MICROBIOLOGY SOCIETY ANNUAL MEETING IN IRELAND

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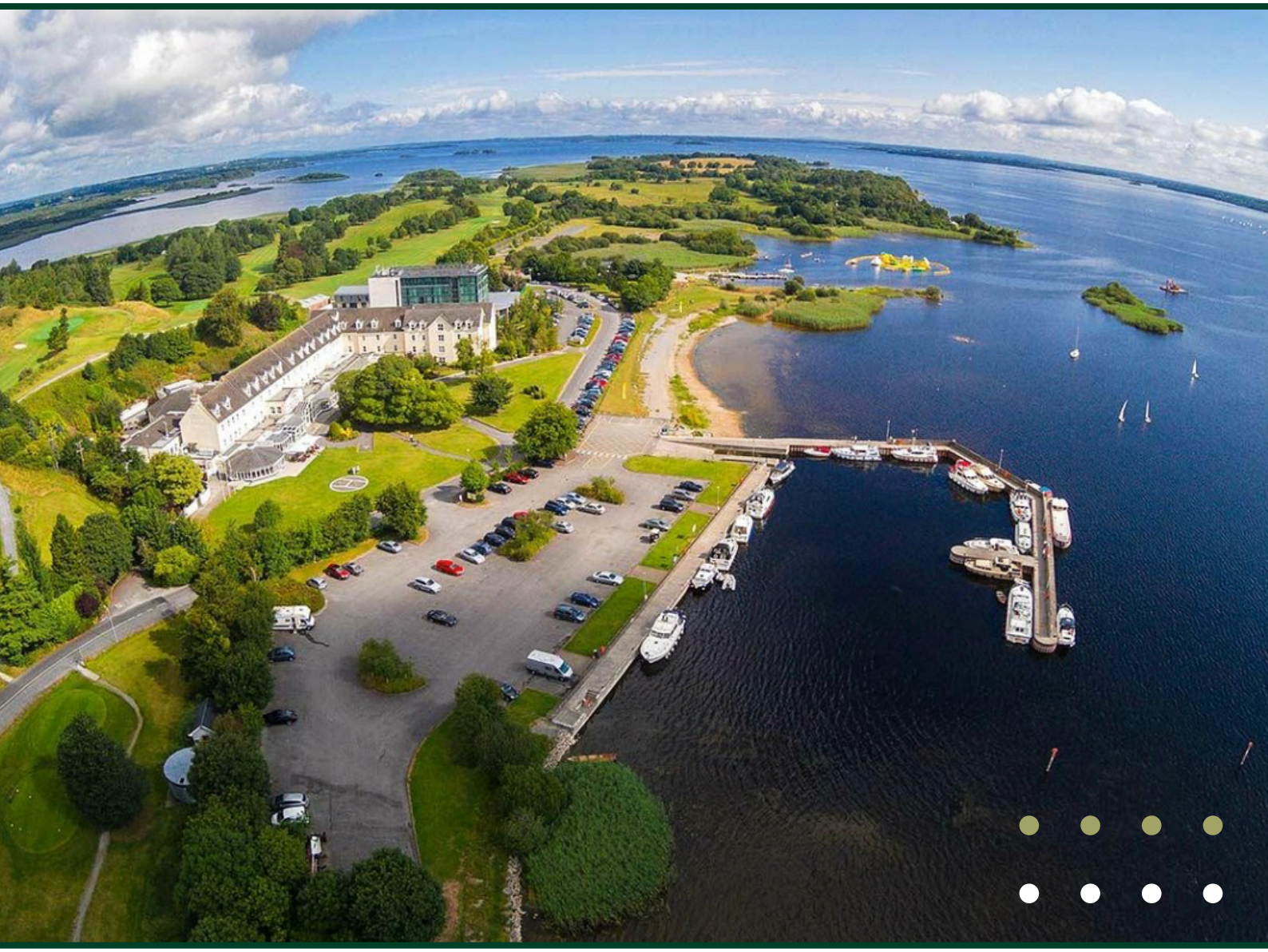
POSTER ABSTRACT BOOK



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MICROBIOLOGY SOCIETY

P001

Crossing Boundaries: The Surge of Multidrug-Resistant Foodborne Pathogens Between Humans and Animals in India

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Abstract

Background:

In India, rising demand for livestock, driven by population growth and changing dietary habits, has led to the adoption of intensive farming practices. Antimicrobial use in animal rearing is rampant and poorly regulated. Due to absence of an integrated surveillance system, true burden of foodborne diseases and antimicrobial resistance from key pathogens in India remains unknown.

Methods:

We present data from WHO-AGISAR India study involving concurrent human and animal sampling across North India, with participation from 14 laboratories. 1,968 community-acquired diarrhea cases (severity assessed by Vesikari score) across all ages were included. Cross-sectional sampling included 487 animal stools and 419 meat samples. Pathogens were identified and tested for antimicrobial susceptibility. Whole-genome sequencing was performed on 117 non-typhoidal Salmonella and 122 Enteroaggregative E. coli isolates from both humans and animals.

