Northern Germany Meeting on Infection, Immunology & Inflammation

Research Center Borstel

Leibniz Lung Center

Manor House

08.10.2025

Contents

Welcome Note

Dear Colleagues and Friends,

It is our great pleasure to welcome you to the joint 2025 symposium of NDI³ and NordInfect.

This special edition brings together two established scientific communities: New Developments in Immunology, Inflammation & Infection (NDI³), now in its 47th year, and NordInfect, a Northern German initiative dedicated to bacterial pathogenesis. By combining the strengths of both, this year's meeting fosters interdisciplinary dialogue and scientific exchange across institutions and career stages.

NDI³ has a long tradition of being organized by second-year PhD students at the Research Center Borstel. It serves as a platform by and for early-career researchers, offering opportunities to present research, gain feedback, and build networks. Through the collaboration with NordInfect, the symposium broadens its reach and brings together an even more diverse community of scientists working in immunology, inflammation, and infection research.

The 2025 program reflects this spirit of collaboration. Highlights include keynote lectures by internationally renowned experts, oral presentations representing a diverse range of research backgrounds and career levels, and interactive poster sessions. We warmly encourage you to actively participate in the discussions, exchange ideas, and make the most of the networking opportunities.

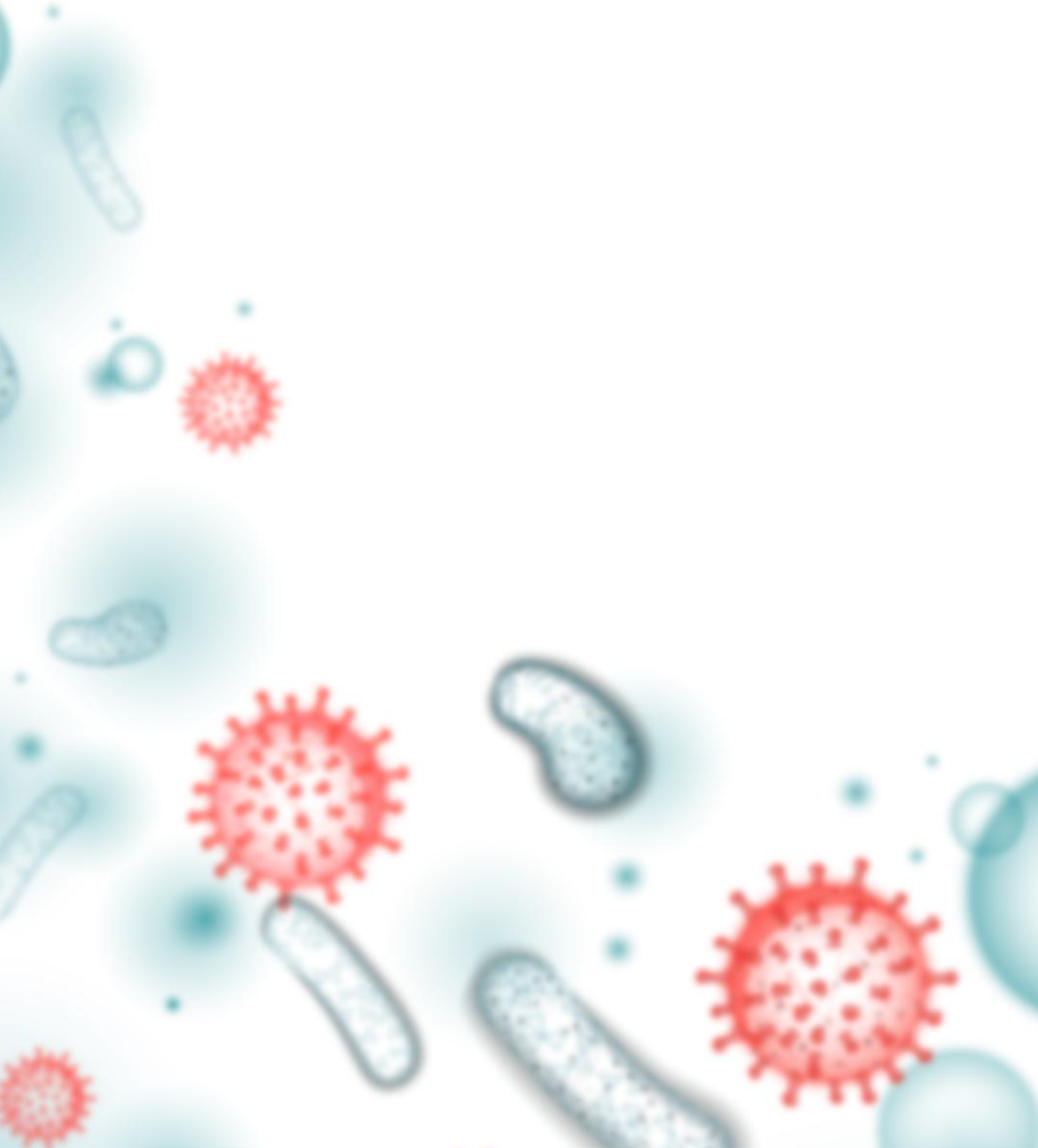
None of this would be possible without the enthusiasm and support of our speakers, sponsors, and attendees — thank you!

We hope you enjoy this year's symposium and the collection of abstracts that follow. We look forward to welcoming you in person to Borstel for an inspiring and memorable event.

Warm regards,

The NDI³ & NordInfect 2025 Organizing Committee

Program



Northern Germany Meeting on Infection, Immunology & Inflammation

08:30 – 09:00 Registration with coffee

09:00 – 09:15 **Welcome** *(Caroline Barisch)*

Infection session

***Chair**: Norbert Reiling*

09:15 – 09:50 **Olivier Neyrolles**, *Toulose (Keynote)*
Effluxosomes and the arms race for metal homeostasis in Mycobacterium tuberculosis

09:50 – 10:05 **Bidiepta Saha**, *Greifswald – Insel Riems, Talk #1*
Tunneling Nanotubes (TNTs): An export/import strategy for Chlamydia via direct cell-to-cell communication

10:05 – 10:20 **Martino Morici**, *Hamburg, Talk #2*
Fighting antimicrobial resistance with new weapons: Saskemycin, a potent antimycobacterial agent targeting a unique site on the ribosome

10:20 – 10:30 Group picture in front of the manor house

10:30 – 11:00 Coffee break

***Chair:** Daniel Meissner*

11:00 – 11:35 **Maximiliano Gutierrez**, London *(Keynote)*
Membrane damage and repair dynamics in tuberculosis

11:35 – 11:50 **Lena Gonner**, *Hamburg, Talk #3*
TARGET-MYCO peptide-enhanced targeting and eradication of pathogenic mycobacteria

11:50 – 12:05 **Michelle Savickis**, *Hamburg, Talk #4*
Function and spatio-temporal patterning of the Small basic protein (Sbp) in Staphylococcus epidermidis biofilm formation.

12:05 – 12:25 Coffee break

***Chair:** Holger Sondermann*

12:25 – 13:00 **Susanne Häußler**, Braunschweig *(Keynote)*
Functional genomics in Pseudomonas aeruginosa

13:00 – 14:00 Lunch break

Immunology & Inflammation session

Chair: *Aileen Kerfin and Lennart Bartels*

14:00 – 14:30 **Sponsor pitches**

Biomol (Anneliese Fuhrmann), ibidi (Carla Seegers), Novogene (Lena Danckert), BioLegend (Agata Widera), MACHEREY-NAGEL (Cordt Westensee)

Chair: *Bianca Schneider*

14:30 – 15:05 **Silke Meiners**, *Borstel (Keynote)*
The immunoproteasome at the crossroad of infection & autoimmunity

15:05 – 15:20 ***Marco Trujillo**, Hamburg, Talk #5*
Exocyst subunit links immune signalling and autophagy

15:20 – 15:35 ***Jakia Khan**, Borstel, Talk #6*
Targeting IL-13 signaling pathway as a potential therapeutic approach in experimental models of pulmonary arterial hypertension

15:35 – 16:00 Coffee break

Chair: *Katarzyna Duda*

16:00 – 16:35 **Christian Karsten**, *Lübeck (Keynote)*
The complement system: Its critical role in Infection control and the challenges of infection risk in patients receiving anti-complement therapies

16:35 – 16:50 ***Daniel Anton Myburgh**, Kiel, Talk #7*
Tracing the pre-antibiotic osteomyelitis pathogens behind today's hospital superbug epidemics

16:50 – 17:05 ***Ann-Cathrin Hofacker**, Kiel, Talk #8*
Effects of smoking, physical activity and obesity on respiratory and organismal health across generations

Chair: *Linda Zemke*

17:05 – 17:40 **Speed and / or "edutainment" talks**

***Maria Lerm**, Linköping, Talk #9*
Quantum biology – an emerging field of science that matters to everyone

***Thierry Cottineau**, Greifswald – Insel Riems, Talk #10*
Hypoxic persistence of spore-like particles (SLPs) of the zoonotic pathogen Coxiella burnetii

***Judith Bossen**, Kiel, Talk #11*
JAK/STAT signalling controls proliferation, apoptosis and migration of progenitor cell types in the Drosophila airways

***Liisa Knipp**, Borstel, Talk #12*
Sex-specific regulation of immunoproteasome function determines response to infection

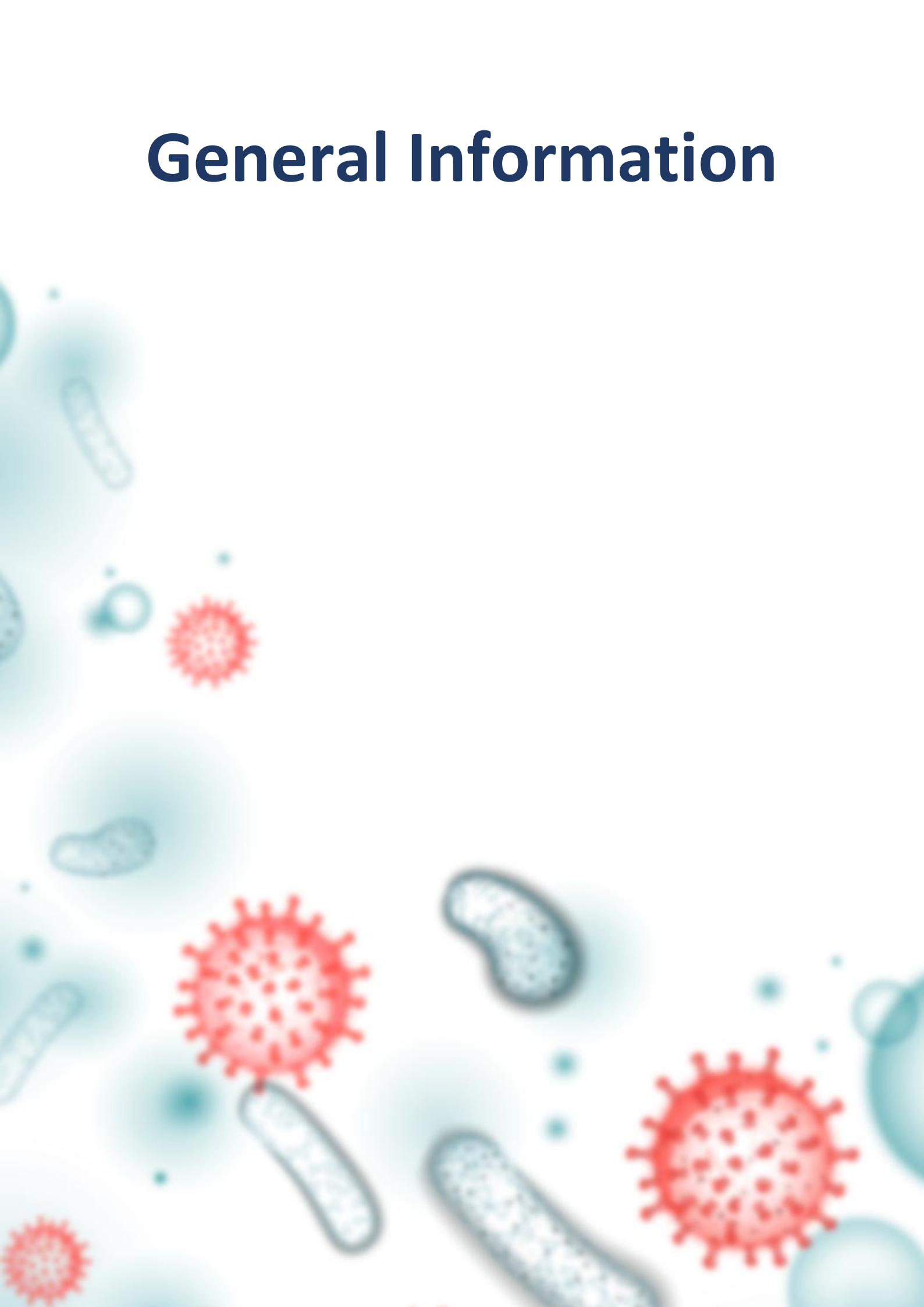
***Kai Guo**, Borstel, Talk #13*
Dissecting immunoproteasome function in lung regeneration using mouse lung organoids

***Fabian Lüttchens**, Hamburg Talk #14*
Synthesis and structure-activity relationships of novel amino acid-based LpxC-inhibitors

Closing session

17:40 – 18:15	**Poster session 1** *(with drinks and snacks)* – even numbers
18:15 – 18:50	**Poster session 2** *(with drinks and snacks)* – odd numbers
18:50 – 19:15	**Awards and conclusion**
19:15 – 20:30	**Dinner**

General Information



VENUE

Research Center Borstel, Leibniz Lung Center

Parkallee 1 (Manor House)

D-23845 Borstel

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via Lübeck
Take the B206 from Lübeck to Bad Segeberg, then continue on the B432 towards Hamburg. After approximately 15 km, turn left at the Borstel junction. The Research Center is on your right-hand side.

via Kiel
Take the B404 from Kiel to Bad Segeberg, then follow the B432 towards Hamburg for about 15 km until you reach the Borstel turn-off. Turn left. The Research Center is located on the right-hand side.

via Bad Oldesloe
From Bad Oldesloe, follow the main road towards Borstel via Grabau and Tönningstedt. The Research Center will be on your left-hand side.

Arrival by public transport

via Hamburg

Start at Ochsenzoll station (U1). From there, take bus 7550 in the direction of Bad Segeberg and get off at “Borstel (Sülfeld), B432”. From the bus stop, it’s about a 15-minute walk to the venue. As the bus runs hourly, we suggest checking the schedule in advance.

via Bad Oldesloe

Start at Bad Oldesloe train station. From there, take the bus 7141 in the direction of Henstedt-Ulzburg and get off at “Borstel (Sülfeld), Borsteler Hof”. From the bus stop, it’s about a 10-minute walk to the venue. As the bus runs hourly or less frequently, we suggest checking the schedule in advance.

