and in vivo. However, available compounds mostly target stem-loop II, whose relatively simple structure may lead to off-target effects. Additionally, the IRES structure has largely been predicted but has not yet been assessed in infected cells. Therefore, this project begins by refining IRES structural predictions through probing viral RNA in infected cells and characterizing its secondary structure using DMS-MaP. The resulting structure enabled the design of a library of LNA-ASOs (locked nucleic acid antisense oligonucleotides), that either unfold or induce degradation of the target RNA. Among the tested ASOs, four exhibit nanomolar activity against RV8 without detectable toxicity in vitro and in human derived respiratory models. Further optimization of these LNA-ASOs, including viral resistance, synergy evaluation and testing across different enteroviruses (including EV68 and EV71), is currently ongoing.

**Student Paper*

MT11∗/P022∗ Development and validation of a LAMP assay for rapid detection of Escherichia coli associated UTIs with increased risk of urosepsis

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Background: Current culture-based diagnostics for UTIs require up to 24 hours for pathogen identification and do not assess the risk of potential invasiveness. Thereby, clinicians rely on empirical antibiotics before results are available. Inappropriate treatment, however, contributes to the development of antimicrobial resistance (AMR). Uropathogenic *Escherichia coli* (UPEC) are the most common cause of UTIs and virulence factors, e.g., PapGII (adhesin of pyelonephritis-associated pili) are associated with higher risks of urosepsis. We aimed to develop and validate a Loop-Mediated Isothermal Amplification (LAMP)-based assay for rapid detection of *papGII*-positive *E. coli* directly from urine.

Methods: We designed specific primers targeting conserved regions of the *E. coli uidA* and *papGII* genes. Serial dilutions of targets were used to evaluate the limit of detection. To validate the assay, we prospectively collected 191urine specimen samples over a three-day period from routine diagnostics and performed fluorescent LAMP. Culture results from routine diagnostics were used as reference for *E. coli* detection and whole genome sequencing for *papGII* detection. For each target we calculated the optimal thresholds to maximize sensitivity and specificity.

Results: For this assay, the limit of detection was found to be 10^3 CFU/ml. Among the clinical urine samples tested, the assay showed 87% sensitivity and 95% specificity for *E. coli,* and 100% sensitivity and specificity for *papGII*, compared to the reference methods. Notably, all results were obtained within 40 minutes and without extensive sample preparation.

Conclusion: This LAMP-based assay was intentionally simplified to demonstrate the requirements of minimal hands-on work and lab equipment. It shows strong to excellent diagnostic performance for the detection of *E. coli* and *papGII*, respectively, highlighting the potential of early UPEC identification and invasiveness risk assessment. To enhance clinical applicability, future optimization will focus on reducing false positives and negatives.

∗Student Paper

MT12/P024 Spontaneous recapitulation of endogenous colonization resistance following antibiotic therapy and the mechanisms underlying microbiota-mediated community resistance to Clostridioides difficile

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Disruptions to the gut microbiota are widely recognized as the primary driver of susceptibility to *Clostridioides difficile* infection (CDI). Although fecal microbiota transplantation (FMT) effectively treats recurrent CDI, the specific microbial components and molecular mechanisms required to confer resistance remain poorly understood. This hinders the rational development of defined, scalable live biotherapeutic products as alternatives to FMT. Notably, most patients with primary CDI achieve clinical cure following first-line antibiotics, demonstrating that spontaneous restoration of colonization resistance is typically sufficient. Using shotgun metagenomics data from a longitudinal CDI clinical trial, we hypothesized that identifying and experimentally replicating the functional traits of these resistant communities using well-characterized human microbes could inform the design of synthetic communities (SynComs) that mimic this protective phenotype.

We implanted gnotobiotic mice with human microbiota derived from CDI patients post-antibiotic treatment, and subsequently challenged them with *C. difficile*. Susceptibility depended reliably on the specific implanted microbiota. Engrafted communities clustered primarily by donor and showed low inter-mouse variability, enabling reproducible functional phenotyping. Post-therapy microbiomes conferred a broad spectrum of colonization resistance, ranging from minimal to complete pathogen suppression, with post-fidaxomicin communities exhibiting greater resilience and community diversity. These resistance phenotypes were highly donor-specific, reproducible across replicates, and correlated with distinct taxonomic configurations.

Our study establishes a reproducible experimental platform to iteratively manipulate specific taxa and functional pathways, allowing us to untangle the mechanisms driving spontaneous microbiome recovery following CDI therapy. These insights reveal patient-specific recovery trajectories that drive the restoration of colonization resistance and the findings provide a structural framework for the mechanistically informed design of simplified microbial consortia engineered with targeted colonization resistance functions.

PF01*/P001* Development and implementation of an evolutionary algorithm for engineering microbiomes via environmental modulation

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The availability and composition of environmental resources – particularly carbon – has a profound effect on the structure and function of microbial communities. This connection presents a promising avenue for engineering communities with specific functions through targeted environmental manipulations.

However, as even slight alterations in resource availability and composition can yield drastically different ecological outcomes, such an approach to community engineering would require testing community responses in a combinatorially intractable space of environmental compositions. To address this challenge, here we develop and experimentally implement a search algorithm to identify combinations of carbon sources that produce bacterial communities with desired properties.

This algorithm navigates combinatorial spaces of carbon sources through an iterative process inspired by evolutionary theory. Communities are assembled in candidate environments, their resulting functions are compared to a desired target, and new candidate environments are generated by recombining carbon sources from the top scoring environments. Thus, with every cycle, the search space is reduced until carbon source compositions that induce the desired function are identified. Utilizing simulated growth data, we have confirmed the efficacy of this approach, assessed the navigability of the search space, and identified optimal parameters for execution *in vitro*. We now begin *in vitro* implementation, selecting for precise abundances of individual community members in a soil-derived microbial community.

Beyond assessing the capability of evolutionary optimization to induce desired properties in microbial consortia, this work will generate a large-scale mapping of environmental compositions to community function, enabling opportunities to examine metabolic mechanisms underlying community assembly patterns.

**Student Paper*

MT01*/P002* Spatiotemporal interactions between biocontrol bacteria and pathogenic oomycete

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Biocontrol bacteria employ a sophisticated, multi-layered strategy to suppress pathogenic fungi. However, relying solely on antifungal metabolites rarely achieves complete hyphal inhibition and often allows hyphae to breach weak points in antagonistic activity, leading to wider infection. Moreover, certain antifungal metabolites may pose potential risks to humans, animals, and the environment, which is why the EFSA (European Food Safety Authority) considers Pseudomonas fluorescens to be an effective biocontrol agent that is "not presumed safe by default."

When genes associated with the production of these antifungal metabolites are knocked out, the reduced growth burden enhances the response rate to chemical signals released by pathogens and improves motility. The bacteria actively track and colonize hyphal surfaces using flagella and pili. This targeted bacterial motility often outpaces the spread of diffusible metabolites, thereby effectively restricting hyphal expansion.

**Student Paper*

PF02/P003 Impact of OprF porin deletion on the efficacy of imipenem therapy in a murine sepsis model caused by Pseudomonas aeruginosa producing NDM-1 carbapenemase

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Background: Carbapenem-resistance in *Pseudomonas aeruginosa* infection is driven by multiple mechanisms, including carbapenemase production (particularly NDM-1) and altered outer membrane permeability (OprD defect). OprF, the major outer membrane porin, plays a central role in membrane integrity and possibly virulence. This study evaluated whether OprF disruption could restore carbapenem efficacy in NDM-1-producing strains during experimental sepsis.

Methods: Isogenic PAO1 and PA14 *oprF*-deficient mutants, including NDM-1-producing derivatives, were analyzed *in vitro* by motility and growth competition experiments, and a murine peritoneal sepsis model. Groups of CD-1 female mice were infected intraperitoneally (ip) with the minimum lethal dose (MLDs) previously determined for each strain. Treatment lasted 24 hours and started two hours post-inoculation and animals were randomly grouped into: i) controls, infected, untreated; ii) suboptimal treatment of imipenem (30mg/kg/q4h/ip/24h). Survival was monitored over 24 hours. Bacterial loads in spleen (log_{10}CFU/g) and blood (log_{10} CFU/mL), and bloodstream-infection (BSI) rates were assessed. Comparisons were performed using Mann-Whitney U and Fisher's exact tests.

Results: Although *oprF* disruption reduced motility and fitness, *in vivo* virulence remained preserved, with similar MLDs among strains. In mice infected with NDM-1-producing PAO1 strain, imipenem treatment failed to improve survival or BSI rates despite slight reductions in spleen and blood bacterial concentration. In contrast, disruption of *oprF* in the NDM-1-producing strain is associated with increased expression of OprD, restored imipenem efficacy, resulting in significant reductions in bacterial concentrations in spleen and blood, complete clearance of BSI, and significantly improved survival, reaching outcomes comparable to those observed with the imipenem-susceptible PAO1 strain.

Conclusions:

1. Loss of OprF porin restores imipenem efficacy against NDM-1-producing *P. aeruginosa*, improving bacterial clearance and survival.
2. Outer membrane permeability alterations may increase susceptibility to antibiotics and enhance therapeutic responses in MBL-producing *P. aeruginosa*.
3. These results highlight the interaction of two specific outer membrane porins, OprF and OprD, in carbapenem resistance.

MT02∗/P004 exploring the ecological and evolutionary roles of persisting bacteriophages in traditional food fermentation

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Traditional fermented foods are simple microbial ecosystems shaped by long-term human influence. Multiple microorganisms interact, adapt, and evolve under dynamic environmental conditions. While the diversity and functional roles of bacteria in these systems are well understood and form the basis of current knowledge on fermentation processes, the ecological and evolutionary contributions of bacteriophages (phages) remain largely underexplored. Recent research shows that phages and their bacterial hosts can stably coexist in fermented foods without negatively affecting fermentation outcomes, challenging the traditional perception of phages as purely disruptive agents. This project aims to advance a comprehensive understanding of the diversity, ecological roles, and evolutionary dynamics of phages in traditional food fermentation systems. In my PhD, I will analyze large-scale genomic and metagenomic datasets to construct an extensive catalogue of phages persisting in these environments. These data will underpin the development of predictive models of phage–host interactions and their ecological functions within microbial communities, while also guiding targeted phage isolation strategies to establish a collection of persistent phages for experimental characterization. By integrating computational and experimental approaches, this work seeks to uncover how phages influence microbial community structure and ecosystem functioning. Ultimately, it aims to reshape current perspectives on phages in food fermentation and reveal beneficial phenotypic impacts that could be leveraged to improve fermentation processes and product quality.

∗Student Paper

PF03*/P005* Host specificity depends on spatial scale: insights from micromammal gut microbiota

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Gut microbial communities (microbiota) and their animal hosts are deeply interconnected. Hosts play a role in shaping microbiota composition, and in turn, gut microbiota contribute to host fitness. This tight interplay is reflected in patterns of host specificity, whereby microbiota composition clusters by host species: composition is more similar among individuals of the same species than among individuals from different species.

Quantifying host specificity precisely is therefore a powerful tool for assessing how tightly interdependent a given host species and its gut microbiota are. Surprisingly, the degree of host specificity measured varies substantially across studies, even within the same groups of animals (e.g. rodents). In this study, we hypothesize that this discrepancy originates from the spatial scale at which specificity is measured. Studies focusing on small spatial scales may underestimate within-species variability because sampled individuals are more likely to be closely related. This, in turn, inflates the measured degree of host specificity, making it appear higher than in studies conducted at broader spatial scales.

Using micromammals as a model, we measure the effect of spatial scale on host specificity by sampling feces across multiple wild host species at both local and regional levels. Our preliminary results show that sampling design is an important determinant of the degree of host specificity observed. We confirm that sampling at small spatial scales does indeed inflate host specificity measures. In addition, we test the extent to which host diet (assessed by plant metabarcoding) mediates the host specificity effect we observe.

This study sheds light on potential causes of discrepant observations of host specificity. It therefore helps bridge the gap between host specificity studies and contributes to our understanding of host–microbiota interactions.

**Student Paper*

MT03*/S63*/P006* Pyoverdines as antimicrobial agents against human opportunistic pathogens

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The emergence and spread of pathogens that are resistant against antibiotics has sparked interest in the development of novel or alternative treatments, including bacteriophages, antimicrobial peptides and new antibiotics. One way to discover new antibacterials is to explore the plethora of secondary metabolites bacteria produce themselves to interact with their environment and other microbes. One group of such potential antimicrobial agents are siderophores. Siderophores are a chemically diverse group of iron-chelating agents secreted by microorganisms to scavenge iron, which is essential for bacterial growth. Certain siderophores are highly species specific and can therefore lock iron away from competitors with incompatible uptake systems.

In our previous work, we showed that environmental *Pseudomonas spp.* produce chemically different variants of pyoverdine – a group of siderophores with strong iron affinity and high specificity. Some of these pyoverdines strongly inhibited the growth of human pathogens *in vitro*, increased survival of wax moth larvae upon infection and showed low potential for resistance development.

In our current work, we examine the clinical potential of pyoverdines as novel antimicrobial agents. We found that pyoverdine 3G07 (our top candidate) effectively inhibited the growth of multiple antibiotic-resistant clinical isolates, including key ESKAPE pathogens like *Acinetobacter baumannii*, *Staphylococcus aureus* and *Klebsiella pneumoniae*. Furthermore, we observed no toxicity following injection of 3G07 into zebrafish larvae, indicating its suitability for *in vivo* applications. In addition, net growth modelling based on time-kill data revealed that pyoverdine acts bacteriostatically. In a next step, we aim to test whether pyoverdine synergies with bactericidal antibiotics or antimicrobial peptides as we hypothesize that a combination therapy may be most potent.

Taken together, our work provides first key insights into the therapeutic potential of pyoverdines and aims to contribute to the global efforts in developing new innovative antimicrobial treatment.

**Student Paper*

PF04*/P007* Establishing genetic modification in primary-like human airway epithelial cultures

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Influenza A viruses cause a respiratory disease in humans that poses a major burden on human health. With the long-term goal of developing antiviral strategies, influenza virus-host interactions are mostly studied on a cellular and molecular level using cancer-derived cell lines. While these cell lines are well suitable for genetic modifications and thus allow for mechanistic studies, they do not recapitulate the complexity of the human airway epithelium. This limitation can be overcome by using primary human airway cultures, a physiologically relevant model of airway epithelia. However, genetic modification of these cultures has thus far been difficult. In this study, we aim to establish a protocol to genetically modify human airway epithelial cultures to assess and understand virus-host interactions, using the immortalized primary-like cell line BCi-NS1.1. We show that these cells can differentiate into the different cell types that are found in human airway epithelia, that they can be infected with several influenza A virus strains and are also amenable to genetic modification via lentiviral transduction. Ongoing work aims at generating and characterizing airway cultures deficient of the type I interferon receptor to validate our protocol. Our approach should thereby enable the study of influenza A virus-host interactions in a physiologically relevant model.

**Student Paper*

MT04*/S03*/P008* Exploration of novel mechanisms of manogepix resistance in Candidozyma auris

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Context: *Candidozyma auris* is an emerging multidrug-resistant yeast responsible for invasive infections. Manogepix is a novel antifungal agent that inhibits Gwt1, a key enzyme in the glycosylphosphatidylinositol anchor biosynthesis pathway required for proper cell wall protein localization and fungal viability. However, *C. auris* can develop reduced susceptibility to manogepix. *TAC1b* mutations resulting in overexpression of *CDR1* efflux pump and reduced susceptibility to both manogepix and fluconazole have been reported.

Aim: In this study, we plan to explore *TAC1b*-independent genetic mechanisms contributing to reduced manogepix susceptibility in *C. auris*.

Methods: A *TAC1b* deletion strain was subjected to *in vitro* evolution under manogepix exposure. Evolved strains were characterized by antifungal susceptibility testing to both manogepix and fluconazole, whole-genome sequencing, and RT-qPCR of *CDR1*.

Results: Five evolved strains with reduced susceptibility to manogepix were obtained. Two strains harbored mutations in the manogepix target gene *GWT1*, while two other strains carried a mutation in a gene encoding an enzyme involved in phospholipid biosynthesis. All four strains did not show cross-resistance to azoles and did not exhibit *CDR1* overexpression. The last strain carried a mutation in *UBR2* (a ubiquitin-ligase involved in control of *CDR1* expression), which was associated with increased *CDR1* expression and reduced manogepix and fluconazole susceptibility.

Conclusion and perspectives: These findings provide new insights into mechanisms contributing to reduced manogepix susceptibility in *C. auris*. We showed for the first time that *C. auris* can develop mutations in the manogepix target gene *GWT1* and in a lipid biosynthesis gene, under manogepix pressure. The function of these mutations needs to be confirmed by genetic modifications.

**Student Paper*

PF05∗/P009∗ Chimerophore antibiotics: combining host defense peptides for multimodal antimicrobial activity

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Ribosomally synthesized host defense peptides (HDPs) represent a promising yet clinically underexploited reservoir of antimicrobial substances. Non-membranolytic HDPs offer high specificity and low host-cell toxicity by targeting intracellular machinery rather than disrupting bacterial membranes. However, their translation is often hindered by limited potency and low activity in physiological fluids. To overcome these barriers, we employed a semi-rational peptide chimerization strategy, fusing the pharmacophores of non-lytic HDPs into diverse chimeric architectures to trigger intramolecular synergy and enforce multimodal MOAs within a single molecule.

While previous efforts to explore chimeric HDPs have been constrained by the high costs and low throughput of chemical synthesis, here we circumvent these bottlenecks by ribosomally synthesizing of a combinatorial library of 99,235 DNA-encoded *chimerophores* (chimeric pharmacophores). Using the high-throughput self-screening platform (Me^X) in *Escherichia coli* for intracellular activity assessment, we identified over 30,300 active variants, representing a significant expansion of the known functional space of chimeric HDPs. We isolated potentiated broad-spectrum candidates that remain active in serum, maintain low host-cell toxicity, use multiple membrane-transport modalities and simultaneously act on multiple intracellular targets. By integrating these orthogonal pathways, our lead candidate cp9 successfully suppressed the evolutionary escape of *Pseudomonas aeruginosa*. This work establishes a scalable, high-throughput platform for the systematic design of next-generation antibiotics capable of eradicating multidrug-resistant pathogens through enforced multimodality.

∗Student Paper

MT05*/P010* Whole-genome enrichment of Mycobacterium tuberculosis from clinical samples: a sensitivity assessment

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Tuberculosis (TB), caused by *Mycobacterium tuberculosis* (MTb), remains a major global health concern, with more than 10 million active cases annually. The rise in drug-resistant strains necessitates their rapid detection for appropriate treatment selection. Existing commercial tests for second-line drug resistance in multidrug-resistant (MDR) or extensively drug-resistant (XDR) TB are largely unavailable or have limited sensitivity. Culture-based diagnostics, regarded as the gold standard, are very slow due to MTb's 20-hour doubling time. Whole-genome enrichment directly from clinical material offers a promising approach to rapidly enabling resistance detection while providing valuable information on lineage and transmission dynamics. We aimed to assess whether target enrichment, also referred to as sequence capture, could be implemented as a culture-free genome sequencing method for routine clinical diagnostics of TB. Using serially diluted sputum spiked with MTb (H37Rv and MDR strains) and clinical samples from the Swiss National Reference Centre for Mycobacteria (NZM), we compared amplicon-based Deeplex® Myc-TB (GenoScreen) and two whole-genome target enrichment approaches, QIAseq® xHYB *Mycobacterium tuberculosis* Panel (Qiagen) and Illumina DNA Prep with Enrichment (IDPE) with custom pan-genome MTb baits, to gold standard diagnostic tests performed at the NZM, including sequencing from culture and phenotypic drug susceptibility testing (pDST). Sensitivity calculations based on genome coverage and mean read depth were derived from read mapping against the H37Rv reference genome using snippy. Lineage determination and drug susceptibility testing used TBProfiler and tbtAMR, and core genome multilocus sequence typing used MBioSEQ Ridom Typer. Preliminary data show that for Deeplex® Myc-TB, only 50% of clinical samples report results with quality score +, and their predicted drug resistances from the easy-to-use web application match pDST results. For QIAseq® xHYB and IDPE, higher bacterial loads produced results with better correlations across the bioinformatic tools. We show promising results for whole-genome target enrichment of MTb, with procedure optimisation needed.

**Student Paper*

PF06∗/P011∗ Decoding Type IV Pilus Recognition of Pseudomonas aeruginosa Phages

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Pseudomonas aeruginosa is a leading pathogen responsible for healthcare-associated infections and presents a significant clinical challenge due to its widespread multidrug resistance. Bacteriophages, viruses that selectively infect and kill bacteria, offer a complementary approach to traditional antibiotic therapies. Type IV pili (T4P) are important virulence factors of *P. aeruginosa* that mediate motility, adhesion, biofilm formation, and host colonization. They also serve as receptors for a diverse range of bacteriophages, making them important drivers of phage-host specificity and co-evolution. Understanding the molecular basis of pilus-mediated viral recognition is therefore essential for the development and selection of effective phages for therapy.

In this study, we isolated and characterized a collection of novel T4P-targeting phages infecting *P. aeruginosa*. Phage-pilus interactions were analysed using integrated genomic analyses, phenotypic assays, structural biology, and electron microscopy. Our results identified distinct receptor-binding modules shared across genetically diverse phages, suggesting that T4P recognition relies on a limited set of conserved yet adaptable molecular strategies. To investigate their role in receptor recognition, we first analysed the host range of phages encoding homologous but distinct pilus-binding modules across a diverse collection of clinical *P. aeruginosa* isolates expressing different T4P variants. We subsequently exchanged homologous receptor-binding modules between phages with divergent host ranges using genetic engineering. Our findings proved the role of these modules in pilus binding and directly linked them to phage host range.

Together, these findings do thus not only broaden our knowledge of phage-pilus interactions but also provide a foundation for the rational application of phage therapy to treat multidrug-resistant *P. aeruginosa* strains.

∗Student Paper

MT06*/P012* Lineage and microenvironment effects drive heterogeneous quorum sensing activation in Pseudomonas aeruginosa microcolonies

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Quorum sensing (QS) is a bacterial cell-cell communication process in which bacteria produce, detect, and respond to extracellular signalling molecules called autoinducers. QS enables bacteria to sense population density and coordinate collective behaviours such as swarming and biofilm formation. Although QS has traditionally been considered a population-wide and homogeneous response, recent studies have revealed substantial heterogeneity in QS gene expression even among clonal cells. We hypothesized that this heterogeneity originates from stochastic activation events at the onset of QS and is amplified in spatially structured environments through limited autoinducer diffusion and local cell-cell interactions.

To investigate QS heterogeneity, we grew single-copy fluorescent reporter strains of *Pseudomonas aeruginosa* on agarose pads and monitored microcolony formation and QS activation using time-lapse fluorescence microscopy at single-cell resolution. Deep learning-based cell segmentation and tracking tools were used to analyse temporal, spatial, and lineage effects on QS activation and propagation in the two homoserine lactone-based QS systems, Las and Rhl.

We observed strong heterogeneity in the expression of the autoinducer synthesis genes *lasI* and *rhlI*, whereas the cognate receptor genes *lasR* and *rhlR* and the effector genes *lasB* and *rhlAB* were expressed more uniformly across cells. This pattern suggests stochastic activation of autoinducer production in a subset of cells, followed by limited local propagation that generates hotspots of QS activity within colonies. Random forest regression models trained on the single-cell dataset predicted *lasI* and *rhlI* expression levels with an accuracy of 76% and 81%, respectively, and revealed that lineage effects explained ~95% of the observed variation, while spatial proximity accounted for only ~3%. Together, these findings indicate that QS signal production is primarily driven by a few highly expressing lineages, whereas induction of neighbouring cells through signal diffusion is relatively rare.

**Student Paper*

PF07/P013 Integrated metagenomic and metabolomic features Predict opportunistic pathogen expansion in allogeneic hematopoietic cell transplant recipients

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Allogeneic hematopoietic cell transplantation (allo-HCT) is a potentially curative therapy for blood cancers. However, receipt of conditioning chemotherapy and prophylactic antibiotics during allo-HCT disrupts the gut microbiota, whose composition is associated with survival, infection risk, and transplant-related complications. Opportunistic bacterial and fungal pathogens can expand in the gut, leading to invasive infections, yet the ecological and pharmacological drivers of these expansions remain poorly understood.

We analyzed 1,279 samples from 156 allo-HCT patients, integrating bacterial composition, pathways, and gene abundances (shotgun sequencing), fungal composition (ITS1 sequencing), fecal metabolomics, and medication records. We developed a dual framework to bridge mechanistic discovery and patient risk stratification: (1) to identify pre-expansion features associated with the subsequent expansion of five key pathogens (*Enterococcus, Klebsiella, Escherichia coli, Candida albicans,* and *Candida parapsilosis*) and (2) to build robust predictive models for these events.

Using generalized linear models and feature selection approaches, we identified 2-62 bacterial pathways, genes, metabolites, and medications associated with subsequent expansion for each of the studied species. In parallel, machine-learning models trained on pre-expansion samples predicted pathogen-specific expansion, explaining 39% and 63% of the variance of *C. albicans* and *Enterococcus* relative abundance, respectively. A classification model predicted *Klebsiella* expansion with 98.1% specificity and 100% sensitivity. Notably, a methyltransferase gene emerged as a strong predictor of *Klebsiella* expansion, and alone outperformed *Klebsiella* pre-expansion abundance.

To ensure robustness, we expanded our dataset with an additional 702 samples for independent *in silico* validation. This comprehensive pipeline serves as a foundation for future *in vitro* and *in vivo* experiments to functionally validate the identified variables and establish causal links between specific microbial features and pathogen expansion.

Overall, our preliminary results show that future opportunistic pathogen expansion after allo-HCT is partially encoded in pre-expansion microbial pathways, fecal metabolites, and medication history, enabling both accurate clinical prediction and biological insight.

MT07/S68/P014 Mapping the temperate phage landscape and integration hotspots across Staphylococcaceae

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Temperate bacteriophages are ubiquitous in staphylococci, yet their diversity and integration patterns remain under-characterized beyond *Staphylococcus aureus*. This study establishes a genomic–experimental framework to map the prophage landscape, integration hotspots, and mobilization potential within non-aureus members of the Staphylococcaceae family.

Using an in-silico pipeline to analyze 114 bacterial genomes (41 coagulase-negative-staphylococci), we identified intact prophages in 68% of strains, and 45% enclosed phage-like elements (incomplete or questionable phages). Of these lysogens, 47, 42 and 14% carried one, two or three intact prophages, respectively. Notably, 37% and 9% of lysogens carried antimicrobial resistance (AMR) and virulence genes, respectively, while 10% harbored AMR-carrying plasmids.

Experimental induction yielded novel phages from *S. epidermidis* (ILRIcamel_95, ILRIcamel_96) and *Mammaliicoccus sciuri* (EG97). These phages integrate into specific sites: tRNA-Ser-Asn, the FAD-coding gene (*merA*), or a VOC-family protein gene, respectively. These attachment sites were present in 72%, 6% and 1% of our Staphylococcaceae lysogenic collection, respectively, with 16%, 10%, 63% truncated by integrase-leading elements, identifying those loci as integration hotspots for phage-related elements. While ILRIcamel_95 and ILRIcamel_96 exhibit *S. epidermidis*-specific infection range in-vitro, EG97 shows sporadic cross-species activity. Furthermore, in-silico prediction of integration sites was performed on *S. simulans* lysogens (n=11). These phages were predicted to integrate into *sufB*, RNA chaperone or tRNA-Ser-Glu. These attachment sites were present in 61%, 10% and 35% of our Staphylococcaceae lysogenic collection, with 20%, 64%, 40% truncated by integrase-leading elements. These prophages could not be recovered on the tested host panel, and alternative in vitro approaches are underway to assess their ability to excise and induce lysis independently of detectable propagation in indicator strains.

This integrated genomic–experimental framework provides a scalable strategy to uncover hidden temperate phage diversity and integration hotspots within Staphylococcaceae, providing insights into bacterial evolution and new opportunities to recover and study experimentally elusive functional temperate phages.

P015* Harnessing SpA-targeting synthetic nanobodies for detection and enrichment of Staphylococcus aureus

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Staphylococcus aureus is a major human pathogen associated with invasive and chronic infections, including endocarditis, osteomyelitis, and biofilm-associated disease. Although rapid pathogen identification is critical for targeted therapy, current diagnostic workflows often remain limited by slow culture-based methods or insufficient species specificity in microscopy-based approaches. Here we show that synthetic single-domain antibodies (sybodies) targeting *Staphylococcal* surface protein A (SpA) enable robust detection and enrichment of *S. aureus* under clinically relevant conditions.

SpA is a highly abundant surface virulence factor that represents an attractive target for species-specific detection. We evaluated the targetability of SpA by a sequence conservation analysis across a large set of *S. aureus* genomes. Based on this target conservation analysis, synthetic nanobodies were selected and characterized, demonstrating high-affinity and specific binding to SpA.

Binding studies on intact bacterial cells showed strong fluorescence staining and efficient magnetic enrichment of S. aureus from cell suspensions. Importantly, the nanobodies retained binding despite repeated washing and in the presence of competing human immunoglobulins. We further evaluated performance in clinically relevant environments, where pathogen staining remained detectable.

These findings establish SpA-targeting sybodies as modular tools for pathogen detection and isolation in translational microbiology workflows. Their compatibility with fluorescence and bead-based platforms will support the development of rapid microscopy-compatible diagnostics and improved pathogen enrichment strategies for downstream molecular and proteomic analyses. Broadly, this work highlights the potential of synthetic nanobody technologies for studying bacterial pathogens directly in clinically relevant environments, allowing the community to understand *S. aureus* infections better.

**Student Paper*

MT08*/P016* In situ characterisation of corynephage infection in mycomembrane-bearing minicells.

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The increase of multidrug-resistant (MDR) pathogens within the Corynebacteriales order, e.g. *Mycobacterium tuberculosis*, demands the development of alternative antimicrobial strategies. Corynebacteriales harbour a mycolate-rich outer membrane, the mycomembrane, which forms a low-permeability barrier contributing to their MDR. *Corynebacterium glutamicum*, a BSL1, genetically tractable model organism, used across the food, pharma, and cosmetics industries, enables the investigation of this complex cell envelope. Here, I use a non-contractile tailed corynephage to study how they overcome the mycomembrane. This forms a powerful system to elucidate the molecular details underlying bacteriophage-host interactions and mycomembrane remodelling mechanisms. To visualise this process at high-resolution, I perform *in situ* cryo-electron tomography (cryo-ET) on *C. glutamicum* minicells. These minicells are generated through the deletion of plasmid segregation genes, producing mutant miniature cells. During my master's project I have established a method to obtain near-homogeneous minicell populations and confirmed their morphology and cell envelope integrity by widefield fluorescence microscopy. Their small size and electron transparency make them highly suitable for cryo-ET, without requiring sample thinning by cryo-focused ion beam (cryo-FIB) milling. This increases the observable infectious events and improves the achievable resolution. I have therefore established these minicells as a robust proxy for cryo-ET analysis of corynephage infection, aiming to visualise infection stages from attachment to lysis at nanometer resolution. My first goal is to characterise the conformational changes within the corynephage baseplate which enable tail tube opening and DNA injection into the host. This work will provide the first mechanistic insights into corynephage infection in Corynebacteriales, advancing our understanding of mycomembrane biology and enabling industrial and antimicrobial applications. The minicell model will further support high-resolution studies of bacterial division, mycolate cell envelope remodelling, and mycobacterial biology, laying the groundwork for potential phage-based antimicrobials strategies.

**Student Paper*

P017 Communicating the Invisible: Using Comics to Explain Microbiology and Antibiotic Resistance

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Communicating the complexity of microbiology to lay audiences remains a challenge, especially when dealing with invisible processes such as microbial evolution and antibiotic resistance. Antimicrobial resistance is rising globally, and both citizens and healthcare professionals play a role in slowing its spread. Clear, engaging communication tools are therefore essential.

This comic book offers an alternative to traditional formats by using narrative, metaphor, and visual storytelling to make microbial life tangible and relatable. Guided by LUCA, the Last Universal Common Ancestor, readers explore the microbial world — from early bacteria to their great diversity and evolutionary strategies.

Scientific mechanisms are translated into intuitive visual metaphors: molecular defences become armour, biofilms turn into fortified cities, dormant bacteria appear as “Sleeping Beauty”, and immune cells take the form of patrolling policemen. These story-based representations give audiences an accessible entry point into the invisible worlds of microbiology.

Beyond content, the project aims to demonstrate the potential of comics to confront assumptions about how science is communicated, providing an imaginative, inclusive medium that engages readers of all ages.

MT09*/P018* Effects of combinations of antibiotics on antibiotic resistance selection in aquatic environments

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The emergence of antimicrobial resistance (AMR) is a major global health threat. It is increasingly recognized that AMR selection is not limited to clinical settings but also occurs at low antibiotic concentrations in environmental systems. However, the effects of environmentally relevant antibiotic mixtures on AMR selection remain poorly understood.

This study investigates whether combinations of antibiotics lead to non-additive selection of AMR in wastewater microbial communities. In controlled microcosm experiments, wastewater inocula were grown in artificial wastewater medium. Communities were exposed to single antibiotics and mixtures of antibiotics representing four classes: macrolides, quinolones, tetracyclines, and dihydrofolate reductase inhibitors. Antimicrobial resistance genes (ARGs) enrichment was quantified using quantitative PCR, while shifts in community composition were assessed through 16S rRNA gene sequencing.

We will present initial data and analysis on comparisons between single antibiotic and mixture exposures to evaluate how interactions of antibiotics influence resistance selection and microbial community dynamics. These results will contribute to improving environmental risk assessment frameworks and support the development of water quality guidelines that account for AMR under realistic mixture exposure scenarios within a One Health context.

**Student Paper*

P019 Rapid Phage Susceptibility Testing Directly from Sputum via electrical field sample processing and Nanomotion Machine Learning Analysis.