Results:

Over 80% of diarrhoeal samples were from moderate to severe cases, showing EAEC (5%), ETEC (4.84%), EPEC (4.32%), Campylobacter(2%), and NTS (1.2%). Food animals showed high carriage of EPEC (32.11%), Campylobacter (24.72%), and NTS (10.15%), mostly atypical EPEC (84.52%). MDR was high, ranging from 28% in NTS to 59.8% in ETEC. Human strains were more resistant to β-lactams and co-trimoxazole; animal strains had higher resistance to ciprofloxacin, aminoglycosides, and tetracycline. A high diversity of serovars, with considerable overlap in serovar and sequence types, was found between human and animal isolates.

Conclusions

This extensive study reveals a high burden of MDR foodborne pathogens in animals, underscoring need for sustained One Health surveillance to address zoonotic transmission and antimicrobial resistance.

P004

Surveillance of sanitary ware in three Irish hospitals for carbapenem resistant Enterobacterales

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Abstract

Sanitary ware in three geographically distinct hospitals in the Republic of Ireland were screened for carbapenemase producing *Enterobacteriaceae* (CPE) over a three-month period with one-off sampling occurring six and ten months after initial sampling. Swab samples from the sink drain, shower drain and toilet, and wastewater from the shower drain were taken by hospital staff and delivered to the university for processing. Culture-based methods were utilised to isolate presumed CPE using EMB agar and Simmons citrate agar with 1% myo-inositol impregnated with imipenem according to the EUCAST guidelines to isolate presumed carbapenem resistant *Escherichia coli* and *Klebsiella* spp. respectively. The species were identified using 16S rRNA PCR amplification and subsequent sequencing. 933 isolates were obtained of which 170 were confirmed to be Enterobacterales with the majority being of the order Xanthomonadales. Other orders identified were Burkholderiales, Pseudomonadales, Bacillales, Lactobacillales, Rhodocyclales, Altermonadales, Kinetoplastida and Actinomycetales. 95 samples were not identified due to unsuccessful PCR reactions and are currently being speciated by MALDI-TOF. Of the confirmed Enterobacterales there were 22 *Citrobacter* spp., 67 *Enterobacter* spp., 26 *Klebsiella* spp., 1 *Leclercia* spp., 1 *Kosakonia* spp., 8 *Morganella morganii*, 3 *Salmonella enterica*, 14, *Proteus mirabilis*, 1 *E. coli* and 27 *Raoultella ornithinolytica*. CPE were identified in all 3 hospitals and they are capable of colonising sanitary ware in the Irish hospital environment. This data also highlights the limitation of culture-based methodology as most of the isolates obtained were not Enterobacterales.

P005

Mycosporine-Like Amino Acids from *Desmonostoc* Species: A Promising Source for Sunscreen Formulation

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Abstract

Mycosporine-like amino acids (MAAs) are secondary metabolites known for their strong ultraviolet (UV) absorbing properties (310–365 nm range), providing photoprotection in various microorganisms, including cyanobacteria. Due to chemical UV filters' adverse effects, MAAs are gaining attention as natural and environmentally friendly UV-protective alternatives. In arid regions, cyanobacterial species are exposed to intense solar radiation, making them potential sources of MAAs. Hence, in this study, we use the cyanobacterial species isolated from hot, arid region for the analysis of MAAs. The experimental organism *Desmonostoc* sp. EERGCy083, isolated from a water sample collected from the Sri Ganganagar canal bordering the Thar Desert in northwestern India, was used for the extraction of UV-protective molecules. The isolates were identified based on morphological and molecular analyses (16S rRNA sequencing). Furthermore, MAA screening was performed using a conventional solvent extraction method, followed by spectrophotometric analysis and high-performance liquid chromatography (HPLC) to confirm the presence of MAAs. The spectroscopic analysis showed the presence of MAA with a UV absorption maximum of 320 nm. On HPLC analysis, the occurrence of multiple MAAs is detected. Further, ESI-MS analysis identified mycosporine ethanolamine as the predominant MAA, along with other detected MAAs. The mass spectrum gave the corresponding molecular ion peak at m/z 232 for mycosporine ethanolamine, m/z 451 corresponding to hexose-bound palythine threonine and m/z 303 for mycosporine-2-glycine. From the experimental findings, it was found that the cyanobacterial strain *Desmonostoc* species EERGCy083 will be a potential source for natural sunscreen formulation.