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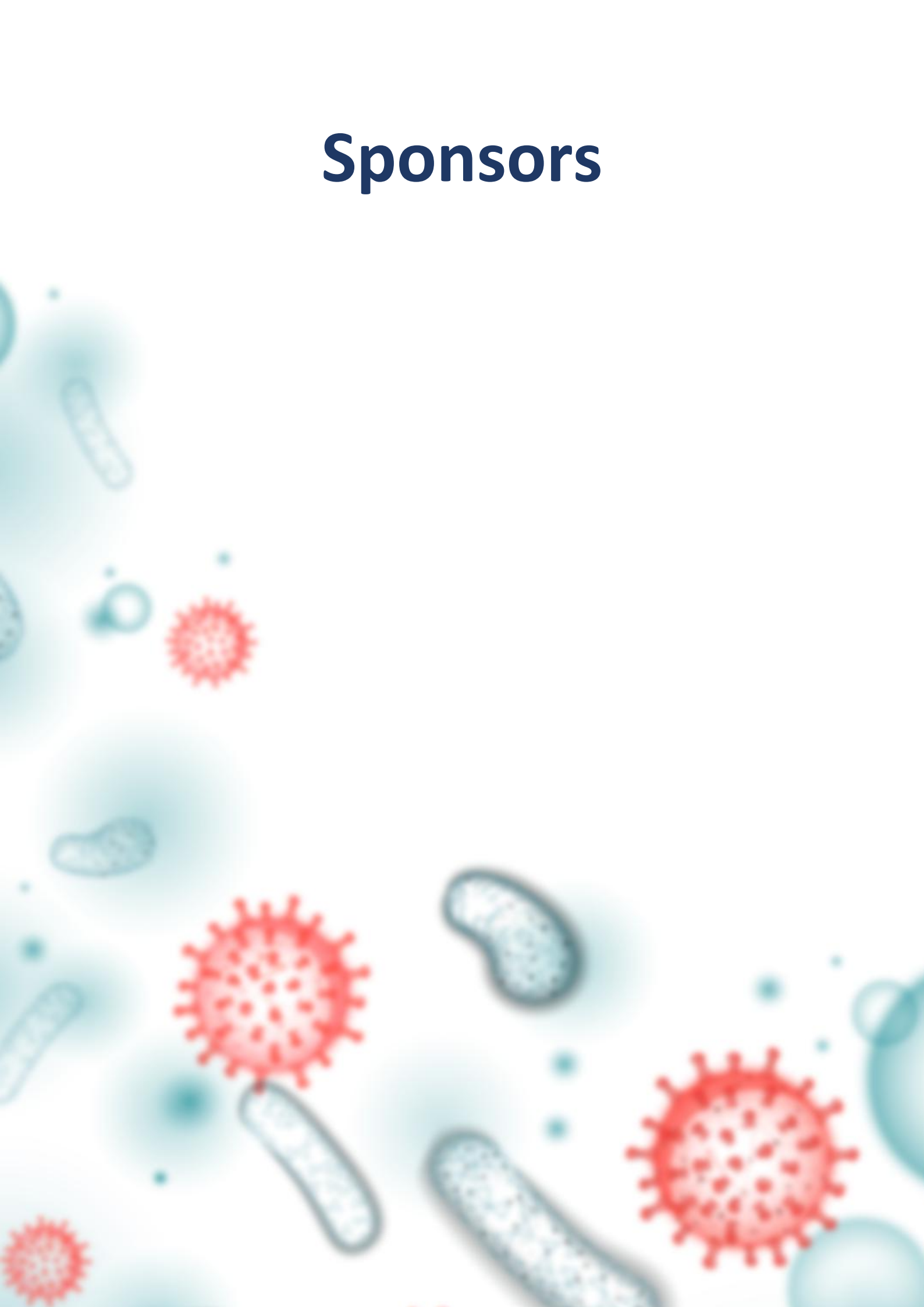

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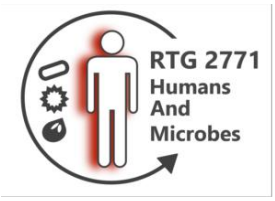

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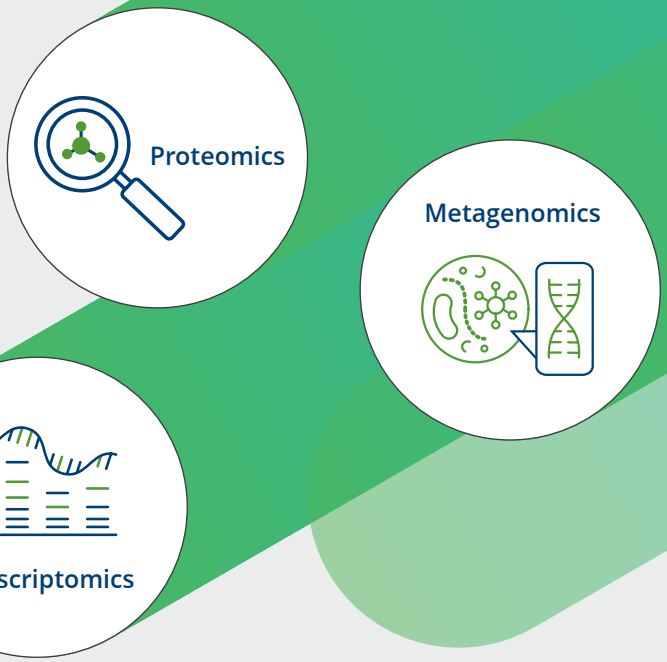

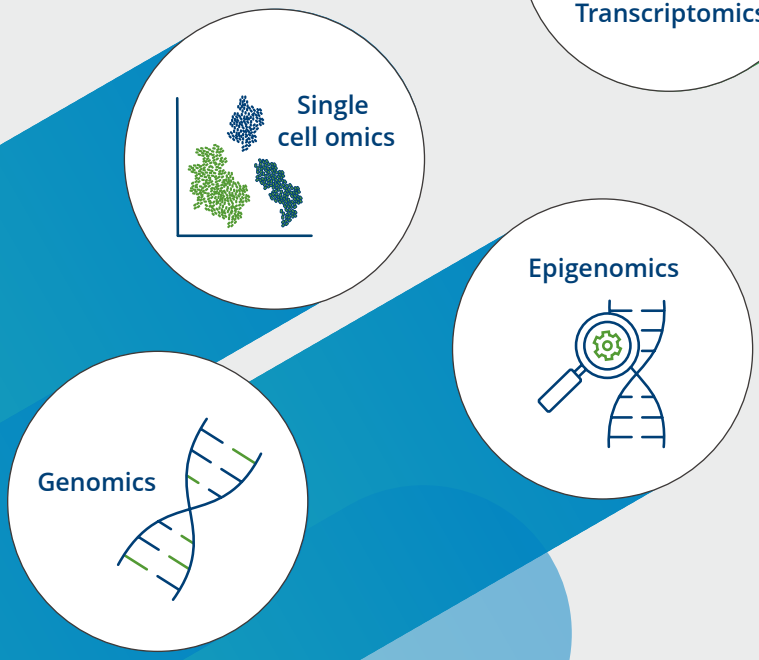

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Keynote Speakers

Olivier Neyrolles, Institute of Pharmacology and Structural Biology, France

Effluxosomes and the arms race for metal homeostasis in *Mycobacterium tuberculosis*

Pathogenic bacteria face a formidable challenge inside host immune cells: surviving the toxic onslaught of transition metals such as zinc, copper, and cadmium. In this talk, I will explore how *Mycobacterium tuberculosis* has evolved sophisticated strategies to counteract this metal-based immunity. Drawing on a body of work spanning molecular microbiology, cell biology, and high-resolution imaging, I will discuss how the bacterium senses, responds to, and exports toxic metals via specialized membrane structures. Central to this process are P-type ATPases, metal efflux pumps, functionally stabilized and organized by PacL proteins into dynamic membrane nanodomains we term effluxosomes. These assemblies coordinate multi-metal detoxification and adapt to environmental stress. I will trace the evolution of this concept from the initial discovery of zinc intoxication in infected macrophages, through the identification of PacL1 as a critical chaperone-like partner of CtpC, to the recent characterization of multimeric effluxosomes involving multiple PacL/Ctp pairs. Our findings reveal a conserved, modular system for metal resistance that supports bacterial survival in hostile intracellular environments and opens new avenues for antimicrobial targeting.

Maximiliano Gutierrez, Francis Crick Institute, United Kingdom

Membrane damage and repair dynamics in tuberculosis

The endomembrane system associated with the endocytic and phagocytic pathways is crucial for cell homeostasis. Multiple human pathogens, such as *Mycobacterium tuberculosis*, can damage this membrane-enclosed compartments leading to the leakage of luminal components. This leakage represents a danger response, and cells have developed repair mechanisms to restore endomembrane integrity and contain the leakage. Here, I will discuss the recent advances in new imaging modalities and highlight their potential applications for studying membrane damage and repair highlighting new concepts regarding how human macrophages sense damage after infection with *M. tuberculosis*.

Susanne Häußler, Helmholtz Centre for Infection Research, Germany

Functional genomics in *Pseudomonas aeruginosa*

Whole Genome Sequencing (WGS) of bacterial pathogens will provide detailed insights not only into the bacterial resistance profile, but also shed light on pathogenicity potential and the phylogenetic relatedness of nosocomial pathogens. This, in turn, holds the promise of serving as the foundation for a more precisely targeted treatment approach and the effective implementation of infection control measures. To fulfil this vision, we work on the following objectives: i) establish a pan-genomic database encompassing all conceivable sequence variations within a bacterial species as a prerequisite for robust and automated extraction of sequence information, ii) correlate phenotypes including antibiotic resistance with genotypes, with the ultimate aim of “reading” the bacterial genomes to predict bacterial behaviour, and iii) develop a user-friendly tool for visualizing the outcomes of genotyping. In essence, our undertaking seeks to harness the power of WGS to not only comprehensively document bacterial genetic variations but also to enhance our ability to predict and combat antibiotic resistance, all while advancing our understanding of the intricacies of nosocomial pathogen dynamics.

Silke Meiners, Research Center Borstel, Leibniz Lung Center, Germany

The immunoproteasome at the crossroad of infection and autoimmunity

Immunoproteasomes are specialized types of proteasomes that degrade intracellular proteins in immune cells and upon inflammatory induction in non-immune cells. They play a key role in MHC class I antigen presentation by shaping CD8+ T cell responses upon virus infection. Immunoproteasome activity is also crucial for immune cell function and has been linked to autoimmune responses. Previously, we demonstrated that cigarette smoke impairs baseline immunoproteasome expression in immune cells of the lung and is reduced in its activity in chronic obstructive pulmonary disease (COPD), a detrimental smoke-related lung disease.

Here, we focus on the key function of immunoproteasomes in infection of parenchymal cells and how this might be affected by cigarette smoke and aging. We show that cigarette smoke impairs the induction of the immunoproteasome in non-immune cells of the lung thereby attenuating activation of CD8 T cells by virus infection. In aging, immunoproteasomes are upregulated in non-immune cells of the lung. In a mouse model of mitochondrial dysfunction-related premature aging, we show that immunoproteasomes and CD8+ T cell activation are upregulated by the cGAS/STING signaling pathway. We provide evidence that the induction of such a novel adaptive type I interferon response occurs in aberrant epithelial cells of patients with idiopathic lung fibrosis (IPF), a devastating lung disease with a survival rate of 3-5 years, which links to the activation of CD8+ T cells in lungs of these patients. Our findings suggest that dysregulated immunoproteasome function in parenchymal cells of the lung contributes to the development of chronic lung diseases.

Christian Karsten, Institute for Systemic Inflammation Research, Germany

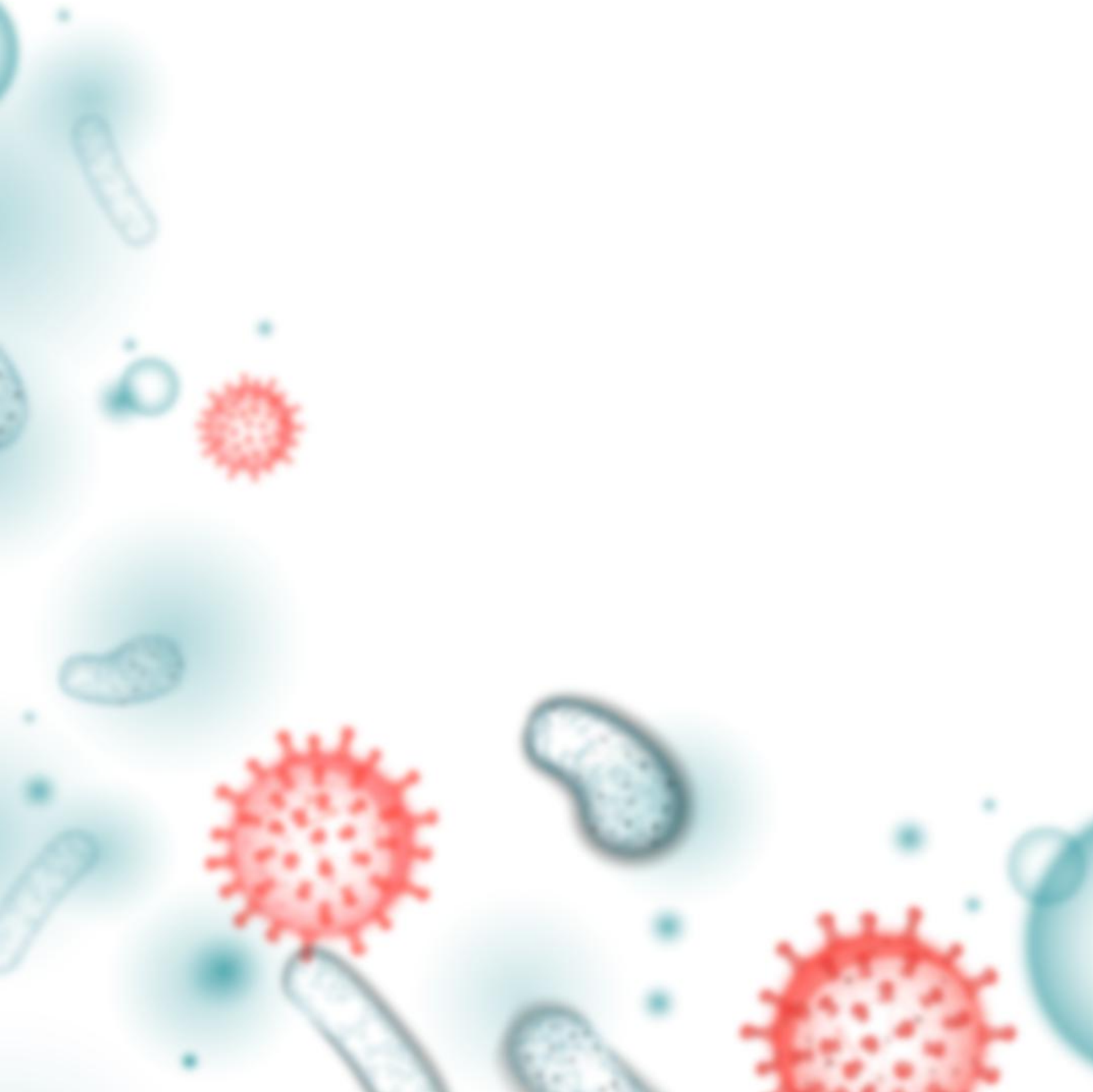
The Complement System: Its Critical Role in Infection Control and the Challenges of Infection Risk in Patients Receiving Anti-Complement Therapies

The complement system represents a fundamental pillar of innate immunity, orchestrating pathogen recognition, immune cell recruitment, and bacterial elimination through multiple effector mechanisms. This ancient cascade operates via three pathways - classical, alternative, and lectin - converging at C3 activation and culminating in membrane attack complex formation. Beyond opsonization and phagocytosis enhancement, complement provides essential serum bactericidal activity against encapsulated bacteria, particularly Neisseria meningitidis, Streptococcus pneumoniae, and Haemophilus influenzae.