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Rapid and reliable phage susceptibility testing (PST) is essential for the clinical implementation of personalized bacteriophage therapy against multidrug-resistant Pseudomonas aeruginosa infections, particularly in cystic fibrosis (CF) and non-CF bronchiectasis (NCFBE). Conventional PST methods remain growth-dependent, labor-intensive, and require 24–72 h to generate results. We developed an ultra-rapid phenotypic PST workflow combining detergent-free electric-field sputum processing with nanomotion-based sensing and machine learning analysis for direct testing from sputum samples.

Clinical *P. aeruginosa* isolates and sputum samples obtained from CF/NCFBE patients at Lausanne University Hospital were exposed to five lytic bacteriophages. Sputum samples were processed using an electric-field microfluidic platform to reduce host-cell background while preserving bacterial viability. Bacterial nanomotions were recorded using cantilever-based sensors following phage exposure. Machine-learning models incorporating spectral quantile feature extraction were trained on 197 nanomotion recordings and evaluated on an independent validation dataset including 44 cultured isolate recordings and 10 direct sputum recordings. Reference PST was performed using turbidity-based growth inhibition assays.

Electric-field processing effectively reduced sputum complexity without significantly affecting bacterial viability. Nanomotion responses enabled discrimination between phage-susceptible and phage-resistant isolates within 2.5–3.75 h following phage exposure. Machine-learning models achieved up to 90% classification accuracy on the independent validation dataset. The optimized sputum-derived model demonstrated 100% sensitivity for detection of susceptible samples. Importantly, PST could be successfully performed directly from polymicrobial sputum samples without prior bacterial isolation or culture.

This proof-of-concept study demonstrates the feasibility of combining electric-field sputum preparation with nanomotion-based PST for same-day phenotypic evaluation of phage susceptibility directly from sputum. This workflow represents a promising step toward rapid, culture-independent, and clinically actionable PST to support personalized phage therapy.

MT10*/S23*/P020* targeting Rhinovirus IRES as new approach for antivirals

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Picornaviridae is a family of viruses that includes different known human pathogens such as Polioviruses, Enteroviruses and Rhinoviruses. Their symptoms range from mild common cold to paralysis and even death. The IRES sequence is a conserved RNA element present in this family that plays a crucial role in the viral life cycle. Targeting the IRES is feasible, as demonstrated by compounds showing activity against EV71 in vitro and in vivo. However, available compounds mostly target stem-loop II, whose relatively simple structure may lead to off-target effects. Additionally, the IRES structure has largely been predicted but has not yet been assessed in infected cells. Therefore, this project begins by refining IRES structural predictions through probing viral RNA in infected cells and characterizing its secondary structure using DMS-MaP. The resulting structure enabled the design of a library of LNA-ASOs (locked nucleic acid antisense oligonucleotides), that either unfold or induce degradation of the target RNA. Among the tested ASOs, four exhibit nanomolar activity against RV8 without detectable toxicity in vitro and in human derived respiratory models. Further optimization of these LNA-ASOs, including viral resistance, synergy evaluation and testing across different enteroviruses (including EV68 and EV71), is currently ongoing.

**Student Paper*

P021 Bioactivity-guided identification of clostrenol, a polyketide with antimicrobial activity, from the psychrophilic Candidatus Clostridium mucoides CF011

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The World Health Organization lists antibiotic resistance among the top ten most urgent global health threats and has recommended intensification of efforts aimed at the discovery of novel antibiotics to tackle the challenge. To find new classes of antibiotics, proposals have been made to explore underexploited microorganisms for promising natural products with antimicrobial activity. Therefore, the aim of the current study was to identify novel bioactive natural products from members of the psychrophilic and anaerobic *Clostridium estertheticum* complex (CEC). A bioactivity-guided approach led to the identification of clostrenol, a novel polyketide with broad antimicrobial activity against Gram-positive bacteria. Clostrenol was produced by *Candidatus Clostridium mucoides* CF011. Structural analysis involving mass spectrometry, nuclear magnetic resonance and stable isotopic labelling revealed an unusual scaffold comprising distal α-hydroxy-β-methyl-γ-lactone and α-methyl-β-enol-δ-lactone moieties interconnected by a monounsaturated alkyl chain, representing a previously unreported chemical scaffold. Further studies are being carried out to for indepth characterization its antimicrobial and physiochemical properties. The identification of clostrenol from CF011 highlights the CEC as an underexplored reservoir of chemically unique natural products with promising candidates for future development as antibiotics.

MT11*/P022* Development and validation of a LAMP assay for rapid detection of Escherichia coli associated UTIs with increased risk of urosepsis

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Background: Current culture-based diagnostics for UTIs require up to 24 hours for pathogen identification and do not assess the risk of potential invasiveness. Thereby, clinicians rely on empirical antibiotics before results are available. Inappropriate treatment, however, contributes to the development of antimicrobial resistance (AMR). Uropathogenic *Escherichia coli* (UPEC) are the most common cause of UTIs and virulence factors, e.g., PapGII (adhesin of pyelonephritis-associated pili) are associated with higher risks of urosepsis. We aimed to develop and validate a Loop-Mediated Isothermal Amplification (LAMP)-based assay for rapid detection of *papGII*-positive *E. coli* directly from urine.

Methods: We designed specific primers targeting conserved regions of the *E. coli uidA* and *papGII* genes. Serial dilutions of targets were used to evaluate the limit of detection. To validate the assay, we prospectively collected 191urine specimen samples over a three-day period from routine diagnostics and performed fluorescent LAMP. Culture results from routine diagnostics were used as reference for *E. coli* detection and whole genome sequencing for *papGII* detection. For each target we calculated the optimal thresholds to maximize sensitivity and specificity.

Results: For this assay, the limit of detection was found to be 10^3 CFU/ml. Among the clinical urine samples tested, the assay showed 87% sensitivity and 95% specificity for *E. coli,* and 100% sensitivity and specificity for *papGII*, compared to the reference methods. Notably, all results were obtained within 40 minutes and without extensive sample preparation.

Conclusion: This LAMP-based assay was intentionally simplified to demonstrate the requirements of minimal hands-on work and lab equipment. It shows strong to excellent diagnostic performance for the detection of *E. coli* and *papGII*, respectively, highlighting the potential of early UPEC identification and invasiveness risk assessment. To enhance clinical applicability, future optimization will focus on reducing false positives and negatives.

**Student Paper*

P023 Enhancing treatment against Pseudomonas aeruginosa infection in the bladder tissue

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Despite being among the most prevalent and recurrent diseases worldwide, urinary tract infections (UTIs) are still heavily understudied. They are also major boosters of the current antimicrobial resistance (AMR) crisis and associated deaths, due to massive and successive antibiotic prescription and misuse. However, research in the field is still dominated by animal and cancer cell models that struggle recapitulating urinary tract structure and physiology, impairing effective drug design/application. The situation is even more critical regarding infections by underexplored uropathogens, such as by *Pseudomonas aeruginosa*.

Filling this gap, I am using a 3D urothelial model that recapitulates the human bladder mucosa, combining differentiation and stratification (~60 µm thickness), barrier function, human urine tolerance and tissue innate responses, as testbed for antivirulence approaches. Regarding *P. aeruginosa*, we observed that lectins A and B are important adhesive factors to the bladder tissue surface. Using a glycommimetics approach with the carbohydrate-based molecules ZAA184 and DH181a (100 µM), we were able to inhibit LecA/B binding and efficiently reduce *P. aeruginosa* adhesion up to 3-fold depending on the strain. Additionally, blocking these lectins also significantly decreases cytotoxicity and barrier disruption caused by all P. aeruginosa strains. Although we observed that the isolated lectins are cytotoxic in tissue, we are also revealing whether they can also facilitate the action of other cytotoxic virulence factors. Moreover, *P. aeruginosa* can form large biofilms after longer infection periods (> 8 hours), which tolerate high antibiotic doses, e.g. ciprofloxacin at 50x MIC. To reduce amount/volume of these biofilms we are testing Disperazol®, which promotes c-di-GMP degradation in the bladder tissue. This molecule was particularly effective against larger biofilms (>80 µm3), while enhancing ciprofloxacin action in tissue.

Overall, this human bladder tissue model is a robust human platform for understanding uropathogenesis and as testbed for development of novel antibiotic-sparing therapeutics.

MT12/P024 Spontaneous recapitulation of endogenous colonization resistance following antibiotic therapy and the mechanisms underlying microbiota-mediated community resistance to Clostridioides difficile

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Disruptions to the gut microbiota are widely recognized as the primary driver of susceptibility to *Clostridioides difficile* infection (CDI). Although fecal microbiota transplantation (FMT) effectively treats recurrent CDI, the specific microbial components and molecular mechanisms required to confer resistance remain poorly understood. This hinders the rational development of defined, scalable live biotherapeutic products as alternatives to FMT. Notably, most patients with primary CDI achieve clinical cure following first-line antibiotics, demonstrating that spontaneous restoration of colonization resistance is typically sufficient. Using shotgun metagenomics data from a longitudinal CDI clinical trial, we hypothesized that identifying and experimentally replicating the functional traits of these resistant communities using well-characterized human microbes could inform the design of synthetic communities (SynComs) that mimic this protective phenotype.

We implanted gnotobiotic mice with human microbiota derived from CDI patients post-antibiotic treatment, and subsequently challenged them with *C. difficile*. Susceptibility depended reliably on the specific implanted microbiota. Engrafted communities clustered primarily by donor and showed low inter-mouse variability, enabling reproducible functional phenotyping. Post-therapy microbiomes conferred a broad spectrum of colonization resistance, ranging from minimal to complete pathogen suppression, with post-fidaxomicin communities exhibiting greater resilience and community diversity. These resistance phenotypes were highly donor-specific, reproducible across replicates, and correlated with distinct taxonomic configurations.

Our study establishes a reproducible experimental platform to iteratively manipulate specific taxa and functional pathways, allowing us to untangle the mechanisms driving spontaneous microbiome recovery following CDI therapy. These insights reveal patient-specific recovery trajectories that drive the restoration of colonization resistance and the findings provide a structural framework for the mechanistically informed design of simplified microbial consortia engineered with targeted colonization resistance functions.

P025 The Causes and Consequences of Early-Life Malnutrition

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Perturbations in early-life nutrition program host metabolic physiology and increase susceptibility to metabolic disease later in life. While the gut microbiota is a key regulator of host metabolism, its causal role in linking early-life malnutrition to metabolic dysfunction remains unclear. We used a neonatal mouse model of *in utero* undernutrition or overnutrition followed by post-weaning dietary challenge to investigate host–microbe interactions in metabolic programming. Offspring born to undernourished dams exhibited a sex-dependent increase in post-weaning weight gain and elevated metabolic biomarkers, including liver damage, compared to control and overnourished groups. These phenotypes were accompanied by reduced intestinal compartmentalisation, indicative of altered gut physiology. Notably, germ-free offspring recapitulated the increased weight gain phenotype, suggesting that catch-up growth is microbiota-independent. In addition, 16S rRNA amplicon sequencing revealed distinct microbial signatures associated with maternal malnutrition and early-life succession. Our findings indicate that host-intrinsic mechanisms may predominate in shaping early growth trajectories following prenatal undernutrition.

P026* phagemals – homing endonuclease mediated engineering of bacteriophages

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Bacteriophage engineering has the potential to overcome challenges of phage therapy. This is particularly relevant to *Pseudomonas aeruginosa*, a pathogen renowned for its extensive drug resistances and previous attempts at phage treatement. However, reliable tools to genetically engineer phages are lacking outside of a few model organisms.

Homing endonuclease genes (HEGs) can be found in phage genomes, either as free standing genes or as part of introns. Their proteins induce double-stranded breaks in a target sequence, promoting repair through homologous recombination. This then allows the gene's horizontal spread through a genetic population and across closely related phages. In this study, we identified novel HEGs in the genomes of *Escherichia coli* and *P. aeruginosa* phages and demonstrated their capability of integrating into non-HEG expressing phages at a high efficiency. We have coupled this homing ability with genetic payloads to allow for a simple, one-step insertion of the payloads into the phage's genome without counterselection. We showed that it is possible to efficiently engineer payloads larger than 3000 bp in size with this technology, termed the PhageMal. Furthermore, we have shown that it is possible to decouple the HEG from the payloads, allowing for a one-time insertion of the payload without the HEG, thus preventing further dissemination.

PhageMals present an innovative method of engineering enhanced phages simply and efficiently. Future work will focus on expanding the applicability of this technology to a wider assortment of phages.

**Student Paper*

P027* Kaposi sarcoma associated herpesvirus exploits the m6A demethylase ALKBH5 during lytic infection

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Kaposi sarcoma-associated herpesvirus (KSHV) is a human gammaherpesvirus associated with several malignancies, including Kaposi sarcoma, primary effusion lymphoma, and multicentric Castleman's disease. During lytic infection, KSHV induces a profound remodeling of host gene expression referred to as Host Shutoff. This remodeling is largely driven by the viral endonuclease SOX, which promotes widespread degradation of cytoplasmic mRNAs. Intriguingly, a subset of transcripts escapes this degradation by encoding a cis-acting RNA element known as the SOX-resistant element (SRE). While the SRE protects transcripts from multiple viral endonucleases, the molecular mechanisms underlying this resistance remain poorly understood.
Recent evidence suggests that transcripts that escape SOX-mediated decay may harbor N6-methyladenosine (m6A) RNA modifications. m6A is a highly prevalent epitranscriptomic mark known to regulate RNA stability and metabolism and is frequently exploited by viruses during infection. In KSHV-infected cells, SOX-resistant transcripts appear to undergo m6A specifically during the lytic phase. We set out to understand how m6A modifications regulate SRE-bearing transcripts during KSHV infection.
Our preliminary data indicate that the m6A demethylase ALKBH5 plays an important role during KSHV infection. In contrast to other gammaherpesviruses, ALKBH5 is not degraded during the lytic phase and appears to be required for efficient viral replication, as ALKBH5 depletion results in a marked reduction in virion production. Although ALKBH5 is predominantly localized in the nucleus, it relocalizes to the cytoplasm during lytic infection. Furthermore, mapping of ALKBH5 interactions during lytic infection revealed interactions with several viral proteins. Our work will define the mechanistic role of ALKBH5 in regulating RNA stability and nuclear export during KSHV lytic infection and uncover how KSHV may hijack the m6A erasers to promote infection.

**Student Paper*

P028* Uncovering the diversity of metabolic strategies among soil pseudomonads

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Microbial communities assemble through processes of niche expansion and niche partitioning, in which host-produced and microbially-derived metabolites create a niche space where species compete for resources. Models of microbial interactions and metabolic niche overlap have proven successful at predicting strong qualitative interaction outcomes, but have historically only considered ability over the entire theoretical niche – the space of all possible metabolic functions a species can perform. However, a growing body of knowledge on microbial physiology indicates that rather than simultaneously, resources may be used sequentially in certain contexts, and thus there can be said to be preferences between metabolites, which combine to form an overall metabolic strategy the organism uses to succeed in a community. Understanding this behavior in greater depth - which species compete over which resources in specific contexts - may allow us to better model and predict interaction outcomes and community assembly processes.

In this project, we will map the preferences and metabolic strategies of taxonomically diverse rhizosphere-derived Pseudomonads by profiling their capacity for simultaneous and sequential resource utilization in vitro. By developing high-throughput assays to determine both carbon source utilization and context-dependent preference profiles, we aim to reveal the variety of metabolic strategies within this model genus and inform the generation of improved models of interspecies interactions, thereby increasing our predictive power in complex communities and supporting our ability to engineer the microbiome.

**Student Paper*

P029 Evaluation of Trientine Analogues Trientine Dihydrochloride and Trientine Tetrahydrochloride as Novel NDM Metallo-Beta-Lactamase Inhibitors

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Background: The global dissemination of New Delhi Metallo-β-Lactamase (NDM) represents a major public health threat, driving critical carbapenem resistance in high-priority Gram-negative pathogens. NDM is a metallo-carbapenemase that relies on zinc ions for its catalytic activity. Currently available β-lactamase inhibitors show no activity against NDM. We investigated the antibacterial activity of trientine analogues trientine dihydrochloride (T2H) and trientine tetrahydrochloride (T4H), known copper chelators used to treat Wilson's disease, as novel NDM inhibitors, hypothesizing that they bind to or displace the essential zinc ions.

Methods: NDM variants: NDM-1, -2, -4, -5, -7, -9, -14 and -19 were cloned into vectors pUCP24 or pVRL1, and transformed into *Escherichia coli* MG1655, *Pseudomonas aeruginosa* PAO1, and *Acinetobacter baumannii* CIP70.10 strains. Minimal inhibitory concentrations (MICs) were determined following EUCAST methodology. Synergistic activity between meropenem and trientine analogues was evaluated via checkerboard assays. Time-kill kinetic studies were performed on *E. coli*-producing NDM-1. IC_{50} values were determined for NDM-1, NDM-5, IMP-1 and VIM-1 against both analogues.

Results: In *E. coli*, synergy was observed at 40 mg/L for 7/8 NDM-variants with T2H, and 50 mg/L for 6/8 NDM variants for T4H. For *P. aeruginosa*, synergy was observed at 250 mg/L in 7/8 and 8/8 NDM variants with T2H and T4H, respectively. Finally, in *A. baumannii*, synergy was observed at 60 mg/L for 6/8 NDM-variants with T2H, and 75 mg/L for 5/8 NDM variants for T4H. Time-kill curves showed that meropenem in combination with T2H at a concentration of 20 mg/L and T4H at a concentration of 50 mg/L exhibited a synergistic effect over a 8 to 24 hour period. Enzymatic inhibition assays confirmed NDM-specific inhibition.

Conclusions: T2H and T4H, successfully restored meropenem activity against high-priority multidrug-resistant pathogens. Given their established safety profiles, these chelators represent a promising strategy for combination therapies to combat carbapenem-resistant infections in clinical settings.

P030 Beneficial impact of acquired AmpC CMY-type β-lactamases on bacterial fitness and pathogenicity: a new paradigm

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Background

AmpC-type β-lactamases share structural similarities with penicillin-binding proteins, supporting the view that both enzyme families evolved from a common ancestor. Considering the widespread dissemination of acquired AmpCs in Gram-negative bacteria, we hypothesize that, besides their established role in β-lactam resistance, they may also influence essential bacterial mechanisms. Our study aimed to elucidate whether acquired CMY enzymes, corresponding to the most commonly-identified plasmid-encoded AmpCs in Enterobacterales, could modulate bacterial physiology and fitness beyond conferring resistance to β-lactam antibiotics.

Materials

E. coli MG1655 was used as the model organism to construct isogenic recombinant strains using pTOPO-Blunt plasmid bearing a series of clinically-relevant CMY-type *ampC* genes, namely bla_{CMY-2}, bla_{CMY-42}, $bla_{CMY-145}$. A series of experiments were performed to better understand the impact of these clinically relevant acquired AmpCs on growth, competition, motility, biofilm formation, and in vitro and in vivo pathogenicity (serum resistance and *Zophobas morio* larvae, respectively), coupled with transcriptomic profiling.

Results

Our experiments showed that some CMY variants (notably CMY-42 and CMY-145) conferred an unexpected in-vitro growth advantage upon measurement of growth curves. Those variants also provided advantages during competition assays performed with isogenic counterpart harboring the same plasmid but without ß-lactamase gene and induced increased biofilm production. Transcriptomic assays showed downregulation of a master regulator of a flagellar biosynthesis cluster. Motility assays and electron microscopy indeed confirmed loss of flagellar biosynthesis upon production of CMY-42 and CMY-145, subsequently generating energy saving. On the other hand, even though showing an in-vitro growth disadvantage, production of CMY-2 induced flagellar production and subsequently enhanced pathogenicity using in-vitro (serum resistance) and in-vivo (larvae infection).

Conclusions

These findings establish a new paradigm in which some AmpCs act as both resistance determinants and regulators of bacterial fitness and virulence, through modulation of flagellar biosynthesis.

P031* Heterotrophic Bacteria Facilitate Methanogen Survival under Oxygen Stress through Metabolic Cooperation

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Methanogens are key contributors to global carbon cycling but are generally considered highly sensitive to oxygen. In natural environments, however, methanogens frequently coexist with facultative aerobic heterotrophs under fluctuating redox conditions. Here, we investigated whether heterotrophic bacteria facilitate methanogen survival under oxygen stress through metabolic cooperation.

Using co-cultures of *Methanococcus maripaludis* and the facultatively aerobic bacterium *Vibrio alginolyticus*, we found that methanogenesis could be maintained during repeated low-level O_2 exposures. Co-cultures rapidly consumed oxygen and sustained methane production under intermittent 2% O_2 pulses, whereas higher oxygen concentrations caused partial or strong inhibition. Notably, methanogenesis and oxygen consumption persisted in the absence of externally supplied organic carbon, suggesting a stable metabolic interaction between the two organisms. Transcriptomic analyses revealed substantial transcriptional changes in both partners during co-culture. In *V. alginolyticus*, pathways related to sulfur assimilation, hydrogen sulfide biosynthesis, central carbon metabolism, and oxidative stress responses were significantly enriched, supporting a role for sulfur exchange and cooperative stress adaptation.

To directly trace sulfur transfer between the two organisms, we performed stable isotope labeling experiments using ^{34}S-labeled sulfate. *V. alginolyticus* was first cultivated with ^{34}S sulfate and subsequently co-cultured with *M. maripaludis* in sulfate-limited conditions. NanoSIMS imaging revealed clear ^{34}S enrichment within *M. maripaludis* cells, providing direct evidence that reduced sulfur derived from *V. alginolyticus* was transferred to the methanogen. Interestingly, no external organic carbon source was supplied to sustain *V. alginolyticus* in the co-culture system, raising the possibility that carbon compounds released *by M. maripaludis* supported heterotrophic bacterial growth. This suggests that the consortium may establish a largely self-sustaining carbon and sulfur exchange network under microoxic conditions.

These findings demonstrate that facultative heterotrophic bacteria can protect methanogens from oxygen stress while simultaneously supplying essential reduced sulfur metabolites, highlighting the ecological importance of interspecies metabolic cooperation in sustaining methanogenesis in microoxic environments.

**Student Paper*

P032 Machine-Learning-Enabled Peptide Discovery Coupled to Translational Infection Models for Antimicrobial Therapeutic Development

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Drug-resistant bacterial infections require therapeutic strategies that combine molecular precision with biologically meaningful validation. This work presents an integrated antimicrobial peptide-discovery pipeline for designing immune-effector-derived peptides, with particular focus on granzyme A, granzyme B and granulysin. The central advance is the coupling of protein-guided peptide design with multi-stage computational filtering and infection-relevant experimental models.

The pipeline integrates curated antimicrobial and non-antimicrobial peptide datasets with descriptor-based analysis and sequence-level learning. Candidate peptides are produced through protein fragmentation, functional-domain mining, motif grafting, template mutation, Markov assembly, genetic algorithms, simulated evolution and neural sequence generation. Each sequence is assessed for features linked to antimicrobial performance and host compatibility, including length, net charge, amino acid composition, amphipathicity, hydrophobicity, membrane interaction, stability, aggregation tendency, novelty and haemolysis risk.

Descriptor-based ensemble models identify patterns associated with activity and selectivity. CNN-BiLSTM and RNN-based classifiers complement this by capturing antimicrobial motifs, residue dependencies and broader sequence organisation. Peptides are ranked using a multi-parameter score that favours predicted activity, structural plausibility, diversity and reduced host-cell liability.

The highest-ranked candidates undergo mechanistic in silico assessment. Docking against selected bacterial protein targets estimates binding affinity, interaction geometry and target engagement, while molecular dynamics simulations examine complex stability, bacterial membrane association and peptide orientation. Computationally favourable candidates progress to in vitro testing against *Listeria* spp., *Staphylococcus aureus*, MRSA, VRSA, *Acinetobacter baumannii* and ESBL-producing bacteria, using MIC, time-kill, antibiofilm, membrane-perturbation and mammalian-cell toxicity assays.

Promising leads advance to organoid-based infection models to assess epithelial compatibility, barrier integrity, pathogen clearance and host-response modulation, followed by animal studies evaluating early efficacy, tolerability and translational feasibility. This staged pipeline prioritises peptides with both predicted antimicrobial potency and a credible biological rationale for preclinical development.

P033* Harmonizing extended-spectrum β-lactamase classification: a systematic re-evaluation of database annotations

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Introduction

Extended-spectrum β-lactamases (ESBLs) represent a clinically important group of β-lactam-hydrolyzing enzymes, but their annotation across reference databases remains inconsistent. A universally accepted ESBL consensus is still lacking, and classification approaches remain challenging due to overlapping phenotypes, evolving nomenclature, and the continuous emergence of new variants. Earlier efforts to expand classification illustrate how definitions have shifted as resistance determinants evolved. To resolve these discrepancies, we systematically re-evaluated all genes annotated as ESBLs in the Beta-Lactamase Database (BLDB) and the NCBI Pathogen Detection Reference Gene Catalog (RefCat) based on primary literature.

Methods

All β-lactamase genes were systematically retrieved from the BLDB and NCBI RefCat. Candidate genes with discordant annotations were evaluated through manual literature review from primary experimental studies. For bla_{ADC}, bla_{GES} and bla_{SHV} genes, additional ESBL prediction was based on previously described amino acid substitutions associated with phenotype conversion. Genes were classified using a standardized, literature-based framework into: classical ESBLs, OXA-ESBLs, ESBL-related enzymes (including inhibitor-resistant ESBLs and minor ESBL families), other third-generation cephalosporin-active β-lactamases, and entries lacking sufficient evidence.

Results

Among 1,400 β-lactamase genes annotated as ESBLs in at least one database, only 63.4% (887/1,400) carried matching ESBL labels in both databases. Manual curation resolved 562 discrepancies, underscoring substantial database discordance and the need for publication-based annotation. Additional review of selected gene families originally annotated as non-ESBL further revealed variants that did not belong to the non-ESBL category. The final curated dataset included 543 classical ESBLs, 30 OXA-ESBLs, 260 ESBL-related enzymes, 64 other third-generation-cephalosporin-active β-lactamases, and 813 non-ESBLs.

Conclusions

Applying a standardized, literature-grounded framework enabled the resolution of major annotation conflicts across reference databases. This curated framework improves the reliability of ESBL annotation for clinical diagnostics, genomic analyses, and molecular epidemiology, and highlights the need for continuous updates as new variants emerge.

**Student Paper*

P034 Deciphering the Non-Canonical Endocytic Trafficking of Chlamydia trachomatis: A Rab14-Dependent Pathway

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Introduction

Chlamydia trachomatis is an obligate intracellular pathogen that evades host degradation pathways by entering and replicating within a membrane-bound compartment known as the inclusion. Unlike many intracellular pathogens, *C. trachomatis* does not rely on the canonical Rab5/Rab7 endosomal maturation cascade. Instead, it traffics via a Rab14-dependent route, bypassing degradative lysosomal compartments. This pathway remains poorly understood.

Objectives

We aim to characterize the Rab14-dependent trafficking pathway exploited by *C. trachomatis* and to identify host factors involved in this alternative endocytic route, with the goal of uncovering potential targets for intracellular therapeutic intervention.

Materials and Methods

CRISPR/Cas9 knockout cell lines and dominant-negative Rab constructs were used to assess the roles of Rab5 and Rab7 in bacterial entry and inclusion formation. Colocalization studies were performed to examine the association of bacterial inclusions with Rab14 and EEA1. To identify host factors regulating this pathway, Rab14 pull-down assays were coupled with mass spectrometry analysis. In parallel, a high-throughput compound screen was conducted to identify molecules capable of reducing *C. trachomatis* infection.

Results

Our results demonstrate that Rab5 and Rab7 are dispensable. Instead, *C. trachomatis* colocalizes with Rab14 and EEA1, even in absence of Rab5. These findings support emerging evidence that early endosomes marked by EEA1 can form independently of Rab5, suggesting the existence of an alternative endocytic pathway. Proteomic analysis identified a network of Rab14-associated host proteins whose depletion significantly reduces bacterial infectivity. Additionally, the compound screen revealed over 90 molecules that decrease infection rates, including drugs not traditionally associated with antimicrobial activity.

Conclusions

These findings uncover a previously underappreciated Rab14-dependent trafficking route that is critical for *C. trachomatis* intracellular survival. By elucidating the mechanisms and host factors involved in this pathway, this work provides new insights into bacterial pathogenesis and highlights potential targets for therapeutic strategies aimed at disrupting infection at early stages.

P035* Quantitative metagenomic sequencing using whole-cell spikes for environmental resistome analysis

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Next-generation metagenomic sequencing (NGS), is quickly becoming a preferred methodology for environmental antimicrobial resistance (AMR) research. Unlike qPCR, NGS does not rely on *a priori* knowledge of targets and can screen for thousands of resistance genes at once. However, NGS is still considered "semi-quantitative" in that it only provides relative quantification. This can lead to misinterpretations as proportional differences do not necessarily translate to "absolute" differences (i.e., concentration per unit volume). We propose to use a whole-cell spike, added at the beginning of the metagenomic sequencing workflow, as a positive reference and recovery standard for calculating absolute abundances of ARGs and understanding biases throughout the workflow.

The ZymoBiomics™ Spike-in Control I was added to wastewater and surface waters samples at a 1% cell/cell ratio based on total cell counts enumerated using flow cytometry. The spike consists of equal concentrations of 2 organisms (*Imtechella halotolerans* and *Allobacillus halotolerans)* exogenous to most environmental microbiomes but detectable in metagenomes based on reference genomes and qPCR. Samples then underwent a typical metagenomic workflow and Illumina sequencing.

Across all samples, sequencing reads assigned to the Zymo spike organisms were detected at less than the expected 1% ratio. Choice of bioinformatic pipeline and the water matrix (wastewater versus surface water) affected abundance and apparent ratio of spike organisms, e.g., Kraken2 + Bracken was biased towards *Allobacillus* compared to Metaphlan4. These results will help clarify and reduce biases originating in the sample processing and sequencing workflow. By quantifying spike retrieval, DNA extraction efficiency can be estimated. The concentrations of targets of interest (e.g., antibiotic resistance genes) can be calculated by comparing target sequencing reads to the ratio of the Zymo spike reads / Zymo spike concentration. This will allow researchers to reap the benefits of metagenomic sequencing without sacrificing quantitative power, better enabling quantitative risk assessment.

**Student Paper*

P036* Legionella pneumophila subverts mitochondrial dynamics and mtDNA quality control to promote intracellular replication

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Intracellular bacteria manipulate and subvert cellular processes in their eukaryotic hosts in a sophisticated manner. While the facultative intracellular pathogen *Legionella pneumophila* naturally replicates in free-living amoeba, inhalation of this pathogen can lead to a severe pneumonia due to the ability of the bacterium to replicate in alveolar macrophages. During the intracellular state, *L. pneumophila* secrets over 300 "effector proteins" into their respective hosts. This enables the formation of a *Legionella*-containing vacuole (LCV) by subverting phagosome maturation and other cellular processes. While LCV formation is essential for the survival and replication of the pathogen, many other host cellular processes are targeted and subverted to further support the survival and replication of intracellular *L. pneumophila*.

Mitochondrial function and dynamics are essential for virtually all cellular processes. Not surprisingly, intracellular bacteria such as *L. pneumophila* specifically target and modulate mitochondrial components and their function. In this study, we identified several *L. pneumophila* effector proteins localizing to and modulating mitochondria. Some *L. pneumophila* deletion mutant strains lacking individual mitochondria-targeting effector proteins are impaired for intracellular replication within host cells and inhibit host cell respiration less efficiently compared to the parental strain.

We are currently investigating the mode of action of a novel secreted mitochondria-targeting *L. pneumophila* effector kinase. The kinase contains an N-terminal mitochondria targeting sequence, which is required and sufficient for mitochondria localization, where it targets the MTFP1 (mitochondrial fission process 1) protein implicated in mitochondria fission. Thus, the *L. pneumophila* kinase subverts critical steps in mitochondrial dynamics and mtDNA maintenance, revealing a novel post translational regulation of a critical host protein.

**Student Paper*

P037 Targeted delivery of vaccine antigens using a safe and programmable bacterial chassis

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Respiratory infectious diseases of livestock impact animal health, productivity and global health. Vaccination remains the most cost-effective control measure to prevent infectious diseases. Of particular interest are vectored vaccines that induce an immune response in the target tissue. Such vectored vaccines are based on genetically attenuated viruses or bacteria, and they offer the possibility to express many antigens in one single vaccine, making them affordable and facilitating vaccination programs. *Mycoplasma* species are safe options for use since they strictly depend on host-derived nutrients and have an atypical genetic code. Additionally, due to their lack of a cell wall, they efficiently express foreign antigens. We aim to develop a fast-growing *Mycoplasma feriruminatoris* strain isolated from Alpine ibex into a vaccine chassis that expresses different viral and bacterial antigens to induce immunity in livestock species.

We engineered the *Mycoplasma feriruminatoris* strain with a landing pad to allow swift and stable integration of synthetic DNA into the bacterial chromosome and to subsequently drive stable surface expression of different viral and *Mycoplasma* antigens. Selected antigens to be expressed include the N-terminal region from matrix 2 protein (M2e) and hemagglutinin (HA) from Influenza A virus, as well as the capsid protein of porcine circovirus 2 (PCV2), and the adhesion protein P97 of *Mycoplasma hyopneumoniae.* Preliminary data confirms surface expression of antigens by using a lipoprotein anchor, as shown by immunoblot, and will be tested with expansion microscopy as well. We will conduct safety and immunogenicity *in vivo* studies in different livestock animals and have started pilot experiments to express selected secreted cytokines (e.g. IFN-λ3) to modulate host immune response. This project, aiming to develop a safe and easy to produce livestock vaccine chassis, could contribute to control programmes against epizootic and zoonotic diseases thus improving animal welfare and global health.

P038 New insights into Lamassu bacterial defense systems

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Bacterial evolution is often shaped by the uptake of extracellular DNA through horizontal gene transfer. While this process can provide adaptive advantages, it can also threaten host survival. To counteract such threats, bacteria have evolved diverse defense systems, including Lamassu (Lmu) systems. A subset of Lmu systems has been shown to protect against both plasmids and phages by triggering abortive infection, thereby sacrificing the infected host cell for the benefit of the surrounding bacterial population (1-4).