The past five years have witnessed unprecedented expansion in complement-targeted therapeutics, with fourteen FDA-approved inhibitors now available. Terminal complement inhibitors (eculizumab, ravulizumab, zilucoplan) targeting C5 demonstrate remarkable efficacy in paroxysmal nocturnal hemoglobinuria and atypical hemolytic uremic syndrome (aHUS). Notably, infection-triggered aHUS represents a critical clinical scenario where complement dysregulation paradoxically increases both thrombotic microangiopathy risk and subsequent infection susceptibility. Proximal complement inhibitors including pegcetacoplan (C3), iptacopan (factor B), and danicopan (factor D) offer alternative therapeutic approaches with potentially distinct infection risk profiles.

However, complement blockade inevitably compromises antimicrobial defense. C5 inhibitor recipients face up to 2,000-fold increased meningococcal infection risk despite vaccination. Current risk mitigation mandates comprehensive meningococcal vaccination (serogroups A, C, W, Y, B) administered ≥2 weeks before therapy initiation, with consideration for antibiotic prophylaxis. Vaccination schedules require modification with accelerated booster intervals every 3-5 years. Emerging evidence suggests alternative pathway inhibitors may confer lower infection risk than terminal complement blockade, particularly in adequately vaccinated patients. Nevertheless, breakthrough infections occur despite optimal prophylaxis, necessitating heightened clinical vigilance and individualized risk-benefit assessments.

Oral Presentations



Talk #1: Tunneling Nanotubes (TNTs): An Export/Import Strategy for *Chlamydia* via Direct Cell-to-Cell Communication

Bidiepta Saha[a], Rico Jahnke[a], Katherina Psathaki[b], Michael R Knittler[a]

[a]Institut of Immunology, Friedrich Loeffler Institute, Insel Riems - Greifswald, Germany
[b]Institut für Mathematik, Universität Osnabrück, Osnabrück, Germany

Chlamydiae are obligate intracellular pathogens causes various diseases in both humans and animals. Earlier research models proposed Chlamydia exit depended on cell lysis, while uptake relied on receptor-mediated interactions. However, *in vivo* evidence rarely shows host-cell lysis, while in vitro uptake of extracellular chlamydiae is inhibited by physiological concentrations of heparin. Another widely studied exit method of Chlamydia is the extrusion pathway which has been described *ex vivo.* However, uptake of these extrusion structures appears to be limited to antigen-presenting cells. This suggests additional pathways for chlamydial transmission in tissues during infection. Our studies show that Chlamydia exploits tunneling nanotubes (TNTs) as an alternative and efficient route of cell-to-cell transmission. TNTs are transient, membrane-bound connections that allow intercellular communication and transport of vesicles, proteins and organelles. Recent studies have shown that various pathogens misuse TNTs for stealthy transmission, enabling them to evade the immune system. Infection models using both immortalized cell lines and primary cells demonstrate the transport of chlamydial reticular bodies (RBs) through these structures. Using high-resolution microscopy, flow cytometry and biochemical assays, we identified the inclusion protein IncA forming filamentous structures within TNTs. Our findings showed a close association between the protein IncA and a key host TNT regulator, LST1. Our discoveries elucidate the roles of bacterial and host proteins in TNT-mediated transmission of Chlamydia, highlighting a strategy for immune evasion. Understanding the mechanism opens opportunities for development of novel therapeutic approaches and vaccines against this widespread pathogen.

Acknowledgements: SPP2225 – EXIT (DFG Project)

References:

1. A. Rico Jahnke, B. Michael R Knittler, Chlamydia trachomatis Cell-to-Cell Spread through Tunneling Nanotubes. **2022***, 10,* 6.

Talk #2: Fighting antimicrobial resistance with new weapons: Saskemycin, a potent antimycobacterial agent targeting a unique site on the ribosome

Martino Morici[a], Dmitrii Travin[b], Michael Cook[c], Min Xu[c], Haaris A. Safdari[a], Nora Vázquez-Laslop[b], Alexander Mankin[b], Gerard Wright[c], Daniel N. Wilson[a]

[a]Institute for Biochemistry and Molecular Biology, University of Hamburg, Martin-Luther-King-Platz 6, 20146 Hamburg, Germany.
[b]Center for Biomolecular Sciences, University of Illinois at Chicago, Chicago, IL, USA.
[c]David Braley Center for Antibiotic Discovery, Michael G. DeGroote Institute for Infectious Disease Research, Department of Biochemistry and Biomedical Sciences, McMaster University, Hamilton, Ontario, Canada.

Tuberculosis is the deadliest bacterial disease on the planet[1]. The months-long regimen of multiple antibiotics required to treat tuberculosis profoundly affects the microbiome and leads to the development of antimicrobial resistance[2]. Furthermore, non-tuberculous mycobacterial infections pose an increasing clinical challenge[3]. Consequently, there is a growing need for new narrow-spectrum mycobacteria-targeting antibiotics with different mechanisms of action[4]. Here, we report the discovery and characterization of a natural antibiotic, saskemycin (SKM), which demonstrates potent and highly selective activity against mycobacteria. Genome sequencing, chemical analysis, and isotope feeding strategies reveal the unique structure and biosynthetic origin of SKM, a cationic glycolipid consisting of a tetra-saccharide 'head' and a 2-*N*-methyl-polyagmatine methyl guanine undecanoic acid 'tail'. SKM binds to the small ribosomal subunit at a site not targeted by clinically relevant antibiotics acting on the ribosome. Bound to the ribosome, SKM blocks the decoding center in a unique way, thereby preventing stable binding of aminoacyl-tRNA in the A site and inhibiting translation in a sequence context-specific manner. Self-resistance in the producing organism is conferred by methylation of a single 16S rRNA nucleotide by SasO and SasN rRNA methyltransferases. These enzymes are orthologs of the ubiquitous RsmC and SpoU methyltransferases found in most bacterial genera but absent in mycobacteria, rationalizing SKM's exquisite selectivity. The discovery of SKM provides an entry to develop selective, microbiome-sparing antimycobacterial antibiotics with a unique structure, binding site, and mechanism of action.

Acknowledgements:

Cryo-EM data collection was performed at the Multi-User CryoEM Facility at the Centre for Structural Systems Biology, Hamburg, supported by the Universität Hamburg and DFG grant numbers (INST 152/772-1|152/774-1|152/775-1|152/776-1|152/777-1 FUGG).

References:

1. World Health Organization (2024). Global Tuberculosis Report 2024. WHO. Oct 29, 2024. https://www.who.int/teams/global-programme-on-tuberculosis-and-lung-health/tb-reports .
2. Naidoo, C.C., Nyawo, G.R., Wu, B.G., Walzl, G., Warren, R.M., Segal, L.N., and Theron, G. The microbiome and tuberculosis: state of the art, potential applications, and defining the clinical research agenda. **2019**, Lancet Respir Med 7, 892-906. 10.1016/S2213-2600(18)30501-0 .
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4. Stanley, S.A., Grant, S.S., Kawate, T., Iwase, N., Shimizu, M., Wivagg, C., Silvis, M., Kazyanskaya, E., Aquadro, J., Golas, A., et al. Identification of novel inhibitors of *M. tuberculosis* growth using whole cell based high-throughput screening. **2012**, ACS Chem Biol 7, 1377-1384. 10.1021/cb300151m .

Talk #3: TARGET-MYCO Peptide-Enhanced Targeting and Eradication of Pathogenic Mycobacteria

Lena Gonner [a,b,c,d], Aby Anand [a,b], Niels Röckendorf [e], Chris Meier [f], Thomas Gutsmann [a,c], Caroline Barisch[a,b,d]

[a]Centre for Structural Systems Biology, Hamburg, Germany.
[b]Division of Host-Microbe Interactome, Research Center Borstel, Leibniz Lung Center, Borstel, Germany.
[c]Division of Biophysics, Research Center Borstel, Leibniz Lung Center, Borstel, Germany.
[d]Biology Department, University of Hamburg, Hamburg, Germany.
[e]Division of Mucosal Immunology and Diagnostics, Research Center Borstel, Leibniz Lung Center, Borstel, Germany.
[f]Chemistry Department, University of Hamburg, Hamburg, Germany.

Tuberculosis, caused by *Mycobacterium tuberculosis*, is the deadliest disease in the world caused by a single infectious agent [1]. The emergence of multidrug-resistant strains has become a major global health concern, highlighting the need for new therapeutic approaches such as host-directed therapies. Antimicrobial peptides (AMPs) are promising candidates due to their ability to disrupt bacterial membranes. However, the lack of specificity of AMPs, as well as the lipid-rich and rigid nature of the mycobacterial membrane, limits their efficiency against mycobacteria [2]. This study aims to improve the effectiveness of AMPs by combining them with a mycobacterial membrane-targeting motif to enhance the direct delivery of the AMPs. The well-established *Dictyostelium discoideum-Mycobacterium marinum* host-pathogen model system will be used to mimic *M. tuberculosis* infection in eukaryotic cells. The first stage of the project involves identifying and characterising potential targeting motifs. This involves synthesising peptides and performing binding assays with mycobacteria using microscopy and flow cytometry. Further optimisation of these motifs will then help develop the most effective combination of motifs and AMPs to eradicate mycobacteria, thereby contributing to novel tuberculosis therapy development.

1. WHO, *Global Tuberculosis Report*. **2024**, WHO Publications.

2. T. Gutsmann, *Interaction between antimicrobial peptides and mycobacteria*. **2016**, Biochim. Biophys. Acta, 1858, 1034–1043.

Talk #4: Function and spatio-temporal patterning of the Small basic protein (Sbp) in *Staphylococcus epidermidis* biofilm formation

Michelle Savickis[a], Samira Weißelberg[a], Thomas Gutsmann[b], Yannic Bischoff[c], Michael Hecht-Bucher[c], Antonio Virgilio Failla[d], Bente Siebels[e], Niclas Dassel[e], Thomas Mair[e], Ali Biabani[e], Jim Monistrol[f], Meytal Landau[f], Holger Rohde[a]

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[b]Division of Biophysics, Research Center Borstel, Center for Systems Structural Biology, Hamburg, Germany
[c]Center for Experimental Medicine, Institute for Biochemistry and Signaltransduction UKE, Hamburg, Germany
[d]Microscopy Imaging Facility (UMIF), UKE, Hamburg, Germany
[e]Core Facility Mass Spectrometric Proteomics, UKE, Hamburg, Germany
[f]Center for Systems Structural Biology (CSSB), Deutsches Elektronen-Synchrotron DESY, Hamburg, Germany

Staphylococcus epidermidis is a common nosocomial pathogen responsible for implant-associated infections. Its pathogenicity is closely linked to biofilm formation, which is supported by an extracellular matrix. This study focused on characterizing Sbp, an 18 kDa small basic protein that is a major matrix component.

CLSM, STED and Minflux microscopy showed that Sbp is produced immediately after adhesion of *S. epidermidis* 1457 to the surface. Interestingly, Sbp initially spreads in a two-dimensional pattern on the substrate before bacterial colonization begins. As the biofilm matures, Sbp becomes integrated into the three-dimensional matrix.

To investigate its structural role and potential interactions, AFM analysis was performed, indicating striking evidence in Sbp's involvement in organizing the cell-matrix architecture. Additionally, affinity pulldown and proximity labeling experiments identified 40 potential interaction partners—primarily oxidoreductases—suggesting a role for Sbp in detoxifying reactive oxygen species (ROS) generated during aerobic metabolism. Supporting this, a *sbp* knockout mutant exhibited increased extracellular ROS levels, along with growth defects and a pronounced stress response upon entering the stationary phase. Importantly, this phenotype could be partially rescued by antioxidant treatment, supporting a protective role of Sbp against oxidative stress.

In conclusion, this study provides dynamic insights into biofilm matrix assembly in S. epidermidis at single molecule resolution. Control of ROS detoxification by biofilm matrix components puts a novel and so far, undiscovered piece to the puzzle of how biofilms ensure bacterial homeostasis to support chronic biomaterial associated infections.

Talk #5: Exocyst subunit links immune signalling and autophagy

Carla Brillada[a,†], Ooi-Kock Teh[b,c,d,†], Franck Anicet Ditengou[a], Chil-Woo Lee[b], Till Klecker[e], Bushra Saeed[a], Giulia Furlan[b], Marco Zietz[b], Gerd Hause[f], Lennart Eschen-Lippold[b], Wolfgang Hoehenwarter[b], Justin Lee[b], Thomas Ott[a,g] and Marco Trujillo[a,b,h]

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[b]Leibniz Institute of Plant Biochemistry, 06120 Halle (Saale), Germany
[c]Department of Biological Science, School of Science, Hokkaido University, 060-0810 Sapporo, Japan
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[e]Institute of Cell Biology, University of Bayreuth, 95440 Bayreuth, Germany
[f]Biozentrum, Martin-Luther-University Halle-Wittenberg, Halle 06120 (Saale), Germany
[g]CIBSS—Centre for Integrative Biological Signalling Studies, University of Freiburg, 79085 Freiburg, Germany
[h]Department of Plant Immune Biology, Institute for Plant Sciences and Microbiology, University of Hamburg, Hamburg, Germany
[†]These authors contributed equally

During immune responses, plant cells need to boost secretion to deliver defense molecules, but they also need to keep secretion under control to maintain balance. The exocyst, a protein complex that directs secretion, is particularly regulated by one of its subunits, Exo70.

In *Arabidopsis thaliana*, we found that one of the Exo70 paralogue acts as a functional part of the exocyst. However, immunogenic such as the bacterial PAMP flg22, as well as the defence hormone mimic BTH, Exo70 is redirected to the vacuole for degradation.