Lmu systems generally consist of a conserved DNA-sensing SMC-like ATPase (LmuB), an optional accessory subunit (LmuC), and an effector protein (LmuA) (2). The diversity of Lmu systems primarily stems from the variability of LmuA effector proteins, with nine subclasses identified through bioinformatic analyses to date. However, most studies so far have focused on Lmu systems associated with Cap4 nuclease effectors, whereas other putative effector proteins remain largely uncharacterized, limiting our understanding of the broader functional diversity of Lmu systems (1-4).

In this study, we initiated the characterization of a novel Lamassu system harboring an unprecedented effector domain that has not previously been described before in any known defense system. Using bioinformatic analyses, we first identified bacterial genomes encoding this system and subsequently screened several homologous candidates for activity in the heterologous host *E. coli.* Protein domain analyses together with AlphaFold structural predictions of the selected LmuA candidates suggested the involvement of putative cofactors. Notably, mutation of amino acids predicted to coordinate these cofactors impaired the toxic activity of the effector protein. Ongoing work aims to further characterize this novel Lamassu system at both the molecular and cellular levels.

P039 Enhancing lysosomal function in bladder epithelium potentiates antibiotic clearance of intracellular uropathogenic Escherichia coli

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Urinary tract infections (UTIs) are among the most common bacterial infections, accounting for 400 million cases globally each year. Treatment of UTIs is complicated by the ability of uropathogenic *Escherichia coli* (UPEC) to invade bladder epithelial cells and persist intracellularly. Within these intracellular niches, bacteria evade killing by host defenses and antibiotics, resulting in high rates of recurrence post-treatment. Despite the central role of the bladder epithelium in these processes, epithelial cell-intrinsic antimicrobial mechanisms remain poorly explored as therapeutic targets.

Here, we show that the bacterial lysate OM-89 (Active Pharmaceutical Ingredient of Uro-Vaxom®), a preventive therapy for recurrent UTIs, directly enhances cell-intrinsic antimicrobial defenses in the bladder epithelium. Using cultured bladder epithelial cells and organoids, we demonstrate that OM-89 stimulates endolysosomal function by increasing lysosome numbers, acidification and protease activity. These changes result in exposure of internalized UPEC to lysosomal compartments with enhanced degradative activities, which could render the bacteria more sensitive to attack by antibiotics.

Consistent with this idea, we show that OM-89 treatment enhances the killing of intracellular UPEC by antibiotics and suppresses bacterial regrowth after antibiotic withdrawal. These effects are reversed by drugs that block lysosome acidification. We further show that OM-89 enhances intracellular accumulation of antibiotics, which may contribute to increased killing of intracellular bacteria. These effects are observed for multiple classes of antibiotics and phylogenetically diverse UPEC strains, including clinical isolates from UTI patients.

In conclusion, our findings uncover a lysosome-centered epithelial mechanism that enhances intracellular antibiotic efficacy and limits post-treatment bacterial regrowth. By providing a mechanistic basis for the clinical activity of OM-89, this work highlights the bladder epithelium as a therapeutically targetable compartment to improve treatment outcomes in recurrent UTIs.

P040 Elucidating cell envelope architecture and remodelling in Corynebacterium glutamicum

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Growth and division of bacterial cells is tightly connected to the assembly of multilayered envelopes containing lipids, glycans and proteins. In contrast to most studied rod-shaped bacterial envelopes, members of the order Corynebacteriales deviate both in envelope architecture and cell wall growth mode. They assemble a waxy outer lipid bilayer – the mycomembrane (MM) - linked to the peptidoglycan (PG) via an arabinogalactan (AG) mesh. In contrast to dispersed elongating bacteria such as *E. coli* or *B. subtilis*, newly synthesized PG is incorporated at polar growth zones. Corynebacteriales divide by V-snapping, a multistage process where first the new envelope layers are synthesized and bisect the entire cell, before being rapidly separating and exposing the new polar caps. The molecular mechanisms driving the remodelling of these cell envelope layers during division remains largely unknown. Furthermore, how these different layers contribute to general morphogenesis in polar elongating bacteria remains unknown.[PN1] Here, we show using a combination of bacterial genetics, live-cell fluorescence microscopy and *in situ* cryo-electron tomography how the absence of different surface layers influences septal architecture, affect the underlying cell morphology, and growth rate in the mycobacterial surrogate model organism *Corynebacterium glutamicum*. Specifically, we focus on mutants lacking key components of the MM, a truncated AG mesh and MM, a PG synthase involved in polar growth, as well as a glycolipid layer that we visualize between inner membrane (IM) and PG. We observe that rod morphology requires fully assembled MM and septum resolution is hindered in cells without the IM-associated glycolipid layer. Our preliminary results demonstrate the complexity of surface layer assembly in Corynebacteriales and allow for further insights on the spatiotemporal remodelling dynamics of the cell envelope during division.

P041* ecological determinants of host specificity in the gut microbiota of social bees

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Gut bacteria often form long-standing and specialized associations with their host. As a result, microbiota composition is more similar within than between host species: a pattern known as host specificity. Yet, the ecological mechanisms underlying this pattern remain poorly understood.

Social bees (honeybees, bumblebees, and stingless bees) have recently emerged as powerful models for gut microbiota research due to their simple, conserved, and experimentally tractable gut microbiota. While similar in overall composition, these bacterial communities are usually composed of host-specific bacterial species and strains, making them an ideal system to study host specificity.

We hypothesized that host filtering is a major driver of the host-specific distribution of bee gut bacteria. To test this hypothesis, we conducted a cross-species microbiota transplant experiment in which we colonized microbiota-free western honeybees (*Apis mellifera*) with non-native gut communities originating from 5 honeybee, 5 bumblebee, and 5 stingless bee species. We found that non-native communities failed to reach the bacterial loads achieved by the native *A. mellifera* community. Moreover, non-native communities experienced a stronger loss of diversity relative to the native community. Strikingly, the strength of host filtering was positively correlated with the phylogenetic distance between donor and recipient hosts and did not systematically affect similar bacterial taxa.

Together, our results provide new insights into the ecological factors that govern the assembly and maintenance of gut microbiota across social bee species and highlight the central role of host filtering in shaping patterns of host specificity.

**Student Paper*

P042 functionally-derivatized nanoantibodies as novel tools for giardia lamblia cyst detection

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Giardia lamblia (also known as *intestinalis* or *duodenalis*) is the pathogen responsible for Giardiasis, a globally significant diarrheal disease that particularly affects young children in unhygienic and resource-limited environments.

Traditional diagnosis relies on light microscopy of stool samples, which is often inadequate. More sensitive ELISA-based fluorescence assays exist but depend on expensive proprietary reagents that are largely inaccessible in the regions most burdened by Giardiasis, restricting their use primarily to affluent areas where the disease is less prevalent and less problematic.

To improve diagnostic equity and advance tools for studying the Giardia cyst wall, we developed and characterized single-domain antibodies (nanoantibodies) derived from alpacas, generated against enriched Giardia cyst-wall preparations.

We tested twelve unique recombinant nanoantibody sequences produced in *E. coli* for their ability to detect Giardia cyst walls, demonstrating their strong binding capacity and highlighting the potential of these versatile protein domains for improved, cost-effective and 3R-inspired Giardia detection and research applications.

These findings were published this year in PLoS-NTDs:

https://journals.plos.org/plosntds/article?id=10.1371/journal.pntd.0013844

P043 Improved Disc Diffusion–Based Detection of ESBL Production in Klebsiella oxytoca complex

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Background: Strains of the *Klebsiella oxytoca* complex carry chromosomal class A OXY-β-lactamases, which share structural and functional similarities with plasmid-mediated ESBLs. Hyperproduction of OXY (hyperOXY) can mimic ESBL-phenotypes, complicating routine interpretation. Importantly, confirmed ESBL-production carries therapeutic as well as infection control implications. We analyzed antimicrobial susceptibility testing (AST) results from genetically characterized *K.oxytoca* complex clinical isolates to facilitate reliable differentiation between hyperOXY- and ESBL-phenotypes using disc diffusion (DD), while reducing unnecessary confirmatory testing.

Methods: A total of 123 clinical isolates collected in Zurich (2009–2024) were analyzed, including 61 *K.oxytoca* (bla_{OXY-2}), 57 *K.michiganensis* ($bla_{OXY-1/-5}$), four *K.grimontii* (bla_{OXY-6}), and one *K.pasteurii* (bla_{OXY-4}). Selection was based on ceftazidime non-resistance to ensure phenotypic diversity. Whole-genome-sequencing identified 26 ESBL producers (bla_{SHV-12}, bla_{CTX-M}, bla_{MUN}), 12 AmpC-producers (bla_{DHA-1}, bla_{DHA-7}, bla_{CMY-4}, bla_{ACC-1}), including six isolates co-producing ESBLs, and 85 isolates lacking ESBL/AmpC genes. DD testing included amoxicillin-clavulanic acid (AMC), piperacillin-tazobactam (TZP), ceftriaxone, cefpodoxime, ceftazidime, cefepime, cefoxitin, and gentamicin. Synergy (AMC with ceftriaxone/ceftazidime/cefepime) and antagonism (cefoxitin–ceftriaxone) were assessed using a 24 mm center-to-center disk distance.

Results: Sixty isolates displayed a typical hyperOXY profile characterized by ceftazidime non-resistance and progressively increasing β-lactam resistance involving TZP/AMC, followed by ceftriaxone, cefpodoxime, and cefepime. HyperOXY-associated inhibition was observed in 10/64 isolates during synergy testing. Three isolates demonstrated phenotypes consistent with inducible plasmid-mediated AmpC production. Twelve ESBL-producers displayed a distinct phenotype requiring no additional testing, while 15 isolates showed combined hyperOXY and ESBL features but remained attributable to ESBL-production based on synergy. Thirty-four isolates resistant to nearly all β-lactams could not be reliably classified and required additional testing.

Conclusion: The proposed DD-based AST reference table provides a practical tool to distinguish hyperOXY- from ESBL-phenotypes in the *K. oxytoca* complex and may reduce unnecessary confirmatory ESBL testing in routine diagnostics.

P044 Locating Escherichia coli in Human Bladder Tissue during Acute and Recurrent UTI

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Urinary tract infections (UTIs) are a major global health burden and a key driver of antimicrobial resistance, with *Escherichia coli (E. coli)* as the predominant causative pathogen. Up to 50% of patients experience recurrent UTIs (rUTIs), often caused by the same bacterial strains, suggesting the presence of persistent bacterial reservoirs. However, the localisation of these reservoirs in human bladder remains poorly defined.

Here, we investigate bacterial persistence and host-pathogen interactions in urinary bladder biopsies from patients with acute or recurrent UTI in absence of antimicrobial therapy. Using immunofluorescence and confocal microscopy, we localised *E. coli* within the bladder tissue, assessed bacterial morphology, aggregation, and filamentation, and distinguished intracellular versus extracellular niches. Bacterial translational activity will be evaluated using RNA fluorescence in situ hybridisation (RNA-FISH) with rRNA-targeted probes. Additionally, we characterise tissue inflammation, relevant host cell types, and epithelial barrier integrity.

Our results demonstrate scattered *E. coli* bacteria across different layers of the bladder wall. Ongoing analyses assess quantitative bacterial distribution, viability, morphology, ribosome content, aggregation state, and relevant host microenvironments. These data will reveal the spatial organisation and physiological state of *E. coli* and corresponding host responses in human bladder tissue. The insights will provide a basis for improving microtissue models for mechanistic investigations.

P045 Dose-Dependent Restoration of Aminoglycoside Activity by Exogenous Hydrogen Sulfide in Carbapenem-Resistant Pseudomonas aeruginosa

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Background

Carbapenem-resistant *Pseudomonas aeruginosa* (CRPA) represents a major public health challenge due to frequent treatment failure when associated with human infections. Currently, the lack of newly developed compounds leaving few therapeutic options for treating these infections.. Aminoglycosides such as tobramycin and amikacin are recommended. Hydrogen sulfide (H_2S), an endogenous signaling molecule produced by several bacterial species such as *P. aeruginosa*, can modulate bacterial physiology, but its impact on antibiotic susceptibility remains unclear. The aim of this study was to evaluate if exogenous H_2S could enhance aminoglycoside activity in CRPA isolates.

Methods

Twenty-seven clinical CRPA isolates carrying diverse β-lactamases and aminoglycoside resistance genes were evaluated. Minimal inhibitory concentrations (MICs) for amikacin and tobramycin were determined in Luria-Bertani medium (LB) alone or in combination with exogenous H_2S supplied as sodium hydrosulfide (NaSH) at 0.1 mM, 0.25 mM, and 1 mM. Synergy was defined as a ≥ 3-fold reduction in MIC (MIC alone / MIC + H_2S). Time-kill assays were performed on a representative resistant isolate to validate the bactericidal effect of the most active combinations.

Results

H_2S alone did not alter MIC values, however, time-kill assays revealed a concentration-dependent impact on bacterial growth dynamics. No significant change was observed at 0.1 mM H_2S when combining with aminoglycosides. At 0.25 mM, sensitivity was restored in 10/27 isolates (37%) for tobramycin and 4/27 isolates (15%) for amikacin. The effect improved markedly at 1 mM H_2S, with significant MIC decrease in 19/27 isolates (70%) for tobramycin and 16/27 isolates (59%) for amikacin. Time-kill kinetics confirmed the concentration-dependent effect of amikacin by H_2S.

Conclusions

Exogenous H_2S significantly enhances aminoglycoside activity against CRPA in a concentration-dependent manner, reversing resistance of clinical isolates. These findings support the potential of H_2S-based adjuvant strategies, particularly for localized applications aimed at preventing bacterial colonization of medical devices.

P046 Whole-genome sequencing-based characterization of human Listeria monocytogenes

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Background: *Listeria monocytogenes* is a foodborne pathogen that poses a significant public health risk due to the high case-fatality rate of listeriosis.

Methods: We used Illumina technology-based sequencing to determine the population structure and virulence traits of 335 *L. monocytogenes* isolates from human cases of listeriosis occurring in Switzerland between 2019 and 2024.

Results: The five major clonal complexes (CCs) were CC1, CC4, CC6, CC8, and CC388. The isolates belonged to 35 clusters, including three large outbreak clusters. Nine smaller clusters matched to food isolates obtained earlier. Multiple singly occurring isolates were genetically related to outbreaks strains reported abroad. Hypervirulence pathogenicity islands (LIPI-3 and LIPI-4) were found predominantly among CC1, CC4, and CC6 and CC388.

Conclusions: Our data provide insight into the genomic characteristics of clinical *L. monocytogenes* in Switzerland.

P047* Spatiotemporal dynamics of cell wall hydrolases in Escherichia coli

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Spatiotemporal dynamics of cell wall hydrolases in *Escherichia coli*

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- Andrea Vettiger (Department of Fundamental Microbiology, University of Lausanne, Lausanne)

Abstract

The Gram-negative bacterium *Escherichia coli* possesses a multilayered cell envelope with a peptidoglycan (PG) cell wall that determines cell shape and provides structural integrity. The cell wall forms an exoskeletal meshwork called the sacculus that protects the cell against osmotic rupture and determines its shape and thus is required for bacterial survival. Therefore, understanding the PG biogenesis is vital for identify target for producing antibiotics and therapeutics.

During cell growth and division PG synthases and hydrolases turn over up to 50% of the existing cell wall material in the sacculus. However, during this process, the cell poles are completely inert to any cell wall remodelling. Why polar PG is not processed by cell wall hydrolases although these enzymes are universally present in the cell remains unknown.

As part of my PhD project, I aim to understand why polar PG is not degraded by cell wall hydrolases. To this end, we are performing muropeptide analysis of mini cells (entirely composed of polar PG), regular rod-shaped cells and filamenting cells (over-representation of sidewall PG) to reveal potential differences in the cell wall architecture and composition between poles and side wall PG. Moreover, I am currently performing *in vitro* assays with a set of distinct classes of purified cell wall hydrolases to test whether polar PG can be digested in principle. Last, we are currently comparing the proteome of rod-shaped cells with mini-cells to identify regulators which are absent or underrepresented at the cell pole.

Overall, I will be presenting our first results, challenges and conclusions since the start of this endeavour.

**Student Paper*

P048 Squiggles to the rescue: Metagenomics-based pathogen surveillance

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Genomics has become an integral part of One Health surveillance for identifying pathogen and resistance outbreaks. Nanopore sequencing technology holds the promise of implementing rapid genomic sequencing at the point of care and potentially globally due to the low upfront investment costs. I will first present our contributions to benchmarking whole-genome sequencing of foodborne and clinical pathogens by nanopore sequencing in comparison with established diagnostics and traditional short-read sequencing technologies. I will then describe our research on metagenomic surveillance by nanopore sequencing, which—unlike culture-based whole-genome sequencing—can be applied rapidly to any sample type and has the potential to holistically assess the presence of any pathogen, their genomic variation, and their viurlence and antimicrobial resistance. By focusing on foodborne and clinical bacterial pathogens, I will show how nanopore sequencing data and AI-informed can address the existing limitations of metagenomics in inferring clinical and public health consequences that currently preclude its uptake for real-world pathogen surveillance—in terms of sensitivity, viability inference, and associations of virulence and resistance genes to their pathogenic hosts. I will, for example, demonstrate how epigenetic information can deliver robust plasmid-host associations, and showcase the potential of nanopore technology-based metagenomics in application to real-world datasets and in direct comparison with established diagnostics.

P049 Investigating SHFL-dependent regulation of RNA and viral gene expression during KSHV infection

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The Kaposi sarcoma-associated herpesvirus (KSHV, HHV-8) is typically asymptomatic, however, under immunocompromised conditions, its infection can lead to lymphoproliferative disorders. To replicate, KSHV extensively reprograms the host gene expression landscape by inducing a widespread degradation of up to 70% of cytoplasmic mRNAs. Yet, we have identified a subset of mRNAs stringently escaping this viral-induced degradation, among which is C19ORF66 (Shiftless, SHFL). This is particularly intriguing, as SHFL is known to be an interferon-stimulated gene with a broad antiviral activity that relies on the modulation of RNA and protein stability. For instance, we showed that SHFL is able to target key viral proteins, such as the latent-to-lytic switch protein ORF50 (RTA), and the KSHV RNA-binding protein ORF57. In addition, SHFL has been implicated in the disruption of RNA processing bodies (PBs), highlighting a potential role in dysregulating the initiation of the KSHV lytic gene cascade. Here, we aim to further characterize the role of SHFL in KSHV infection by identifying host factors that mediate its antiviral activity and by investigating the broader cellular consequences of SHFL-dependent remodeling of PBs. To this end, we are generating a SHFL knockout (SHFL-KO) iSLK cell line to assess the impact of SHFL loss on viral gene expression under both lytic and latent conditions. In parallel, we are examining the interplay between SHFL and KSHV lytic reactivation in B cells, providing a more physiologically relevant model of infection in lymphocytes compared to epithelial systems. Looking ahead, we will compare the interactome of wild-type SHFL with that of a mutant known to lose its anti-KSHV activity, to elucidate the molecular basis of SHFL-mediated PB dynamics. Together, these approaches will provide insights into how SHFL coordinates antiviral defense mechanisms with RNA regulation, and how this interplay influences KSHV reactivation and pathogenesis.

P050 Genetic determinants of Pseudomonas aeruginosa internalization and intracellular survival in host epithelial cells

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Pseudomonas aeruginosa is an opportunistic human pathogen capable of breaching the airway epithelium of the respiratory tract, making it a leading cause of hospital-acquired lung infections and mortality in patients with compromised pulmonary function. Although widely considered an extracellular pathogen, *P. aeruginosa* can invade and internalize into airway epithelial cells. Recent evidence suggests that internalization contributes to epithelial invasion and may promote long-term intracellular persistence, thereby facilitating chronic and recurrent infections. However, the genetic factors driving internalization and intracellular survival in non-phagocytic host cells remain poorly understood. Here, we employed transposon insertion sequencing (Tn-seq) screens to identify key determinants of these processes in A549 lung epithelial cells. Our screen highlighted multiple motility-associated genes, notably those involved in flagellar and type IV pilus (T4P) systems, as major contributors to internalization. Strikingly, disruption of several T4P-associated genes strongly impaired internalization while conferring a selective advantage for intracellular survival, suggesting opposing roles in these processes. Gentamicin protection assays using clean deletion mutants confirmed these contrasting effects. Further genetic analyses suggest that these effects arise from a combination of T4P-mediated twitching motility, surface sensing, and downstream activation of the cAMP–Vfr regulatory network. Together, our findings identify T4P as a key determinant with differential impact on internalization and intracellular survival of *P. aeruginosa* in host epithelial cells.

P051* Deciphering the diversity of Gram-negative bacterial mechanisms of resistance towards antimicrobial peptides

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Antimicrobial peptides (AMPs) are short, cationic molecules produced by many living organisms as part of their innate immunity and represent potential alternatives to antibiotics towards multidrug resistant bacteria. Several AMPs are currently tested in clinical trials, however, the molecular mechanisms used by pathogenic bacteria to gain resistance to AMPs remain largely unknown.

To characterize the diversity of mechanisms Gram-negative bacteria can develop to overcome AMP activity we performed serial passaging of *Escherichia coli* and *Acinetobacter baumannii* in progressively increasing concentrations of a selection of 6 AMPs, representing the diversity of peptides currently under clinical evaluation, as well as 5 Proline-rich AMPs. Resistant isolates obtained after 8 passages were sequenced to identify the underlying resistance mechanisms. We then assessed the potential consequences of resistance development on bacterial fitness, virulence, and cross-resistance to other antimicrobial agents. We identified several genes associated to AMP resistance, most of which are involved in cell envelope biogenesis, in particular lipopolysaccharide (LPS) biogenesis. In general, resistance to AMPs did not affect bacterial fitness and virulence and rarely induced broad-range cross-resistance to other antimicrobial agents. Different mutations in the LPS biogenesis pathway resulted in resistance specific to distinct AMPs, indicating that AMPs can differently interact with LPS. In contrast, the inactivation of the *crp* gene, encoding the cyclic-AMP responsive protein, conferred broad-range resistance to AMPs in *A. baumannii*, but not in *E. coli*, by a yet unknown mechanism. Finally, we observed that acquisition of AMP resistance can have variable effects on bacterial fitness and virulence.

Our findings show that resistance to AMPs can emerge by diverse species- and AMP-specific mechanisms having variable consequences on bacterial fitness. We thus provide new insight into AMP resistance mechanisms in Gram-negative bacteria, highlighting the need for further characterization of these mechanisms to support a safer and more effective development of AMP-based therapies.

**Student Paper*

P052 Complete genomes of Arsenicicoccus dermatophilus strains causing pododermatitis in greater flamingos

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Arsenicicoccus dermatophilus was first reported in 2013 as the causative agent of pododermatitis in greater flamingo, especially affecting birds kept in captivity. Although it is genetically related to the Genus *Arsenicicoccus,* the cells form hyphae which are typical of *Dermatophilus congolensis*, a bacterium that infects skin of terrestrial mammals. Here, we present the complete circular genome of three *A. dermatophilus* strains KM 894/11^T, KM18/12 and KM9/12.

The strains were isolated from the skin lesion of three captive greater flamingos in Switzerland on TSA-SB agar incubated at 37°C for 48h. Genomic DNA was sequenced on Illumina NovaSeq 6000, and with the Oxford Nanopore Technology (ONT) MinION device on a R9.4.1 flow cell. ONT reads were assembled using trycycler v0.5.3 and polished with Illumina reads using Polypolish v0.6.0. Blast distance phylogeny and digital DDH (dDDH) were performed using the Type Strain Genome Server. Genomes were compared to each other with MAUVE. Gene prediction was obtained with Geneious Prime 2026.1.

Each genome contains a circular chromosome of 3.3 Mbp and two to three circular plasmids with sizes of 16.9 to 61.2 kbp with a total high GC content of 72.6%. Protein-coding sequences ranged from 3,109 to 3,158, pseudogenes (33 to 51), and 57 RNA-encoding genes were predicted. Taxonomic analysis confirmed that the strains belonged to the Genus *Arsenicicoccus* and are most closely related to *A. bolidensis* (dDDH, 22.7%). The three genomes differed by 419 to 134,359 SNPs, and by the inversion of a 649,173- and 652,852-bp chromosomal fragment flanked by *mnmA* and *der* genes in KM18/12 and KM894/11^T, as compared to KM9/12, and likely driven by IS elements.

These assemblies represent the first circular genomes of *A. dermatophilus* and may serve as baseline for further genetic characterization of virulence mechanisms and development of DNA-based detection tools for early detection of this pododermatitis pathogen.

P053 Deciphering the mechanisms of tolerance to antimicrobial peptides in Gram-negative bacteria

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Antimicrobial resistance is an increasing concern for medicine: resistant bacteria are insensitive to antibiotics and proliferate in their presence. However, in some conditions, bacteria not intrinsically resistant to an antibiotic can survive antibiotic treatment, possibly resulting in chronic infections. These bacteria are referred to as tolerant bacteria and are thought to exhibit low metabolic activity, resulting in slow growth. Classical antibiotics are inefficient towards these bacteria, since they usually target pathways related to bacterial proliferation. In contrast, antimicrobial peptides (AMPs) are membrane-active compounds that have been hypothesized to efficiently target slow-growing bacteria. However, recent evidence indicates that bacteria can be tolerant to AMPs.

In this project, we investigated the extent of bacterial tolerance to AMPs, testing 6 AMPs from diverse origins and with diverse modes of action and structures on 2 models of non-growing bacteria, in 3 different Gram-negative bacterial species: *Escherichia coli*, *Acinetobacter baumannii* and *Pseudomonas aeruginosa*. In the first model, we used controlled stationary phase bacterial cultures in a well-defined synthetic medium. We observed that stationary phase bacteria were tolerant to all tested AMPs, but at different levels. In a second model, we used serine hydroxamate (SHX), an inducer of the stringent response, to chemically block bacterial proliferation, mimicking amino acids starvation. Surprisingly, the effect of SHX pretreatment on tolerance to AMPs varied strongly depending on the AMP and the bacterial species. In some cases, pretreatment with SHX was even potentiating the efficacy of AMPs. We are currently investigating the mechanisms involved in this tolerance, using both a biased approach – testing known mechanisms of antibiotic tolerance- and an unbiased approach through transposon insertion and subsequent Tn-Seq to identify genes important for tolerance to AMPs.

Understanding mechanisms involved in tolerance to AMPs may allow the design of strategies to improve the efficacy of AMPs, especially towards chronic infections.

P054 Genomic Diversity and Long-Term Persistence of Listeria monocytogenes in Cheese Processing Facilities

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Listeria monocytogenes continues to represent a significant concern for the food sector. Nevertheless, information regarding its genomic diversity and the genetic determinants linked to persistence in food-processing settings remains incomplete. To broaden understanding of the diversity and persistence of *L. monocytogenes* within the cheese production chain, 194 isolates collected over a 24-year span from 50 Swiss cheese-processing facilities were investigated through whole genome sequencing (WGS)-based analyses. The isolates were assigned to 23 distinct clonal complexes (CCs), among which CC3, CC1, and CC101 were the most frequently detected. Importantly, seven isolates were identified for the first time as novel sequence types. In addition, seven CC1 isolates, classified as ST1 and CT5518, grouped closely with the strain implicated in the first major listeriosis outbreak reported in Switzerland during the 1980s, an event associated with numerous fatalities. CC1 strains were predominantly linked to soft cheese and artisanal manufacturing sites, whereas CC3 strains were more commonly recovered from hard and semi-hard cheeses, as well as from industrial plants and ripening environments. Concerning persistence, several CC101 and CC3 isolates were repeatedly recovered from the same facilities over periods reaching 5 and 7 years, respectively. Investigation of persistence-associated markers indicated that survival strategies in cheese-processing environments were varied and mainly reflected CC-specific genomic adaptations to stress conditions, including high salinity and acidic pH. In particular, CC3 and CC517 strains displayed increased stress resistance associated with the presence of Stress Survival Islet 1 (SSI-1), while other CCs achieved similar tolerance through different genetic determinants. Altogether, this study offers a detailed long-term genomic perspective on *L. monocytogenes* circulating in Swiss cheese production and highlights that persistence mechanisms are largely CC-dependent, involving factors such as SSI-1 in selected CCs, while the acquisition of genomic islands driven by selective pressures may further support adaptation and persistence.

P055 Deciphering developmental switching in Waddlia chondrophila through stage-specific effector proteins

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Waddlia chondrophila is an emerging pathogen associated with adverse pregnancy outcomes in humans and abortion in ruminants. It belongs to the same phylum as *Chlamydia trachomatis*, the leading cause of bacterial sexually transmitted infections worldwide. Both bacteria are strict intracellular pathogens that alternate between an infectious elementary body (EB) and a replicative reticulate body (RB), which multiplies inside a host-cell vacuole called an inclusion. The transitions between EBs and RBs are essential for the bacterial life cycle, yet the mechanisms regulating these developmental switches remain poorly understood. These processes however likely involve interactions with the host cell mediated by secreted bacterial effectors.

We compared the transcriptomes of *W. chondrophila* EBs and RBs, identified genes differentially expressed between the two forms and decided to focus our work on EB-associated proteins Wcw_0260 and Wcw_1046, on RB-associated protein Wcw_1915, and on Wcw_0984, which is expressed in both stages.

We first determined their gene expression profiles throughout the bacterial life cycle and validated RNASeq findings. Using a heterologous secretion system in *Yersinia enterocolitica*, we then demonstrated that Wcw_0260 and Wcw_1046 are probable T3SS effectors. In transfected HeLa cells, these 2 proteins localized to the plasma membrane, Wcw_0984 targeted the nucleus, and Wcw_1915 localized mainly to the cytoplasm with occasional Golgi association.

Since *Waddlia chondrophila* is untractable, we chose to overexpress the proteins in *Chlamydia trachomatis* as a model system to determine their role in bacterial development. We evaluated the effects of overexpression by quantifying bacterial replication using quantitative PCR (qPCR) and assessing the infectivity of elementary bodies (EBs) through inclusion-forming units (IFUs) assays.

Future experiments will involve co-immunoprecipitation to identify potential interactions between these proteins and eukaryotic host factors. Ultimately, this approach aims to elucidate the specific roles of these four proteins in the developmental cycle of *W. chondrophila*.

P056 Niche Overlap Determines the Ecological Impact of Phage Predation

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Objective:

Phages are key top-down forces shaping bacterial communities, alongside bottom-up factors such as resource availability. In this study, we aimed to disentangle their relative contributions to bacterial community composition and diversity.

Methods:

We tracked the temporal dynamics of synthetic communities of multiple Bifidobacterium strains grown on simple or complex carbon sources (bottom-up effect), with or without predating phages (top-down effect), in a full-factorial experimental design.

Results:

We found that the ecological impact of phages highly depends on the resource environment. In simple media, strong competition is modulated by phage predation, which reduces dominant strains and increases community diversity and temporal turnover. In complex media, niche partitioning reduces competition, thereby limiting phage-driven bacterial turnover and resulting in minimal changes in diversity.

Conclusion :

There is growing interest in using phages for microbiome engineering and to treat antibiotic-resistant infections, but these applications depend on predicting their ecological effects in complex bacterial communities. Here, we show that the magnitude of phage impact is strongly shaped by the underlying resource environment. Our results highlight that phage–bacteria interactions cannot be understood in isolation from the broader ecological context and provide a step toward a more predictive understanding of phage-based interactions.

P057 Winogradsky columns as model ecosystems to investigate how biosignatures change according to microbial metabolisms for the extra-terrestrial detection of life

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Microbial life developed everywhere on Earth. This ubiquity is largely due to the incredible variety of microbial metabolisms, allowing microbes to use diverse energy sources. Among all these metabolisms some, like the anaerobic photosynthesis and the chemolithotrophy, were key at the early stages of evolution of life on Earth. Accordingly, these primitive metabolisms might be also very relevant for the emergence and evolution of extraterrestrial life in other planets. To detect traces of extra-terrestrial microbial life, we developed an approach based on the homochirality phenomenon, that is the predominance of one molecular handedness in biological systems. This asymmetry is called homochirality and is considered a signature feature of life. Homochirality can be detected by assessing the fractional circular polarization of light upon encountering asymmetric molecules. FlyPol is a highly sensitive spectropolarimeter that can detect circular polarization and could be deployed in space to assess the presence of life on the Earth's surface, or even on the surface of other planetary bodies. To validate its use, we used Winogradsky columns in which microbial communities structured mostly around the sulfur, oxygen, and carbon cycles. We inoculated the columns with biofilms sampled in a natural sulfur-rich terrestrial environment. After five months, the columns developed distinct colored layers corresponding to organisms employing different metabolic strategies. Our preliminary results show that we can assess the presence of these microorganisms using spectropolarimetry and that the signal varies according to microbial type. Such results pave the way to explore microbial life that thrives in a wide range of environmental conditions and towards the remote detection of life beyond Earth.

P058* Experimental evolution in infection mimetic media highlights divergent and pathogen-specific interspecies interactions

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Polymicrobial infections are an urgent public health problem with several hundred thousand deaths being attributed to them. Infections caused by combinations of multidrug-resistant bacteria require strategies to understand the interactions between the infecting pathogens to develop improved treatment options. We focus on three opportunistic and often multidrug-resistant human pathogens: Pseudomonas aeruginosa (PA), Staphylococcus aureus (SA), and Klebsiella pneumoniae (KP) and their interactions with each other in an infection mimetic environment. We competed our pathogens on bovine brain-heart infusion agar supplemented with sheep blood. We then quantified species abundance using selective plating as well as our novel digital PCR assay. Lysates of pathogen communities containing genomic DNA were used as templates to quantify species abundances using a probe-based dPCR assay. We designed probes targeting single-copy genes in each of the focal pathogens (lasI for PA, nucA for SA, and khe for KP), representative of single cells. Our data show that pathogen-pathogen interactions between these focal species are either neutral or antagonistic, with all combinations benefiting PA. However, extended interaction through co-evolution alters these interaction dynamics. We found that antagonistic interactions between PA and SA shift towards more neutral interactions with both species surviving to comparable abundances. However, we also observed that interactions between SA and KP shift from neutral to antagonistic, with SA driving KP to extinction, below detectable limits. The contrasts between short-term and long-term interactions highlight the ability of these pathogens to employ strategies to prevent the colonization and growth of competitors, likely through production of secondary metabolites, contact-dependent inhibition, and modification of the environment. Understanding these changes can yield valuable insights into the infection dynamics of long term polymicrobial infections. This information can be used to improve treatment outcomes for infections caused by multidrug-resistant bacteria by developing strategies aimed at reducing virulence and fitness of drug-resistant pathogens.