This redirection depends on autophagy. Exo70 contains two motifs that allow it to interact with the autophagy protein ATG8. We also discovered that Exo70B2 is phosphorylated by the immune-activated kinase MPK3. Phosphorylation has two effects: it prevents Exo70B2 from working at secretion sites by inhibiting binding to the plasma membrane and it strengthens its interaction with ATG8, enhancing its degradation by autophagy. Interfering with Exo70 phosphorylation results and stronger immune responses.

Together, these results suggest a mechanism in which phosphorylation controls whether Exo70B2 promotes secretion or is degraded via autophagy, thereby fine-tuning the levels of immune receptors and thus, of the immune response.

Talk #6: TARGETING IL-13 signaling pathway as a potential therapeutic approach in experimental models of pulmonary arterial hypertension

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Introduction: Systemic sclerosis (SSc) is a severe autoimmune disease characterized by inflammation, fibrosis, and vasculopathy. Pulmonary arterial hypertension (PAH) caused by progressive vascular remodeling and vascular occlusion, is among the most fatal complications. We recently established a novel PAH model using IL-13 transgenic (IL-13tg) mice, which overexpress IL-13 in activated T cells. In this study, we explored the therapeutic potential of targeting the IL-13 signaling pathway in this model. Materials and Methods: We used three experimental systems: (i) in vivo: IL-13 transgenic (IL-13tg) mice with or without AT1R immunization, (ii) in vitro: human pulmonary arterial smooth muscle cells (PASMCs) for proliferation (xCELLigence, alamarBlue) and receptor blockade (anti-IL-4Rα, antiIL-13Rα2), and (iii) ex vivo: precision-cut lung slice (PCLS) to establish a long-term culture system for vascular pathology. Results: IL-13tg mice developed severe occlusive pulmonary vasculopathy, independent of AT1R immunization, demonstrating IL-13 as a direct driver of vascular pathology. Histology revealed vascular wall thickening due to PASMC hyperplasia rather than endothelial expansion. PASMCs expressed both IL-13Rα1 and IL-13Rα2, and IL-13 induced dose-dependent proliferation. Blockade of IL-4Rα significantly inhibited the IL-13-induced PASMC proliferation, while IL13Rα2 blockade had minimal impact. In the PCLS model, preliminary results showed that culture in medium with 0.1% FCS extended tissue viability to 14 days, providing a feasible time window to study ex vivo vascular remodeling. Conclusion: IL-13 producing T cells plays a pivotal role in driving pulmonary occlusive vasculopathy in mice. Our integrated in vivo, in vitro and ex vivo approaches provides a translational platform for testing targeted inhibitors, suggesting IL-13 signaling as a promising therapeutic target in SScassociated PAH.

Talk #7: Tracing the pre-antibiotic osteomyelitis pathogens behind today's hospital superbug epidemics

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Osteomyelitis is a potentially life-threatening infection of the bone marrow (1), historically associated with high mortality prior to the introduction of antibiotics (2,3). Despite advances in treatment, it remains a major healthcare burden today, with an estimated 50,000 cases annually in the United States (4) and an incidence of ~17 per 100,000 in Germany (5). The condition can be caused by a wide range of pathogens, most notably *Staphylococcus aureus* (1,6,7), as well as other members of the "ESKAPEE" group (1,6). Several of these taxa are organisms of particular concern, due to their rapid acquisition of multidrug resistance. Despite their clear clinical importance and frequent association with life threatening sepsis and osteomyelitis, the evolutionary origins and phylogenetic relationships of these pathogens remains largely unexplored. To address this, we analysed seven autopsy specimens from adults who suffered from osteomyelitis between the mid-1800s and 1920s in Germany. Using untargeted pathogen screening, phylogenetic analysis and investigation of virulence factors, we identified pathogenic strains of *Acinetobacter baumannii, Staphylococcus aureus* and *Streptococcus pyogenes*. Phylogenetic analysis revealed direct links between these historical strains and modern pandemic lineages. Additionally, investigation of virulence factors uncovered that all strains harboured conserved genes for adhesion, biofilm formation, cytotoxicity and immune evasion, suggesting these traits are crucial for establishing bone infections. We also demonstrated that the *S. aureus* strain was methicillin susceptible, whist the *A. baumannii* exhibited early multidrug resistance potential. Our findings provide critical insights for understanding the past and present of hospital-acquired infections.

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Talk #8: Effecs of smoking, physical activity and obesity on respiratory and organismal health across generations

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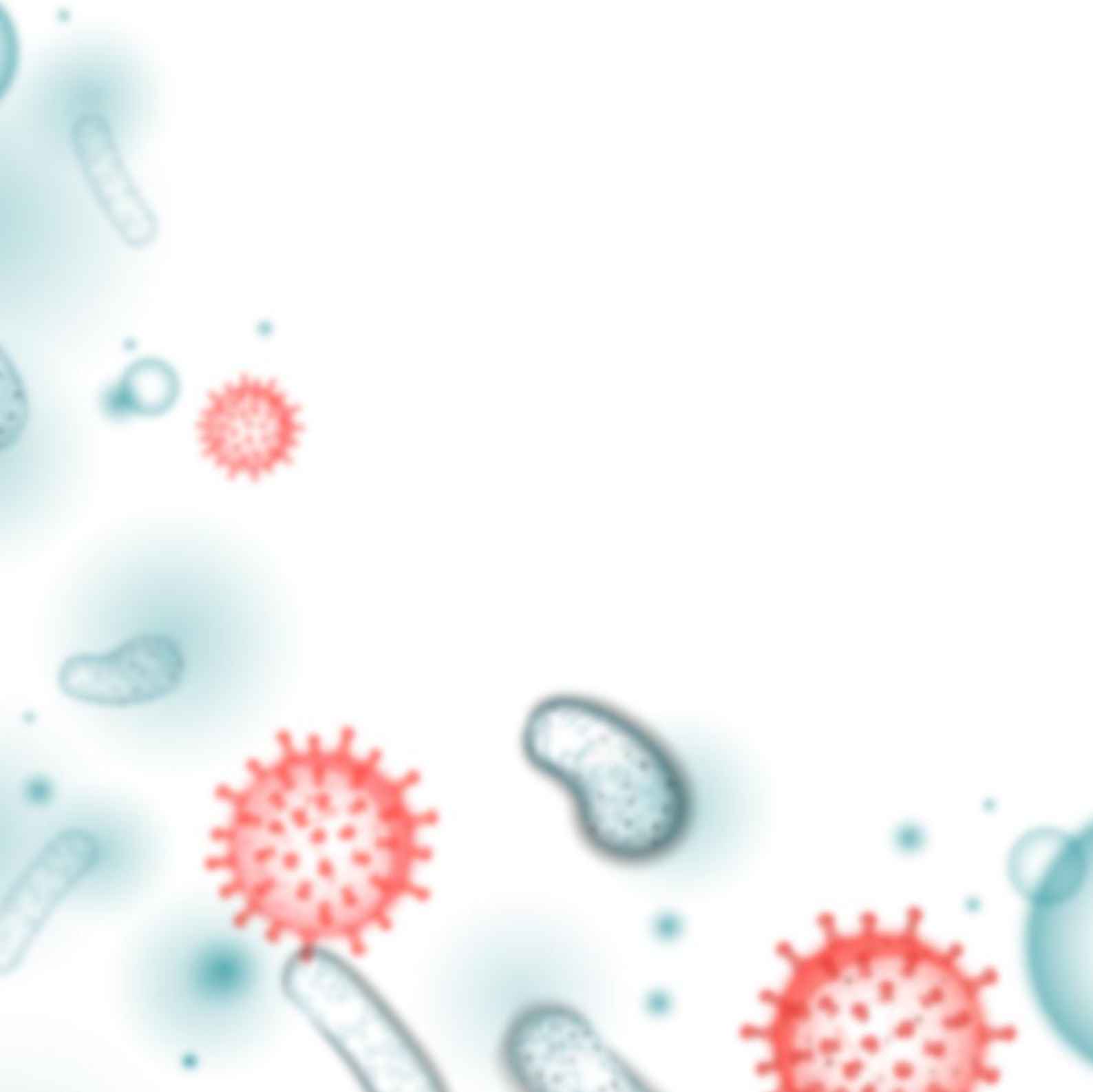
The main characteristics of the so-called Western lifestyle, which is widespread in industrialized societies, are a combination of a sedentary lifestyle and a high-calorie diet that is characterized by high levels of sugar and fat. This unfavorable lifestyle in early and later life impairs lung development and significantly increases the risk of cardiovascular and pulmonary diseases. Moreover, cigarette smoke substantially aggravates these pathological developments. We used the fruit fly *Drosophila* as a model to study the effects of high-calorie diets in combination with cigarette smoking and physical activity. In the intragenerational setting, a high-calorie diet and smoking significantly shorten the lifespan of flies. The combination of smoking and a high-calorie diet aggravated the lifespan shortening. Impaired body composition and fitness parameters induced by a high-calorie diet were usually aggravated by cigarette smoking. Moreover, a high-calorie diet decreased physical activity dramatically, and the combination of a high-calorie diet and cigarette smoking increased the susceptibility to airborne stressors. Interestingly, both interventions increased stem cell proliferation and reduced gut health. The analysis of the effects on the airway structure and the airway progenitor cells in the intra- and transgenerational setting is currently underway.

Speed Talks



Talk #9: Quantum biology –an emerging field of science that matters to everyone

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The United Nations has proclaimed 2025 to be the International Year of Quantum Science and Technology. The ambition is to raise public awareness of the impact of quantum science on all aspects of life and celebrate the 100th anniversary of a number of discoveries in quantum mechanics in 1925. These science-transforming discoveries made by Erwin Schrödinger and others are now the fundament of science in physics and chemistry. In 1944, Schrödinger published a book entitled “What is life?” in which he in theory explores the fundamental question of how physical and chemical processes can account for the phenomena of life, with the ambition to bridge the gap between physics and biology. Today, a growing body of publications provides experimental evidence for non-trivial quantum phenomena in biological systems. These include magnetoreception, enzyme function, mutagenesis, photosynthesis, olfaction and more. In the science slam, I would like to introduce you to the fascinating world of quantum physics and its applications and let you know more of how we want to apply these principles to our research on mycobacteria-phagocyte interaction.

Talk #10: Hypoxic persistence of spore-like particles (SLPs) of the zoonotic pathogen *Coxiella burnetii*

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Coxiella burnetii, the causative agent of Q fever, is an emerging zoonotic pathogen infecting humans and various other animals. Its primary reservoirs are ruminants, such as cattle, sheep and goats, in which infection can cause severe reproductive disorders. Outbreaks typically peak during lambing season and are associated with high bacterial loads in birth products, facilitating transmission to herds and farm workers. Despite rare detection after lambing, coxiella reappears annually on farms, due to its environmental stability and immune evasion strategies. The current absence of safe, long-lasting treatments for Q fever outbreaks highlights the need to understand the mechanisms driving coxiella persistence. Placental tissue is hypoxic (≤5% O_2) during early pregnancy, suggesting that oxygen availability may play a role in shaping infection dynamics. We conducted coxiella infection experiments in cell culture models under hypoxic conditions using a hypoxia chamber system. Interestingly, electron microscopy analyses revealed the formation of intracellular spore-like particles (SLPs), previously described as environmentally stable and infectious forms of coxiella. Flow cytometry and fluorescence microscopy experiments confirmed that hypoxia enhances infectivity while reducing the intracellular bacterial load, which is consistent with bacterial dormancy. Preliminary results also suggest that host interferon-induced transmembrane protein 3 plays a role in hypoxia-mediated susceptibility to coxiella infection, by potentially controlling cholesterol homeostasis. Our findings suggest that SLP formation under hypoxia represents a mechanism of intracellular persistence, enabling coxiella to evade host defenses. Increased placental oxygenation later in pregnancy could reactivate these dormant SLPs, driving bacterial replication and tissue invasion, contributing to stillbirth or miscarriage.

Talk #11: JAK/STAT signalling controls proliferation, apoptosis and migration of progenitor cell types in the *Drosophila* airways

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The Janus kinase (JAK) signal transducer and activator of transcription (STAT) pathway is a conserved signalling system that plays a central role in development, repair and the immune response. Deregulation is associated with many chronic lung diseases. A prominent characteristic is altered stem cell activity, resulting in impaired repair mechanisms. In the lung, different pools of basal cells and the alveolar AT2 cells serve as progenitor cells for epithelial repair and homeostasis. The fruit fly Drosophila has become one of the most suitable model systems for studying stem cell properties. The simple organisation, combined with the wealth of genetic tools available, makes Drosophila a perfect model to study the general role of progenitor cell biology in the respiratory organs and the underlying mechanisms of disease. We aimed to understand the functional role of JAK/STAT signalling in tracheal stem cell proliferation, apoptosis and migration in two existing stem cell lineages. We showed that JAK/STAT is active in all progenitor cells and that blockade of JAK/STAT leads to inhibition of cell division and apoptosis of the affected cells. Activation of the JAK/STAT pathway prevented cell proliferation and migration. The effects of JAK/STAT activation varied significantly between progenitor cell regions. Considering the functional conservation of the signalling pathway between humans and Drosophila, our study implicates the crucial role of the JAK/STAT signalling pathway in the cell fate decision of respiratory progenitors and thus in pathological processes as a potential therapy typically observed in chronic lung diseases.

Talk #12: Sex-specific regulation of immunoproteasome function determines response to infection

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Is 'man flu' a myth, or is there a biological kernel of truth to it? Our research dives deep into the cell to investigate the immunoproteasome, a protein complex that degrades viral components within the cell, enabling the immune system to recognize and combat the attack. Its mission is to dismantle viral intruders in a way that allows our CD8+ T cells to recognize them as the enemy and eliminate infected cells.

Our data from the influenza model points towards a clear pattern: immunoproteasome activity is upregulated more strongly in female mice. This heightened expression suggests an enhanced capacity for viral antigen processing and subsequent epitope generation. We hypothesize that this accelerated processing pathway leads to a more effective and timely activation of the virus-specific CD8+ T-cell response. Therefore, the initial magnitude of the immunoproteasome response may be a key determinant influencing the overall efficacy of antiviral immunity in a sex-specific manner, which could be one reason why the 'man flu' isn't entirely a myth…

Talk #13: Dissecting immunoproteasome function in lung regeneration using mouse lung organoids

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Immunoproteasomes (IP) are specialized proteasomes that play critical roles in multiple immune responses. Their three catalytic subunits (PSMB8, PSMB9, PSMB10) are expressed at low levels in non-immune cells but are induced when cells encounter virus infection or upon stimulation with interferons (IFNs) and inflammatory cytokines. It has recently been shown that type I IFN disrupts repair of influenza virus damaged lung epithelial cells. As induction of the IP is part of the canonical response to type I interferon signaling, we here hypothesize that IPs play a role during injury and regeneration of lung.