**Student Paper*

P059* An engineered phage cocktail enables rapid susceptibility testing and targeted eradication of Uropathogenic Escherichia coli

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Urinary tract infections are among the most common bacterial infections worldwide, with *Escherichia coli* being the leading causative pathogen Though antibiotics remain the standard treatment, high recurrence rates and increasing antibiotic resistance are driving treatment failure motivating the development of alternative interventions such as phage therapy. However, phage therapy is often limited by narrow host range, emergence of resistance to the phage and the time required to identify active therapeutic phages. To address these limitations, we isolated two bacteriophages with naturally broad host ranges, characterized them, and engineered them using CRISPR-assisted methods. We generated a dual-functional cocktail comprising reporter phages carrying Nanoluciferase enabling rapid susceptibility testing. These are complemented by colicin-producing phages to enhance bacterial killing and suppress resistance emergence. We show that our engineered reporter phage cocktail infects 70% of our in-house collection of uropathogenic *E. coli* isolates (n = 60) and demonstrate that the activity of our reporter phages reflects their plaquing host range. Furthermore, colicin-producing phages reduce the emergence of resistance in both LB media and *E. coli* spiked human urine. Together, these results demonstrate that engineered broad-host-range phages can improve both the speed of phage selection and the efficacy of antibacterial treatment compared with their wild-type counterparts. We anticipate that the use of engineered phages will facilitate the implementation and increase the prevalence of phage therapy in healthcare for the treatment of bacterial infections.

**Student Paper*

P060 Prophages and extrachromosomal elements are pervasive among acetogenic bacteria

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Acetogenic bacteria, or acetogens, use the ancient Wood-Ljungdahl pathway to grow at the thermodynamic limit of life. They often couple this pathway with other metabolic modules to utilize a wide range of organic compounds as energy and carbon sources for acetogenesis. This metabolic flexibility allows acetogens to coexist and compete in anoxic ecosystems despite the low energy yield of acetogenesis. The presence of these metabolic modules, as well as the fact that acetogenesis does not follow an exclusive phylogenetic pattern, suggests that the spread and persistence of acetogenesis is attributed to horizontal gene transfer via still elusive extrachromosomal DNA elements (ecDNA).

To address this, we computationally predicted prophages and plasmids in all isolated and publicly available acetogen genomes and found that they are present in 86 out of 111 strains. To assess whether ecDNA is induced in acetogens under laboratory conditions, we analyzed publicly available sequencing datasets using a tool that identifies circular elements from short read paired-end reads. The analysis detected excised prophages in one-fifth of the analyzed strains and a myriad of plasmids and other yet unclassified circular elements. Additionally, four strains were cultured in the laboratory under standard medium conditions and subjected to particle-protected DNA sequencing which revealed prophage regions with several fold higher read coverage in three of the strains. This indicates spontaneous, independent replication and packaging of phage DNA in a subpopulation of the culture.

Together, these findings suggest that circular extrachromosomal elements are present in most acetogenic isolates and that their genomes are more dynamic than previously recognized.

P061 Sticking Around: Characterizing the mechanisms of surface adaptation in Acinetobacter baumannii

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Acinetobacter baumannii (Ab) is a gram-negative nosocomial pathogen that can cause pneumonia, wound, bloodstream and urinary tract infections. Antibiotic therapies often fail to cure *Ab* infections due to its high resistance. Furthermore, *Ab* can form biofilms, which contribute to increased antimicrobial tolerance. To form biofilms, bacterial cells adhere to surfaces. However, it remains unclear whether and how *Ab* actively adapts to surfaces to ultimately establish a persistent sessile lifestyle. While *Ab* relies on extracellular filaments such as Type IV and Csu pili to attach to biotic and abiotic surfaces, we know little about the mechanisms mediating subsequent activation of surface-associated regulatory programs. Here, we investigate how *Ab* phenotypically and transcriptionally adapts to life on surfaces. We compared *Ab* growth on agarose surfaces to liquid cultures at high temporal resolution. *Ab* showed a pronounced reduction in growth during the first 3 hours on surface before recovering liquid-like growth rate. To investigate factors regulating this phenotypic adaptation, we performed RNA-seq on surface-associated population. When growing on surfaces, *Ab* upregulates iron uptake genes associated with the siderophore acinetobactin (*basABCDEFG, barAB, bauABCDEF*). As iron is essential to *Ab* growth, we assessed the relative contribution of iron limitation and surface adaptation in upregulation of these genes. Population-level measurements and single-cell-level fluorescent reporters confirmed that those genes are regulated by iron levels. Supplementation of agarose pads with iron restored the growth rate of surface-associated cells comparable to liquid culture. This supports that iron pathways play a critical role in surface-associated growth. As *Ab* can form biofilms in flow conditions, such as in blood vessels or pipes, we further investigated surface adaptation in microchannels. Under flow, *Ab* formed comet-shaped biofilm phenotypes. Overall, our work identified genetic determinants underlying surface adaptations in *Ab* which could inspire the development of new therapeutic strategies against surface-associated virulence in this pathogen.

P062* Host cell range and intracellular trafficking of the novel species Chlamydia vaughanii

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Bacteria belonging to the *Chlamydiales* order are strict intracellular, Gram-negative organisms that are characterized by a two-stage developmental cycle. Several species of *Chlamydiales,* like *Piscichlamydia salmonis* or *Clavochlamydia salmonicida,* were recently discovered in fish farms. These fish pathogens can cause epitheliocystis, a disease that causes cysts in the gills of the infected animal. Recently, our group isolated a new *Chlamydiales* species from a tropical fish. This new species, named *Chlamydia vaughanii,* belongs to the *Chlamydia* genus, which also encompasses several important human and animal pathogens such as *Chlamydia trachomatis* or *Chlamydia psittaci.* This discovery came about following an event where 95% of the fish in a tropical aquarium died suddenly. *C. vaughanii* was isolated by inoculating McCoy cells with a sample from the head of a dead fish in the presence of antibiotics and antifungals. However, the implication of *C. vaughanii* in these fish deaths has not yet been determined.

Our results showed that *C. vaughanii* successfully replicated in four human cell lines: A549 (human alveolar basal epithelial cells), THP-1 (monocyte-derived macrophages), HeLa, and Ishikawa (human endometrial cells) at 37°C. In contrast, it did not replicate in the fish cell line rainbow trout gonad cells (RTG-2) at 25°C. *C. vaughanii* exhibited the characteristic biphasic developmental cycle of *Chlamydia.* Notably, no host cell lysis was observed in any of the permissive cell lines, despite successful bacterial egress and reinfection of neighboring cells. In HeLa cells, *C. vaughanii*, like other *Chlamydia* species, avoided the canonical endocytic pathway and displayed strong co-localization with endoplasmic reticulum markers calnexin and PDI. Although replication occurred predominantly in the peri-Golgi region, *C. vaughanii* did not induce Golgi fragmentation, a hallmark of *Chlamydia* infection. Nevertheless, replication was impaired when HeLa cells were treated with the Golgi-disrupting agent Brefeldin A, indicating that the Golgi is important for bacterial development.

**Student Paper*

P063* Second messenger signaling in Pseudomonas aeruginosa infection

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The opportunistic human pathogen *Pseudomonas aeruginosa* relies on second messengers like cAMP and c-di-GMP to integrate environmental signals and coordinate behavior during surface colonization and infection. In response to changing host environments, cAMP activates acute virulence, whereas c-di-GMP is associated with biofilm formation and chronic infections. How these signaling pathways are coordinated during infection remains poorly understood. Here, we investigated the interplay between cAMP and c-di-GMP and their regulatory roles during *P. aeruginosa* surface colonization and infection.

Using single-cell fluorescent reporters, we observed that surface-associated *P. aeruginosa* progressively transitioned from a high c-di-GMP state to a high cAMP state. These states rarely overlapped within individual cells, arguing for antagonistic mechanisms driving population diversification. Correlating reporter activity with bacterial behavior linked c-di-GMP to attachment and cAMP to motility and virulence activation, supporting functional diversification and expansion during early colonization.

In a human lower-airway tissue model, disruption of *P. aeruginosa* second messenger signaling impaired epithelial barrier breaching efficiency and perturbed multiple infection-associated programs. Lack of cAMP production or elevated c-di-GMP led to an increase of intracellular pathogens, suggesting that balanced second messenger signaling is required to coordinate different infection steps. Reporter analysis further provided preliminary evidence that cAMP signaling remains dynamically regulated during tissue infection. Overall, our findings support a model, in which coordinated cAMP and c-di-GMP signaling drives functional diversification of *P. aeruginosa* to optimize infection and invasion of human tissue surfaces.

**Student Paper*

P064* Re-sensitizing Antibiotic Resistant Bacteria by Exploiting Acquired Vulnerabilities

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Antibiotic resistance is often viewed as a unidirectional process: once bacteria acquire mutations that confer resistance, treatment options become limited, implying that resistance mutations are uniformly beneficial under antibiotic pressure. However, growing evidence shows that resistance frequently carries broader costs beyond reduced fitness. Adaptive mutations can reshape cellular physiology, rewire regulatory networks, and constrain evolutionary trajectories, potentially exposing hidden vulnerabilities. Collateral sensitivity (CS), where resistance to one antibiotic leads to enhanced susceptibility to another, provides one example of such vulnerabilities, yet most CS studies focus on drug–drug relationships without resolving the underlying resistance mechanisms, assessing whether CS is shared across different resistance pathways, or determining how CS evolves as resistance develops. Here, we evolved 31 independent lineages of *Escherichia coli* to become resistant to 31 antibiotics and systematically profiled collateral sensitivity and cross-resistance interactions against the same antibiotic panel. We identified extensive CS networks and bi-directional CS interactions, although most interactions produced only modest MIC shifts, with a limited number showing strong CS effects (log2FC ≤ −3). To evaluate whether this concept can be used in the clinics, we will investigate the stability of resistance and CS phenotypes in the absence of selective pressure, test whether cycling bi-directional CS pairs can constrain resistance evolution, and determine whether CS interactions are conserved across independently evolved lineages, strains, and species. Whole-genome sequencing will further be used to identify the genetic mechanisms underlying conserved and high-magnitude CS interactions. Exploring deeper into the mechanisms of CS has the potential to establish a generalized framework for converting antibiotic resistance from a devastating problem to an exploitable weakness.

**Student Paper*

P065 Spatially dependent microalgal lipid production in a membrane biofilm reactor

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Microalgae are a promising source of lipids that can be used for fuel and/or human/animal nutrition. However, realization of such microalgae-based processes is hampered by low cell concentrations in standard suspension cultures. Additionally, as lipid production in microalgae is only induced by nutrient limitation, for example nitrogen-depletion, culture conditions must be adjusted on a temporal scale in the form of inefficient batch production. To address these challenges, we present here an immobilized cultivation system wherein *Chlamydomonas reinhardtii* is grown on a transparent, gas permeable, flatsheet membrane within a bioreactor. Light, oxygen and CO_2 (from ambient air) as well as ammonia are provided through the membrane. This system promotes microalgal growth directly on the membrane in the form of a biofilm, which possesses much higher cell density vis-à-vis suspended cultures. This system also results in the formation of various niches (*i.e.*, spatially dependent physiological behavior presented by the algae) that are a result of the concentration gradients of light and nutrients across the biofilm. Ultimately, this spatial heterogeneity enables the transformation of temporal scale lipid production into a spatially dependent continuous system. Specifically, cells show high growth activity close to the membrane where nutrients and light are readily available while cells further away from the membrane would experience nutrient limitation and therefore accumulate lipids. These peripheral cells could then be harvested *in-situ* while the active growth zone towards the membrane remains unperturbed. In this work, we will demonstrate the formation of a nitrogen gradient and the biological response to it by directly measuring the ammonium concentration in the biofilm using microsensors as well as via lipid staining. Besides these experimental results, we will present a computational model of this interdependent dynamic of nutrient gradient formation and cell physiology within our system.

P066 zDB: bacterial comparative genomics made easy

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The analysis and comparison of genomes can be complex, as it requires integrating and visualizing the results from various tools specialized for tasks such as orthology inference, phylogenetic inference, and structural and functional annotation. To simplify this process, we have developed *zDB*, an easy-to-use comparative genomics software integrating an analysis pipeline and a graphical user interface (GUI).

Starting from annotated Genbank files, zDB identifies orthologs and infers a phylogeny for each orthogroup as well as a species phylogeny constructed from shared single-copy orthologs. Pfam, COG, KEGG, antimicrobial resistance and virulence factor annotations can also be added to the genes, while homologs can be identified in the Swissprot and RefSeq databases. Finally, genomic islands can be predicted and their conservation evaluated across genomes.

This wealth of information can then be analysed in the GUI, which allows to search for specific genes or annotations, to perform BLAST queries, to compare genomic regions or whole genomes, to display the presence/absence of orthologs and annotations in subsets of genomes in various ways (heatmaps, presence/absence tables, Venn diagrams, phylogenies, etc.), and to compare metabolic capacities between genomes.

zDB is open-source (https://github.com/metagenlab/zDB), easy to install with conda, and provides a command-line interface allowing to easily download the necessary reference databases, run the analysis pipeline and start the GUI. The underlying technologies, nextflow for the bioinformatics pipelines and python/django for the GUI, allow portability of the software, reproducibility of the results and easy integration of new features.

Overall *zDB* is easy to use while offering a wide range of features, making it useful both for bioinformaticians and researchers more accustomed to laboratory research. A demo of the GUI is available at https://zdb.metagenlab.ch/.

P067 Identifying gut microbiome biomarkers in Alzheimer's disease through shotgun metagenomic meta-analysis

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1. Institute of Microbiology, Lausanne University Hospital (CHUV), Lausanne Context:

Context:

Alzheimer's disease is one of the most prevalent forms of dementia worldwide, affecting approximately 15% of individuals aged 80–89 in Switzerland, making it a major public health burden. Although age is the primary risk factor, this multifactorial disease is characterized by the aggregation of intracellular Tau proteins and extracellular beta-amyloid fibers, leading to neuroinflammation. An emerging research avenue focuses on the gut microbiome, which may fuel this neuroinflammation through the gut-brain axis via pro-inflammatory metabolites. While several studies have highlighted variations in microbial species and functions in the disease, results remain heterogeneous, largely due to dietary and geographical differences across cohorts.

Objectives:

Our aim is to identify taxonomic and functional gut microbiome biomarkers associated with Alzheimer's disease that are consistent across geographical origin.

Methods:

We conducted a meta-analysis integrating 4 cohorts including participants from China, the United States, Japan, and Germany. Raw shotgun metagenomic sequencing data were processed through our ZShoman pipeline to standardize bioinformatic analysis methods and subsequently subjected to statistical analyses at both taxonomic and functional levels, with models accounting for covariates including age, sex, and BMI.

Results:

Preliminary results reveal several taxa consistent with existing literature, including an increase in *Collinsella* species and a decrease in *Bifidobacterium longum*, in AD patients compared to controls. At the gene-derived functional level, we observe a decrease in nitrate assimilation. Furthermore, we detect microbial amyloid-like proteins, broadening our scope beyond the canonical human amyloid Aβ42 and its established role in AD.

Conclusions and perspectives:

This meta-analytic approach provides an integrated view of the gut microbiome's role in Alzheimer's disease. Further characterizing its composition and functional activity could open new diagnostic and therapeutic avenues but also deepen our understanding of the underlying mechanisms of Alzheimer's disease.

P068 Modelling the human immune response in phage therapy of urinary tract infections

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Urinary tract infections (UTIs) constitute an important burden to human health. Phage therapy is a promising strategy to treat uropathogenic *Escherichia coli*, the main causative agent of these infections. However, its applicability is limited by the difficulty in translating *in vitro* results to clinical success. In a previous study, we found that immune proteins in human serum can block phage activity. More specifically, we showed that phage inhibition was mediated by the complement system, a set of proteins present not only in blood but also in the kidneys and the bladder during infection. *In vivo*, this effect could be a potential reason for phage therapy failure. To better understand this problem, we designed an assay where we combine human serum as a source of complement with different culture media, including human urine, to model the environment that therapeutic phages face in the infected bladder. We have studied the infectivity of phylogenetically diverse *E. coli* phages of the BASEL collection in this system, showing that human serum broadly inhibits phages across multiple taxonomic groups. Moreover, our results revealed that complement proteins strongly protect *E. coli* laboratory strains as well as clinical isolates. To overcome this barrier, we evolved different phages in our *in vitro* model of serum challenge through serial passaging. The evolved phages presented mutations in genes coding for the receptor binding proteins which led to enhanced infectivity. In addition, this *in vitro* system allowed us to identify new phages with enhanced activity in serum, demonstrating how this approach may inform the selection of phages for therapy. Our study highlights the importance of testing phages in physiologically relevant media and offers insights into how the immune system and phage characteristics may shape the outcome of therapy in human patients.

P069 Screening and characterization of ESBL-producing Enterobacterales in different food matrices in North-Western Switzerland

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Extended-spectrum beta-lactamase (ESBL)-producing *Enterobacterales* represent a major global public health threat. While food products can act as reservoirs for resistance genes, the prevalence and specific risks across various food matrices require continuous monitoring. With this study we aimed to investigated the presence of ESBL-producing *Enterobacterales* in food products in North-Western Switzerland.

Between January 2025 and January 2026, we screened 956 food samples on selective CHROMagar™ ESBL. The analyzed food matrices comprised heated, mixed, and natural raw ready-to-eat foods; pastries with whipped cream; ice cream; meat preparations; whole, sliced, or portioned heat-treated meat products; dried fruits and nuts; and herbs and spices. Each distinct morphotype was isolated on blood agar and identified via MALDI-TOF mass spectrometry. We next performed antibiotic susceptibility testing (AST) and confirm ESBL production for all *Enterobacterales* strains.

Out of the 956 food samples tested, 89% showed no growth on ESBL-selective media, while 11% (n=106) exhibited initial growth. The highest presumptive positivity occurred in the "Natural, raw, ready-to-eat" category, where 32% of these samples showed growth on ESBL-selective agar, followed by “dried fruits & nuts” (25%) and “herbs & spices” (21%). We identified 218 bacterial isolates using MALDI-TOF MS, predominantly belonging to *Pseudomonadaceae* and *Moraxellaceae*. We performed AST on the 15 *Enterobacterales* strains and confirmed only two true ESBL-producers (0.9% of the presumptive positive strains): a *Klebsiella pneumoniae* isolate from carrots, and a multidrug-resistant *Mixta calida* isolate from sumac, which was resistant to ceftriaxone, cefepime, fluoroquinolones, and trimethoprim-sulfamethoxazole.

With this regional focused surveillance study, we showed that the overall prevalence of ESBL-producing *Enterobacterales* in the sampled food matrices was low (0.9%). We observed that baseline growth of non-*Enterobacterales* on selective media was driven by intrinsic mechanisms. These findings suggest that processed and monitored food chains in North-Western Switzerland present a very low-risk for the dissemination of ESBL-producing *Enterobacterales*.

P070 Mixed-species transcriptomics reveals bidirectional pathogen-commensal and inter-commensal crosstalk

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Staphylococcus aureus is an important human pathogen and frequent coloniser of the nasal microbiome, with colonisation a risk factor for infection. As antimicrobial resistance rises, alternative strategies to prevent colonisation are needed. Certain nasal commensal bacteria inhibit *S. aureus*, yet the mechanisms underlying these interactions remain unclear. Using an *in vitro* colonisation model combined with mixed-species transcriptomics, this study examines bidirectional transcriptional responses between *S. aureus* and inhibitory nasal commensals, and among commensal species. The commensals studied included a commensal *S. aureus* isolate, *Staphylococcus lugdunensis*, *Corynebacterium tuberculostearicum* and *Dolosigranulum pigrum*.

S. aureus exhibited a shared transcriptional response to commensal exposure, alongside changes unique to specific commensal species. Commensals mounted distinct responses to *S. aureus* exposure, notably, *Staphylococcus lugdunensis* upregulated genes involved in cell wall stress response. Beyond pathogen-commensal interactions, commensal co-cultures revealed transcriptional changes consistent with metabolic cross-feeding. Together, these findings advance our understanding of pathogen-commensal and inter-commensal crosstalk in the nasal niche, supporting microbiota-based strategies to reduce *S. aureus* colonisation and reliance on antibiotics.

P071* proMGEflow: recombinase-based detection of mobile genetic elements inbacterial (meta)genomes

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Mobile Genetic Elements (MGEs) are drivers of bacterial adaptation and can increase fitness of microbial communities in the changing environment. Yet the identification of MGEs remains challenging due to the fuzziness of different MGE types and incompleteness of metagenomic assembled genomes (MAGs). Here we present proMGEflow - a Nextflow pipeline designed to annotate full genomes and MAGs with discrete MGE categories: conjugative elements, integrons, phages and transposable elements. In comparison to other tools, proMGEflow takes a top-down approach to harmonize all recombinase-based MGEs from a given (meta)genome in one go. Our pipeline uses subfamilies of recombinases as universal MGE markers, as well as MGE type-specific mobility machinery, e.g. structural phage genes, for fine-grained assignment. MGE boundary estimation is based on the joint bacterial species pangenome from the MAG and its species cluster of high-quality reference genomes from the proGenomes3 database. By decoupling the MGE boundary determination step from the finer grained MGE type assignment, we can further annotate MGEs in user-provided genomic regions via rule-based classification of their machinery and recombinases. In total, we applied proMGEflow to about 200,000 MAGs from the Searchable Planetary-scale mIcrobiome REsource (SPIRE). This did not only result in the discovery of around 3 million MGEs of different types but also helped to gain the first functional insights into a global environmental mobilome. The availability of scalable and reproducible pipelines for unified MGE annotation from metagenomes will improve our understanding of mechanisms of gene mobility as well as the cross-talk with their prokaryotic hosts.

**Student Paper*

P072∗ Investigating the single molecule displacement dynamics of cell wall hydrolases

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Understanding the regulation of bacterial cell wall biogenesis remains an important objective in bacterial cell biology. Coordination between peptidoglycan (PG) synthesis and hydrolysis is essential for maintaining cell shape and viability, yet the mechanisms governing this balance remain poorly understood across both spatial and molecular scales. Advanced live-cell imaging studies have shown that inner membrane-localised PG synthases are guided by cytoskeletal elements during cell elongation and division; however, comparatively little is known about how PG hydrolases are coordinated with these processes.

Here, we investigate the dynamics of the *Escherichia coli* lytic transglycosylase MltG and endopeptidase MepM using single-particle tracking (SPT), for which fluorescent protein fusions were constructed and imaged to characterise their single-molecule behaviour in live cells.

Preliminary SPT analyses reveal that both MepM and MltG transition between diffusive states and immobile states that likely reflect association with the PG layer. In contrast to actively synthesising PG enzymes, these hydrolases do not appear to exhibit persistent directional motion, suggesting that they may not be physically coupled within a larger PG synthesis complex. These observations indicate that hydrolases may be coordinated differently from PG synthases while still contributing to the spatial and temporal coordination of cell wall remodelling.

Additionally, preliminary growth analyses under antibiotic stress conditions suggest that MltG may play a distinct role in maintaining PG precursor homeostasis.

∗Student Paper

P073 Drivers of host-specificity in the gut microbiome across social bee species

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The gut microbiome can play an important role in respect to host health, physiology and food niche exploitation. In turn, the host provides their gut bacteria with a relatively stable environment with diverse ecological niches, supplying access to different nutrient sources. Specific host species traits, such as diet, habitat and immune response, can shape the gut microbial composition both at the community and strain level. Consequently, animal hosts typically harbour specialised gut microbiota, having a host-specific microbiome. However, the processes that shape host specificity and how this evolved with respect to the evolution of their host is not fully understood. Social bees harbour a very specialised yet relatively simple microbiome, making them an ideal study system to investigate the mechanisms underlying host-specificity across many host species. To investigate these mechanisms, we assessed the gut microbiome across three clades of social bees, namely honeybees, bumblebees and stingless bees. We collected the hindgut of over 20 social bee species from different locations including Brazil, France, Kenya, Malaysia and Switzerland. Using a metagenomic approach to assemble the genomes of the bacteria, we are able to assess the gut microbial composition of bees at the strain level and look at their evolutionary history and functional potential. Taken together, our findings can help to identify the key drivers that shape host-specificity across different animal hosts.

P074* Evolutionary Responses of a Clinical Cystic Fibrosis Pseudomonas aeruginosa Isolate to Phage-Based Therapeutic Pressure

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Objective
Phage therapy studies have mainly focused on safety and efficacy, while evolution of acquired phage resistance remains underexplored. We assessed resistance-associated trade-offs in a *P. aeruginosa* cystic fibrosis (CF) respiratory isolate compared with PAO1.

Methods
PAO1 and a chronic CF isolate (Pae_0571) were exposed to a myovirus phage. Survivors were analyzed for resistance stability, genomic alterations, and virulence in *Galleria mellonella*.

Results
PAO1 evolved stable resistance (93% of survivors) via mutations in *pilC* and *oprB* and a complete deletion of *galU*, with no spontaneous phage release and markedly reduced virulence (up to 200-fold increase in LD_{50}). In Pae_0571, no stable resistance was observed; 63% of survivors exhibited transient tolerance via *tolC*-mediated membrane remodelling, spontaneous phage release, and unchanged virulence.

Conclusion
Our results confirm that very distinct genomic modifications can lead to phage-resistance with different implications for bacterial virulence. In addition, spontaneous production of phage particles may indicate a prophage state, contributing to a transient resistance. Overall, this suggests that acquired phage resistance is driven by complex and strain-dependent mechanisms, reinforcing the need for personalized approaches in phage therapy.

Keywords

Pseudomonas aeruginosa, Cystic fibrosis, Resistance dynamics, Superinfection exclusion, Virulence attenuation

**Student Paper*

P075* Biofilm formation and dynamics within porous structures

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Bacteria exist in two primary states: as free-floating planktonic cells or as sessile communities known as biofilms, which are embedded in a matrix of extracellular polymeric substances (EPS). Biofilms confer survival advantages, including nutrient retention, resistance to antibiotics, and facilitation of horizontal gene transfer. While biofilm formation has been extensively studied over flat surfaces (e.g. Petri dishes) and in well-mixed environments, the influence of physical structure on aggregation, resources availability and biofilm dynamics remains poorly understood. We investigate how environmental architecture shapes bacterial aggregation and biofilm development under controlled flow conditions in porous media. Using *Escherichia coli* MG1655, we combine time-lapse microscopy with microfluidic systems to study biofilm growth in porous media. Preliminary results reveal peculiar spatial organization that changes over time. After an initial growth phase, extensive clogging emerges with un-stable river-like flow paths through the biofilm itself, characterised by heterogeneous biomass distribution and dynamic restructuring of flow paths. These observations suggest that structural heterogeneity enhances biofilm plasticity, potentially improving nutrient accessibility under progressive clogging. Future work will explore how quorum sensing could be a potential driver of structural adaptation and apply quantitative metrics to characterize flow-path complexity. Understanding these interactions is critical for predicting biofilm behavior in natural and engineered systems, with implications for health, agriculture, and bioremediation.

**Student Paper*

P076 Exploring the protein interaction network in Tn1549 integrative and conjugative element

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Integrative and Conjugative Elements (ICEs) are transposable genetic elements capable of moving from a donor to a recipient bacteria genome. This genetic material movement combines DNA transposition and conjugation functions that are encoded on the ICEs as well as diverse cargos such as virulence factors, metabolic traits or antibiotic resistances. Their ability to transfer horizontally makes these elements important drivers of genetic diversity and has the potential to give rise to multidrug-resistant bacteria. An example is Tn1549, a clinically relevant ICE that carries and spreads resistance to vancomycin, a last-resort antibiotic. This ICE is found in Gram-positive bacteria and often in pathogenic clinical isolates of multidrug-resistant *Enterococci*, limiting treatment options. In this work we focus on the molecular characterization of Tn1549. We first investigated the interaction network of the so far poorly characterized transposon proteins using the bacterial adenylate cyclase-based two-hybrids method. We found a intricate interaction network in Tn1549 with numerous links within and between the transposition, conjugation and regulatory functions providing insights into how these different processes coordinate to ensure successful transfer of the ICE. We then focussed on several specific subnetworks and identified key regulatory mechanisms involved in Tn1549 activation. Altogether, these results pave the way for a better understanding of Tn1549 movement and will help propose new strategies/targets to slow, prevent or block ICE-mediated antibiotic resistance spread.

P077 Functional metagenomics to uncover candidate enzymes for bioremediation of flourinated compounds

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Fluorinated compounds are used for agrochemical, pharmaceutical, and numerous industrial applications, resulting in global contamination, especially by the per- and polyfluoroalkyl substances (PFAS). These compounds persist due to the exceptional stability of the C-F bonds, currently relying on high energy inputs and excessive additives for degradation. Therefore, enzymatic deflourination would be a preferred and more sustainable alternative. Unfortunately, currently known defluorinase enzymes are often catalytically inefficient. To discover new defluorinase candidates I am using a functional metagenomics strategy which promotes the high-level expression of captured genes. I sourced soil from seven locations across Switzerland and constructed a functional metagenomic library. In preliminary testing, I am screening this library against simple flourinated compounds to find genes capable of providing resistance to toxicity or enabling growth in carbon-deficient media. The aim is to compile a panel of defluorinase candidates which can be characterised and tested with more challenging fluorinated substrates. Ultimately, the goal of this research is to provide enzymatic tools for PFAS bioremediation.

P078∗ Effects of water stress and arbuscular myorrhizal fungi inoculation on pea root rot

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Root rot is a major threat to pea (Pisum sativum) production in Switzerland, caused by a pathogen complex including the oomycetes *Aphanomyces euteiches* and *Pythium ultimum*, as well as several *Fusarium* species. In the absence of fully resistant genotypes, farmers currently rely on partially resistant varieties. An alternative strategy is to harness the protective potential of the root microbiota. Arbuscular Mycorrhizal Fungi (AMF) are known to form beneficial symbioses with host plants and have been shown to reduce disease severity, yet their effectiveness under water stress remains poorly understood.

Here, we study whether artificial AMF inoculation reduces root rot severity more effectively than native soil AMF communities, and how hydric stress modulates plant performance and pathogen virulence while simultaneously facilitating or hindering mycorrhizal colonisation.

To answer these questions, we conducted a controlled pot experiment using naturally pathogen-infested or gamma-ray-sterilised soil. Two pea genotypes with contrasting resistance levels were subjected to three watering regimes: water deficit, optimal watering, and excessive watering; with or without artificial AMF inoculation. After 30 days, plant performance was evaluated through phenotypic measurements, while root DNA was analysed by high-throughput amplicon sequencing to characterise microbiota shifts. Mycorrhizal colonisation success and *A. euteiches* abundance were quantified by qPCR. Preliminary results suggest complex, genotype-dependent responses to the interaction between hydric stress and microbial communities. Water deficit appears to reduce root rot severity, while artificial AMF inoculation does not produce the expected protective effect, showing neutral or even negative outcomes, possibly due to competition with the native soil AMF community. Ongoing microbiota composition analyses will help elucidate the mechanisms underlying these responses.

∗Student Paper

P079* TargetMosaic: A Fast and Scalable Framework for Protein Conservation Analysis

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The growing number of sequenced bacterial genomes offers unprecedented opportunities for the rational development of protein-targeting precision antibiotics and affinity reagents for diagnostics. A key step in evaluating a protein target for its suitability as a therapeutic or diagnostic target is the analysis of its sequence conservation, both within a bacterial species as well as across closely related species. However, performing such analyses requires extensive bioinformatics expertise, which remains a barrier for many experimental researchers. To overcome this hurdle, we developed TargetMosaic, an efficient and easy-to-implement workflow operating via a kmer-based approach for species-wide assessment of protein sequences to assess conservation within a bacterial species. TargetMosaic automates the download of genome annotations from NCBI, identification of homologous sequences from a reference FASTA, and evaluation of gene presence and domain-level conservation. Designed for SLURM-based clusters, it supports parallelized data retrieval, kmer based protein sequence extraction, multiple sequence alignment, and functional domain variant analysis. The tool is particularly useful for the development of protein-based antibiotics as well as antibodies, nanobodies and in-silico designed scaffolds against protein targets that are known to have pre-existing mutations. Applied to representative bacterial proteins, TargetMosaic effectively summarizes conservation patterns across thousands of strains, highlighting its scalability and interpretability.

Availability and Implementation: TargetMosaic is implemented in Python (≥3.10) and runs on ubuntu shell with access to SLURM-based clusters. It is available on GitHub under the MIT License: https://github.com/arpita-sahoo/TargetMosaic

**Student Paper*

P080 "Biocontrol of Plant Pathogen Responsible for Guava Die- Back Disease by Marine Microbial Source"

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Guava (Psidium guajava L.), an important member of the Myrtaceae family and a major fruit crop in tropical and subtropical regions, is critically affected by die-back disease, leading to significant economic losses. In countries like Pakistan, a leading global producer, this disease severely impacts yield and export potential due to a reliance on conventional control methods and a lack of effective postharvest management practices.
The fungal genus Colletotrichum, particularly C. gloeosporioides, is a primary causative agent of guava anthracnose and die-back, responsible for substantial pre- and post-harvest fruit decay. This study aimed to isolate and characterize the specific Colletotrichum species responsible for die-back disease in guava orchards of Sindh, Pakistan. Furthermore, it investigated the biocontrol potential of marine-derived microorganisms against this pathogen.
Marine ecosystems are a rich source of novel bioactive compounds. In this research, marine microbes were isolated and screened for their antagonistic activity against the identified fungal pathogen. The results demonstrate a significant suppressive effect of selected marine microbial isolates on the mycelial growth and sporulation of Colletotrichum spp. in vitro. This study concludes that marine microbial sources represent a promising and sustainable alternative to chemical fungicides for the biocontrol of guava die-back disease, offering a potential strategy to mitigate production losses and enhance fruit yield.
Keywords: C. gloeosporioides, Plant Pathogen, Guava Die-Back Disease, Anthracnose, Biocontrol, Marine Microbial Antagonists.