Single cell RNA sequencing data analysis revealed that the IP is dynamically regulated during lung organoid formation with low baseline expression in alveolar type (AT)2 and AT1 cells that was transiently increasing during AT2 to AT1 differentiation. To validate these data, we established a mouse alveolar organoid system and investigated the role of the IP in lung regeneration. We confirmed upregulation of the IP (PSMB8 and PSMB9) upon AT1 differentiation. Treatment of the organoid at AT2 stage with IFNg impaired organoid formation and strongly stimulated IP expression. Of note, inhibition of the IP using the specific inhibitor LU005i blocked differentiation of AT1 cells from AT2 organoids as revealed by reduced and aberrant AT1 marker gene expression. Our data thus suggest a critical role for the IP in alveolar differentiation. In aging mice, IP triple KO mice showed superior organoid formation efficiency and larger organoid sizes than WT cells at AT2 differentiation conditions implying a role in maintenance of AT2 cell stemness.

In summary, the use of an alveolar organoid culture system allowed detection of the dynamic regulation of the IP and an unexpected function for alveolar differentiation. The significance of IP expression for lung regeneration is currently evaluated in depth.

Talk #14: Synthesis and Structure-Activity Relationships of Novel Amino Acid-Based LpxC-Inhibitors

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With an ever-growing number of antibiotic-resistant bacteria due to the overuse and misuse of common antibiotics, modern medical research must urgently focus on finding new antibiotic classes to tackle this pressing problem [1].

One strategy to accomplish this goal is to target novel bacterial structures that have not previously been addressed by antibiotics. A promising target is the Zn2+-dependent deacetylase LpxC, which catalyzes the first irreversible step of lipid A biosynthesis. Since lipid A is an integral constituent of the outer membrane of all gram-negative bacteria, the inhibition of its biosynthesis is lethal to these organisms. Furthermore, LpxC is a highly conserved, single-copy coded enzyme possessing no mammalian counterpart and is therefore a promising target for the development of gram-negative-selective antibiotics [2].

The aim of the current study was to synthesize novel amino acid-based LpxC inhibitors containing a key 1,4-, 1,5- and 1,4,5-substituted triazole ring, enabling the mild and efficient combination of different terminal and internal alkyne fragments with the core structure exhibiting an azide moiety via transition metal-catalyzed [3+2] cycloadditions [3], yielding a variety of potential inhibitors. These structures were then evaluated for their antibacterial properties as well as their inhibitory activity toward LpxC to deduce structure-activity relationships.

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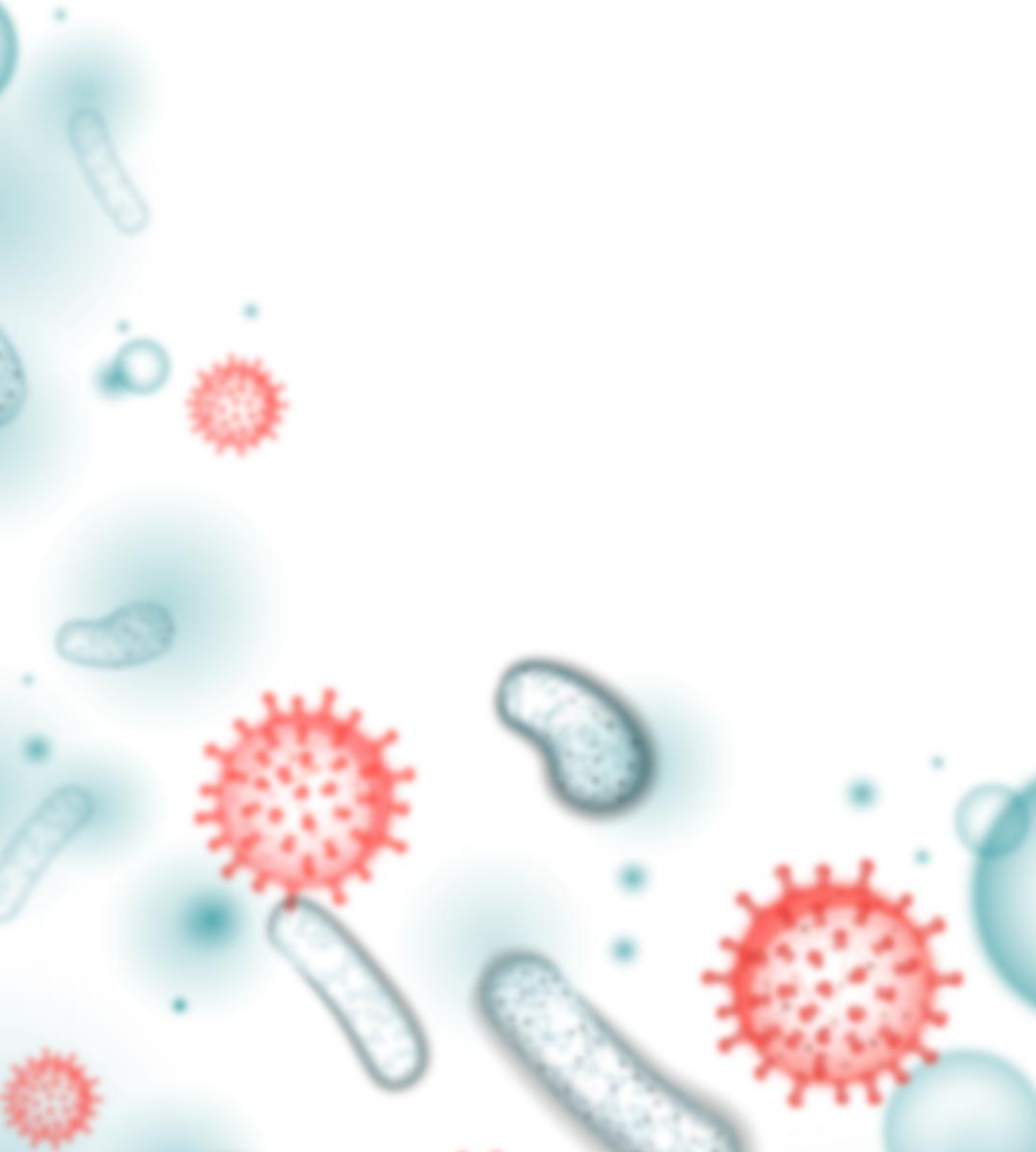
Poster Presentations



P01: Manipulation of VAP-mediated membrane contacts by pathogenic mycobacteria

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Tuberculosis, caused by Mycobacterium tuberculosis (Mtb), remains the deadliest bacterial infectious disease. Once inside macrophages, Mtb resides in a vacuole and manipulates the host lipid metabolism to create a nutrient-rich environment. Mycobacteria utilize outer cell wall protein complexes to take up lipids, such as sterols, which serve as carbon and energy sources. In studying tuberculosis, the Dictyostelium discoideum/Mycobacterium marinum infection model reveals sterol accumulation within the Mycobacterium-containing vacuole (MCV), hinting at a lipid supply route for mycobacteria. Lipid transfer proteins (LTPs) facilitate non-vesicular lipid transport at membrane contact sites (MCS). Tether proteins like the ER-resident (VAMP) associated protein (VAP) stabilize MCS by interacting with proteins containing FFAT motifs. Some pathogens, like Chlamydia, exploit VAP to create lipid pipelines, expanding their vacuoles. Our research identified the recruitment of the LTP oxysterol binding protein 8 (OSBP8) to the infection site with M. marinum due to membrane damage, forming an ER-MCV MCS. We also noted OSBP8 at ER-cytosolic bacteria MCS in later infection stages, possibly due to interactions with the bacterial cell wall. VAP was discovered on the surface of cytosolic mycobacteria, emphasizing its role in MCS formation. We plan to further explore these interactions in the D. discoideum/M. marinum system and infected macrophages.

P02: Host-Driven TB Severity Outweighs Bacterial Load: Insights from GeneXpert and Bandim TB Scoring in Ghanaian Cohorts.

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Tuberculosis (TB) manifests differently across individuals, presenting a dilemma in the control of TB. Precursors that can indicate the trajectory and severity of infection are needed to ensure early treatment measures to reduce morbidity and mortality associated with TB. This study examined how bacterial load and clinical scoring systems can predict the severity of TB disease in diverse patient groups. We collected sputum samples and clinical data from newly diagnosed TB patients across five health facilities in Ghana, categorizing them into three cohorts: TB only, TB with HIV (TB_HIV), and TB with diabetes mellitus (TB_DM). Using the GeneXpert MTB/RIF Ultra Assay, we measured bacterial load through cycle threshold (Ct) values, in conjunction with sputum microscopy, culture time to positivity (TTP), and the Bandim TB scoring system, to assess disease severity. Our findings revealed a significant disease severity across cohorts ($p = 0.0143$), with TB_HIV individuals presenting with more severe disease. Although Ct-values differed across cohorts ($p = 0.0039$), however, when correlated with severity, a weak positive correlation ($r = 0.002193$, $p = 0.9582$) was shown, with an average Ct-value of 20.5. Microscopy amongst the cohorts varied significantly ($p = 0.0289$), with TB_HIV often presenting with scanty or negative smears, indicating lower bacterial loads. TTP differed amongst the groups, with TB_HIV cases showing longer times, suggesting slower bacterial growth. These results show variability in disease presentation amongst different hosts, highlighting that host factors such as HIV coinfection drive TB severity more than bacterial load. This emphasizes the need for more tailored diagnostic and prognostic approaches in the management of TB.

P03: Tunneling Nanotubes (TNTs): An Export/Import Strategy for Chlamydia via Direct Cell-to-Cell Communication

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Chlamydiae are obligate intracellular pathogens causes various diseases in both humans and animals. Earlier research models proposed Chlamydia exit depended on cell lysis, while uptake relied on receptor-mediated interactions. However, in vivo evidence rarely shows host-cell lysis, while in vitro uptake of extracellular chlamydiae is inhibited by physiological concentrations of heparin. Another widely studied exit method of Chlamydia is the extrusion pathway which has been described ex vivo. However, uptake of these extrusion structures appears to be limited to antigen-presenting cells. This suggests additional pathways for chlamydial transmission in tissues during infection. Our studies show that Chlamydia exploits tunneling nanotubes (TNTs) as an alternative and efficient route of cell-to-cell transmission. TNTs are transient, membrane-bound connections that allow intercellular communication and transport of vesicles, proteins and organelles. Recent studies have shown that various pathogens misuse TNTs for stealthy transmission, enabling them to evade the immune system. Infection models using both immortalized cell lines and primary cells demonstrate the transport of chlamydial reticular bodies (RBs) through these structures. Using high-resolution microscopy, flow cytometry and biochemical assays, we identified the inclusion protein IncA forming filamentous structures within TNTs. Our findings showed a close association between the protein IncA and a key host TNT regulator, LST1. Our discoveries elucidate the roles of bacterial and host proteins in TNT-mediated transmission of Chlamydia, highlighting a strategy for immune evasion. Understanding the mechanism opens opportunities for development of novel therapeutic approaches and vaccines against this widespread pathogen.

P04: Synthesis and Structure-Activity Relationships of Novel Proline-Based LpxC Inhibitors

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The ongoing rapid emergence of multidrug-resistant bacteria represents a threat to global health and is associated with high morbidity and mortality. As strains resistant to last-resort antibiotics are already known, the development of novel antibiotic drugs addressing so far unexploited bacterial targets is not only urgent but mandatory.[1,2]

One promising target for the development of such an agent is the Zn2+-dependent deacetylase LpxC. This enzyme is single-copy coded and highly conserved in Gram-negative bacteria. It catalyzes the first irreversible step of lipid A biosynthesis – an essential element of the outer membrane of all Gram-negative bacteria. Thus, the lack of lipid A results in the death of the microorganisms. In addition, LpxC possesses no mammalian counterpart and is not addressed by already established antibiotics, making it a promising target for novel drugs against Gram-negative bacteria.[3]

In this study, novel proline-based LpxC inhibitors were synthesized. These inhibitors were functionalized at the central pyrrolidine nitrogen atom, resulting in a variety of potential inhibitors. The synthesized compounds were tested for inhibitory activity toward LpxC to deduce structure-activity relationships.

Unstructured and structured abstracts are welcomed. Please make sure that the background (including the research question/objective), methods, results, and conclusions are included in the abstract.

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P05: Synergistic effects between membrane active peptides and classical antibiotics on bacterial membranes

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The occurrence of multidrug resistant pathogenic bacteria is increasingly threatening human health globally and may lead to a situation encountered in the preantibiotic era. In addition to the development of antimicrobials against novel targets, the identification of membrane active antimicrobial peptides (AMP) is regarded a valid and promising approach for the treatment of otherwise difficult to treat infections by antimicrobial resistant superbugs. They hold promise as complementary active drugs enhancing conventional antibiotics, yet possess such activity on their own as seen for polymyxins, of which colistin is regarded as one of the last resort antibiotics.

Our current work therefore aims at the identification and analysis of membrane-active peptides whose presence increase the uptake and efficacy of antibiotics. Elucidating their mechanisms of action when used in combination with classical antibiotics and their effect on bacterial membranes may lay the basis for the prediction of novel AMP circumventing resistance mechanisms.

We have therefore established artificial systems employing different membrane models to provide a comprehensive picture of the biophysically complex processes. These include the effects of modified lipopolysaccharide structures (resistant phenotypes) by systematically altering culture growth conditions (e.g. addition of CaCl2 or EDTA) using whole bacteria but also simplified models such as outer membrane vesicles and spheroplasts. In addition, symmetric and asymmetric reconstituted lipid bilayers allow us to identify heterogeneous regions in the membrane critical for peptide dynamics and organization and to tune specific compositions. High resolution techniques such as AFM, cryo-EM/ET and fluorescence microscopy will be used to study specific interactions.

P06: Influence of the biological sex on the immune response to infection with Mycobacterium tuberculosis

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Tuberculosis (TB) remains the most prevalent bacterial infectious disease and the leading cause of death from a single pathogen globally. Epidemiological data consistently demonstrate a higher incidence and mortality in males compared to females, with a male-to-female ratio of approximately 2:1. Both sociocultural (gender) and biological (sex) factors are thought to contribute to this disparity. To specifically address the role of biological sex, we employ murine models, which eliminate many human confounders such as smoking and alcohol consumption. Our results show that disease progression after Mycobacterium tuberculosis (Mtb) infection varies by sex and immune status: In the absence of B and T cells, females are more susceptible, while immunocompetent males are more vulnerable to Mtb infections, reflecting the situation in humans. This highlights how adaptive immunity influences disease progression and immune response in a sex-specific manner.

In immunocompetent animals, increased male susceptibility is linked to reduced B cell follicle formation in the lungs. Although B cells have historically been considered non-essential in TB immunity, emerging evidence highlights their immunomodulatory potential. Beyond antibody production, B cells function as antigen-presenting cells and secrete cytokines that shape the local immune environment. In a recent study, we demonstrated that interleukin 10 (IL-10)-deficient B cells confer enhanced resistance to aerosol Mtb infection. This effect was more pronounced in males, and linked to male-specific immune changes, indicating a sex-specific regulatory function of B cell-derived IL-10 in chronic TB. These findings underscore the importance of incorporating sex as a biological variable in TB research and therapeutic development.

P07: M. avium case study: investigation of genome alterations during resistance development

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Infections due to nontuberculous mycobacteria (NTM) are increasingly recognized worldwide, and one of the most consistently identified NTM species is Mycobacterium avium, which is also among the most prevalent pathogens responsible for NTM pulmonary disease (NTM-PD). Treatment is challenging and requires long-term, multidrug therapy, thereby increasing the risk of developing resistance. In this study, we investigated genomic changes between two M. avium isolates obtained 1.5 years apart from the same NTM-PD patient. Both isolates were whole-genome sequenced (MiSeq, Illumina) at the International Reference Laboratory of Mycobacteriology, Copenhagen, after phenotypic drug susceptibility testing (pDST) indicated emerging resistance to amikacin, clarithromycin, ethionamide, levofloxacin, linezolid, and rifampicin. Sequencing reads were analysed with variant caller tool Snippy to identify single nucleotide polymorphisms (SNPs) in the genomes. Core genomes were mapped to M. avium references downloaded from the NCBI Genbank, identifying the closest relative to be M. avium 104 (NCBI: txid243243) in a phylogenetic analysis. Comparative analysis revealed four SNPs unique to the resistant isolate, located in coaBC, a cutinase family protein, a metal-dependent transcriptional regulator, and an RNA polymerase sigma factor. Subsequently, a resistance prediction with Abricate produced four hits in the CARD database, though these did not overlap with the newly detected SNPs. With this study, we identified four SNPs in an extensively resistant M. avium strain, which could contribute to resistance. This study underscores that NTM resistance remains poorly understood and highlights the need for improved genotypic detection methods, as current prediction tools show substantial limitations.

P08: Liposome-based microfluidic platform for standardised analysis of antimicrobial peptides

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Antimicrobial resistance is a rapidly increasing global concern and has the potential to become a significant public health crisis. Antimicrobial peptides (AMPs) offer a promising alternative to conventional antibiotics. We utilized a genetic algorithm to systematically design, rank, and optimise a series of synthetic AMPs targeting specific bacterial strains.

In our study, we investigate the membrane-lytic activity of the top-ranked peptides using a microfluidic platform. The platform allows individual immobilisation of hundreds of liposomes for real-time observation of AMP–liposome interactions.

We generate fluorophore-filled liposomes in a controlled manner using octanol-assisted liposome assembly. The lipid composition mimics bacterial membranes. After trapping the liposomes and injecting peptides, we observe and analyse the dynamics of fluorescence decrease in a classical dye release assay with single-liposome resolution.

With this approach, we (a) evaluate the permeabilising efficacy of AMPs, (b) classify their mode of action in combination with other methods, and (c) use the microfluidic platform to optimise standardised and routine compound testing. We compare our results with microbiologically determined fitness values of AMPs.

P09: Investigation of the structural diversity of lipoteichoic acids in Streptococcus suis

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Streptococcus suis is an important porcine pathogen associated with meningitis, sepsis, arthritis, and endocarditis. Additionally, *S. suis* has a high zoonotic potential and causes hundreds of human infections yearly.[1] *S. suis* strains are classified into 29 serotypes based on their capsular polysaccharide[2] and belong to numerous sequence types (STs).[3] Lipoteichoic acid (LTA) have been described as a major molecule of the *S. suis* cell wall and its contribution to virulence has been suggested .[3,4,5]

Previous structural studies focused on the LTA of serotype 2, as this is the predominant serotype worldwide. A deviating structure was found in strains P1/7 (ST1, from Europe) and SC84 (ST7, from China) compared to strain 89-1591 (ST25, from Canada).[4] The investigated strains produced two distinct types of LTA, a type I LTA with a poly-glycerol phosphate chain, and a second, more complex LTA containing glycosylated repeating units in addition. However, the glycosylation pattern of this second LTA type was different in the ST25 strain compared to the other two strains.[4]

The aim of the presented study is to explore LTA structures in other serotypes on the molecular level. We structurally characterize LTA from six selected *S. suis* strains belonging to the serotype 1 (ST1), 1/2 (ST1), 1/2 (ST28), 7 (ST29), 9 (ST16), and 14 (ST6). These findings will expand our current knowledge of the diversity of LTA structures in *S. suis*, especially since many of the LTA biosynthesis genes in *S. suis* are still elusive to date, so far precluding a genome-based investigation of this.

Acknowledgements:

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P10: Reprogramming of human macrophages in response to Mycobacterium tuberculosis complex (MTBC) infection

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Tuberculosis (TB), caused by Mycobacterium tuberculosis complex (MTBC) bacteria, continues to pose a significant global health threat. MTBC employs various strategies to persist in the human host, including epigenetic reprogramming through altered gene expression and potential DNA methylation changes in innate immune cells.

In this study, we analyse host gene expression changes in response to MTBC infection using an early-stage human macrophage model. We focus on how distinct MTBC lineages (Lineage 4 and Lineage 6) drive specific gene expression shifts in host cells and relate these changes to potential DNA methylation changes. Eventually, by correlating our in vitro findings with epigenetic profiles from patient-derived peripheral blood mononuclear cells (PBMCs), we aim to uncover lineage-specific host immune signatures linked to TB progression. Our results might offer new insights into how MTBC modulates the host immune response through epigenetic mechanisms, advancing our understanding of TB pathophysiology and host-pathogen interactions

P11: Exploring lineage effects in "TB Sequel 1" data at time of Tuberculosis diagnosis

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Background: Tuberculosis (TB) disease severity at diagnosis may be influenced by host factors, comorbidities, and Mycobacterium tuberculosis complex (Mtbc) lineage. Understanding these associations is essential for guiding prognosis and identifying high-risk groups.

Methods: Whole-genome sequencing and clinical data from 862 patients enrolled in the *TB Sequel 1* study were analysed to identify baseline predictors of TB disease severity, with a primary focus on the potential role of Mtbc lineage. A Bayesian multivariate linear model was used to assess associations between severity and predictors, including Mtbc lineage, age, sex, body mass index (BMI) category, HIV status, and study site.

Results: Mtbc lineage and sub-lineage distributions varied substantially across the four study sites (Gambia, Tanzania, Mozambique, South Africa). In the full cohort model, underweight status was associated with significantly increased severity, and marked differences were observed between study sites. Site-specific models indicated variable influence of risk factors, but no consistent or statistically significant effect of Mtbc lineage on severity profiles.

Conclusion: Baseline TB disease severity was primarily associated with host factors, particularly underweight status and study site, suggesting local or unmeasured influences. Mtbc lineage did not show a consistent effect. It is important to note that these findings reflect baseline assessments only. The ultimate goal is to evaluate long-term outcomes and disease trajectories, which will be analysed in subsequent phases of the study.

P12: Novel Insights into the Chemistry of TolC-binding Compounds

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Tripartite efflux pumps are integral membrane complexes that convey antimicrobial resistance in Gram-negative bacteria by extruding antibiotics. Targeting efflux systems with inhibitory small molecules represents a promising strategy for restoring antibiotic susceptibility in multidrug-resistant bacteria. However, no efflux pump inhibitors have been approved so far. In *Escherichia coli*, the AcrAB-TolC efflux pump is responsible for the extrusion of a broad set of antibiotics. Except for the outer membrane factor TolC, several inhibitory small molecules have been identified targeting the periplasmic adapter protein AcrA and the efflux pump AcrB [1]. Due to its role in the assembly of many different efflux pumps in *E. coli*, blocking TolC should lead to efficient efflux inhibition [2].

In this work, we employed a microbiological high-throughput screen using an in-house repurposing library and biophysical validation to identify efflux pump inhibitors targeting TolC. We discovered a cytostatic drug that potentiates linezolid in a *tolC*-dependent manner. Subsequently, we characterized analogues of this drug in spectral shift assays and identified crucial structural features for TolC-binding. With this information we plan to identify analogues with higher or equal binding affinity to TolC, while decreasing their cytostatic potential.

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P13: Synthesis and Structure-Activity Relationships of Novel Triazole-Based LpxC Inhibitors

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Fighting multi-drug resistant Gram-negative bacteria appears to become one of the biggest challenges in a post-antibiotic era, with structurally novel antibiotic drugs getting significantly scarcer.

The bacterial Zn^{2+}-dependent deacetylase LpxC is a promising target for new antibiotics, as it is highly conserved in different Gram-negative bacterial species and shows no sequence homology to any human enzyme. LpxC catalyzes the first committed step in the biosynthetic pathway of Lipid A, which is the membrane anchor for the lipopolysaccharides in the outer membrane of Gram-negative bacteria. Thus, the inhibition of LpxC leads to a dysfunctional cell wall, and is therefore lethal for Gram-negative bacteria.[1]

Building upon the structure of potent LpxC inhibitors such as CHIR-090 (**1**), the main goal of this project is to synthesize novel LpxC inhibitors bearing a triazole-based fragment addressing the UDP-binding pocket of the target enzyme. As most of the already described LpxC inhibitors leave the UDP-binding pocket unoccupied, is it is of particular interest to further explore this site. The presented novel triazole-based LpxC inhibitors were synthesized via CuAAC reactions.[2,3]

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P14: Hypoxic persistence of spore-like particles (SLPs) of the zoonotic pathogen *Coxiella burnetii*

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Coxiella burnetii, the causative agent of Q fever, is an emerging zoonotic pathogen infecting humans and various other animals. Its primary reservoirs are ruminants, such as cattle, sheep and goats, in which infection can cause severe reproductive disorders. Outbreaks typically peak during lambing season and are associated with high bacterial loads in birth products, facilitating transmission to herds and farm workers. Despite rare detection after lambing, coxiella reappears annually on farms, due to its environmental stability and immune evasion strategies. The current absence of safe, long-lasting treatments for Q fever outbreaks highlights the need to understand the mechanisms driving coxiella persistence. Placental tissue is hypoxic (≤5% O_2) during early pregnancy, suggesting that oxygen availability may play a role in shaping infection dynamics. We conducted coxiella infection experiments in cell culture models under hypoxic conditions using a hypoxia chamber system. Interestingly, electron microscopy analyses revealed the formation of intracellular spore-like particles (SLPs), previously described as environmentally stable and infectious forms of coxiella. Flow cytometry and fluorescence microscopy experiments confirmed that hypoxia enhances infectivity while reducing the intracellular bacterial load, which is consistent with bacterial dormancy. Preliminary results also suggest that host interferon-induced transmembrane protein 3 plays a role in hypoxia-mediated susceptibility to coxiella infection, by potentially controlling cholesterol homeostasis. Our findings suggest that SLP formation under hypoxia represents a mechanism of intracellular persistence, enabling coxiella to evade host defenses. Increased placental oxygenation later in pregnancy could reactivate these dormant SLPs, driving bacterial replication and tissue invasion, contributing to stillbirth or miscarriage.

P15: TRIP6 as a host target of *Cryptosporidium* parasites in intestinal pathology

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Cryptosporidiosis is a leading cause of diarrheal disease and infant mortality in the Global South, yet effective therapeutic strategies to combat the disease are lacking. Using single-cell RNA sequencing analysis in a neonatal mouse model revealed a dynamic regulation of the NF-κB pathway during *Cryptosporidium parvum* infection. We hypothesize that disruption of this pathway is linked to the interaction of a novel parasite dense granule protein and the host adherens junction-associacted protein TRIP6, which can shuttle to the nucleus and interact with NF-kb. To dissect this interaction, we generated TRIP6 knockdown and overexpression Caco-2 cell lines using CRISPR interference and the Sleeping Beauty transposon system. We also established a suitable *in vitro* infection model with Caco-2 cells expressing a Gaussia luciferase NF-kB reporter, enabling time sensitive monitoring of host responses in the presence or absence of TRIP6. By uncovering how *Cryptosporidium* parasites manipulate host signaling and junctional proteins, this work aims to identify the molecular basis of the severe intestinal pathology and therapeutic targets to reduce the burden of cryptosporidiosis in children.

P16: Structural Validation and Characterization of Herpesviral Amyloids

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Viral infections such as SARS-CoV-2, HCV, herpesviruses, HIV, and Influenza A are increasingly linked to amyloid-associated diseases, including Alzheimer's disease (AD) and other neurodegenerative disorders. Among them, herpesviruses are strongly implicated in promoting neuroinflammation, amyloid-β accumulation, and neuronal dysfunction through alterations in synaptic protein expression and dendritic architecture (1). These viruses persist latently in neural tissues and can reactivate under stress or immune decline, generating chronic inflammatory states that may accelerate AD pathogenesis. Amyloids, traditionally regarded as pathological aggregates, are now recognized as multifunctional molecules, including antimicrobial agents in innate immunity. Several herpesviral proteins exhibit high amyloidogenic propensity; however, their ability to cross-seed human amyloidogenic proteins and contribute to fibril formation remains poorly understood (2). While extensive research has focused on human amyloids, studies on viral amyloid structures and mechanisms remain scarce. To address this gap, we employed the HerpesFold computational toolbox to predict amyloid-like folds in herpesviral proteins. Candidate proteins were purified through a two-step process and subjected to structural and biophysical characterization of fibrillation. Further analysis will involve cross-seeding assays and *in cellulo* experiments using live-cell microscopy, correlative light and electron microscopy (CLEM), and cryo-electron tomography (cryo-ET). This integrated approach will provide insights into viral amyloid formation both in vitro and in situ, elucidating their potential contribution to neurodegeneration and clarifying the molecular interplay between viral infection and amyloid-driven pathology.

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P17: Deciphering the Lipidome of M. tuberculosis: Culture-Dependent Variations in Virulence Lipids and Membrane Components

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The infectious disease Tuberculosis (TB), caused by *Mycobacterium tuberculosis* (*Mtb*), has afflicted humans for thousands of years and continues to be a major health emergency, causing over a million deaths annually. The fact that approximately one-third of the world's population is infected with *Mtb* highlights the pathogen's well-adapted interaction with its host. Especially *Mtb*-specific lipids of the outer mycobacterial membrane (MOM) play a direct role in intracellular survival, host-pathogen interaction and virulence[1,2]. Phthiocerol dimycocerosates (PDIMs) are the essential virulence lipids and known for its role in the pathogenicity of the bacterium[3]. The loss of PDIM in *Mtb* cultured in vitro is a well-known phenomenon and poses a challenge in research, as PDIM-deficient mutants exhibit reduced virulence and can distort experimental outcomes[4]. To overcome this problem, the supplementation of propionate or vitamin B_{12} during bacterial growth in vitro enhances the production of PDIMs[5].