P081 Metaproteomic analysis reveals stable methanogenic communities during anaerobic digester reconfiguration and glycerol amendment

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Anaerobic digestion relies on complex microbial consortia that convert organic substrates into methane through syntrophic interactions between bacteria and methanogenic archaea. Understanding how microbial communities respond to operational changes is essential to improve reactor stability and methane production. In this study, we investigated microbial community dynamics and metabolic functions in anaerobic digesters during a transition from parallel reactor operation to serial configuration, followed by glycerol amendment as an additional carbon source.

Metaproteomic analysis was used to characterize protein abundance profiles, taxonomic composition, and metabolic pathway activity across multiple time points during reactor operation. Principal coordinate analysis was used to assess temporal shifts in microbial community structure and functional metabolism, while grouped comparisons of parallel, primary, secondary, and glycerol-amended reactors were used to evaluate changes in dominant taxa and metabolic modules.

Taxonomic analysis revealed that the microbial community was dominated by methanogenic archaea, particularly members of the family *Methanotrichaceae* (order *Methanotrichales)*, representing approximately 30–40% of the detected community. Several syntrophic bacterial groups, including *Syntrophales* and *Propionibacteriales*, were also abundant and likely contribute to methane production through degradation of fatty acids and fermentation intermediates. Multivariate analysis demonstrated gradual temporal shifts in microbial community composition and metabolic profiles as the reactors transitioned from parallel to serial operation. Moderate differentiation between primary and secondary reactors was observed, while glycerol addition caused only minor shifts in metabolic activity.

Overall, the results demonstrate the resilience of methanogenic microbial communities and highlight the stability of methane-producing pathways despite operational changes. These findings emphasize the importance of syntrophic microbial networks in maintaining stable anaerobic digestion performance under varying reactor configurations and substrate conditions.

P082* DecoloniX: Phage-Based Strategies for Non-Typhoidal Salmonellosis (NTS) Decolonization.

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Background: Acute non-typhoidal salmonellosis is the second most common cause of gastroenteritis in Switzerland. Antibiotics are not recommended due to limited efficacy and the perturbation they cause to the gut microbiota, a natural barrier to the infection. Thus, untreated carriers represent an important reservoir for transmission to vulnerable people, in whom the infection can become life-threatening. Using bacteriophages, specific bacterial viruses, is proposed as a decolonization strategy.

Methods: A collection of Salmonella clinical isolates was assembled in collaboration with NENT, UZH. Bacteriophages were isolated from local wastewaters. Their host ranges were characterized through diluted drop tests and their efficacy by turbidity assays. *In silico* analyses of the phage genomes allowed evaluation of their suitability for clinical intervention.

Results: We retrieved seventeen genetically distinct lytic phages. None carried genes coding for known virulence or antimicrobial resistance determinants. The collection efficiently infected and lysed 99 % of 106 *Salmonella* clinical isolates from the six most dominant serovars (Enteritidis, Typhimurium, Monophasic Typhimurium, Napoli, Infantis, and Derby). Infantis and Derby remained resistant, suggesting lack of receptors or anti-phage mechanisms. Single phage turbidity assays showed prolonged bacteriostatic effects (up to 6-8 h) while phage combination managed to increase the effect up to 14h.

Conclusions: These results identify genetically diverse *Salmonella* phages from Swiss wastewaters with promising clinical potential. Future work will assess the impact of anti-phage systems in bacteria, GMP production at CHUV, effect on commensal flora of the gut microbiota, and *in vivo* performance in non-typhoidal salmonellosis models such as *G. mellonella*, SICs and mice models.

**Student Paper*

P083 Nutrient environments dictate antibiotic susceptibility across gut bacteria

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Antibiotic treatment disrupts gut microbiome composition, potentially leading to dysbiosis and disease. The extent of this disruption is strongly influenced by microbial resistance, antibiotic class, and mechanism of action. While antibiotic- and compound-specific effects on microbial tolerance have been documented, the role of other environmental factors, particularly the nutrient environment, in shaping microbial metabolism and community structure remains underexplored.

In this study, we systematically screened 32 representative gut bacterial strains across 11 undefined growth media and found that minimum inhibitory concentrations (MICs) varied markedly in a species- and medium-dependent manner. Several species within the genera Bacteroides and Parabacteroides exhibited distinct responses to a range of mono- and disaccharides, resulting in saccharide- and species-specific changes in ciprofloxacin sensitivity. In most species tested, the presence of glucose, mannose, or sucrose decreased the MIC, whereas maltose had little or no effect. While most Bacteroides and Parabacteroides species responded to at least three of the tested sugars, Bacteroides thetaiotaomicron showed no detectable change in ciprofloxacin MIC at sugar concentrations up to 6 g/L.

The observed species-specific differences in nutrient-dependent MIC modulation suggest opportunities for the targeted protection or selective elimination of bacteria through manipulation of the nutrient environment.

Overall, our results highlight the importance of considering the nutrient environment when assessing the impact of antibiotics, particularly in gut microbes that are continuously exposed to fluctuating nutrient conditions.

P084* metabolic signatures of microbial consortia for biofungicide design against r. solani

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Fungal pathogens are responsible for significant crop losses worldwide and represent a major threat to global food security. Developing sustainable approaches to control these pathogens is therefore essential, particularly to reduce reliance on synthetic fungicides, which can have detrimental effects on soil health, the environment, and human health. Biological control methods represent a promising alternative and can be effectively integrated into Integrated Pest Management (IPM) strategies.

Switzerland is not exempt from these challenges. The soil-borne fungal pathogen *Rhizoctonia solani*, responsible for severe damage to various horticultural crops, is a key target for the development of sustainable biocontrol solutions. In this context, the ENHANCE project was established, building on the Microbial Consortium Approach (MCA) concept. The project aims to encapsulate a consortium of beneficial fungi and bacteria within alginate-based microcapsules to develop a novel biofungicide. The efficiency of the consortium relies on the fungal highway concept, whereby bacteria are delivered to the infection site by travelling along fungal hyphae through the soil.

This study contributes to this broader initiative by developing experimental methods to investigate the interactions between the microbial consortium, the fungal pathogen, and the host plant. In particular, this work investigates the modes of interaction by analysing chemical profiles and secondary metabolites produced by the organisms when cultivated individually or in co-culture conditions. A central challenge of this work was designing setups that simultaneously establish suitable environmental conditions for biological interactions and remain compatible with metabolite recovery for HPLC–MS analysis.

The insights gained from this research contribute to a deeper understanding of the biological and chemical mechanisms underlying consortium-mediated biocontrol, and support the future development of effective, environmentally friendly crop protection solutions.

**Student Paper*

P085* real-time analysis of polymicrobial infections in vivo: interactions between pseudomonas aeruginosa, klebsiella pneumoniae, and acinetobacter baumannii

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Polymicrobial infections, defined by the coexistence of multiple pathogens within the same host, are a major cause of treatment failure and poor clinical outcomes. Among the multidrug-resistant ESKAPE pathogens (*Enterococcus faecium, Staphylococcus aureus, Klebsiella pneumoniae, Acinetobacter baumannii, Pseudomonas aeruginosa*, and *Enterobacter* spp.), *P. aeruginosa, K. pneumoniae,* and *A. baumannii* are frequently co-isolated in pulmonary and bloodstream infections, particularly in intensive care unit patients. Despite the growing clinical recognition of polymicrobial infections, most studies rely on in vitro systems, leaving major gaps in our understanding of pathogen interactions in vivo.

In this study, we used the zebrafish (*Danio rerio*) larval model, whose optical transparency enables real-time visualization of infection dynamics. By microinjecting bacteria in pairwise combinations at different inocula, we investigated how polymicrobial interactions influence larval survival, bacterial growth dynamics, innate immune cell recruitment, and host-pathogen proteomic responses.

At high inocula, larvae infected with *P. aeruginosa* alone exhibited lower survival than polymicrobial infections, indicating that infection outcomes are shaped not only by pathogen burden but also by interspecies interactions that alter disease dynamics. Real-time imaging during the first 24 hours post-infection further revealed that *P. aeruginosa* growth remained spatially restricted while selectively inhibiting *A. baumannii* between 7 and 24 hours post-infection, without significantly affecting *K. pneumoniae*. These findings suggest that *P. aeruginosa* virulence and competitive behavior do not depend on extensive bacterial expansion, but rather on the production of virulence-associated and antagonistic factors that suppress competing pathogens within the host environment.

Ongoing experiments aim to further determine whether polymicrobial infections alter innate immune system recruitment and reshape the host-proteomic response. By integrating live imaging, immune profiling, and proteomic analyses, this work seeks to identify bacterial traits induced during co-infection that may represent novel therapeutic targets to reduce virulence and improve infection control alongside conventional antibiotics.

**Student Paper*

P086 Nanopore Sequencing for Foodborne Pathogen Surveillance

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Background: Whole-genome sequencing (WGS) is a cornerstone of modern pathogen surveillance and cross-border outbreak detection. Oxford Nanopore Technologies (ONT) rapid barcoding sequencing is gaining popularity as an accessible, fast, and cost-effective alternative to Illumina-based workflows. However, sequencing of native DNA may be affected by DNA modifications that compromise basecalling accuracy and downstream genotyping, raising concerns about the reliability of ONT-only approaches.

Methods: We applied both Illumina and ONT sequencing (R10.4.1 flow cells, RBK114.96 kit, dorado SUP v5.2 bacterial model) to 294 genetically diverse isolates from ten major foodborne pathogens, including *Salmonella enterica*, *Listeria monocytogenes*, and others. Isolates originated from clinical, food, and environmental surveillance in Switzerland. To improve quality control of ONT-only workflows, we developed alpaqa, a lightweight computational tool to detect systematic nanopore assembly errors without requiring reference genomes or short-read data.

Results: At 50× coverage, 97.3% (286/294) of ONT-only assemblies produced identical or near-identical cgMLST profiles (≤3 allelic differences) compared to Illumina-polished references. Most traditional ONT-associated errors, such as those in homopolymer or methylated regions, were largely resolved. However, elevated error rates were observed in a subset of isolates, notably *S. enterica* serovar Kentucky and *L. monocytogenes*, associated with phosphorothioate (*dnd*) or methylation systems, with up to 38 mismatching alleles. Alpaqa successfully identified assemblies affected by such systematic errors. Masking low-quality regions improved cgMLST accuracy, albeit with reduced locus completeness.

Conclusion: Current ONT rapid sequencing workflows enable highly accurate ONT-only WGS for foodborne pathogen surveillance. Nonetheless, rare DNA modifications, particularly phosphorothioation, can compromise accuracy. Tools such as alpaqa provide an effective safeguard, supporting the robust integration of ONT-only sequencing into routine genomic surveillance frameworks.

P087 Geophysical imaging and radiocarbon age models link peat depth and age to microbial community transitions in sub-alpine peatlands

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Peatlands are the largest terrestrial stores of organic soil carbon and significant sources of methane and carbon dioxide, yet the microbial communities in sub-alpine peatlands are poorly understood. We investigated six peatland sites to depths up to 5.5 m in the UNESCO Biosphere Reserve Entlebuch, Switzerland via 16S amplicon sequencing, radiocarbon dating, and electrical resistivity tomography (ERT). Peat age and depth were highly influential in determining microbial community composition, with a distinct inflection point at ~250 cm, corresponding to uncalibrated radiocarbon age of ~2,500 years BP, where methanogenic Rice Cluster II gave way to a predominance of Bathyarchaeia. Furthermore, deep peat profiles were also characterized by higher abundances of rare phyla including Asgardarcheota, CPR, and DPANN superphyla as well as the putative methanotroph Methylomirabiliaceae. Surface peat communities were also strongly dominated by methanogens, although driven primarily by land use history and hydrological state. The overall abundance of archaea, particularly methanogens, was substantially higher than that of well-studied pristine northern peatland sites, especially in surface peat, indicating a distinct community composition with important implications for methane cycling and peatland function. In ongoing analysis, we compare the microbial depth composition, particularly archaea, with ERT-deducted peat structure to infer microbial communities across other peat sites. Our results establish sub-alpine peatlands as compositionally distinctive and vertically stratified methane-cycling ecosystems with depth-driven ecological niches that underscore the need to incorporate depth- and age-resolved microbial data into greenhouse gas emission models.

P088 Competition between co-localized symbionts underlies inter-individual variation for bacterial gut colonization in Apis mellifera

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Microbes in the animal gut compete to colonize spatially restricted host niches. However, whether competition drives inter-individual variation among hosts, and which factors determine the outcome of such competition, remain poorly understood. Here, we investigated competitive interactions of the western honeybee gut symbiont *Frischella perrara* with other members of the bee gut microbiota. Shotgun metagenomics analysis of individual bees revealed that *F. perrara* is negatively correlated with a specific species of the genus *Gilliamella*. Co-colonization of microbiota-depleted bees with these two bacteria resulted in their competitive exclusion. The outcome of this competition depended on the relative number of bacteria each bee received and benefited *Gilliamella* when one of the two type VI secretion systems of *F. perrara* was mutated. Using fluorescently tagged strains, high-resolution microscopy, and gut region–specific quantification, we show that both bacteria localize to the same host niche in the ileum of mono-colonized bees, indicating competition for space. This competition provides an explanatory mechanism underlying variation in the occurrence of *F. perrara* across honeybee colonies and highlights the importance of gut spatial structure and microbial competition in shaping microbiome composition and inter-individual variability.

P089 Investigation of comEC as a potential driver of competence in Chlamydia spp.

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The *Chlamydiaceae* family comprises host-adapted pathogens that cause various diseases in animals and humans. Their adaption to a unique obligate intracellular niche resulted in the reduction of their genomes including genes pertaining to horizontal gene transfer (HGT), such as the conjugation machinery. However, exemplified by the presence of *tetA*(C)-induced tetracycline resistance in porcine *Chlamydia (C.) suis*, HGT events are rare but possible for the chlamydiae. Although the precise uptake mechanism remains unclear, competence-induced genetic transformation appears to be the most plausible uptake pathway. The only chlamydial competence gene identified to date is *comEC*, located on the chromosome of *C. trachomatis*, in which initial functional studies demonstrated its importance for DNA exchange. Little is known about the presence of *comEC* in other chlamydiae, its expression level during the chlamydial developmental cycle and its effects on transformation efficiency. In this present study, we examined the conservation level of *comEC* across all recognized *Chlamydia* spp., characterized its expression during the developmental cycle, and assessed its function through knock-out and overexpression approaches. *In silico* analysis showed that *comEC* is present in all known *Chlamydia spp.* including the recently described *Chlamydiifrater volucris*. The level of conservation was low to moderate across species but high within *C. suis* with signs of homologous recombination. Preliminary data has further shown that *comEC* is expressed during the replicative phase of *C. suis* (mid-cycle gene). Moreover, it appears that transformation is prevented upon gene inactivation by lamda Red recombineering, but that its constitutive overexpression limits rather than promotes chlamydial competence. Overall, this study revealed that *comEC* is a tightly regulated, non-essential mid-cycle gene required for chlamydial HGT. Next, we aim to unravel the complex regulation of *comEC* by its conditional overexpression in *C. suis* and *C. trachomatis* at different times of the developmental cycle.

P090 Antimicrobial resistance in Steinach river impacted by combined sewer overflow events

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The spread of antimicrobial resistance (AMR) is a major public health concern, and under the One Health framework, aquatic environments are considered as dissemination pathways and sites of AMR evolution. Combined sewer overflows (CSOs) are recognized as important pathways for the release of AMR into surface waters. However, the impacts of untreated overflow discharges during precipitation events remain poorly characterized. This study investigates the occurrence, dynamics, and environmental implications of AMR associated with sewer overflows in the Steinach River (St. Gallen, Switzerland), a unique urban river system that frequently receives untreated overflow discharges but no treated wastewater effluent. The study aims to develop transferable methodologies for monitoring AMR during episodic sewer overflow events and to assess the extent to which these discharges influence river water, sediments, and epilithic biofilms. Event-based sampling upstream and downstream of overflow locations will be combined with cultivation based analyses of antibiotic-resistant bacteria, quantitative PCR targeting antibiotic resistance genes, and capture-enrichment metagenomic characterization of riverine resistomes. The project will further evaluate whether sewer overflow events lead to transient or persistent AMR contamination in aquatic river habitats. By evaluating micropollutants alongside AMR it will identify critical sources and provide new practical guidelines for CSO situational assessment as a basis for decisions on mitigation opportunities, including retention systems and blue-green infrastructure. By integrating microbial, chemical, environmental, and catchment-scale analyses, the project will provide a scientific basis for incorporating micropollutants and AMR into future Swiss sewer overflow management guidelines and support the development of evidence-based monitoring and mitigation strategies for urban water systems. Concepts and initial results will be presented.

P091 INSSIGHT: A modular in silico EQA framework for benchmarking of bacterial WGS bioinformatics pipelines

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Bacterial whole-genome sequencing (WGS) is now routinely performed in microbiology laboratories worldwide. Ensuring reproducibility and quality control (QC) of downstream bioinformatics pipelines remains a major challenge. Analytical outputs can differ across laboratories, software and versions, and QC-thresholds. To address this, we developed INSSIGHT, an international, in-silico EQA framework designed to evaluate and harmonise WGS analysis pipelines. The platform offers a standardised approach for assessing core bioinformatics functions, failure-handling logic, and consistency in genomic interpretation. A taxonomically diverse genome dataset was generated covering 141 relevant clinical, foodborne, and environmental relevant bacterial and yeast species. Illumina (PE150) sequencing data included high-quality, borderline, and deliberate failures, selected to reflect real-life variability.

The EQA scheme was structured into four modules: Module1: preprocessing, assembly, contamination detection, and species identification; Module2: antimicrobial resistance (AMR) gene detection; Module3: clustering and outbreak inference; Module4: edge cases assessing QC thresholds and interpretive decision-making.

In a pilot phase, the dataset was distributed to eight research or clinical microbiology laboratories across seven centres in six countries and independent outputs were collated and analysed. Species identification was largely concordant, but discrepancies occurred within species complexes, closely related species and for yeast. The number and identity of detected ARGs varied between laboratories, with inferred resistance mechanisms being imilar but not fully consistent. Most laboratories correctly identified expected clusters, but parameter choices influenced interpretation. Labs using the same cgMLST scheme produced identical outputs, whereas different cgMLST schemes or SNP analyses correlated but did not match exactly, underscoring the need for harmonisation when comparing outbreak analyses. Heterogeneity in QC cutoffs used to determine acceptable coverage, contig number, or contamination levels, affected whether sequences were accepted or rejected by the different labs.

P092* monitoring putative desulfonation genes involved in PFAS degradation by Gordonia sp. NB4-1Y using quantitative PCR assays

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Perfluoroalkyl and polyfluoroalkyl substances (PFAS) are anthropogenic compounds that are considered environmental contaminants of concern due to their persistence, toxicity, and bioaccumulative potential. PFAS are characterized by long fluorinated carbon chains containing strong carbon-fluorine bonds, which promote chemical stability and resistance to biodegradation. The use of certain PFAS, including 6:2 fluorotelomer sulfonamidoalkyl betaine (6:2 FTAB) and 6:2 fluorotelomer sulfonate (6:2 FTSA), as ingredients in aqueous film-forming foams (AFFFs) used for Class B hydrocarbon fire suppression, has contributed to widespread environmental contamination. The aerobic soil bacterium *Gordonia* sp. strain NB4-1Y, a Gram-positive bacterium originally isolated from vermicompost, has been shown to degrade 6:2 FTAB and 6:2 FTSA to utilize them as sulfur sources for growth. In this study, quantitative chip digital polymerase chain reaction (dPCR) assays for six genes of interest, selected based on published transcriptome data and recently acquired proteome data, and a reference gene were designed and validated. Growth kinetics of *Gordonia* sp. strain NB4-1Y were analyzed in a time course experiment conducted in biological triplicate where NB4-1Y was provided with 6:2 FTAB, 6:2 FTSA, a non-fluorinated analog of 6:2 FTSA (1-octanesulfonate), or $MgSO_4$ as sole sulfur source. Optical density (OD_{600nm}) and fluoride ion release were monitored, and cell pellets were collected for RNA extractions at 13 time points over a period of 22 days. Quantitative PCR assays and dPCR was used to quantify gene expression across treatments at four time points. One-way analysis of variance (ANOVA), followed by Tukey's Honest Significant Difference (HSD) post hoc test, was used to determine whether gene expression differed with sulfur source. Higher expression for genes of interest was hypothesized when NB4-1Y was exposed to 6:2 FTSA or 6:2 FTAB compared to $MgSO_4$. Results presented here contribute to further understanding of the mechanisms involved in microbial PFAS degradation.

**Student Paper*

P093 Bioprospection of maize rhizoplane-associated bacteria for consortia construction across different Moroccan agro-systems for plant growth and P acquisition

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Sustainable improvement of agricultural productivity is crucial for addressing food security challenges. Beneficial plant-associated microbes, particularly beneficial microbial consortia play crucial role in enhancing crop resilience and yield. In the present study, rhizoplane-associated bacteria were isolated form maize plant across seven different Moroccan agroecosystems using a novel consortia-oriented isolation approach (niche conservatism) aiming to capture a range of functional species belonging to the same niche with the potential to promote plant growth. Genetic fingerprinting using randomly amplified polymorphic DNA-generated profiles allowed elimination of duplicates before bacterial identification using 16s sequencing. Phylogenetic analysis of 16S rRNA sequences revealed the diversity of the isolates form which a total of 36 BC were constructed with, 28 intra-zone consortia, seven intra-region and one global consortia. Assessment of the effect of BC inoculation on the above and below plant growth parameters of maize plant under green-house conditions revealed increased overall plant biomass production mainly plant shoot and root dry weight , improved plant nutrients uptake (N, P, and K) , as well as inducing root morphological traits which was associated with enhanced phosphorus (P) solubilization in the soil. Quantification of genes contributing to P cycling in soil revealed the abundance of *gcd* gene in most of the BC, followed by the *pqqC* gene, while the *phoD* gene which was present in only one BC. Our findings highlight the efficiency of the adopted consortia construction approach for designing potential BC with practical applications in crop improvements.

P094* microbial growth and community responses to antiscalants in seawater desalination systems

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Water desalination removes salts and other substances from seawater to produce water suitable for human consumption and industrial use. Reverse osmosis (RO) is one of the most widely applied desalination technologies, but its sustainability is challenged by membrane fouling, particularly scaling and biofouling. To control scaling, antiscalants are routinely added to feedwater; however, growing evidence suggests that these chemicals can unintentionally promote or inhibit microbial growth. This is of concern given their discharge via desalination brine and the potential disruption of marine microbial and biogeochemical cycles.

To investigate bacteria–antiscalant interactions under environmentally relevant conditions, we developed a growth potential assay using natural seawater and its autochthonous microbial community. We evaluated the microbial response to several commercially used antiscalants, including phosphonates, polymers, and blended formulations. Bacterial growth was monitored by flow cytometry, and microbial community composition was assessed using 16S rRNA gene sequencing. Complementary analyses included nuclear magnetic resonance (NMR) to determine antiscalant chemical identity, measurements of total organic carbon (TOC) and phosphate in seawater and antiscalant formulations, and iron quantification before and after antiscalant addition using mass spectrometry.

Our results show that microbial responses to antiscalants are governed by chemical composition, concentration, and ambient seawater conditions. Phosphonate-based formulations stimulated bacterial growth when supplying bioavailable phosphate, but only under phosphorus-limited conditions. Similarly, some polymeric antiscalants enhanced growth in a concentration-dependent manner, suggesting alternative carbon-driven pathways. In contrast, growth-inhibiting antiscalants consistently displayed bacteriostatic effects, supporting a mechanism linked to sequestration of essential trace metals such as iron.

Overall, this study supports a systematic and environmentally informed approach to antiscalant selection that balances effective fouling control in RO systems with minimization of impacts on marine microbial processes.

**Student Paper*

P095 Non-Antimicrobial Approaches to Control MDR Salmonella Kentucky: Role of Synbiotics and Probiotics

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Multidrug-resistant (MDR) *Salmonella* Kentucky has emerged as a global concern, highlighting the need for non-antimicrobial strategies to control non-typhoidal *Salmonella* (NTS) infections. This study evaluates the therapeutic potential of synbiotic and probiotic interventions against MDR *S*. Kentucky.

Clinical samples from children ≤5 years with acute gastroenteritis (2017–2024, Dr. B.C. Roy Hospital, Kolkata, India) were processed for NTS identification. Isolates were characterized using antimicrobial susceptibility testing (AST), ARG profiling, plasmid analysis, Multilocus sequence typing (MLST), pulse-field gel electrophoresis (PFGE), and whole genome sequencing (WGS) of representative MDR strains. The efficacy of synbiotic and probiotic treatments was assessed through in vitro and in vivo assays.

A total of 90 (1.18%) *Salmonellae* were recovered after screening 7567 samples. *S*. Typhimurium (31; 34.44%) was predominant, followed by *S*. Kentucky (14; 15.55%) and *S*. Bareilly (7; 7.77%). MDR was predominantly observed in *S*. Kentucky (n=12/14; 85.71%) with a high rate of resistance towards ampicillin, nalidixic acid, and ciprofloxacin (100% each), tetracycline (91%), & cephalosporins (61.53%). WGS revealed the presence of $bla_{\text{TEM-1}}$, $bla_{\text{CTX-M-15}}$, $bla_{\text{CTX-M-55}}$, $bla_{\text{OXA-9}}$, *tetA, aadA1* and *aadA2, sul1, and mphE. A* double mutation in *gyrA* (D87Y, S83F) and a single mutation in *parC* (S80I) are responsible for resistance in FQs. PFGE showed *S*. Kentucky isolates are heterogeneous with a similarity coefficient of 19.38%. Seven-day synbiotic/probiotic treatment significantly reduced ileal colonization and liver dissemination of *S*. Kentucky compared to the infection-only group. Infection caused villus damage and crypt hyperplasia, which were attenuated by synbiotic treatment. Proinflammatory cytokine levels were markedly elevated in the infection group but reduced in synbiotic- and probiotic-treated groups.

The tested synbiotic can be used to reduce the severity of the infection and to limit the use of reserve antibiotics. Besides this, continuous monitoring of AMR profile is recommended for preventing the spread of resistant organisms.

P097 Impact of the ripening cellar environment on the microbial composition of Ticino alpine cheese

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Cheese production is one of the oldest fermentation-based preservation methods used today. Written evidence of alpine cheese production in southern Switzerland dates back to the 12th century. A representative example of such traditional production is the Ticino alpine cheese (*Formaggio* d'alpe ticinese), a Protected Designation of Origin (PDO) product. PDO guidelines delimit pasture geography, origin of the milk mix, and ripening conditions, yet the final product from each alpine farm retains its own distinct organoleptic and physical properties. During maturation, bacteria, yeasts, and filamentous fungi naturally colonize the cheese surface. Ripening cellars and their environmental conditions can favor the growth of specific taxa, which contributes to matured cheese characteristics.

To investigate the influence of ripening cellars on the microbial composition of Ticino alpine cheese, we placed cheese wheels produced at three PDO sites (Lucomagno Croce, Pertusio, Piora) in their original cellars and the other two facilities. We sampled one month after cheese production and at the end of the maturation period in the Alps (2.5 months). Cheese rind samples were analyzed by 16S rRNA gene and ITS region metabarcoding to characterize their microbiota. The results allow us to: i) compare the microbial communities of cheese ripened in different cellars, providing insights into the influence of the ripening environment on the final microbial composition; ii) assess temporal changes in microbial composition during ripening (early versus late stages); iii) expand upon previous preliminary findings regarding the microbiota of traditional Ticino alpine cheese.

P098 Phenotypic and biochemical characterization of the class C β-lactamase, PAC-1

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Background The class C β-lactamase, PAC-1, was originally described as acquired in Pseudomonas aeruginosa clinical isolates from South Asia, the corresponding gene being chromosomally located and embedded within a Tn1721-like transposon. Despite its association with high-level resistance to cephalosporins and β-lactam/inhibitor combi- nations, data on its full hydrolytic profile and kinetic parameters remain scarce. The aim of this study was to characterize the susceptibility profile and hydrolytic kinetics of PAC-1, with a particular focus on cefiderocol hydrolysis, and the activity of novel β-lactam/β-lactamase inhibitor combinations. Materials The blaPAC-1 allele was cloned into vector pUCP24 and expressed in Escherichia coli TOP10 and P. aeruginosa PAO1, respectively. MICs were performed by broth microdilution and interpreted according to EUCAST guide- lines. PAC-1 was purified and steady-state kinetic measurements of selected β-lactams were determined. IC50 values were determined for the beta-lactamase inhibitors avibactam, taniborbactam, xeruborbactam, and zide- bactam. Whole-genome sequencing was performed to characterize the genetic environment of blaPAC-1. Results PAC-1 conferred broad resistance to penicillins, broad-spectrum cephalosporins, and cefiderocol, while car- bapenems remained unaffected. Kinetic analyses confirmed high catalytic efficiency against cefotaxime and aztreonam, but no hydrolysis of imipenem. Avibactam failed to inhibit PAC-1 (IC50 >1,000 µM), whereas xerubor- bactam and zidebactam showed potent inhibitory activities (IC50 = 1.0 and 0.6 µM, respectively). A chromoso- mally located Tn1721-like transposon was identified carrying the blaPAC-1 in P. aeruginosa ST4936. Conclusions PAC-1 is not inhibited by avibactam and hydrolyzes a broad range of β-lactams including the last resort an- tibiotic cefiderocol. The combination cefiderocol/zidebactam was the most active against PAC-1. Transposon- mediated dissemination across distinct lineages warrants continued epidemiological surveillance of PAC-type β-lactamases.

P099 Establishing a standardized diagnostic pipeline for phage susceptibility tests

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Background

Antibiotic-resistant bacteria have renewed interest in bacteriophage therapy for the personalized treatment of multidrug-resistant infections. Phage susceptibility tests (PSTs) such as spot assays (SAs) and turbidity reduction assays (TRAs) are essential for screening and selecting therapeutic bacteriophages. However, most laboratories rely on in-house protocols and readouts, which limits comparability and implementation of phage diagnostics in routine microbiology. Here, we aim to develop and evaluate a standardised PST workflow that can be integrated into diagnostic processes similar to antibiotic susceptibility testing.

Methods

We evaluated SA and TRA reproducibility using Escherichia coli K-12 and three lytic bacteriophages (BASEL-38, BASEL-48 and BASEL-54). SAs on LB soft-agar were scored semi-quantitatively based on plaque morphology and lysis patterns. TRAs were performed in 96-well plates, recording OD600 over time. Bacterial inhibitions were summarised as percentage inhibition using area under the curve. The workflow was piloted on clinical *E. coli* isolates, including UTI89, exploring strain-specific challenges for assay standardisation.

Results

For *E. coli* K-12, SAs with BASEL-38, -48 and -54 produced consistent plaque phenotypes and categorical readouts across all replicates, indicating high intra-assay robustness. TRAs generated highly reproducible growth-inhibition curves for all three phages, with narrow variation in percentage inhibition. Together, these data support the feasibility of using K-12 and selected BASEL phages as internal controls for PST implementation. Preliminary application of the workflow to the clinical strain UTI89 confirmed the overall functionality of the assays but revealed increased variability in TRA readouts, suggesting that host strain characteristics and media conditions can strongly influence quantitative PST results.

Conclusions

Our findings confirm that standardized SA and TRA protocols are achievable and robust for reference strains. However, clinical implementation requires strict control of environmental variables. This workflow represents a step towards harmonized phage diagnostics, facilitating the integration of personalized phage therapy into routine clinical microbiology.

P100 Culture-based methods underestimate antimicrobial resistance burden in retail fresh produce

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Antimicrobial resistance (AMR) poses a critical threat to human and animal health, undermining our ability to treat infectious diseases. While food is a pathway for AMR transmission, its presence in fresh produce remains underexplored. Consuming raw fruits and vegetables may expose individuals not only to food pathogens but also to antibiotic-resistant bacteria. In this study, we combined traditional culture-based techniques with modern molecular approaches to assess AMR on carrots, lettuce, parsley, basil, and tomatoes, purchased from two big Swiss retailers. Enterobacterales resistant to third-generation cephalosporins were most prevalent in herbs and lettuce but absent in carrots and tomatoes, whereas carrots harboured a high abundance of vancomycin-resistant enterococci, which were not detected in lettuce or tomatoes. Across all samples, the abundance of nine selected antibiotic resistance genes was highest in herbs, followed by carrots, lettuce, parsley, and basil, and lowest in tomatoes. Nearly all samples tested positive for beta-lactam, tetracycline, macrolide-lincosamide-streptogramin, sulfonamide, and aminoglycoside resistance genes, as well as the class 1 integron. While we observed a positive correlation between total Enterobacterales counts and 16S rRNA gene abundance, molecular methods detected more positive samples than traditional culture-based techniques. Phenotypic analysis revealed that the highest resistance rates were exhibited by Enterobacterales isolates from herbs and lettuce, with numerous isolates displaying resistance to the third-generation cephalosporins and carbapenems. Whole-genome sequencing confirmed the presence of cephalosporin and quinolone, but not carbapenem resistance genes. Furthermore, multidrug-resistant plasmids encoding resistance to colistin, chloramphenicol, sulfonamides, and streptomycin were identified in *Enterobacter* isolates, with similar plasmid-borne resistance genes also detected in isolates of *Escherichia* , *Klebsiella* , and *Citrobacter* species. A significant proportion of these multidrug-resistant bacteria originating from herbs or tomatoes. Together, these findings demonstrate that fresh produce is a critical reservoir for multidrug-resistant bacteria and highlight the urgent need for robust AMR surveillance in retail supply chains.

P101 Can we manipulate the gut of Zophobas morio larvae (ZmL) to obtain a stable human-like microbiota?