In this study, we analysed a genetically well-defined *Mtb* strain under growth conditions with and without propionate as a PDIM enhancing additive to the standard growth media. In this context, it is of particular interest whether the supplementation influence the overall lipidome and/or lead to the formation of additional PDIM species. In this perspective we adapted our shotgun lipidomics workflow to analyse also PDIMs. This included adaptation on the data acquisition strategy using the Q Exactive Plus Hybrid Quadrupole-Orbitrap mass spectrometer (Thermo Scientific) and in-depth analysis of the tandem mass spectrometric fragmentation pathways. Finally, we automated identification routines using LipidXplorer. We additionally determined the lipid profiles of the pathogen including phosphatidyl-*myo*-inositols (PI), lyso-phosphatidyl-myo-inositols (LPI), cardiolipins (CL), phosphatidylethanolamine (PE), lyso-

phosphatidylethanolamine (LPE), phosphatidylglycerol (PG), (lyso-)phosphatidylglycerol (LPG) and phosphatidyl-*myo*-inositiol mannosides (PIMs), as well as neutral lipids. It is also of interest, whether *Mtb* grown with propionate do have a higher growth rate and therefore an infection advantage.

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P18: Lipocalin-2 is secreted by primary bronchial epithelial cells in response to viral infections

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Background: Cigarette smoke (CS) is a major risk factor for chronic obstructive pulmonary disease (COPD). Viral infections are more frequent and severe in smokers. Our previous findings showed that lipocalin-2 (LCN2), a mediator of innate immune defence, was strongly upregulated in both murine cell-free BALF upon smoking and in lung tissue from COPD patients. Here we aimed to study whether LCN2 is secreted by airway epithelium in response to CS exposure and viral infection.

Methods: Primary bronchial epithelial cells from normal (NHBE) and COPD (CHBE) donors were grown at the air-liquid interface and exposed to smoke (1R6F at 1 puff/min, ExpoCube) for 14 days, followed by infection with Human Rhinovirus (HRV-16) for 4 hours. Analysis was performed 24 hours post-infection.

Results: HRV-16 infection led to a significant LCN2 release in both NHBE and DHBE cells. Moreover, we found that CS further exacerbated LCN2 levels from CHBE, compared with air-exposed controls. In contrast, no significant difference in LCN2 release was observed between CS-exposed and air-exposed NHBE cells after HRV-16 infection.

Conclusions: Together, our data suggest that HRV-16 infection induces LCN2 secretion from airway epithelium. Also, we found that CS exacerbate LCN2 secretion in HRV-infected CHBE cells, potentially contributing to COPD progression and exacerbation. Moving forward, we aim to investigate the role of the epithelial-derived LCN2 and its potential as biomarker for COPD.

P19: Investigation of a putative Fic/DOC toxin-antitoxin system in Staphylococcus epidermidis: Importance for biofilm formation and infection

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Staphylococcus epidermidis is a nosocomial human pathogen often linked to infections of medical implants such as prosthetic joints and intravenous catheters.

Its invasive potential correlates with specific lineages, such as sequence type 2, which uniquely harbors a chromosomally integrated SPβ-like prophage1 encoding a putative type II toxin–antitoxin (TA) system. The toxin contains a Fic domain, a catalytic module known to mediate post-translational modifications such as AMPylation and phosphorylation, often contributing to virulence. The precise function of this Fic domain—and consequently of the TA system—remains unknown.

Here we combine biochemical, bioinformatic, and microbiological approaches to investigate its structure, function, and role in infection.

Confocal microscopy showed that overexpression of toxin and antitoxin enhances biofilm formation. In clinical isolates, qPCR revealed low TA expression during growth but strong induction under antibiotic and mitomycin C treatment as well as phage exposure. A GFP reporter indicated activation in ~15% of cells under basal conditions, rising to 75% under stress conditions.

In silico screening of the bacterial proteome with an AlphaFold-based pulldown pipeline identified potential toxin substrates. Functional assays confirmed the toxin phosphorylates a key protein biosynthesis factor using ATP as co-substrate. Computational analysis of the TA complex, via FoldSeek and surface charge calculations, suggested a dimerization/DNA-binding domain in the antitoxin. Size-exclusion chromatography revealed concentration-dependent dimerization of TA complexes, absent when the domain was removed.

These findings suggest the TA system mediates stress adaptation by modulating translation and may contribute to *S. epidermidis* pathogenicity in implant-associated infections.

P20: Functional Amyloid Formation in Staphylococcal Hemolytic Peptides

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While amyloids are classically associated with neurodegenerative diseases, in bacteria, they can serve adaptive roles, such as providing structural support and stabilizing biofilms, leading to enhanced virulence. Here, we investigate whether hemolytic peptides from diverse Staphylococcus species form amyloid-like fibrils and how this conformational change might contribute to bacterial pathogenicity. Building on previous findings that Staphylococcus aureus δ-toxin forms amyloid fibrils in vitro, we hypothesized that amyloidogenesis could be a conserved feature among staphylococcal hemolytic peptides. We screened 20 naturally occurring peptide variants from six Staphylococcus species using transmission electron microscopy and amyloid-binding dyes, and found that a subset of seven peptides formed fibrillar aggregates, suggesting that this structural property is both widespread and potentially regulated. We will also compare the cytotoxic effects and hemolytic activity of the fibrillar aggregates versus their monomeric counterparts to assess how amyloid formation influences peptide function. This comparative study aims to explore the functional consequences of amyloid formation during infection. Previous research on bacterial amyloids indicates that fibrillar assembly may increase peptide stability against proteolytic cleavage or slows it down and contribute to biofilm resilience. Pathogen-derived amyloid fibrils have also been shown to modulate host immune responses. In addition, bacterial amyloids have the potential to influence host processes by interacting with human amyloidogenic proteins, raising the possibility of cross-seeding events that could link infection to chronic inflammatory or neurodegenerative conditions. This work contributes to a broader understanding of bacterial pathogenesis and highlights the possible intersection between microbial infection, immune activation, and long-term host pathology.

P21: Precision Utilizing Lung Multi-Omics (PULMO) –A data-driven approach to identify epigenetic biosignatures of lung disease

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In cancer research, epigenetic changes are increasingly elucidated to inform development of new therapies and diagnostic tools. However, today there are no generalized tool that differentiate lung diseases. This project aims to leverage DNA methylation (DNAm) data for detecting serious respiratory conditions like tuberculosis (TB) and lung cancer through a data-driven approach. The specific aims are to i) To identify disease signatures in DNAm data from airway samples of TB and lung cancer patients. ii) To elucidate unique DNAm patterns of airway macrophages from healthy subjects and patients with lung cancer or TB and iii) To develop an DNAm-informed XAI disease classifier algorithm for early diagnosis of patients with TB or lung cancer. Lung diseases are a leading cause of morbidity and mortality worldwide. TB and lung cancer present similar symptoms, complicating diagnosis and often requiring invasive procedures. Misdiagnosis i leads to treatment delays and unfavourable outcomes, both in low- and high-resource settings. This novel approach to lung disease classifiers has the potential to benefit patients globally. This project addresses this need by integrating multi-omics data (genomics, microbiomics, and DNAm) with clinical evaluations and patient-reported symptoms using XAI models. Developing a precision health tool based on this, yet unexplored approach, has the potential to diagnose serious lung disease while also breaking boundaries for applications in other fields.

P22: Refining HLA Divergence Metrics Through Functional Sites

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The Human Leukocyte Antigen (HLA) system is a keystone of adaptive immunity, characterized by extreme allelic polymorphism. Quantifying genetic divergence within this system is central not only to understanding disease associations and transplant compatibility but also to elucidating evolutionary pressures that have shaped immune recognition and pathogen defense. The Human Leukocyte Antigen Evolutionary Divergence (HED) Index, building on the divergent allele advantage framework and formalized by Pierini & Lenz (2018) [1], quantifies divergence using Grantham distances within peptide-binding domains and correlates with breadth of predicted peptide binding. However conventional implementations relying on full-length sequences of these domains may obscure functionally critical variation. We propose an improved HED framework that emphasizes functionally informative residues. Grantham distances were computed from sequence alignments limited to specific residue subsets defined by structural and evolutionary criteria. The number of predicted HLA-bound peptides was then derived with the software *NetMHCpan/NetMHCIIpan*, which estimate peptide–MHC binding affinities in silico, from a pathogen-derived peptide dataset [1] to evaluate the functional relevance of the refined divergence metric. Comparisons reveal that restricting divergence calculations to targeted subsets increases sensitivity relative to full-sequence methods. This focused HED approach thus captures functionally relevant variation more effectively and provides a more precise measure of HLA divergence. It has methodological implications for evolutionary studies and supports improved frameworks in disease-association research and transplant compatibility assessment.

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P23: Regulation of the proteasome system in response to mitochondrial import stress

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Mitochondria produce the bulk of cellular ATP and are involved in the metabolism of, e.g., amino acids, lipids and iron-sulfur clusters. 99% of the mitochondrial proteins are nucleus-encoded and synthesized at cytosolic ribosomes. Mitochondrial precursor proteins are then transported through the outer mitochondrial membrane via the TOM complex and through the inner membrane via the TIM23 complex. Impaired protein import causes acute mitochondrial stress with stalled, misfolded and mislocalized mitochondrial proteins accumulating. Degradation of these proteins by the proteasome system and adaptation of cytosolic protein translation machinery are directly coupled and essential to maintain cellular viability. However, how these two processes are regulated and connected with each other is poorly understood in the human system.

We previously observed that inhibition of the TIM23 import complex led to the accumulation of mitochondrial precursor proteins, upregulation of ribosomal and proteasomal proteins. Among them was the proteasomal activator PA28αβ which is known to upregulate proteasomal protein degradation. We are currently investigating the effect of stendomycin, a small lipopeptide that inhibits import through TIM23, on the regulation of the proteasome system by this import stress. For that, human HEK293T cells are treated with stendomycin in a kinetic analysis (0h, 1h ,2h, 5h, 16h, 24h and 48h) and changes in the proteasome system are determined using expression, proteasome activity and composition analyses. This analysis will be extended to import stress due to clogging of the outer mitochondrial membrane complex TOM and accompanied by detailed anaylsis of mitochondrial cell fractions. Furthermore, we aim to pull down the 20S proteasome after stendomycin treatment and subsequent cross-linking followed by interactome analysis by mass spectrometry (MS). This will provide us with additional information on the interacting proteins and the interaction with the mitochondrial import machinery.

The aim of this research is to deepen our understanding on the proteasome regulation upon mitochondria dysregulation. The project is part of a collaborative DFG-funded project and we

will closely interact with Sven Dennerlein' lab from the University Göttingen on import stress and proteasome staining, and Bettina Warscheid's group at the University Würzburg on advanced MS analysis.

P24: Female preconception vaping of nicotine induces severe airway remodeling in their offspring via progenitor cell depletion in D. melanogaster

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Background: Adolescent vaping raises concerns that it may induce respiratory health risks in offspring conceived years later. Using Drosophila melanogaster, we aimed to explore how female preconception nicotine exposure affects the offspring airway architecture and identify underlying molecular pathways. We further investigated the function of airway progenitor cells.

Methods: Virgin female flies were exposed to vaporized nicotine (e-nicotine) or water (controls). F1 larval tracheae were assessed for structural defects, epithelial remodelling and their mRNA sequenced. Airway stem cell niches (tracheoblasts) were quantified and their proliferative capacity assessed by testing pH3.

Results: E-nicotine exposure of future mothers induced severe airway malformations in F1 offspring, with shortened tracheae (segment Tr9: $p<0.001$), increased tracheal deformity rates and thickened epithelia ($p<0.0001$). Tracheoblast areas in T4/T5 airway segments were reduced by 35% ($p<0.0001$), with lower capacity to proliferate (pH3+ cells). mRNA sequencing of isolated airways identified significant downregulation of Blimp-1 (stem cell regulator), methuselah-like 10 (ECM homeostasis receptor), and Relish (immune-epithelial signaling factor).

Conclusion: Maternal preconception e-nicotine exposure triggers airway remodeling, stem cell depletion, and dysregulation of Blimp-1/mthl10/Rel critical for airway development and epithelial signaling in Drosophila. These genes may drive the airway maldevelopment induced by their mothers earlier “vaping”. Ongoing studies focus on mRNA sequencing of T4/T5 tracheoblast niches to uncover deeper molecular mechanisms linking maternal e-nicotine exposure to impaired airway development in offspring. Our findings highlight the potential intergenerational harms of adolescent vaping which warrants further investigations in humans.

P25: Impact of Mycobacterium tuberculosis complex strain diversity on tuberculosis transmissions in a cosmopolitan low-incidence setting: a longitudinal population-based molecular epidemiological study

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Some of the *Mycobacterium tuberculosis* complex (Mtbc) phylogenetic lineages, such as L2 and L4 are widespread (generalists) while others, such as L1, L3, and L5-L10, are regionally restricted (specialists). These differences suggest variations in virulence and transmissibility across host populations. A key question is whether more virulent generalist strains can cause larger outbreaks or alter population structures when introduced into low-incidence tuberculosis (TB) settings.

To explore this, we performed whole genome sequencing (WGS) on 3,131 Mtbc strains collected from patients with TB in Hamburg, Germany, over 25-years (1997-2021). We analysed the population structure and transmission dynamics in relation to patients' self-reported geographical origins.

Infections with strains of the major Mtbc lineages L1-6 and *M. bovis* were detected, with L4 strains being the most prevalent (74.6 %, n = 2,337). Over time, the lineage distribution shifted: L4 strains decreased from 83.5 % in the first five years to 63.0 % in the last five years, while L3 strains increased from 4.8 % to 18.1 %. These changes correlated with an increasing number of foreign-born patients rather than altered transmission. L4 strains accounted for the majority of clusters (81 %, 218/269). In-depth analysis revealed a differential potential of

L4 sublineage strains to be transmitted within the local population, highlighting the importance of considering sublineages.

Migration influences the population structure of Mtbc in low-incidence settings. However, this is not driven by the increased transmission of introduced generalist strains such as L2.

P26: Lineage-dependent rifampicin tolerance in Mycobacterium tuberculosis complex strains

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Treatment failure remains a critical challenge in the global fight against tuberculosis (TB), leading not only to poor clinical outcomes but also to continued disease transmission. While antibiotic resistance has long been recognised as a major contributor, increasing evidence points to drug tolerance as an additional, underappreciated factor. Drug-tolerant Mycobacterium tuberculosis complex (MTBC) bacteria exhibit prolonged survival during antibiotic treatment. We hypothesise that tolerance mechanisms are greater in strains from certain lineages (or sub-lineages), which may explain variations in antibiotic resistance rates as well as epidemiological success.

In this study, we examine the tolerance of 12 strains representing MTBC lineages 2.2.1 (Beijing Central Asia) and 4.1.2.1 (Euro American Haarlem) plus H37Rv, using time-kill assays with increasing concentrations of rifampicin. Our results show that strains of Lineage 4 exhibit slower drug killing, thus greater tolerance as compared to Lineage 2 strains and H37Rv. This suggests that Lineage 4 strains employ a tolerance-based survival strategy, as opposed to the resistance-based strategy observed in Lineage 2 strains.

These findings underscore the need to integrate lineage-specific tolerance profiles into TB treatment strategies and diagnostics.

P27: ”Seeing is revealing”: Lipid Droplet Dynamics During Mycobacterial Infection

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Tuberculosis remains the leading cause of death from a single infectious agent, *Mycobacterium tuberculosis* (*Mtb*). *Mtb* reprograms host lipid metabolic pathways to establish an optimal niche for its survival and replication. Notably, *Mtb* has perfected many ways to utilize host fatty acids (released from lipid droplets (LDs)) and sterols as carbon and energy source to support its persistence within host cells. However, the underlying molecular mechanisms by which mycobacteria exploit host LDs remain poorly understood. To address this, we employ *M. marinum*-infected human macrophages and *Dictyostelium discoideum* as complementary model systems, along with advanced imaging approaches such as live cell imaging, transmission electron microscopy and correlative imaging strategies. In both *D. discoideum* and macrophages, LDs interact with the *Mycobacterium*-containing vacuole (MCV) during early stages of infection, subsequently translocating into the MCV lumen. Strikingly, we have identified a host protein involved in lipophagy that is recruited to the MCV during infection. We are currently investigating its role in lipid droplet turnover and its potential impact on intracellular mycobacterial survival through genetic perturbation and quantitative imaging. To visualize these organelle interactions and membrane dynamics at high resolution, we perform cryo-correlative light and electron microscopy (cryo-CLEM) combined with electron tomography. By elucidating how pathogenic mycobacteria manipulate host lipid metabolism, our work aims to uncover fundamental host-pathogen interactions and reveal new avenues for therapeutic intervention.

P28: Investigation of *Listeria monocytogenes* colonizing corn salad

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Listeria monocytogenes is one of the most important foodborne pathogens due to its high hospitalization and mortality rates. While meat, fish, and dairy products are recognized as common sources, leafy greens such as corn salad have also been identified as vectors. A major concern is the pathogen's ability to attach to plant surfaces and form biofilms, which allows it to persist over time. This study aimed to investigate the attachment, colonization, and biofilm formation of *L. monocytogenes* on corn salad and to identify the genes involved in these processes. Three isolates, two of plant origin and one clinical, were characterized by whole-genome sequencing. Biofilm formation was quantified using crystal violet assays in different media (BHI, LB, and corn salad medium), revealing peak biofilm development after 12–14 h.

L. monocytogenes strains were fluorescently labelled using a chromosomally integrated plasmid vector. Colonization and biofilm formation on corn salad leaves were analysed using confocal laser scanning microscopy (CLSM) and scanning electron microscopy (SEM). CLSM and SEM revealed that *L. monocytogenes* predominantly colonizes the stomata of corn salad leaves. Dense clusters and string-like structures were observed in the stomatal regions, while microcolony and biofilm formation was also detected on surrounding surfaces. Three-dimensional CLSM imaging of the GFP-expressing strain, combined with quantitative analysis using the "BiofilmQ" software, showed that the architectural characteristics of the biofilm differed depending on the growth medium. These findings suggest that stomatal colonization may offer *L. monocytogenes* protection from external stressors, posing a specific challenge to food safety.

P29: Exploring possible quantum coherence in vault particles and its contribution to host infection defence

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This project aims to explore possible quantum coherence of vault particles and their role in cellular defense against mycobacterial infections. *Dictyostelium,* a unicellular model organism for phagocyte function, expresses highly conserved vault particles, which we have shown to be upregulated during mycobacterial infection. The high symmetry and organized tryptophan networks of vault particles suggest that, like recently demonstrated for microtubules, these structures may exhibit quantum coherence at ambient temperature. The aims of the project are 1) to study quantum coherence and superradiance in purified vault particles, 2) to assess whether vault particles contribute to *Dictyostelium* defense against mycobacteria, and 3) to investigate a possible link between superradiance and anti-bacterial defense. We will measure quantum yield of purified Dictyostelium-derived vaults as well as study the role of vaults in *Dictyostelium* anti-mycobacterial defense. By systematically acquiring data from the described systems, we aim to determine whether vaults use quantum principles to interfere with mycobacterial survival, making the bacteria more vulnerable to host cell clearance mechanisms. The project thereby pioneers the study of quantum mechanism in host antimicrobial defence.

P30: Fate Amenable To Change: Chameleon Characteristics of Antimicrobial Peptides from Wasp Venom and Frog Skin

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Bacterial infections remain a critical global health threat, further aggravated by the rapid spread of antimicrobial resistance. Rising mortality rates and treatment failures emphasise the urgent need for alternative therapeutic strategies grounded in fundamental molecular research. Among others, antimicrobial peptides (AMPs) emerged as a particularly promising class of molecules, given their potent biological activity.

To further expand our understanding of the AMPs structural diversity, we investigated two natural AMPs: Aurein 3.3, derived from the frog skin (*Ranoidea raniformis)*, and Polybia-CP, isolated from the venom of wasp *Polybia paulista*. Using Cryo Electron Microscopy and X-Ray crystallography, we studied structures of the peptides and observed that both peptides display unusual structural plasticity. Specifically, each of the peptides is capable of adopting two distinct supramolecular organisations - α-helical assemblies and canonical amyloid-like fibrils composed of stacked β-sheets. We also evaluated an antimicrobial potency of the AMPs (MIC assay), cytotoxicity toward mammalian cells (LC50, LDH Assay) and interactions of the peptides with bacterials membranes (TEM).

Our findings expand the repertoire of peptides exhibiting secondary-structure switching behaviour and highlights the potential of the AMPs for the design of peptide-based antimicrobials with improved stability, selectivity, and bioavailability. The AMP-based drugs could serve as valuable candidates in the development of innovative therapies against multidrug-resistant pathogens.

P31: Keratinocyte-targeting functional autoantibodies in systemic sclerosis are associated with pulmonary fibrosis

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Objective: Systemic sclerosis (SSc) is a chronic autoimmune connective tissue disease characterized by multiple organ involvement and heterogenous clinical manifestations. Pulmonary fibrosis is a common complication and a major cause of SSc-related mortality. In addition to autoantibodies against nuclear antigens, SSc patients produce autoantibodies targeting cell surface receptors, which not only serve as biomarkers but also play an active role in disease pathogenesis. This study aimed to identify novel functional autoantibodies that may contribute to the development of SSc.

Methods: Serum samples from SSc patients and healthy controls were analyzed for IgG binding to the surface of normal human epidermal keratinocytes (NHEK) using the On-Cell Western assay. Associations between anti-NHEK IgG levels, SSc susceptibility, and clinical manifestations, were assessed using GraphPad Prism software. Functional effects of these autoantibodies were evaluated using cytokine array analysis.

Results: IgG autoantibodies against NHEKs were detected in human sera, with significantly higher levels in SSc patients compared to healthy controls. Notably, SSc patients with pulmonary fibrosis exhibited higher levels of these autoantibodies than those without, suggesting a potential link to fibrotic progression. Moreover, stimulation of NHEKs with purified IgG isolated from anti-NHEK positive SSc sera induced a marked increase in the release of pro-fibrotic cytokines such as PDGF-BB.

Conclusion: These findings support the presence of functional autoantibodies in SSc that target keratinocytes and induce the secretion of pro-fibrotic mediators. This mechanism may contribute to fibroblast activation, myofibroblast differentiation, and the development of fibrosis in SSc.

P32: The immunoproteasome as a prognostic biomarker for NSCLC progression

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Non-small cell lung cancer (NSCLC) is the most common subtype of lung cancer, representing one of the deadliest forms of malignant tumors. Immune checkpoint therapies that aim at the reactivation of exhausted anti-tumor CD8 T cells represent a promising treatment strategy for NSCLC patients. The immunoproteasome plays a crucial role in CD8+ T cell activation as it generates MHC class I antigen presented to CD8+ T cells. In tumors, RNA signatures related to the overexpression of the immunoproteasome positively associate with the response of cancer cells to immune checkpoint blockade. Protein expression data and spatial information on immunoproteasome and CD8+ T cell interaction, however, is missing.

Here, we analyzed early-stage NSCLC tissue using multiplex immunofluorescence (mIF) for determination of the spatial characteristics of the immunoproteasome and T cells to correlate them with patient tumor subtypes, disease severity and progression. For that, we first optimized a panel of markers for the immunoproteasome, CD8 T cells and cytokeratin to identify CD8 T cells and the immunoproteasome in tumor and stroma areas. In addition, we used PD-1 to identify exhausted CD8 T cells. This mIF panel was applied to NSCLC tissue micro arrays containing 1383 images from >300 NSCLC patients. Machine-learning based image analyses were conducted to segment the tissue into tumor and stroma compartment as well as to perform cell classifications and distance calculations on single-cell level.

This study thereby evaluates immunoproteasome-CD8 T cell interactions as a prognostic biomarker for NSCLC progression.

P33: Identification of the ICAM-2/ICAM-1 ratio as a treatment-insensitive biomarker for SSc-associated ILD and pulmonary inflammation

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Objectives: This study aimed to characterize the serum profile of soluble cell adhesion molecules (sCAMs) in systemic sclerosis (SSc) and identify treatment-insensitive sCAM biomarkers for the disease.

Methods: A two-phase study design was employed. The discovery phase enrolled 103 SSc patients and 86 healthy controls, while the validation phase recruited an independent cohort of 43 SSc patients and 29 healthy controls. High-resolution computed tomography (HRCT) was utilized to assess interstitial lung disease (ILD)-associated features, including ground-glass opacity (GGO), reticulation, traction bronchiectasis, honeycombing. Serum levels of 13 sCAMs were quantified using LEGENDplex™ Multi-Analyte Flow Assay Kit.

Results: Compared to healthy controls, SSc patients exhibited dysregulated sCAM characterized by altered levels and correlations among CAMs, ICAM-2/ICAM-1 and the E-selectin/P-selectin ratios. Analysis comparing healthy controls, untreated SSc patients, and treated SSc patients identified the ICAM-2/ICAM-1 ratio as a treatment-insensitive biomarker for SSc. Stratification based on organ involvements revealed that the ICAM-2/ICAM-1 ratio was significantly lower in SSc patients with ILD compared to those without ILD (8.92 vs 17.05, $p=0.002$). Multivariate regression analysis further showed that ICAM-2/ICAM-1 ratio was independently associated with SSc-ILD. HRCT assessment showed that the ICAM-2/ICAM-1 ratio was significantly lower in SSc patients with GGO compared to those without GGO (9.31 vs 16.57, $p=0.007$), and it was negatively correlated with GGO score ($r = -0.354$, $p < 0.01$). These findings were validated in the independent cohort, confirming the associations of the ICAM-2/ICAM-1 ratio with SSc, ILD and GGO.

Conclusion: Correctively, the study identifies the ICAM-2/ICAM-1 ratio as a treatment-insensitive biomarker for SSc. Furthermore, its association with ILD and GGO suggests that the ICAM-2/ICAM-1 ratio may contribute to the development and progress of pulmonary inflammation in SSc.

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P34: Association of systemic sclerosis-related interstitial lung disease with tumor-associated antigens and their autoantibodies

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Background: Interstitial lung disease (ILD) is the leading cause of systemic sclerosis (SSc) patients' morbidity and mortality. The role of tumor-associated antigens (TAAs) and their autoantibodies in SSc-ILD remains unclear.

Methods: A total of 124 SSc patients were enrolled at the First Affiliated Hospital of Xiamen University, China. Clinical and serological data, including serum levels of TAAs, were retrospectively collected from medical records. Single-cell RNA sequencing data were obtained from the public GEO database (GSE128169) to evaluate the expression of TAAs in SSc-ILD patients lung tissue. Additionally, 15 SSc patients' sera were tested the concentrations of autoantibodies targeting TAAs using microarray (PA003, GeneCopoeia).

Results: Compared with SSc-non-ILD (n=38), SSc-ILD patients (n=86) showed significantly elevated serum levels of carcinoembryonic antigen (CEA), cytokeratin 19 fragment (CYFRA21-1), pro-gastrin-releasing peptide (proGRP) and carbohydrate antigen 153 (CA153). Further subclassification of the 86 SSc-ILD patients into those with fibrosing ILD (SSc-F-ILD, n=62) and non-fibrosing ILD (SSc-non-F-ILD, n=24) revealed that serum levels of CEA, neuron-specific enolase (NSE) and CA153 were significantly higher in the SSc-F-ILD subgroup. Single-cell analysis exhibited increased expression of CEA and CA153 related genes in alveolar and bronchial epithelial cells. Antibody microarray demonstrated elevated serum levels of anti-CEA, anti-NSE and anti-CA199 IgG autoantibodies in SSc-ILD compared with SSc-non-ILD patients.

Conclusions: Elevated TAAs and their autoantibodies are associated with SSc-ILD, especially the fibrosing subtype. Single-cell analysis indicates that lung epithelial cells are a major source of these antigens, likely linking epithelial injury with autoantibody production.

P35: Dissecting immunoproteasome function in lung regeneration using mouse lung organoids

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Immunoproteasomes (IP) are specialized proteasomes that play critical roles in multiple immune responses. Their three catalytic subunits (PSMB8, PSMB9, PSMB10) are expressed at low levels in non-immune cells but are induced when cells encounter virus infection or upon stimulation with interferons (IFNs) and inflammatory cytokines. It has recently been shown that type I IFN disrupts repair of influenza virus damaged lung epithelial cells. As induction of the IP is part of the canonical response to type I interferon signaling, we here hypothesize that IPs play a role during injury and regeneration of lung.

Single cell RNA sequencing data analysis revealed that the IP is dynamically regulated during lung organoid formation with low baseline expression in alveolar type (AT)2 and AT1 cells that was transiently increasing during AT2 to AT1 differentiation. To validate these data, we established a mouse alveolar organoid system and investigated the role of the IP in lung regeneration. We confirmed upregulation of the IP (PSMB8 and PSMB9) upon AT1 differentiation. Treatment of the organoid at AT2 stage with IFNg impaired organoid formation and strongly stimulated IP expression. Of note, inhibition of the IP using the specific inhibitor LU005i blocked differentiation of AT1 cells from AT2 organoids as revealed by reduced and aberrant AT1 marker gene expression. Our data thus suggest a critical role for the IP in alveolar differentiation. In aging mice, IP triple KO mice showed superior organoid formation efficiency and larger organoid sizes than WT cells at AT2 differentiation conditions implying a role in maintenance of AT2 cell stemness.

In summary, the use of an alveolar organoid culture system allowed detection of the dynamic regulation of the IP and an unexpected function for alveolar differentiation. The significance of IP expression for lung regeneration is currently evaluated in depth.