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BACKGROUND. The human gut is a major reservoir for multidrug-resistant pathogens, yet scalable *in vivo* models for evaluating decolonization strategies remain limited. We previously established *Zm*L as an alternative host model in alignment with the 3Rs. Here, we tried to humanize the gut of *Zm*L to mimic the effects of new decolonization strategies in humans.

METHODS. A pool of human stool was incorporated into the diet administered to *Zm*L every 48-h for 28-days, followed by a 28-day washout phase with non-contaminated food. A parallel control group received the baseline diet throughout the entire 56-day experiment. Gut microbiome dynamics were profiled weekly (T0–T56) using 16S rRNA sequencing of whole larval homogenates.

RESULTS. The humanized diet significantly altered gut microbiota composition compared with the control group between T7 and T28, as assessed by a Bray-Curtis distance matrix (PERMANOVA, R^2 = 15.0%, p= 0.017). This compositional shift occurred without a significant change in alpha diversity; the Shannon Diversity Index remained stable (5–6) in both groups throughout the experiment, suggesting one-to-one niche substitution rather than net community expansion. This was driven by *de novo* engraftment of highly abundant human taxa (top 10; e.g., *Bacteroides*, *Bifidobacterium*, *Akkermansia*, *Blautia*) completely absent in controls, alongside enrichment of native genera (e.g., *Enterococcus*, *Latilactobacillus*, *Pediococcus*). During the washout phase, engrafted human taxa dropped rapidly within 7-days of dietary cessation (T35), indicating transient rather than permanent colonization.

CONCLUSIONS. Our exploratory humanized *Zm*L system functions as a transient, dietary-dependent *in vivo* model rather than a stable colonization platform. Engrafted human taxa rapidly washed out upon removal of the fecal diet, allowing the native microbiota to re-establish dominance. Therefore, a refined strategy to achieve stable human microbiome colonization in *Zm*L is required. This might include prior depletion of the native flora to create a vacant ecological niche before fecal administration.

P102 Evaluation of a rapid immunochromatographic assay for the detection of mcr-mediated colistin resistance in Gram-negative bacilli

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Background: Colistin remains a critical last-line antibiotic for the treatment of severe infections caused by multidrug-resistant (MDR) Gram-negative bacilli. The emergence of plasmid-mediated colistin resistance, driven by *mcr* genes represents a major public health concern due to its rapid worldwide dissemination. To date, ten *mcr* variants (*mcr-1* to *mcr-10*) have been identified, highlighting the need for rapid and reliable detection methods in order to adapt antibiotic treatments quickly and limit the spread of these resistance mechanisms. *mcr*-mediated colistin resistance has been widely reported in Asian countries, particularly China and India, in both human and animal isolates. The RESIST-1 MCR Multi immunochromatographic assay (Coris BioConcept) is designed to detect the most common *mcr*-mediated mechanisms of colistin resistance.

Methods: We evaluated the performance of the RESIST-1 MCR Multi assay using a panel of 100 Gram-negative bacilli isolates from the laboratory collection as well as some of the National Center for Emerging Antibiotic Resistance, Switzerland. The collection included 63 *mcr*-positive isolates carrying variants from *mcr-1* to *mcr-9*, 18 colistin-resistant isolates without *mcr* genes, and 19 colistin-susceptible isolates used as negative controls. The tested species belonged predominantly to the order *Enterobacterales*, with additional non-fermenting Gram-negative bacilli, including *Acinetobacter baumannii* and *Pseudomonas aeruginosa*.

Results: All isolates harboring *mcr-1*, *mcr-2*, and *mcr-6* in each species were correctly detected, whereas isolates carrying the other tested variants (mcr-3, -4, -5, -7, -8 and -9) were not detected. For the variants claimed in the technical data sheet of the manufacturer, sensitivity and specificity were both 100%.

Conclusion: Overall, the RESIST-1 MCR Multi test is a rapid, simple, and reliable method for detecting the main *mcr* variants included in the assay, particularly the most prevalent variant, *mcr-1*. Its principal limitation is that it only detects *mcr*-mediated colistin resistance and therefore does not identify resistance mechanisms unrelated to *mcr* genes.

P103* Characterization of volatile organic compounds produced by benthic cyanobacteria

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Cyanobacteria are photosynthetic microorganisms capable of producing toxins that can be detrimental to ecosystems as well as human and animal health. Planktonic cyanobacteria are known to produce a variety of volatile organic compounds (VOCs) which play important ecological roles during blooms, including allelopathic interactions with algae and aquatic plants and dissuasion of herbivores. Moreover, VOC production has been suggested to contribute to the shaping of microbial communities within the phycosphere. While VOCs produced by planktonic cyanobacteria have been extensively studied, VOC production by benthic cyanobacteria remains largely unexplored.

Because benthic cyanobacteria inhabit a specific ecological niche growing on substrates at the bottom of aquatic ecosystems, their VOC profiles may differ substantially from those of planktonic species. In addition, benthic mats can detach from substrates due to accumulation of oxygen produced by photosynthesis and by the combined effect of water currents. Floating mats are then exposed to changing environmental conditions that can potentially result in different VOC emissions from those of attached benthic mats.

In this study, we used gas chromatography–mass spectrometry (GC–MS) and proton-transfer-reaction mass spectrometry (PTR-MS) to characterize the VOCs produced by nine cyanobacterial strains from the Laboratory of Microbiology at the University of Neuchâtel. These strains include dihydroanatoxin-a-producing *Microcoleus anatoxicus* and an axenic strain derived from it. Both floating and benthic mats were also analyzed to assess VOC production not only from isolated strains but also from mixed communities that are more representative of natural environments.

**Student Paper*

P104* Understanding and hacking the transport across the outer membrane in Caulobacter crescentus

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Nowadays, bacteria continuously adapt to new measures designed to eliminate infection. The available pharmaceutical arsenal contains many molecules able to target Gram-positive bacteria, but few molecules capable of killing Gram-negative bacteria due to the impossibility to reach the target. Indeed, the outer membrane of diderm bacteria is a selective barrier that prevents the entry of large unwanted compounds, while allowing the passage of nutrients. Our project investigates the principles of antibiotic permeation by rendering Gram-negative bacteria sensitive toward Gram-positive specific antibiotics. For our investigation, we are using the Gram-negative bacterium, Caulobacter crescentus, which has the ability to thrive in nutrient-poor environments mainly due to a vast collection of outer membrane transporters called TonB-dependent receptors (TBDRs) that can internalize large molecules. C. crescentus encodes 66 different TBDRs, harboring various levels of expression, differential regulation and many structural architectures. In recent years, we identified TBDRs as entry doors for large antibiotic molecules such as vancomycin or bacitracin while their precise role and regulation remain mysterious. Our project aims to characterize possible substrates of the main TBDRs encoded by C. crescentus using vast antibiotic libraries. Moreover, we will dive into the understanding of the specificity of TBDR's structures and the interaction with their ligands, either natural or artificial, by modifying the internal or surface structure of our TBDRs of interest. By understanding how molecules are granted access through some TBDR, we hope to hack bacterial mechanism of import to force entry of antibiotic molecules.

**Student Paper*

P105 Genomic and phenotypic imprints of microbial domestication on cheese starter cultures

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Domestication – the artificial selection of wild species to obtain variants with traits of human interest – was integral to the rise of complex societies. The oversupply of food was probably associated with the formalization of food preservation strategies through microbial fermentation. While considerable literature exists on the antiquity of fermented food, only few eukaryotic microbes have been studied so far for signs of domestication, less is known for bacteria. Here, we tested if cheese starter cultures harbour typical hallmarks of domestication by characterising over 100 community samples and over 100 individual strains isolated from historical and modern traditional Swiss cheese starter cultures. We find that cheese starter cultures have low genetic diversity both at the species and strain-level and maintained stable phenotypic traits. Molecular clock dating further suggests that the evolutionary origin of the bacteria approximately coincided with the first archaeological records of cheese making. Finally, we find evidence for ongoing genome decay and pseudogenization via transposon insertion related to a reduction of their niche breadth. Future work documenting the prevalence of these hallmarks across diverse fermented food systems and geographic regions will be key to unveiling the joint history of humanity with fermented food microbe

P106 Investigating the interaction dynamics between Pseudomonas aeruginosa and other ESKAPE pathogens on polyethylene medical tubing

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2. Department of Quantitative Biomedicine - University of Zurich Emerging antibiotic resistance and the potential to become endemic in hospital environments makes pathogens, Zurich, Switzerland

Emerging antibiotic resistance and the potential to become endemic in hospital environments makes pathogens in the ESKAPE group some of the greatest modern threats to public health and safe healthcare operations. The ability to adhere to and persist on polyethylene tubing (PET) – a material common in medical devices such as catheters and ventilators - is a key trait for the establishment and endemic circulation of bacterial pathogens in healthcare settings. *Pseudomonas aeruginosa* poses a significant threat to vulnerable patients and is particularly adept at colonizing medical tubing. This pathogen often co-occurs with other ESKAPE pathogens forming polymicrobial infections. With few effective antibiotics available to treat it and its ability to quickly form treatment-resistant biofilms it is of high medical relevance. Therefore, we seek to gain a deeper understanding for how *P. aeruginosa* interacts with other microbes on PET surfaces. We present a simple method for studying polymicrobial interactions on PET where bacteria populations are cultured inside cross-sections of tubing mounted on solid growth media. We use this method to investigate how *P. aeruginosa* colonizes this material and its ability to invade established populations of other species. Future directions of this work will include macroscopic analysis of the biogeography of multispecies biofilms on PET, investigation of the specific mechanisms driving species interactions, and experimental evolution if *P. aeruginosa* and other ESKAPE species to observe how interactions change over time. Altogether, findings from this work will contribute to foundational knowledge about microbial species interactions that can be utilized to prevent the spread and establishment of difficult to treat pathogens in healthcare settings.

P107 Oral administration of Bdellovibrio bacteriovorus, alone and in combination with INTESTI phage cocktail, for gut decolonization of Salmonella spp. in Zophobas morio larvae

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Background: Repeated administrations of *INTESTI* bacteriophage cocktail (*INTESTI*bc) led to overgrowth of an OXA-48-producing *Salmonella enterica* strain (*Sk*-1) used to colonize the gut of *Zophobas morio* larvae (*Zm*L) [PMID:41131385]. Here, we investigate whether predatory bacterium *Bdellovibrio bacteriovorus* (*Bdv*) HD100, alone or combined with *INTESTI*bc, improves the decolonization outcomes.

Methods: Gut colonization in *Zm*L was established by 3 days of *Sk*-1-contaminated feeding, followed by a non-contaminated diet until day 14 (T14). At T3, larvae were randomly assigned to HEPES-BASE (CaHEPES, pH=7.2), HEPES-FRESH (fresh CaHEPES, pH=7.6), HEPES-pH9 (NaOH-adjusted, pH=9), or *INTESTI*bc (pH=6.8) treatments, each tested without or with co-administering *Bdv*. Treatments were given orally at T3 and T5 (10 µL). Sampling was performed at T0, T2, T3, T5, T7, T10, and T14, with 4 larvae per group analyzed for *Sk*-1 CFU/mL after homogenization. *Bdv* predation of *Sk*-1 was assessed *in vitro* at room temperature and 29°C, and tissues underwent 16S rRNA sequencing.

Results: Rapid *Zm*L colonization with *Sk*-1 was observed (T3: $1.1'10^7$ CFU/mL). CFU/mL was stable in HEPES-BASE and unchanged by *Bdv* (range T5–T14: $1.6–6.3\times10^5$ *vs*. 1.4×10^5-4.6×10^6), with the same pattern in HEPES-FRESH. In HEPES-pH9, CFU/mL was 0–4.5×10^1 with *Bdv* and 0 without *Bdv* at T10-T14. In *INTESTI*bc, CFU/mL increased at T7 without *Bdv* (1.5×10^6 CFU/mL) and at T10 with *Bdv* (1.7×10^6 CFU/mL), followed by a significant decrease between conditions at T14 to 2.3×10^5 and 2.5×10^4 CFU/mL, respectively (p=0.036). *Bdv* predation was confirmed *in vitro*, and both *Salmonella* and *Bdv* were detected by 16S rRNA sequencing.

Conclusions: *Zm*L remained stably colonized with *Sk-1* under all conditions except HEPES-pH9, which achieved decolonization by T10, including the control without *Bdv*. *Bdv* alone did not affect *Sk-1* CFU/mL across buffers. Similarly, *INTESTI*bc+*Bdv* did not improve outcomes, despite a significant difference at T14, with *Zm*L exhibiting the same *Sk*-1 overgrowth as *INTESTI*bc alone.

P108* Novel tetracycline resistance gene in Brucella pseudintermedia isolated from cattle

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The genus *Brucella* consists of classical, intracellular pathogens that cause brucellosis, alongside atypical species such as *Brucella pseudintermedia*. Unlike the classical *Brucella* species which are strictly regulated zoonotic pathogens capable of causing severe human and animal diseases, these atypical species are commonly found in different environments. However, they are increasingly recognized as emerging opportunistic human pathogens and are frequently associated with multi-drug resistance. Because tetracyclines are first-line agents in human *Brucella* infections, the emergence of resistance within this genus presents a significant clinical concern. This study aimed to characterize a novel tetracycline resistance gene discovered in *B. pseudintermedia* and demonstrate its functionality.

Tetracycline-resistant strains of *B. pseudintermedia* were isolated from fecal samples of cattle (n=4). Following identification by MALDI-TOF MS, complete genomes were obtained by hybrid assembly of Illumina and Oxford Nanopore reads using Autocycler v0.5.2. The new gene was cloned into vector pBBR1MCS-2 and transformed into tetracycline-susceptible strains of *B. intermedia* and *O. chromiisoli*. MICs were determined by broth microdilution. Efflux was demonstrated by the addition of the efflux pump inhibitor carbonyl-cyanide-*m*-chlorophenylhydrazone (CCCP), and transmembrane protein structure and phylogenetic position were analyzed.

The novel gene is integrated in the chromosome and preceded by a *tetR* repressor in all four strains. Protein structure prediction revealed a 395-amino-acid protein belonging to the Major Facilitator Superfamily (MFS) with 12 transmembrane segments. The gene shares 50.7 % amino acid identity with its closest known related tetracycline efflux protein, *tet*(62). Expression in *B. intermedia* and *O. chromiisoli* increased the MICs of tetracycline, doxycycline and minocycline, up to 128 µg/ml, 64 µg/ml, 16 µg/ml, respectively. In growth assays, the resistance could be reversed by the addition of CCCP, indicating its function as an efflux pump.

A novel chromosomal tetracycline MFS efflux protein, capable of mediating broad-spectrum tetracycline resistance, is present in *B. pseudintermedia*.

**Student Paper*

P109* Identification and characterization of a putative anti-phage defense system in a clinical isolate of Vibrio cholerae

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Due to the constant exposure of bacteria to predatory bacteriophages, both organisms are engaged in a continuous evolutionary arms race. This ongoing conflict has driven the evolution of diverse defense systems that protect bacteria against phage infection. One of the best-studied species in this context is *Vibrio cholerae*, the causative agent of cholera. However, previous studies have primarily focused on the *V. cholerae* lineage responsible for the ongoing seventh pandemic, while comparatively little is known about the defense strategies of other members of the species. Recently, Meyer *et al.* 2024 described a collection of non-pandemic *V. cholerae* isolates obtained from travelers returning to Switzerland [1], providing an opportunity to expand our understanding of the defend systems used by non-pandemic strains to protect themselves against bacteriophage infection.

In this work, we focused on one isolate that displayed receptor compatibility with a panel of commonly studied vibriophages. Using this strain, we first performed basic bioinformatic analyses and identified a highly diverse repertoire of mobile genetic elements encoding known, predicted, or putative defense systems. Based on these analyses, we generated 21 deletion mutants targeting either short gene clusters or entire genomic islands. We subsequently assessed the phage susceptibility of these mutants and compared their phenotypes to that of the parental strain.

This initial screen led to the identification of a putative defense system encoded by a two-gene operon, which we further characterized using bacterial genetics as well as growth- and microscopy-based phage infection assays. In parallel, AlphaFold-based structural predictions revealed no detectable homology to previously described defense systems. Collectively, our preliminary data suggest that this gene cluster encodes a novel anti-phage defense system that protects bacteria against the tested bacteriophages through direct phage inactivation rather than through an abortive infection mechanism that relies on population-wide protection.

**Student Paper*

P110 Retargeting the R2-type pyocin of P. aeruginosa PAO1 towards Salmonella

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The R2-type pyocin of *P. aeruginosa* PAO1 is a phage tail-like bacteriocin that is produced upon stress and burst released from the producing cell. The native tail fiber protein (prf15/PA0620) was exchanged by tail spike proteins (TSPs) from phage KCK6, a Kuttervirus infecting *Salmonella enterica*. Here we present the efficacy of the newly designed pyocins for control of *Salmonella*. In addition, specificity of the TSPs for *Salmonella* LPS O-antigen binding and processing is described. Engineered pyocins represent an ecologically friendly alternative for the specific control of pathogenic Gram-negative bacteria.

P111 Tracking anatoxin-producing cyanobacteria in Switzerland: a molecular approach enabled by citizen-science sampling

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Since 2020, Switzerland has experienced incidents of dogs dying after ingesting toxic benthic cyanobacterial mats floating on river surfaces. These events have been linked to the neurotoxins anatoxin-a and dihydroanatoxin-a, produced by the filamentous cyanobacterium Microcoleus anatoxicus. This species has been observed year-round in the Areuse river, where several dog fatalities occurred. To improve our monitoring scheme, we developed molecular markers targeting genes within the anatoxin biosynthetic operon, and for the detection of this toxigenic species. The aim of this research was to better understand the geographical distribution of M. anatoxicus across Switzerland. We started a citizen science project involving local authorities and volunteers. The participants were trained to identify and collect samples of floating mats. The program proved successful, yielding approximately 35 samples from rivers and lakes throughout Switzerland. Among these samples, 29 contained filamentous cyanobacteria, and 16 tested positive for anatoxin-related genes. In addition, we observed Tychonema spp. in Switzerland, a second toxic cyanobacterium known to produce the same neurotoxins. Furthermore, we also discovered a strain of M. anatoxicus in a lake environment without the anaK gene, and thus potentially putatively lacking the ability to convert anatoxin-a into dihydroanatoxin-a. This research highlights the value of citizen science in environmental monitoring and underlines the widespread distribution of anatoxin-producing cyanobacteria across Swiss aquatic ecosystems.

P112* Re-evaluating bacteriostatic and bactericidal classification of antibiotics across diverse bacterial species

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Antibiotics are traditionally classified as bacteriostatic or bactericidal according to their ability to inhibit bacterial growth or kill bacterial populations. However, increasing evidence suggests that this distinction is not an intrinsic and stable property of antibiotics, but instead depends strongly on the physiological and environmental context. Here, we evaluated the robustness of the bacteriostatic/bactericidal classification by investigating the influence of antibiotic concentration, exposure time, and bacterial context on antibiotic activity. To address this, 27 pathogenic strains representing multiple bacterial species were exposed to 20 antibiotics belonging to different antibiotic classes. Antibiotic activity was experimentally determined using survival assays and each antibiotic–strain combination was classified as bacteriostatic or bactericidal based on bacterial survival. Additionally, antibiotic activity was characterized in a collection of commensal gut bacterial strains to compare activity patterns between pathogenic and commensal bacteria. Our results revealed that the experimental classification differed substantially from the traditional textbook classification. Antibiotic activity was highly context dependent and appeared to be strain dependent, with variability observed even within the same bacterial species. Exposure time also influenced activity, particularly for β-lactam antibiotics, which displayed increased bactericidal activity after prolonged exposure. Antibiotic concentration also had an impact, with lower concentrations generally associated with more bacteriostatic behavior, whereas higher concentrations shifted activity towards predominantly bactericidal outcomes. Furthermore, pathogenic strains displayed overall higher resistance levels than commensal strains exposed to the same antibiotics. Altogether, these findings challenge the traditional bacteriostatic/bactericidal classification and demonstrate that antibiotic activity is strongly shaped by exposure conditions and bacterial context.

**Student Paper*

P113* Global distribution and temporal dynamics of ESBL-producing non-Escherichia coli Enterobacterales

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Background
Extended-spectrum β-lactamase (ESBL)–producing Enterobacterales are major drivers of antimicrobial resistance worldwide. While *Escherichia coli* has been extensively studied, the genomic epidemiology and dissemination dynamics of other clinically relevant ESBL-producing Enterobacterales across One Health reservoirs remain poorly characterized.

Methods
Species were selected based on their representation among ESBL-producing Enterobacterales isolated at the University Hospital Basel. We constructed a harmonized global genomic database by integrating publicly available assemblies and metadata from the National Center for Biotechnology Information and Pathogenwatch. Following deduplication, metadata harmonization, and species-specific quality control, ESBL genes and plasmid replicons were identified using ResFinder and PlasmidFinder. Host metadata were standardized into 14 One Health–relevant categories encompassing human, animal, food-chain, wildlife, and environmental reservoirs.

Results
The final dataset comprised 139,379 genomes from 26 non-*E. coli* Enterobacterales species collected over two centuries (1819 to 2025) from 140 countries and 493 host sources. The collection includes major pathogens (*Klebsiella pneumoniae, Enterobacter cloacae* complex*, Citrobacter freundii, Proteus mirabilis*) and underrepresented species. Samples were heavily skewed toward human-associated isolates, highlighting substantial representation biases in publicly available sequencing data. Approximately 60,000 genomes habour ESBL genes, with marked differences in the proportion of ESBL across species, hosts, geographic regions, and years. Temporal analyses showed a progressive shift from SHV-type ESBLs toward dominance of the CTX-M-1 group across multiple species and reservoirs.

Conclusions
This harmonized resource enables large-scale analyses of ESBL-producing non-*E. coli* Enterobacterales across One Health sectors. The observed geographic, host-associated, and temporal patterns provide insights into the evolving dissemination of resistance genes and mobile genetic elements worldwide.

**Student Paper*

P114 Influence of lake water temperature on the decay of wastewater-borne antimicrobial resistance

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Wastewater treatment plant effluent is a known source of antimicrobial resistance genes (ARGs) and resistant bacteria (ARB), in receiving water bodies. As lakes are sources of drinking water and recreation, understanding the fate and decay of ARGs and ARB in lakes is crucial for assessment of risks to public health. Temperature has been shown to be an important driver of microbial decay, e.g. of fecal indicator bacteria, bacteriophages, and viruses. However, little work has focused on ARB/ARGs, and published work was often conducted under highly controlled laboratory conditions and bacteria isolates. Here we studied the influence of temperature on the decay of ARGs and ARBs in wastewater inocula using permeable membrane microcosms allowing passage of water and small ions, as an experimental system that is more representative of the complex ecological interactions of wastewater introduced into lakes.

Dialysis bag microcosms containing 10% wastewater and 90% lake water were incubated in lake water for 30 days at 4 different temperatures (6 °C, 12 °C, 18 °C, 24 °C). 11 ARGs were quantified by qPCR and total and cefotaxime-resistant *E. coli* were quantified on mTEC media with and without cefotaxime. Decay rates of ARGs and *E. coli* were modeled with first-order and biphasic kinetics.

Preliminary results show that the decay behavior of (ARGs) are target-specific and influenced by temperature. For instance, *ermF* and *sul1* had similar initial concentrations, but the rate of decay of *ermF* was generally higher and was more heavily dependent on temperature. Cefotaxime-resistant *E. coli* remained cultivable for longer in cooler temperatures (4 days at 24 °C versus 15 days at 4°C), highlighting a positive effect of temperature on ARB decay. This work will provide an improved understanding of the environmental fate of potentially harmful ARGs and ARB and contribute to future environmental modeling and risk assessment.

P115 A novel Real-time Immunoassay for Differentiation of TBE Infection from Vaccination

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Tick-borne encephalitis (TBE) remains a significant public health concern in Switzerland, with 200–450 cases reported annually. Although an effective vaccine exists, vaccination failure can occur in approximately 3% of individuals. In such cases, serological interpretation becomes challenging because vaccine-induced IgM/IgG profiles cannot be differentiated from those following an infection with the available assays. The non-structural protein NS1, essential for viral replication, is only present in trace amounts in inactivated vaccines and therefore might use as an antigen to distinguish vaccine-induced immunity from infection.

In this study, we evaluated an innovative real-time immunoassay (ElioDX™, Elionova AG) for the detection of NS1-specific IgM and IgG antibodies in over 200 human serum and plasma samples. TBE vaccination status was documented for all participants and categorized as unvaccinated-uninfected, infected, or vaccinated with one to more than 3 doses. The assays demonstrated a clear separation between infected and unvaccinated or vaccinated individuals. Using a predefined cut-off, the anti-NS1 IgG and IgM assays demonstrated excellent diagnostic performance, with IgG showing 93.3% sensitivity and 90.9% specificity, and IgM showing 90% sensitivity and 90.9% specificity, each achieving an area under the curve (AUC) of 0.95. Combining IgM and IgG further improved sensitivity (96.7%) and specificity (96%), yielding an AUC of 0.99.

These findings indicate that combined detection of anti-NS1 IgM and IgG provides a robust and reliable approach for differentiating natural TBE infection from vaccine-induced antibody responses. ElioDX™ real-time immunoassays offer a rapid and sensitive method for anti-NS1 detection and could be extended to other flaviviruses, such as dengue and West Nile virus.

P116∗ Investigating the Virulence Potential of Klebsiella pneumoniae Species Complex Isolates from Indian Arable Land

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While the *Klebsiella pneumoniae* species is a notorious global driver of nosocomial infections, its ecological success spans diverse non-clinical habitats, including agroecosystems. Understanding these environmental reservoirs is critical from a One Health perspective, particularly as multidrug-resistant and hypervirulent lineages cross ecological boundaries. This study investigates the occurrence, virulence profiles, and colonization dynamics of *Klebsiella pneumoniae* species complex (KpSC) isolated from agricultural matrices. Soil and compost samples collected from fifteen geographically distinct agroclimatic regions in India were subjected to targeted enrichment and isolation. Presumptive KpSC isolates were validated via species-specific PCR screening. To evaluate their pathogenic and ecological fitness, the isolates were profiled for key virulence-associated genes and characterized using robust phenotypic assays, including biofilm architecture, mucoviscosity production, and siderophore secretion. Crucially, environmental KpSC isolates displayed phenotypic virulence traits comparable to clinical reference strains. Furthermore, in vitro hydroponic assays demonstrated a strong capacity of select environmental isolates to successfully colonize the tomato rhizosphere, highlighting their potential to persist in close association with food crops. In conclusion, our findings demonstrate that arable lands harbour KpSC strains with potent virulence reservoirs. These insights emphasize the critical need to track environmental dissemination dynamics to comprehensively understand and mitigate public health risks at the human-agriculture interface.

∗Student Paper

P117 National survey on antimicrobial susceptibility practices in Swiss clinical microbiology laboratories

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Background: Harmonized antimicrobial susceptibility testing (AST) is essential to ensure comparable reporting, effective antimicrobial resistance surveillance, and optimal patient management across Swiss laboratories and hospitals. The Swiss AST Committee (SAC) conducted a national survey on current AST practices to support future discussions, guideline development, harmonization efforts, and educational activities, enabling anonymous benchmarking.

Methods: An online questionnaire was distributed in early April 2026 to Swiss clinical microbiology laboratories performing bacterial AST.

Results: Responses were received from 41 laboratories, including private laboratories (17/41, 41.5%), cantonal hospital laboratories (13/41, 31.7%), university hospital laboratories (5/41, 12.2%) and others (6/41, 14.6%). Daily AST workload ranged from 10–60 antibiograms in 27/41 laboratories (65.9%), while 5/41 (12.2%) performed more than 100 per working day. VITEK 2 was the most used first-line AST method (27/41, 65.9%), followed by disc diffusion (10/41, 24.4%), whereas other commercial broth microdilution-based systems were used by a minority of laboratories. Overall, DD was used by 37/41 laboratories (90.2%), gradient diffusion by 34/41 (82.9%), and VITEK 2 by 28/41 (68.3%). EUCAST breakpoints were used either exclusively by 17/41 laboratories (41.5%) or in combination with alternative breakpoints when EUCAST criteria are unavailable in the remaining 24/41 (58.5%). The EUCAST "I" category was reported as "susceptible, increased exposure" by 37/41 laboratories (90.2%). EUCAST updates were implemented within 3 months by 21/41 laboratories (51.2%). ESBL

confirmation relied predominantly on double-disk synergy testing (30/41, 73.2%). Anaerobic AST was performed by 27/41 laboratories (65.9%), mostly using gradient diffusion (23/27, 85.2%) or DD (14/27, 51.9%). ATU results were mostly managed by repeat testing with an alternative method (20/41, 48.8%). Rapid AST directly from positive blood cultures (RAST) was implemented by 19/41 laboratories (46.3%).

Conclusion: Swiss laboratories show broad adherence to EUCAST recommendations, although relevant variability persists in selected AST practices. These findings support future national harmonization efforts and guideline development.

P118 Impact of Occupational Exposure on the Human Skin Microbiome

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The human skin microbiome is a specialized ecosystem shaped by anatomical topography and host physiology. Extrinsic environmental forces can disrupt this homeostatic equilibrium. While urban pollution impacts are documented, how localized industrial or service workplace bioaerosols affect specific cutaneous microenvironments remains poorly characterized.

We explored the skin microbiota composion of professionals across five occupational sectors (industrial cooking, metal recycling, room cleaning, health care, welding) and a non-exposed control group using PacBio Full-Length 16S rRNA gene sequencing. Swabs were collected from five distinct anatomical niches representing dry, moist, and sebaceous microenvironments. Distance-based PERMANOVA models and longitudinal alpha-diversity tracking were stratified by site and sequentially adjusted for host factors, including biological sex.
Preliminary analyses indicate that community-level alterations induced by occupational exposure are highly restricted to specific anatomical niches. Open-exposed microenvironments, including the lipid-rich cranial regions and the highly exposed dry surface of the anterior forearm, demonstrate a heightened susceptibility to workplace environmental shifts, revealing noticeable variations in microbial composition across professional sectors. Conversely, occlusion-shielded niches or protected mucous membranes, exhibit greater structural resilience, displaying more stable alpha-diversity profiles (Observed richness and Shannon index) between sampling intervals.
Occupational bioaerosols and chemical exposures do not induce a uniform reshuffling of the cutaneous flora, but rather print a localized ecological footprint that is strictly dependent on anatomical site exposure and the degree of clothing coverage protecting the skin surfaces. This targeted susceptibility emphasizes the vulnerability of uncovered body parts (such as the FAC and RAC) and highlights the need for site-specific dermatological protective strategies in occupational health.

P119 Soil microbiome responses to organic fertilization in a subtropical banana agroecosystem

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Organic fertilizers are increasingly applied in intensive banana production to improve soil fertility and support sustainable crop management. However, their long-term effects on soil microbial communities in subtropical systems remain poorly characterized. In this study, we evaluated the impact of four organic amendments commonly used in the Canary Islands—chicken manure, cow manure, compost, and pelletised compost—on soil bacterial and fungal communities during a banana pot experiment spanning almost two years and covering a complete crop cycle.

Microbial community dynamics were assessed using long-read amplicon sequencing of bacterial 16S rRNA genes and fungal ITS regions. The different amendments produced distinct microbial responses over time. Chicken manure caused the strongest shifts in both bacterial and fungal community composition and was associated with increased soil electrical conductivity and reduced soil pH. In contrast, compost-based amendments and cow manure promoted more progressive changes in microbial structure and were linked to higher organic matter content and nutrient availability throughout the experiment.

Differential abundance analyses identified amendment-specific microbial responses, including reductions in several bacterial taxa under chicken manure treatment and an enrichment of saprotrophic fungi in compost-amended soils. Comparisons between culture-dependent methods and sequencing data revealed strong consistency for bacterial communities but lower agreement for fungal taxa.

These findings highlight the importance of fertilizer composition and nutrient release patterns in shaping soil microbial communities in banana production systems. Understanding these microbial responses can support the development of more sustainable fertilization strategies for subtropical agriculture.

P120 Comparison of Zybio EXS2600/Ex-Accuspec and Bruker MALDI Biotyper® Sirius MALDI-TOF MS systems for microbial identification

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MALDI-TOF MS is a cornerstone of routine clinical microbiology, enabling rapid identification of bacteria and fungi. The emergence of new platforms such as the Zybio EXS2600/Ex-Accuspec requires rigorous validation against established systems such as the Bruker MALDI Biotyper® Sirius, since performance may vary according to the instrument, organism group, consumables, target plate, and workflow.

Zybio and Bruker MALDI-TOF MS workflows were compared for bacterial and fungal isolates, and microorganisms recovered from positive blood cultures. Bacterial workflows included direct spotting and formic acid-assisted preparation on disposable and/or reusable target plates. Fungal workflows included formic acid preparation and extraction-kit protocols. Blood culture workflows used reusable target plates and system-specific extraction kits. Scores were interpreted using thresholds: >2.0 for species-level identification, 1.7–2.0 for genus-level identification, and <1.7 for no reliable identification.

Overall, bacterial performance was high for both platforms: species-level identification ranged from 70% to 91% with Bruker and from 93% to 97% with Zybio. Formic acid improved Bruker reusable target performance from 70% to 90%, while Zybio reusable targets reached 93% species-level identification by direct spotting.

For fungi and yeasts, performance was more variable and group-dependent. Species-level identification ranged from 24% to 37% with Bruker and from 48% to 69% with Zybio. Dermatophytes were more challenging with Bruker, whereas Aspergillaceae showed more comparable performance, with Bruker formic acid reaching 61% species-level identification and Zybio up to 77%.

In positive blood cultures, Zybio showed higher overall performance than Bruker, with 74% versus 45% species-level identification. This difference was most marked for Gram-positive cocci, while Gram-negative bacilli showed comparable results.

These data suggest that Zybio provides excellent identification performance across several applications, with potential to simplify workflows and reduce time to results and costs.

P121 Genome Analyses of Campylobacter isolates from Human in Switzerland in 2024 and comparison to isolates from 2010-2024

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Bacterial gastroenteritis is frequently caused by *Campylobacter*, mostly via intake of contaminated food. Sixty-two *C. jejuni* and 9 *C. coli* isolates from campylobacteriosis cases in Switzerland in 2024 were sequenced. Core genome MLST analysis revealed that the isolates were diverse, although 9 isolates belonged to the complex ST475-CT827 and four to ST19-CT2633. Virulence factors were typical for *Campylobacter* and differed between isolates mainly in factors involved in immune modulation. Cytolethal distending toxins CdtA, CdtB, and CdtC were found in but one *C. jejuni*. Campylobacteriosis can lead to colorectal cancer, which is linked to the presence of the virulence genes *cstIII* and *neuABC* and these were present in 18 of the 62 C. jejuni strains. Further, Group 1 sialylated lipo-oligosaccharides (LOS) are associated with an enhanced risks for Guillain–Barré and Miller-Fischer syndromes and the LOS of forty-two *C. jejuni* isolates were assigned to this Group 1.

Sixty-two percent of the human isolates had a GyrA T81I mutation conferring quinolone resistance and 37% harbored *tet*(O) conferring tetracycline resistance.
The isolates from this study were further compared with sequences in our database comprising 880 Campylobacter isolates collected from various sources between 2005 and 2025. Thirty of the isolates formed clusters with isolates from a poultry slaughterhouse, from chicken meat collected at retail level, from human 2005-2020, and/or from wild birds. The chicken meat isolates were sampled in the same period as the strains in this study, thus providing a direct link between food and human infection. Remarkably was clonal type ST21-CT2570 which contained 4 strains from a this study and 28 isolates from all other sources, showing that this single clone is ubiquitous in the Swiss C. jejuni population.

Our data provide inside in *Campylobacter* in human in Switzerland and the exchnage between human isolates and the whole *Campylobacter* population.

P122 Added value of a new quantitative real-time PCR for Neoehrlichia mikurensis, an underdiagnosed emerging tick-borne pathogen

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Background

Neoehrlichia mikurensis is an emerging tick-borne intracellular bacterium causing underdiagnosed infections, particularly in immunocompromised patients. Clinical manifestations include FUO, inflammatory syndromes, cytopenias, thromboembolic events, and severe systemic disease. Current molecular diagnostics have limited sensitivity or specificity or only provide qualitative results. We implemented a quantitative real-time PCR (qPCR) assay to improve diagnosis and longitudinal monitoring of bacterial loads following antimicrobial treatment.

Methods

A qPCR assay targeting the *groEL* gene of *N. mikurensis* was developed on our automated molecular diagnostic platform [1, 2]. Primers and probes were adapted and optimized for TaqMan technology using LNA and MGB modifications. Validation included determination of the LOD, reproducibility, specificity, and testing of previously characterized clinical samples. Quantification was performed using serial dilutions of a synthetic plasmid standard and expressed as DNA copies/mL.

Results

The assay demonstrated high analytical sensitivity with detection down to 10 copies/reaction and occasional detection at 1 copy/reaction. No cross-reactivity was observed with related tick-borne pathogens, including *Anaplasma phagocytophilum*, *Rickettsia* spp., and *Coxiella burnetii*. Clinical validation confirmed detection in previously characterized positive specimens. The assay provided clinically relevant quantification of bacterial load, ranging from a few hundred to several million DNA copies/mL. In two immunocompromised patients presenting severe disease, bacterial loads exceeded 10^6 copies/mL in EDTA blood. Serial measurements demonstrated a rapid decline in blood DNA after treatment initiation, supporting quantitative monitoring of microbiological response for treatment-duration decisions.

Conclusion

Implementation of a quantitative real-time PCR assay expands the diagnostic toolbox for *N. mikurensis*. Beyond rapid and sensitive detection, bacterial load quantification provides clinically actionable information for disease assessment and therapeutic monitoring. These findings illustrate the growing role of quantitative molecular microbiology and diagnostic stewardship in the management of emerging tick-borne infections.

[1] Greub G et al. Future Microbiol. 2016;11(3):403-25.

[2] Jenkins A, et al. BMC Microbiol. 2019 Aug 28;19(1):199.

P123 Sexually transmitted infections do not significantly alter the rectal microbiome in men who have sex with men

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Chlamydia trachomatis (CT) and *Neisseria gonorrhoeae* (NG) are the most frequently reported sexually transmitted bacterial infections (STI) among men who have sex with men (MSM). Coinfections are common, and epidemiological evidence suggests that CTNG coinfections may be associated with higher bacterial loads and increased transmissibility compared with single infections. This study compared bacterial loads and rectal microbiome profiles across CT, NG, and CTNG coinfections.

Rectal swabs from PrEP-MSM positive for CT and/or NG were collected prior to treatment between December 2021 and June 2025. The study included 34 single CT infections, 34 single NG infections, 9 CTNG coinfections, and 9 samples with negative rectal but positive pharyngeal or urethral results. Bacterial loads were quantified using real-time PCR, and the microbiome composition was assessed using PacBio full-16S long-read sequencing.

Eighty-six MSM were included. Bacterial loads did not differ significantly between single and coinfected samples (CT vs CTNG, p=0.26; NG vs CTNG, p=0.49). No differences in alpha diversity (Chao1, Shannon, or Pielou indices) were observed between infection groups or by symptom status. Beta diversity analyses (Jaccard and Bray-Curtis) showed similar community composition across groups, except for a difference between NG and uninfected individuals using Bray-Curtis dissimilarity (p=0.006). Symptomatic status was not associated with differences in bacterial load or microbiome diversity. Differential abundance analysis revealed that the CT profile showed *reduced Peptoniphilus asaccharolyticus, Neisseria gonorrhoeae,* and a Cryptobacteroides species; the NG profile was enriched in *Gamella* spp.; and uninfected samples showed reduced *P. asaccharolyticus* and *Dialister micraerophilus.*

CTNG coinfection was not associated with higher bacterial burden or distinct overall microbiome diversity compared with single infections. However, subtle differences in microbial composition were observed between groups. Further longitudinal studies incorporating behavioral and clinical metadata (e.g. sexual practices, PrEP use, and other habits) are needed to better understand recurrent infections and microbiome changes over time.

P124* From PCR to NGS: automated extraction and sequencing workflows for clinical mycobacteriology

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Background

Molecular identification of *Mycobacterium tuberculosis* complex (MTBC) and non-tuberculous mycobacteria (NTM) relies on PCR and sequencing workflows. However, DNA extraction from positive cultures often remains manual, labor-intensive, and performed in biosafety level 3 (BSL-3) environments. We evaluated a streamlined workflow combining automated nucleic acid extraction in BSL-2 conditions with downstream PCR, Sanger sequencing, and exploratory next-generation sequencing (NGS).

Methods

DNA extraction using the MagNAPure automated instrument (Roche) in a BSL-2 environment was compared with the routine manual QIAamp DNA Mini-Kit (Qiagen) for positive mycobacterial cultures (MGIT). Ten MTBC and NTM isolates representing a broad taxonomic spectrum were analyzed, including *M. tuberculosis*, *M. bovis*, *M. avium*, *M. abscessus*, *M. kansasii*, *M. xenopi*, and *M. marinum*. Downstream analyses included PCR amplification and Sanger sequencing of 16S rDNA, *rpoB*, *hsp65*, *gyrB*, and *pncA*. Amplification performance, sequence quality, identification and resistance-associated mutations were compared. Inactivation process was validated, allowing safe transfer from BSL-3 to BSL-2. Preliminary shotgun metagenomic sequencing (Illumina) was also investigated.

Results

The inactivation protocol demonstrated complete absence of subculture positivity enabling safe downstream processing outside of BSL-3 facilities. Species identification was fully concordant between manual and automated extraction methods. Amplification performance and Sanger sequencing quality were equivalent across targets. Sequence identity ranged from 96% to 100%. No discrepancy were observed for *pncA* resistance-associated single nucleotide polymorphism detection. Automation substantially reduced hands-on time from 1h to <5min per extraction while improving standardization and traceability. Preliminary NGS analysis demonstrated compatibility of the automated extraction workflow with high-throughput sequencing applications. NGS identified two mixed infections undetected by Sanger sequencing: MTBC/*M. intracellulare* and *M. pseudokansasii*/ *M. gordonae* coinfections.

Conclusion

Automated MagNAPure extraction demonstrated equivalent performance to manual extraction for molecular identification and resistance-associated sequencing of MTBC and NTM. This workflow improves safety, efficiency, and standardization. Preliminary results further support its use for NGS-advanced mycobacterial diagnostics.

**Student Paper*

P125* Exploring microbial community- and 14C-variability along the chronosequence of the Witenwasseren Glacier in Valais, Switzerland.

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The cryosphere is changing at unprecedented rates due to rapid climate change; however, the impact that these changes have on the microorganisms inhabiting the cryosphere are largely unknown. This project investigates how microbial diversity, carbon sources, and carbon sinks vary along a high alpine deglaciating valley chronosequence in Valais, Switzerland. Ice, water, sediment and soil samples were collected from the Witenwasseren Glacier, its forefield and outflow into the Reuss River. The microbial community composition and abundance were characterized through 16S rRNA gene sequencing and quantitative PCR, respectively, and paired with measurements of carbon content and radiocarbon age to provide a timeline of glacial retreat. Together, this data will be used to map the development of microbial communities over time. Preliminary data shows a general trend of increasing biomass and higher $F^{14}C$ of inorganic carbon as the distance from the glacier increases, despite minimal changes in dissolved organic carbon (0,42-1,13 mg C/L). With more developed microbial communities further from the glacier, it is expected that they will have a higher microbial diversity than those first colonizing the newly exposed glacial material.

**Student Paper*

P126* maphelios: mapping-based high-throughput exploration of sequencing data

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High-throughput functional genomic screens such as gain- or loss-of-function screens can be used to identify responsible gene products for queried functions in bacteria or communities of interest, resulting in the generation of large amounts of sequencing data. User-friendly tools for systematically exploring and visualising this kind of data are currently missing. Here, we present Maphelios, a generalised workflow for systematic exploration of sequencing data which offers the opportunity to not only map data in an easy user-interface, but also generate customisable visualisations on two levels: genome- and sequence level. Maphelios integrates several mapping algorithms, enabling the mapping of both short- and long-read sequencing data in the same tool. It has embedded quality control plots and offers optional pre-processing of sequencing data, including quality filtering and trimming. Users obtain detailed mapping information that can be used for downstream bioinformatics analysis, next to publication-ready plots. Maphelios can either be used in a web-app where users can interactively submit and explore their sequencing data or as a python package. Maphelios bridges the gap between high-throughput screens and systematic data exploration to identify and study functions encoded in bacterial genomes, while broadening the access to such analytical workflows to a larger public.

**Student Paper*

P127 WGS-based characterization of methicillin-susceptible and methicillin-resistant Staphylococcus aureus from cheese

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Staphylococcus aureus is one of the major opportunistic pathogens associated with hospital- and community-acquired infections in humans. It is also causing infections in animals like mastitis in cattle. Contamination with *S. aureus* originating either from contaminated milk or humans may occur during cheese making, with strains further persisting in raw milk cheese.

A total of 152 raw milk soft and semi-soft cheeses were tested for the presence of methicillin-resistant and -susceptible *S. aureus* (MRSA, MSSA) using an 18-h enrichment step at 37°C in Müller-Hinton broth containing 5% NaCl followed by an overnight selection on BioRad SAselectAgar and BD CHROMagar MRSA II. Isolates were identified with Maldi-Tof MS (Bruker) and sequenced using long read sequencing technologies (ONT and seqWell LongPlex). Complete circular genomes were obtained using Autocycler v.0.6.2. Phylogenetic analysis, antimicrobial resistance and virulence gene detection were performed using MBioSEQ Ridom Typer software v.12.

Seven *S. aureus* (3 MSSA, 4 MRSA) were detected in 6 different cheeses with a prevalence of 3.9% [95%CI 1.5-8.6%]. The strains were genetically diverse belonging to ST5, ST97, ST398 (MSSA) and ST398, ST8, ST6697 (MRSA). In addition to the methicillin resistance gene *mecA* in MRSA, additional genes were detected in both MRSA and MSSA like e.g. those for resistance to aminoglycosides (*aac(6')-Ie–aph(2'')-Ia*), beta-lactams (*blaZ*), macrolides (*erm*(T)), lincosamides (*lnu*(B), *lsa*(E)), phenicols (*fexA*), trimethoprim (*dfrG*, *dfrS*), and mutations in topoisomerases (GyrA_S84L, ParC_S80Y) for fluoroquinolone resistance. Virulence factors associated with immune evasion, secretion, adhesins, exoenzymes and enterotoxins were detected in different proportions. Antimicrobial resistance and virulence genes were identified on the chromosomes.

Raw milk cheese may harbor antimicrobial-resistant *S. aureus* containing virulence factors. The diversity and type of strains suggest different human and animal origins emphasizing the need for increased hygiene and bacteriological control in this sector to avoid *S. aureus* dissemination in the population through cheese consumption.

P128 Tuberculosis at the animal–human–environment interface

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Tuberculosis remains one of the deadliest infectious disease globally, with *Mycobacterium bovis* representing the major zoonotic component at the human–livestock–wildlife interface. Genomic surveillance in resource-limited settings is often constrained by cost and infrastructure requirements.

We applied targeted Next-Generation Sequencing (tNGS) using Oxford Nanopore Technologies (ONT) sequencing to detect *M. bovis* in human sputum samples from communities surrounding a South African national park. In addition, ONT adaptive sampling was used to enrich pathogen DNA and generate whole-genome sequences from *M. bovis* recovered culture-independently from wildlife and cattle tissue samples, as well as from environmental water samples.To benchmark performance, we compared ONT-derived genomes with Illumina short-read and Pacific Biosciences (PacBio) long-read assemblies from culture isolates. ONT assemblies showed comparable SNP-level concordance across platforms, with improved genome completeness relative to Illumina assemblies and similar structural resolution to PacBio. While PacBio provided high-quality long-read assemblies, ONT offered substantially greater accessibility and lower cost, enabling scalable field-deployable genomics.In addition, ONT sequencing enabled detection of epigenetic modifications, providing methylation profiles not accessible through Illumina-based workflows and only indirectly inferred in PacBio datasets. This added layer of information may contribute to understanding strain adaptation and host–pathogen interactions.

Overall, our results demonstrate that ONT-based workflows provide a robust, cost-effective alternative for high-resolution *M. bovis* genomics in One Health settings, combining accurate genome reconstruction with the unique advantage of direct epigenetic detection. This approach enables integrated surveillance across humans, livestock, wildlife, and environmental reservoirs using locally deployable sequencing infrastructure.

P129* Plasmid-mediated linezolid resistance in multidrug-resistant Staphylococcus aureus from pigs and cattle

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Staphylococcus aureus is one of the major opportunistic pathogens associated with infections in humans. It can also colonize and cause infections in animals. Use of antibiotics in animals may select bacterial resistance to human last-resort antibiotics (e.g. linezolid) through co-selection of one single multidrug-resistance gene. For instance, the *cfr* gene encoding 23S rRNA methyltransferases confers resistance to linezolid as well as to phenicols and pleuromutilins, which are used in veterinary medicine.

Two hundred fifty-four caecum contents from pigs and 233 from bovine calves were tested for linezolid-resistant *S. aureus* using an 18-h broth enrichment at 37°C followed by an overnight selection on CHROMagar LIN-R (Mast Diagnostica). Isolates were identified with Maldi-Tof MS (Bruker) and sequenced using Oxford Nanopore Technologies. Complete circular genomes were obtained using Autocycler v.0.6.2. Phylogenetic analysis, antimicrobial resistance gene detection were performed using MBioSEQ Ridom Typer software v.12. Plasmid comparison was performed using clinker v.0.0.32.

Linezolid-resistant *S. aureus* were isolated in pigs (n=2) and calves (n=7) with a prevalence of 0.8% [95%CI 0.9-2.8%] and of 3.0% [95%CI 1.2-6.2%], respectively. The strains detected in pigs and in one calf belonged to ST398 *spa* type t899 and were methicillin-resistant *S. aureus* (MRSA) whereas the other strains from calves were methicillin-susceptible *S. aureus* (MSSA) of ST352-t267. Five *cfr*-containing plasmids with different sizes and structures were identified among the 7 *S. aureus* strains indicating different acquisition events. Additional antimicrobial resistance genes to macrolides, lincosamides, tetracyclines, phenicols and streptomycin were co-located on the *cfr* plasmids of ST398-t899 [*fexA*, *tet*(L), *erm*(B), *erm*(C)] and of ST352-t267 [*tet*(L), *erm*(B), *erm*(C), *lsa*(B), *str*].

Food-producing animals may harbor MSSA and MRSA which contain plasmid-mediated linezolid resistance gene *cfr*. Plasmids were likely selected in animals using veterinary antimicrobials, creating a reservoir of *S. aureus* strains which are resistant to last-resort antibiotics, and which may be transferred to humans.

**Student Paper*

P130 Unraveling the impact of protein synthesis inhibitors on Bacteroides uniformis

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Protein synthesis inhibitors (PSIs), such as macrolides like erythromycin, are textbook bacteriostatic antibiotics. They have been associated with strong dysbiosis within the human gut microbiome and have also been shown to induce lysis in certain gut bacterial species. In this study, we aim to clarify the molecular mechanisms by which macrolides impact *Bacteroides uniformis* and broaden our understanding to encompass other strains and additional PSIs. Our findings indicate that various PSIs can induce lysis in *B. uniformis*. Survival assays revealed that there is a concentration and time-dependent effect for each antibiotic and uncovered activity differences, with more efficient killing observed using e.g., clindamycin. PSI treatment of *B. uniformis* leads to DNA compaction and the formation of phase-light areas, which contain cytoplasmic content, as well as membrane defects like blebbing and rounding up. Additionally, we are investigating the transcriptional response of *B. uniformis*, to elucidate the potential stress induced by erythromycin treatment. Finally, we will utilize a panel of 48 different *B. uniformis* strains to assess survival for each strain. By integrating the obtained results, we want to determine whether the genes identified in the type strain can be used to predict the effects of PSIs. Altogether, our results will shed light on the actual mode of action of PSIs in gut microbes, which might advance our understanding of why these antibiotics induce long-term dysbiosis in the gut microbiota of patients.

P131 Selecting and formulating beneficial bacterial strains as biocontrol agents against the late blight causing agent Phytophthora infestans

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Crops face different challenges leading to yield losses. One of those challenges are plant pathogens, such as the oomycete *Phytophthora infestans*, the causing agent for late blight in potatoes and tomatoes. Thus, repeated treatments across the season are necessary when culturing those crops. Currently in organic farming copper-based and in conventional farming synthetic, chemical fungicides are used, leading to an accumulation of those products in the soil. On the market, there is a lack of alternative solutions, but studies indicate that the use of microorganisms could be an efficient solution. However, microbial biocontrol strains that show protective effects in controlled conditions often fail to confer protection under real field conditions. One reason for this lack of efficiency is the poor survival of strains in the harsh environment of plant leaves. The aim of this project is to develop a protective and activity-boosting formulation that will allow biocontrol agents to provide efficient and reliable control of late blight under field conditions. First results have led to the selection of 23 potential biocontrol candidate strains due to their good performance in several *in vitro* and leaf disc assays. These strains belong to well-known biocontrol genera such as *Pseudomonas*, *Bacillus*, and *Streptomyces*, while others belong to other genera like *Acidovorax* and *Arthrobacter*, which are less studied in the biocontrol context. Future experiments will investigate those strains' ability to use different carbon and nitrogen sources, as well as their suitability for mass production. In addition, a formulation based on alginate will be developed to improve their attachment onto plant leaves and their survival in this challenging environment.

P132* Exploration of Achromobacter spanius B5 : A New Free-Living Nitrogen Fixer from Bamboo Rhizosphere

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Abstract

Free-living nitrogen fixing bacteria (FLNFB) play a crucial role in enhancing nitrogen availability particularly in low-input farming systems. To explore this ability in an underexplored niche, FLNFB were isolated from the bamboo rhizosphere in Nepal using nitrogen-free bromothymol blue medium (NFBBM). Among the isolates, strain B5 exhibited efficient **biological nitrogen fixation (BNF), producing 32.757 ± 1.86 ppm of fixed nitrogen within 24 hours and** achieving a nitrogen-fixing index of 7.36 ± 0.78 by day 5. The increase in pH from 6.0 to 6.96 ± 0.16 indicated substantial ammonia excretion. It also exhibited ammonium production confirmed by Nesslerization, with ammonium accumulation reaching 76.33 ± 2.81 ppm at 72 hours. Phylogenetic analysis of the 16S rRNA gene and biochemical characterization identified B5 as *Achromobacter spanius,* representing the **first global report of nitrogen fixation by this species and the first isolation of a FLNFB from the bamboo rhizosphere** integrating a **geographically and microbiologically underexplored Himalayan rhizosphere** in Nepal. Seed biopriming with B5 did not show positive effect on germination rate or radicle length, however, it significantly increased total seedling length by 25% and plumule length by 86% at 8 days post-germination. The novel *A. spanius* strain B5, with its dual capacity for both BNF from atmosphere and ammonia production from peptone water, emerges as a promising candidate for developing biofertilizer to support sustainable, low-input agriculture.

Keywords: *Achromobacter spanius*, biological nitrogen fixation, biofertilizer, sustainable agriculture

**Student Paper*

P133 Paradoxical oral cleaning effect in frail institutionalized adults

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Background: In frail older adults, the oral cavity can transform from a symbiotic ecosystem into a reservoir for systemic pathogens associated with pneumonia and neurodegeneration. This is increasingly recognized as a key aspect of oral frailty, which is closely linked to general frailty and increased mortality. We have characterized this reservoir and investigated the long-term effects of professional dental care.

Methods: This single-center monitoring study (MuGes-KtZH) enrolled 66 residents (mean age 84·4 years) in Swiss nursing homes. We employed a dual-diagnostic approach: automated, standardized culture to identify viable aerobic and enteric pathogens, and multiplex PCR for 11 anaerobic periodontal species. Bacterial growth and PCR C_t-values were semi-quantitatively categorized to assess colonization density.

Findings: Widespread dysbiosis of enteric pathogens was observed. Notably, all detected *Escherichia coli* and 75 % of *Enterococcus faecalis* showed abundant growth (> 10^5 CFU/mL). In a longitudinal subgroup (n = 29), a paradoxical increase in periodontal pathogen prevalence and abundance was observed six months after professional dental cleaning. Simultaneously, protective commensals declined.

Interpretation: The oral cavity in care-dependent adults serves as a stabilized reservoir for pathogens linked to aspiration pneumonia and neurodegeneration. Current biannual cleaning protocols appear insufficient and may inadvertently trigger the growth of virulent anaerobes by disrupting superficial commensal layers while leaving deep subgingival reservoirs intact. The transition to more frequent, specialized oral care protocols is recommended.

P134* Comparison of Bruker and MSI-2 database for MALDI-TOF MS identification of clinical moulds

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Background: Mould infections are difficult to diagnose and treat, and associated with high mortality in immunocompromised patients. Mould identification in clinical mycology relies on microscopy, culture characteristics, matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS), and molecular methods such as PCR and sequencing. Major challenges include slow growth, overlapping morphological features, limited spectral databases and need for specialized expertise. This study aims to determine the suitability of MALDI-TOF MS as a rapid and reliable method for routine identification of moulds in our laboratory and to assess the impact of the database choice on diagnostic accuracy.

Methods: Twenty mould isolates, including *Aspergillus* spp., *Mucorales* and dermatophytes were analyzed by MALDI-TOF MS. All strains were grown at 25°C on Sabouraud agar. The identification performance of different mycelium stages and sample preparation methods (i.e. direct on-target protein extraction Mycelium Transfer (MyT), and full protein extraction as described by Bruker) and different commercial (Bruker, Germany) and non-commercial (MSI-2, Normand 2021.) spectral databases was compared. Identification results obtained by MALDI-TOF MS were compared with those achieved by morphological identification and sequence analysis of the internal transcribed spacer (ITS) region.

Results: Using the Bruker Database, correct identification was achieved for 60.0% of isolates with MyT and 48.3% with full protein extraction, whereas the MSI-2 Database achieved rates of 55.0% and 81.7%, respectively. Overall, 18 of 20 strains were correctly identified at the genus level at least twice across three independent trials. The MSI-2 Database showed superior performance for dermatophytes.

Conclusion: MALDI-TOF MS has been an established method for the identification of bacteria and yeasts for more than a decade. In contrast, mould identification remains challenging, with performance strongly influenced by fungal species, sample preparation protocols, operator expertise, and the quality of the reference database.

**Student Paper*

P135 Rapid delineation of Acinetobacter baumannii dose responses to zosurabalpin based on machine-learning assisted interpretation of bacterial nanomotions

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Background: Carbapenem-resistant Acinetobacter baumannii (CRAB) is a critical-priority pathogen associated with high mortality and limited therapeutic options, underscoring the urgent need for both novel antibiotics and rapid antimicrobial susceptibility testing (AST) methods. Zosurabalpin (ZAB) is a first-in-class antibiotic targeting the $LptB_2FGC$ complex of the lipopolysaccharide transport pathway in A. baumannii for which clinical breakpoints have not yet been established. We present a rapid, growth-independent AST approach combining nanomotion recordings with machine learning (ML) to characterize phenotypic responses of A. baumannii to ZAB within two hours.

Method: Nanomotion responses were obtained from five A. baumannii strains with a broad range of minimum inhibitory concentrations (MICs), measured across multiple ZAB concentrations with 90 minutes exposure. Recordings were labelled as susceptible or non-susceptible according to reference MIC values and antibiotic exposure concentrations. In parallel, the effect of horse serum supplementation was evaluated with the aim to align nanomotion responses to the reference MIC, performed according to CLSI-approved methodology, previously reported for ZAB. Quantile-based spectral features extracted from the nanomotion signals were used to train a logistic regression classifier.

Results: Across 120 experiments, the ML models achieved >90% classification accuracy relative to reference susceptibility profiles, reaching up to 100% accuracy at the sample-aggregated level in serum-supplemented media. This method allowed early detection of cellular responses to ZAB prior to measurable bacterial killing in classic time-kill kinetic studies, enabling rapid phenotypic assessment of a novel antibiotic.

Conclusion: These findings establish nanomotion AST as a promising platform for accelerating the clinical integration of emerging antimicrobials and supporting next-generation rapid AST workflows against multidrug-resistant pathogens such as CRAB.

P136 Microaerophilic, Alkalitolerant and Ammonia-tolerant Methanotrophy

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Methane (CH_4) is a potent greenhouse gas with approximately 80 times the warming potential than CO_2 over a 20-year period. About 70% of all anthropogenic and natural methane emissions to the atmosphere are the result of methanogenesis by anaerobic archaea. Microbial methanotrophs are predicted to remove over 90% of the methane produced by methanogenesis and, consequently, there is a growing interest in enhancing methanotrophic activity for the biological mitigation of CH_4 emissions. Additionally, methanotrophs present promising platforms for converting CH_4 into valuable bioproducts, such as biofuels, bioplastics, and single-cell protein.

An important source of methanogenic CH_4 emissions arises from manure management - the handling, storing, disposing, and recycling of slurry from livestock. This contributes about 8% of anthropogenic CH_4 emissions globally. Thus, understanding and enhancing methanotrophy in these environments could provide an important mitigation strategy. However, little is known about the presence and function of methanotrophs in slurry systems as traditional methanotrophs are likely thought to be inhibited by the high soluble ammonium concentration, combined with high pH (typically pH 8 to 10) and low O_2 availability.

Despite this, limited observations suggest that methanotrophs could survive in the low-O_2 and high-ammonia conditions experienced in slurry. Therefore, we set out to investigate whether methanotrophs naturally inhabit slurry and what their potential roles could be in mitigating CH_4 emissions from these systems. Using targeted enrichments and microcosms generated from slurry samples, we identified persistent methanotrophic activity. We further characterised these communities with shotgun metagenomics to identify putative methanotroph MAGs, then investigated their genetic potential to perform methane oxidation and adapt to the slurry environment. Further work will include attempting isolations and characterisation of organisms of interest.

P137* Characterization of BEM-1, a Novel Chromosomal Metallo-β-Lactamase (MBL) from Paenimyroides ceti

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Paenimyroides ceti strain MC-ZM-24 was isolated from *Zophobas morio* larvae. It was resistant to ceftazidime (CAZ) but susceptible to ceftazidime (CAZ)/clavulanate (CL) and carbapenems. The Blue-Carba test indicated carbapenemase production. Whole-genome sequencing identified a new chromosomal subclass B1 bla_{MBL} gene named bla_{BEM-1}.

bla_{BEM-1} was cloned using the GeneArt Gibson Assembly HiFi Cloning Kit into the pCR Blunt II TOPO vector and transformed into TOP10 *E. coli*. Susceptibility tests were performed using Sensititre panels in the presence or absence of $ZnSO_4$, and interpreted according to the EUCAST 2026. The activity of CAZ in combination with CL was analyzed using the checkerboard broth microdilution method to determine the FICI.

The expression of BEM-1 in TOP10 was confirmed by a positive Blue-Carba test, which was inhibited in the presence of EDTA. BEM-1 conferred reduced susceptibility to 1st-, 2nd- and 3rd-generation cephalosporins (GCs). However, the transformant remained susceptible to cefepime (MIC≤1 µg/mL), aztreonam, and carbapenems. BEM-1 activity was zinc-dependent, as the addition of $ZnSO_4$ led to an increase in MICs for tested β-lactams. For instance, MICs for CAZ and CAZ with $ZnSO_4$ were 32 and 128 µg/mL, while those of imipenem and imipenem with $ZnSO_4$ were ≤0.5 and 1 µg/mL, respectively. While MC-ZM-24 was susceptible to CAZ/CL (MIC≤0.12 µg/mL), the TOP10 clone producing BEM-1 exhibited an MIC of 32 µg/mL. FICI confirmed no synergy between CAZ and CL in either strain (FICI=1). Notably, the clone remained susceptible to CAZ/avibactam (MIC=2 µg/mL).

This study provides the first characterization of the chromosomal MBL BEM-1, which is responsible for reduced susceptibility to $1^{st}/2^{nd}/3^{rd}$ GCs. Notably, the original *P. ceti* isolate remained highly susceptible to CL despite the production of BEM-1. This suggests that *P. ceti* possesses one or more Penicillin Binding Protein (PBP) inhibited by CL, as already observed (e.g., *Neisseria* and *Haemophilus* spp.).

**Student Paper*

P138* Co-infection of human host cells with Chlamydia trachomatis and Waddlia chondrophila

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Obligate intracellular bacteria of the order *Chlamydiales* rely on host-derived nutrients and extensive remodeling of host cell organelles to complete their developmental cycles. *Chlamydia trachomatis*, the leading bacterial cause of sexually transmitted infections worldwide, forms an inclusion that hijacks host trafficking pathways, including the Golgi apparatus, and can transition into aberrant bodies (ABs) under stress. In contrast, *Waddlia chondrophila*, recruits mitochondria around the inclusion and can also adopt an ABs state, suggesting distinct strategies of host exploitation. Although viral co-infections with *C. trachomatis* have been explored, interactions between bacterial members of the *Chlamydiales* within the same host cell remain unclear.

Here, we establish a co-infection model combining *C. trachomatis* and *W. chondrophila* to investigate intracellular niche organization and responses to iron limitation. Human A2EN epithelial cells were infected under synchronized conditions, and iron starvation was induced at 8 h post-infection using 2,2'-bipyridyl, followed by continuous exposure or recovery. Samples were collected across the infection cycle and analyzed by confocal immunofluorescence microscopy and inclusion-forming unit assays.

Co-infection revealed that both bacteria occupied the same host cell but remained segregated within distinct inclusions, each maintaining association with its characteristic host organelle. *W. chondrophila* developed larger inclusions, whereas *C. trachomatis* formed smaller inclusions. Upon iron restriction, both species exhibited persistence-associated morphologies. Following BPDL removal, *W. chondrophila* resumed a typical developmental cycle, with evidence of reversion from persistence, including transitions from ABs to RBs, and regained infectivity. In contrast, *C. trachomatis* was no longer detectable during co-infection, despite recovering and retaining infectivity in single infection. These observations were supported by inclusion-forming unit assays, confirming differential impacts on bacterial fitness.

Together, these findings demonstrate that *Chlamydiales* co-infection leads to spatial segregation and asymmetric responses to nutrient limitation, with competition predominating. The study was performed during practical laboratory sessions with bachelor students, integrating research-driven hands-on training.

**Student Paper*

P139 Evaluation of two commercial molecular multiplex assays detecting fungi of medical importance

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Introduction

Dermatophytes are common etiologies of superficial mycoses. Diagnosis can be long and fastidious as culture can take up to 28 days and microscopy requires expertise for species identification. Commercial PCRs detecting dermatophytes, yeasts and other filamentous fungi of medical importance have emerged on the market and are promising tools to bring faster diagnosis of superficial mycosis. We conducted a preliminary retrospective study of 3 molecular assays.

Methods

Three commercial PCRs were evaluated on 9 clinical samples: Dermagenius 3.0 Complete (Pathonostics), DermaScreen (Eurobio) and EuroArray Dx (Euroimmun). Nucleic acids were extracted with EZ1 tissue kit (Qiagen) in 50 µl elution volume with and without internal controls of DermaGenius 3.0 Complete and DermaScreen PCR kits at the Department of Dermatology from the University Hospital of Zürich. PCRs were performed according to the manufacturers on CFX96 thermocycler (Biorad) at the Microbiology Department, admed. A qualitative evaluation of the results was determined.

Results

The qualitative evaluation showed that the results were concordant for the 9 samples. Some DNA traces (Ct>35) of *C. parapsilosis* were found in 3 samples with the DermaScreen kit. According to the Instruction for Use (IFU), samples showing Ct>35 should be considered as negative. The DermaGenius kit only detected *T. violaceum* DNA in one sample. This result was expected because DermaScreen is not able to detect it.

Conclusion

DermaGenius, DermaScreen and EuroArray Derma Dx kits allow the rapid detection of *T. rubrum/T. soudanense* and *T. interdigitale/T. mentagrophytes* DNA. These preliminary results validate the feasibility of a complete prospective study aiming to compare the sensitivity of these 3 commercial molecular assays on 300 samples. The results will be presented at the Swiss Society of Microbiology Congress in next August.

P140* Iron Limitation-Induced Modulation of Transcription in Chlamydia trachomatis

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Chlamydia trachomatis (*C. trachomatis*) is an obligate intracellular bacterial pathogen responsible for trachoma and sexually transmitted infections. Like other Chlamydiota members, *C. trachomatis* undergoes a biphasic developmental cycle alternating between infectious elementary bodies (EBs) and replicative reticulate bodies (RBs). Under stressful conditions, RBs differentiate into non-dividing aberrant bodies (ABs), a reversible state associated with persistence. Here, we investigate how early iron deprivation affects transcriptional regulation in *C. trachomatis*.

We performed RNA sequencing to compare transcriptional profiles between EBs and RBs and between RBs and ABs induced by iron chelation with 2,2′-bipyridyl at 8 hpi.

In EBs, 29% of genes were downregulated and 30% upregulated compared to RBs, revealing extensive transcriptional remodelling. At 24 hpi, ABs displayed downregulation of TCS genes (*atoS*, *atoC*, and *chxR*). Comparative analysis across five *Chlamydiales* species revealed a conserved phosphotransfer core but variable sensory and output domains, indicating lineage-specific diversification. Following removal of the iron chelator, transcription of TCS genes resumed to pre-stress levels, highlighting their dynamic regulation during exit from persistence.

These findings show that *C. trachomatis* adapts transcriptionally to iron deprivation, revealing stress- and time-dependent changes in metabolism, stress responses, and host–pathogen interactions. Persistence alters the developmental cycle while maintaining inclusion integrity and modulating host interactions, contributing to chronic infection. To determine TCS function, we generated *C. trachomatis* strains with inducible TCS overexpression. Preliminary results indicate that TCS-overexpressing bacteria fail to recover infectivity after stress removal, and ongoing experiments aim to define the precise role of TCS regulation in exit from persistence.

**Student Paper*

P141 Penicillin or no penicillin? A real-time PCR for the detection of blaZ in clinical isolates of Staphylococcus aureus

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Invasive infections with methicillin-susceptible (MSSA) and/or phenotypic penicillin-susceptible *Staphylococcus aureus* (PSSA) have steadily increased in recent years. For treatment, penicillin represents a very good therapeutic alternative to other commonly used antibiotics, such as flucloxacillin or cefazolin, due to its lower rate of side effects and favourable pharmacokinetic/pharmacodynamic properties. A prerequisite for the safe therapeutic use of penicillin in infections caused by MSSA or PSSA is the absence of the *blaZ*-encoded *β*-lactamase in the pathogen. In accordance with EUCAST guidelines, detection or exclusion of *blaZ* is assessed by the appearance of the penicillin inhibition-zone edge ("beach-like", *blaZ*$^-$; "cliff-like", *blaZ*$^+$). However, this interpretation method is observer dependent and misclassification may occur, especially for isolates displaying penicillin inhibition zone diameters close to the breakpoint (≥ 26 mm).

We have established a real-time PCR for detecting all four subtypes (A – D) of the *blaZ* gene from clinical MSSA/PSSA culture isolates. The assay was validated using >200 phenotypic penicillin-resistant and susceptible *S. aureus* clinical isolates. As compared to the EUCAST method, the *blaZ* PCR showed excellent performance[TR1] . WGS data of a sub-set of the tested isolates (N=120) was 100% accordant with PCR results with respect to *blaZ* presence or absence.

The *blaZ* PCR is a valuable molecular addition to conventional penicillin inhibition zone analysis. Above all, it allows quick and reliable resolution of ambiguous cases, thereby providing an additional level of security, especially in the context of finding optimal treatment for critically ill patients. Moreover, PCR confirmation is highly recommended when antimicrobial susceptibility testing merely relies on automated systems, such as e.g. Vitek 2, since *blaZ*-positive isolates may appear phenotypically susceptible to penicillin.

P142* Characterizing Nuclear-Cytoplasmic Shuttling of mRNAs during KSHV Infection

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Kaposi's Sarcoma-Associated Herpesvirus (KSHV) is a gamma herpesvirus that leads to lifelong infections. To co-opt host resources for its own translational needs, KSHV triggers a phenomenon known as host-shutoff whereby 70% of the host transcriptome is degraded by the viral endoribonuclease SOX. This completely remodels the host transcriptional landscape within the cell and leads to the release of RNA binding proteins (RBP) from their mRNA targets. The localization of these targetless RBPs can thus be widely different upon infection and one such protein is Poly(A) binding protein cytoplasmic (PABPC), a critical stabilization regulator of polyA tails in the cytoplasm. Upon KSHV-associated host shutoff, it was observed that PABPC relocates to the nucleus, where its presence leads to stabilization of polyA tails within the nucleus, triggering aberrant hyper-polyadenylation of nascent transcripts and their subsequent nuclear retention. However, we know that 30% of host transcripts resist SOX-induced decay and remain stable in the cytoplasm, suggesting that they also manage to escape nuclear retention associated with hyperadenylation.

My PhD research project aims to gain an understanding of how the escaping transcripts manage to make their way out of the nucleus. We previously showed that, while these SOX resistant mRNA can escape nuclear retention, they do not avoid hyperadenylation. To investigate how these mRNAs can still be exported, we are exploring the role of Chromosome Region Maintenance 1 (CRM1) (or Exportin 1 (XPO1)) - a mRNA nuclear export protein involved in the export of a subset of non-canonical transcripts. We are characterizing the contribution of CRM1 to helping SOX resistant mRNAs traffic out of the nucleus, its specificity toward host and viral transcripts and how CRM1 can overall shape KSHV infection. This work will provide insights into how the hijacking of the gene expression machinery may be critical to control the outcome of infection.

**Student Paper*

P143* Target enriched long-read sequencing for culture-independent genomics of sexually transmitted bacterial pathogens

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Whole-genome sequencing has transformed the surveillance of sexually transmitted bacterial infections (STIs), enabling high-resolution strain typing, transmission analysis and prediction of antimicrobial resistance (AMR). However, many STI pathogens are difficult or impossible to culture from routine clinical samples. This is particularly relevant for *Chlamydia trachomatis*, *Treponema pallidum* and *Mycoplasma genitalium*, while *Neisseria gonorrhoeae* is often not recoverable after routine nucleic-acid-based sampling. Our project addresses this limitation by developing target-enrichment sequencing approaches for direct pathogen genome recovery from clinical material. Using short-read sequencing for such quasi-metagenomic samples, with large amounts of DNA from host and microbiota as well as the enriched pathogens, assigning genes of interest such as those involved in AMR to relevant species can be impossible. In addition, read depth variation caused by the multiple PCR cycles reduces the ability to assemble data, reducing the continuity and the resolution of repetitive or structurally complex genomic regions. This can obscure important genomic context surrounding AMR determinants, recombination events and mobile genetic elements. To address this, we have adapted our existing target-enrichment workflow for long-read sequencing using Oxford Nanopore Technologies (ONT) platforms.

Target enrichment was performed using Agilent's SureSelect Max with adaptations for long-read sequencing. To generate fragments of appropriate length, a variety of methods were tested for DNA shearing. Library preparation used the ONT Native Barcoding Kit, followed by sequencing on MinION Mk1D with R10 Flowcells. Sequencing was performed with/ without adaptive sequencing to further reduce human contamination.

Results analyse the enrichment level of the target enrichment on both spiked samples and real clinical samples, as well as the ability to improve assemblies compared to short read sequencing.

Combining target-enrichment with long-read sequencing has the potential to generate more complete pathogen genomes directly from clinical samples, improving culture-independent genomic surveillance and enhancing detection of emerging AMR mechanisms in STI pathogens.

**Student Paper*

P144 Responses of human monocytes to intracellular Staphylococcus aureus infection in vivo

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Deep-seated *Staphylococcus aureus* infections are difficult to treat: antibiotics often fail and recurrence is common, making them a significant clinical challenge. In previous work, we demonstrated that *S. aureus* is present both intra- and extracellularly in soft tissue biopsies from patients, predominantly within non-classical monocytes and macrophages. Proteomic analysis of whole biopsies revealed bacterial adaptation to the tissue microenvironment and broad tissue inflammation and coagulation; however, these approaches lack cellular resolution and cannot reveal host responses at the single-cell level.

Here, we investigate the response of individual host cells to *S. aureus* infection within patient-derived tissue. Using AI-guided fluorescence microscopy, we identified infected and non-infected host cells and confirmed the intracellular presence of bacteria within monocytes and macrophages. Target cells are then isolated using laser microdissection for downstream mass spectrometry proteomic analysis. This approach enables the comparison between infected and non-infected cells to understand host-pathogen interactions in vivo. The results will guide our efforts to build physiologically relevant in vitro models for drug testing and mechanistic analysis.

P145 Phenotypic screening for carbapenemases and ESBLs in Pseudomonas aeruginosa using the VITEK-2 AST-N443 and Phoenix NMIC-486 cards

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Background: *Pseudomonas aeruginosa* can develop carbapenem resistance through chromosomal mechanisms or by acquiring plasmid-mediated carbapenemases and/or extended-spectrum β-lactamases (ESBLs). Conventional β-lactam susceptibility testing does not reliably distinguish carbapenemase/ESBL-producing carbapenem-resistant *P. aeruginosa* (CP/ESBL-CRPA) from non-carbapenemase-producing isolates, here referred to as AmpC-hyperproducers. However, these groups exhibit distinct susceptibility patterns to novel β-lactam/β-lactamase inhibitor (BL/BLI) combinations, including ceftolozane/tazobactam (C/T), ceftazidime/avibactam (C/A), imipenem/relebactam (I/R), and meropenem/vaborbactam (M/V). We investigated potential screening cut-offs for detecting CP/ESBL production in CRPA using the VITEK-2 AST-N443 and Phoenix NMIC-486 cards.

Methods: Antimicrobial susceptibility testing was performed using the VITEK-2 AST-N443 and Phoenix NMIC-486 cards on 126 whole-genome-sequenced international CRPA isolates resistant by disc diffusion to ceftazidime, cefepime, imipenem, and/or meropenem. The collection included 70 metallo-β-lactamase (MBL)-producing isolates, of which eight co-produced ESBLs, as well as two OXA-48-, one KPC-2- and one GES-5-producing isolate. Twelve isolates carried ESBLs only, while the remaining 40 represented AmpC-hyperproducers. MIC distributions were analyzed to identify discriminatory cut-offs for carbapenemase and ESBL detection.

Results: C/T was the most discriminatory BL/BLI combination for differentiating CP/ESBL-CRPA from AmpC-hyperproducers. For the VITEK-2 AST-N443, a C/T MIC ≥32 mg/L achieved 100% sensitivity for MBL±ESBL detection (70/70) and 91.7% sensitivity for ESBL-only producers (11/12), with 88.1% specificity (37/42). For the Phoenix NMIC-486, a C/T MIC >8 mg/L achieved 100% sensitivity for both MBL and ESBL detection, with 83.3% specificity (35/42). Combining additional BL/BLI agents did not further improve specificity.

Conclusions: Elevated C/T MICs represent reliable phenotypic screening markers for MBL and ESBL production in CRPA using both the VITEK-2 AST-N443 and Phoenix NMIC-486 systems.

P146 Should our medicine go viral? A dialogue with Swiss society about phage therapy

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Bacteriophage therapy is increasingly used around the world as an innovative treatment option for patients with antibiotic-resistant or otherwise difficult-to-treat bacterial infections. However, access to phage therapy in Switzerland remains limited because important regulatory, ethical, clinical, and societal questions are still unresolved. The SNSF Agora project "Should our medicine go viral?" creates a forum that brings together stakeholders and Swiss society to promote a dialogue around these questions. Through several moderated discussions with active audience engagement across Switzerland, we connect patients, clinicians, researchers, regulators, policymakers, and the public to explore what responsible access to phage therapy could mean in Switzerland. Our central goal is not to promote phage therapy as a simple solution, but to support an informed and balanced conversation about its opportunities, limitations, and possible routes towards patient access in the real world. At the Swiss Society for Microbiology meeting, we present the concept and first experiences of this project. Beyond reporting our specific activities, we showcase how an informed dialogue between the public and diverse stakeholders may help shape the responsible implementation of innovative therapies in Switzerland and beyond.

P147 Integrating metabolomics with susceptibility assays reveals context-dependent antimicrobials under in vivo-like conditions.

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Conventional antibiotic discovery platforms typically screen compound libraries under standardized *in vitro* conditions that fail to recapitulate the physiological constraints encountered by bacteria at infection sites. As a result, promising candidates often fail during *in vivo* validation, while compounds with potential *in vivo* efficacy are overlooked. Classical susceptibility screens primarily assess bacterial growth inhibition without elucidating the drug's mode of action, leading to the rediscovery of compounds targeting pathways already exploited by existing antibiotics. To address this gap, we combined growth inhibition assays with metabolomics to quantify drug-induced metabolic perturbations in bacteria. We applied this approach to *Escherichia coli* challenged with human-targeted compounds in media designed to mimic host-like physiological conditions.

We found that *in vivo*-mimicking conditions strongly influence the antimicrobial activity of these compounds. As an illustrative example, we discovered that a purine analogue approved as an immunosuppressive agent exhibits antimicrobial activity exclusively under osmotic stress conditions. Metabolomics analysis revealed that osmotic stress induced the downregulation of nucleotide biosynthetic enzymes, thereby increasing bacterial vulnerability to nucleotide analogue compounds. Furthermore, the phosphatase UmpH, a key enzyme in nucleotide metabolism, dephosphorylated toxic drug metabolites, thus attenuating the drug's antimicrobial effects. Consistently, UmpH-deficient strains exhibited increased susceptibility to purine analogues compared with wild-type controls. Together, these findings reveal a dual mechanism, in which osmotic stress dysregulates nucleotide metabolism and compromises bacterial detoxification capacity. Our results suggest the potential for repurposing nucleotide analogues in the treatment of infections associated with high-osmolarity environments.

In summary, our findings demonstrates that screening under *in vivo*-like conditions combined with metabolomics can uncover context-dependent antimicrobial activities and reveal stress-induced metabolic vulnerabilities, thereby providing a rational framework for drug repurposing and antibiotic discovery.

P148 GRAMUR—Evaluating the Gram Stain in Urinary Tract Infections: A Retrospective Analysis of Discordance Determinants and Clinical Impact

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Background and Objectives

Urinary tract infections (UTIs) are frequent infections, but urine Gram staining has declined due to reliability concerns and doubtful clinical utility, only performed on physician request at Lausanne University Hospital (CHUV) since 2020. The primary objective of this study is to evaluate the clinical impact of Gram staining. The secondary objective is to assess the reliability of urine Gram staining and to identify the determinants of discordance.

Materials and Methods

This retrospective observational study used data from CHUV. For the primary objective, patients with a Gram request are compared against a control group, assessing clinical outcomes such as antibiotic modifications and additional interventions within six hours after Gram reporting. For the secondary objective, urine samples with both Gram staining and culture results between 2012 and 2025 are analyzed to identify factors associated with discordant findings.

Results

A total of 299,965 urine samples were identified, of which 33,932 had both Gram stain and culture results. After February 2020, 40 urine Gram stains were requested. For Gram-negative bacteria, Gram staining showed an accuracy of 86.1%, sensitivity of 76.6%, and specificity of 94.6%. Corresponding values were 84.1%, 69.8%, and 88.3% for Gram-positive bacteria, and 97.8%, 46.4%, and 99.8% for yeasts. Discordant Gram stain and culture results occurred in 13.8% of Gram-negative and 13.3% of Gram-positive cases. Among Gram-negative discordances, 21.2% were negative culture with positive Gram findings, potentially attributable to prior antibiotic treatment, while 78.8% showed positive culture with negative Gram, reflecting the lack of sensitivity of Gram staining. To answer the primary objective, the clinical data is currently being analysed.

Conclusions

Urine Gram staining demonstrated high specificity but moderate sensitivity, making it useful for confirming infection but insufficient for excluding it. Ongoing analyses will determine whether Gram staining demonstrates clinical impact, such as empirical antibiotic prescribing and antimicrobial stewardship.

P149* Impact of Heavy Metals on Antibiotic Resistance of Escherichia coli from Slum Wastewater

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Slum dwellers face significant infrastructure challenges which facilitate prevalence of antibiotic resistance. This study evaluated the impact of heavy metals on antibiotic resistance patterns of *Escherichia coli* in wastewater from slums of Bwaise II, Bwaise III, Kazo, and Makerere III in Kawempe division, Kampala.

Levels of heavy metals (lead, mercury, cadmium, chromium, and arsenic) in wastewater were determined using inductively coupled plasma mass spectroscopy. *Escherichia coli* were isolated from wastewater using MacConkey agar and their susceptibility to 50 µl of stock antibiotics (tetracycline, amoxicillin, ceftriaxone at 30 µg/ml, and ciprofloxacin at 5 µg/ml) determined. The potential of heavy metals to induce antibiotic resistance in Escherichia coli was determined by culturing susceptible isolates in 200 µl of Luria-Bertina broth containing stock antibiotics (10 µl), or stock antibiotics (10 µl) and stock heavy metals (10 µl).

Detectable levels of heavy metals were reported in wastewater from Bwaise II, Kazo and Makerere III only. Lead, cadmium and arsenic, mercury and chromium, were highest in Bwaise II, Kazo, and Makerere III, respectively. The occurrence of *Escherichia coli* resistant to at least an antibiotic was 72.8% (169 of 232) and resistance to tetracycline, ceftriaxone, amoxicillin, and ciprofloxacin were 34.1%, 28.9%, 35.3%, and 34.5%, respectively. Study findings further revealed a positive correlation (R^2 = 0.371–0.985) between the presence of heavy metals in wastewater and antibiotic resistance patterns of Escherichia coli. Also, heavy metals; lead (77.41 µg/ml), mercury (1.44 µg/ml), and cadmium (10.21 µg/ml) significantly ($p < 0.05$) induced antibiotic resistance in susceptible *Escherichia coli*.

Findings highlight the role of heavy metals in prevalence of antibiotic resistance in bacteria. There is a need for proper disposal of wastes that contribute to the build-up of heavy metals in the environment, hence contributing to management of antibiotic resistant bacteria.

**Student Paper*

P150 NANOMUR—Accelerating Urine Antibiotic Susceptibility Testing via Nanomotion Technology and Machine Learning: A Prospective Validation Study

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Background

Urinary tract infections (UTIs) are very prevalent and can become life-threatening in cases of complications such as pyelonephritis or sepsis. However, urine antibiotic susceptibility testing (AST) requires more than 30 hours, potentially leading to suboptimal antibiotic treatment. This delay complicates UTI management, especially amid rising antibiotic resistance.

The NANOMUR study aims to shorten the time-to-results for urine AST using an innovative technology called “nanomotion”, which measures bacterial vibrations, bypassing the need for culture.

Methods

Turbid urine samples from the CHUV were selected when positive for leukocytes and nitrites on dipstick testing. Time-to-result and nanomotion performance were compared with the routine diagnostic workflow—culture and VITEK2. Several machine-learning approaches were compared: the first framework was trained on spiked blood samples using logistic regression, whereas the other models were trained and evaluated directly on the urine sample dataset using 3-fold cross-validation.

Results

Nanomotion’s median time-to-result was significantly shorter than routine diagnostic, with 4.4 hours, instead of 37.5 hours.

Nanomotion performance was first evaluated on 81 monomicrobial urine samples using a model trained on spiked blood sample. The categorical agreement (CA) was 82.72%, Major Errors rate (ME) 14.1%, and Very Major Errors rate (VME) 40%. Subsequently, performance was assessed on urine samples using a new framework and multiple machine-learning approaches with 3-fold cross-validation. The best-performing model was a multilayer perceptron, achieving a 95.06% CA, 2.82% ME, and 20% VME.

Nanomotion experiments are now conducted at 37°C for 2 hours, and preliminary results demonstrate a reduction in VME down to 10.53%.

Conclusion

Nanomotion demonstrated great performance when using an algorithm tailored to urine samples. More experiments at 37°C are conducted to conclude the analysis. The main advantage of nanomotion is its short time-to-result, which could allow earlier narrow-spectrum antibiotic with potential such as better antibiotic stewardship or even better patient outcomes.

S41*/P151* Hemagglutinin-associated stability of H5N1 influenza A virus in exhaled respiratory droplets

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The stability of influenza A virus (IAV) in exhaled respiratory droplets is a key determinant of airborne transmission, as virions are rapidly exposed to inactivating conditions such as low pH, and high salinity. Different strains of IAV vary in their ability to transmit via aerosols and the viral hemagglutinin protein (HA) plays a central role in transmissibility. To assess whether HA contributes to viral stability during the airborne phase, we characterized a pair of biosafe replication-incompetent H5N1 reporter viruses. Both viruses are based on the A/Vietnam/1203/2004 isolate but differ in their HA protein: While one (Viet/04-Renilla) carries the HA of the non-airborne transmissible A/Vietnam/1203/2004 isolate, the other one (Viet/04-Renilla-Tx) carries HA of A/Texas/37/2024, which belongs to the currently circulating H5N1 2.3.4.4b clade associated with airborne transmission in the ferret model. Our data indicate that Viet/04-Renilla-Tx displays increased thermal stability compared with Viet/04-Renilla. Under high-salinity conditions in bulk solutions, no discernible difference in stability was observed between the two viruses. However, in drying 0.5 and 5uL droplets, which mimic efflorescence and salt supersaturation during aerosol evaporation, Viet/04-Renilla-Tx exhibited increased stability relative to Viet/04-Renilla. In a next step, controlled aerosol chamber experiments are planned to assess viral stability in aerosols. Together, our findings indicate that Viet/04-Renilla carrying HA from A/Texas/37/2024 exhibits greater resistance to inactivating conditions during the airborne phase, which may contribute to its increased transmissibility.

**Student Paper*

P152 Biofilm Formation and Antimicrobial Resistance in Uropathogenic Staphylococcus aureus: Molecular and Phenotypic Insights from Casablanca, Morocco

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Introduction

Antibiotic resistance and biofilm formation in *Staphylococcus aureus* represent a major public health concern, particularly in urinary tract infections (UTIs), where they complicate treatment outcomes. This study investigated the relationship between antimicrobial resistance and biofilm-forming capacity in uropathogenic *S. aureus* isolates from Casablanca, Morocco.

Methods

Antibiotic susceptibility was assessed using disc diffusion and broth microdilution methods. Resistance genes, including aph(3′)-IIIa, aac(6′)/Ie-aph(2″)-Ia, ant(4′)-Ia, blaZ, and mecA, were detected by PCR. Biofilm formation was evaluated using Congo red agar and microtiter plate assays. Genes associated with adhesion and biofilm production (spa, fnbA/B, clfA, bap, icaADB, and icaR) were also screened.

Results

Among 20 isolates, 55% were multidrug-resistant and methicillin-resistant. All strains exhibited biofilm-forming ability, with 55% moderate and 35% strong producers. Significant associations ($p < 0.01$) were observed between multidrug resistance and enhanced biofilm production. All isolates were susceptible to linezolid, while 20% showed constitutive clindamycin resistance. Virulence genes spa, fnbB, and clfA were detected in all strains, followed by bap (95%) and fnbA (90%). The icaA gene was present in 90% of isolates, and the coexistence of icaA/icaADB/icaAD was significantly associated with MRSA strains, whereas icaR was more frequent in methicillin-susceptible isolates.

Conclusion

This study demonstrates a significant association between biofilm formation and antimicrobial resistance in uropathogenic *S. aureus*, emphasizing their combined role in treatment failure and their relevance as a public health concern.

S45∗/P153∗ A genome editing pipeline in human lung cells enables functional dissection of epithelial barrier protection against P. aeruginosa

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The opportunistic pathogen *Pseudomonas aeruginosa* (*PA*) is a leading cause of life-threatening respiratory infections. During airway colonisation, the bacterium uses diverse strategies, including surface adherence, virulence induction, and tissue dissemination, to adapt to the respiratory niche. However, studying these complex dynamics in murine models is limited by the lack of high temporal and spatial resolution, while many cell models fail to faithfully recapitulate human airway physiology. To provide a more translatable and physiologically relevant understanding of the interaction between *PA* and the airway epithelium, we adapted a Transwell-based air-liquid interface (ALI) model from primary human bronchial epithelial stem cells. This microtissue faithfully recapitulates the native airway epithelium, showing proper epithelial barrier function and mucociliary differentiation.

In order to define the host–pathogen interactions that facilitate or restrict *PA* invasion, we established a highly efficient, virus- and plasmid-free CRISPR-Cas9 genome editing pipeline. This platform allows us to systematically deconstruct the epithelial defence layers, such as mucus production and ciliary function, to determine their individual roles in bacterial infection. As a functional validation, we targeted FOXJ1, a master regulator of ciliogenesis, which led to marked changes in airway epithelial morphology and loss of ciliation. By enabling the precise genetic manipulation of the human airways, this approach allows for a detailed dissection of host-pathogen dynamics with a level of precision difficult to achieve *in vivo*.

∗Student Paper

P154 Molecular Characterisation of Multidrug-Resistant Salmonella Typhi Isolated from Stool Samples of Patients with Typhoid Fever

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The emergence of Multi-Drug-Resistant (MDR) *Salmonella typhi* to commonly used antibiotics has posed greater challenges in the treatment of typhoid fever. The aim of this study was to characterise MDR *Salmonella* Typhi isolated from stool samples of patients using molecular techniques. One thousand and twenty-two (1022) stool samples were collected from patients attending secondary health centres within Benue South Geographical Zone. *Salmonella* Typhi was isolated using classical cultural method. The isolates were purified on Bismuth Sulfite agar and further characterized using biochemical techniques. The isolation rate of *S.* typhi in the study area was 43.7% (n=447). Prevalence of *S.* typhi with regards to location (χ^2 = 54.293), age (χ^2 = 15.316), sex (χ^2 = 5.263) and month of sampling (χ^2 = 94.090) differed significantly ($p<0.05$). *S. typhi* infection was most prevalent in subjects aged ≤10 years (52.7%, n=243). Male subjects had a significantly (χ^2=5.263; $p< 0.05$) higher isolation rate (46.8%; n=583) than female (39.6%; n=439). Highest isolation rate was in the month of May (61.8%, n=76). The antibiotic susceptibility pattern of the *S*. typhi isolates showed that 64% (n=286) were resistant to one or more class(es) of antibiotics. The isolates demonstrated the highest resistance to the fluoroquinolone (ciprofloxacin) (55.6%; n=159). All the isolates were however, susceptible to carbapenem and aminoglycoside. In addition, 27.3% of the isolates exhibited multi-drug resistance (MDR). There was a significant difference in the occurrence rate of MDR *S. typhi* in the various locations (χ^2 = 27.459, $p < 0.05$). Most of the isolates showed plasmid mediated resistance. Most MDR *S.* typhi isolates were positive for ESBL production. The MDR isolates harboured *bla* SHV, *bla* TEM and *QnrA* but lacked *Imp*A and *aac*. There is high prevalence of *S.* typhi in the study area and the fact that most of them are MDR calls for concern.

S36*/P155* Sublethal pesticide exposure and the honey bee gut microbiota: interactions and impacts on bee health

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Bees are essential pollinators whose health is increasingly threatened by agricultural intensification and widespread agrochemical exposure. While pesticide risk assessment largely focuses on acute toxicity, chronic sublethal exposure can also impair immune competence, pathogen resistance, and foraging behavior. Because these same functions are partly supported by the specialized honey bee gut microbiota, pesticide-induced perturbation of this community may contribute to sublethal effects on the host. Conversely, the gut microbiota may modulate pesticide toxicity through biotransformation or by altering their persistence and bioavailability.

Here, we use the honey bee gut microbiota to investigate reciprocal pesticide–microbiome interactions. Building on a high-throughput *in vitro* screen of agrochemical effects on bee-associated bacterial isolates, we exposed gnotobiotic honey bees colonized with a defined synthetic community to selected agriculturally relevant pesticides at sublethal concentrations corresponding to 1% of the reported bee LD50. Microbial community dynamics were followed across exposure and recovery using qPCR and full-length 16S rRNA amplicon sequencing.

Overall, the synthetic gut community was largely resilient at these concentrations, although selected pesticides caused subtle strain-specific shifts in community assembly. Follow-up experiments comparing mono-colonization with *Gilliamella apicola*, a conserved honey bee gut bacterium, to full-community colonization at higher pesticide doses further suggest that community context can buffer pesticide sensitivity of vulnerable strains. In parallel, targeted and untargeted LC-MS analyses revealed pesticide-dependent differences in gut metabolite profiles and pesticide persistence, providing a basis to explore microbiota-mediated pesticide biotransformation and broader metabolic changes *in vivo*. Finally, pathogen challenge experiments with *Serratia marcescens* showed that some pesticide treatments increased infection frequency, suggesting that sublethal exposure may weaken microbiota-mediated colonization resistance. This provides an initial readout for exploring how pesticide-induced microbiota perturbations may affect broader bee health outcomes. Together, our results support the gut microbiota as a key factor in modulating the sublethal impacts of agrochemical exposure on bees.

**Student Paper*

S75*/P156* Individual bacterial taxa drive colonisation resistance to methicillin-resistant Staphylococcus aureus in human nasal microbiome samples

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Nasal carriage of the pathobiont *Staphylococcus aureus* represents a major risk factor for infections, yet the ecological factors that determine *S. aureus* colonisation success within healthy nasal microbiomes remain poorly understood. Our aim is to identify and characterise *S. aureus*-antagonistic interactions in the nasal microbiome and quantify the contribution of pairwise interactions to community-level outcomes.

Using an in vitro microcosm approach to cultivate nasal microbiome samples from healthy donors, we find that samples differ markedly in their ability to inhibit growth of incoming methicillin-resistant *S. aureus* (MRSA), and that these differences are associated with community composition. By assembling nasal model communities reflecting our samples and applying targeted drop-in and drop-out designs, we show that individual taxa can drive colonisation resistance to MRSA. Together, our results establish causal connections between microbiome composition and clinically relevant community-level outcomes, thereby bridging the gap between associative, sequencing-based microbiome studies and mechanistic work on pairwise bacterial interactions.

Ongoing work investigates the molecular mechanisms underlying these inhibitory interactions, with the aim of informing both our understanding of nasal microbiome ecology and the development of novel therapeutic strategies.

**Student Paper*

P157* Excitation Emission Matrix enables Screening of Proteolytic Activity in Fungal Solid-State Fermentation

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Fungal solid-state fermentation (SSF) is increasingly applied to improve the nutritional quality of plant-based foods particularly by enhancing protein digestibility through fungal enzymatic activity. However, optimization of SSF processes remains limited by the lack of rapid, extraction-free tools to monitor proteolytic activity across strains, substrates, and fermentation conditions. Here, we explore Excitation Emission Matrix (EEM) fluorescence spectroscopy as a scalable approach to monitor fungal proteolysis during SSF of plant-based substrates. EEM fingerprints of fermented materials reveal characteristic and time-dependent transformations of intrinsic protein fluorescence, including systematic changes in the protein region and the emergence of amino acid peaks. Quantitative EEM-derived descriptors, such as peak intensities and tyrosine-to-tryptophan fluorescence intensity ratios, show strong correlations with the degree of protein hydrolysis measured using the o-phthaldialdehyde (OPA) assay. Protein quantification by BCA was used to correct protein extraction yields across samples, enabling valid comparison of OPA results. Based on these correlations, EEM fluorescence enables semi-quantitative tracking of proteolytic activity over fermentation time and across different fungal strain-substrate combinations. In a funnel-based workflow, selected fermentation conditions representing distinct proteolytic behaviors are subsequently prioritized for nutritional assessment. Ongoing work applies the standardized INFOGEST in vitro digestion protocol to evaluate how fermentation-induced proteolysis, as monitored by EEM and OPA, relates to protein solubility and digestibility in the bioaccessible fraction. Overall, this study highlights the potential of EEM fluorescence to bridge fermentation kinetics and nutritional assessment, offering a rapid screening strategy to guide strain and process selection in fungal SSF of plant-based foods.

**Student Paper*

P159* Exploring actinomycetes as biocontrol agents for late blight management in potato and tomato

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The widespread use of chemical pesticides for crop protection has raised significant environmental and health concerns, driving the urgent need for sustainable alternatives. Potato (*Solanum tuberosum*), one of the world's most important staple crops, is highly susceptible to various pathogens, leading to severe yield losses. Notably, *Phytophthora infestans*, the causative agent of late blight disease, remains one of the most devastating threats to global potato production. Traditional late blight management relies heavily on fungicides, which pose environmental risks and are increasingly ineffective due to the rise in pathogen resistance. Actinomycetes are filamentous Gram-positive bacteria that have demonstrated potential as biocontrol agents due to their ability to produce specialized metabolites with antimicrobial properties and to enhance plant growth and resilience.

This study aims to explore the biocontrol potential of 238 actinomycete strains isolated from the rhizosphere and phyllosphere of potato plants. Their antagonistic activity was assessed *in vitro* against 6 major potato pathogens, namely *Dickeya solani*, *Pectobacterium carotovorum*, *Rhizoctonia solani*, *Alternaria solani*, *Phytophthora infestans*, and *Pythium ultimum*, using a high-throughput screening approach, with particular emphasis on *P. infestans*. Approximately 71% of the strains exhibited antagonistic activity against at least 1 pathogen. Inhibition rates varied among the tested pathogens, with 5% of the strains active against *D. solani*, 8% against *P. carotovorum*, 41% against *A. solani*, 39% against *R. solani*, 30% against *P. ultimum*, and 59% against *P. infestans*. Additionally, 15% of the strains showed activity exclusively against *P. infestans*.

Promising isolates will subsequently be evaluated *in planta* for their ability to reduce late blight infection in potato and tomato plants and to enhance plant resistance responses, with the most effective candidates being further assessed under field conditions. This work highlights the potential of potato-associated actinomycetes as sustainable biocontrol agents, supporting their use as an alternative to chemical pesticides in modern agriculture.

**Student Paper*

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