P006

PerfringicinS, a novel circular bacteriocin produced by *Clostridium perfringens* with potent antimicrobial activity

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Abstract

Bacteriocins have emerged as potential alternative or adjuncts to traditional antibiotics given the problem with resistance to classic antibiotics. These peptides generally exhibit potent activity against a narrow spectrum of bacterial species that are closely related to the producer, which is desirable as it may allow for targeted removal of bacteria/ pathogens from complex microbiomes. Therefore, to develop targeted treatment strategies we mined for bacteriocin producers from different sources including a number of animal fecal microbiomes.

Antimicrobial isolates recovered after primary selection were verified by overlays, spot and well-diffusion assays against several bacterial and/or pathogenic indicators. The potential candidates were then sequenced using the Illumina MiSeq platform. Among the resultant potential antimicrobial isolates, a *Clostridium perfringens* strain exhibited potent inhibition against another *C. perfringens* — the causative agent of foodborne illness. Interestingly, the 67 amino acid bacteriocin peptide revealed a short leader sequence and percent identity of 37.5% with circularin A from *Clostridium beijerinckii* ATCC 25752. The mass of the structural peptide revealed by colony MALDI-TOF analysis was found to be 6084 Da. Peptide purification and minimum inhibitory concentration (MIC) assays will be performed to validate the bacteriocin activity. Furthermore, heterologous expression of the peptide in a GRAS strain will be carried out.

Given the clinical importance of the target pathogen, this potentially novel bacteriocin producer from *Clostridium* sp. may have potential as a targeted biotherapeutic in food applications and/or clinical settings.

P009

Impact of External Factors on EV Production and RNA Cargo Profiles in Lactobacillus acidophilus

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Abstract

Extracellular vesicles (EVs) produced by Gram-positive bacteria, including probiotic *Lactobacillus* species, have emerged as promising mediators of host–microbe interactions. Despite their potential in inter-kingdom communication and probiotic function, significant gaps remain in understanding how EVs form, their molecular cargo, and how environmental and physiological factors influence their production, composition and RNA content.

This work systematically investigates the factors influencing EV yield and RNA content in *Lactobacillus acidophilus*. Specifically, this study evaluates: (i) the impact of different EV isolation methods on vesicle yield and size distribution, and (ii) how stress-related growth conditions such as temperature shifts, pH variation, antibiotic exposure, and radiation influence EV production and RNA cargo profiles. Although research in other Gram-positive bacteria suggests that environmental conditions can alter EV yield and composition, their specific effects on probiotic strains remain uncharacterised.

To explore this, *L. acidophilus* cultures are grown under varied conditions. EVs are isolated using precipitation, differential ultracentrifugation, and high-speed centrifugation to capture a broad size range. Nano flow cytometry, nanoparticle tracking analysis (NTA), and microscopy are used to characterise vesicle size distribution and concentration. These methods will support the optimisation of isolation protocols and provide a robust basis for downstream RNA sequencing to catalogue stable and abundant RNAs packaged within these vesicles under varying conditions.

The outcomes of this work-in-progress will contribute to a deeper molecular understanding of probiotic EV biogenesis and support future functional studies on how probiotic-derived EV-RNA contribute to microbiome–host interactions.

P011

Proteomic analysis of Methicillin Resistant Staphylococcus aureus in the Presence of Oxacillin

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Abstract

Background

Staphylococcus aureus is a gram-positive bacterial species found on human skin and is most often associated with skin infections such as abscesses and cellulitis. S. aureus has emerged as a worldwide nosocomial pathogen acquiring resistance to antibiotics such as methicillin (MRSA). New methods of treating antibiotic-resistant infections must be developed as it is predicted that by 2050, global deaths related to antibiotic-resistant infections could rise to 10 million annually.

Methods

An MRSA clinical isolate and an oxacillin-susceptible Staphylococcus aureus strain, RN4220, were grown in M9 minimal media supplemented with oxacillin. The concentration of oxacillin and incubation time needed to illicit a pharmacodynamic kill response was determined via a kill curve assay. Following incubation, protein extraction was carried out and samples were prepped for LC-MS in a Q Exactive Spectrometer. Data was processed using MaxQuant and AMICA. Enriched and reduced abundance protein networks were detected using KEGG Mapper.

Results/Discussion

Protein networks with differential abundances were detected in MRSA when grown with oxacillin. Enriched abundance networks included those involved in peptidoglycan synthesis and carbon metabolism. Reduced abundance networks included those involved in Staphylococcus aureus infection and glycolysis.

Conclusion

Identifying protein networks with differential abundance in MRSA grown in oxacillin expands our understanding of the resistance mechanism. This could lead to the identification of novel targets which, when targeted, could disrupt the resistance mechanism.

P013

Understanding microbial communities and the resistome of hospital wastewater samples from Reunion Island

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Abstract

Hospital sanitary ware can serve as reservoirs for pathogenic bacteria and their associated antimicrobial resistance genes (ARGs), contributing to the spread of resistance via mobile genetic elements. These reservoirs have been linked to outbreaks, particularly involving extended-spectrum beta-lactamase (ESBL)-producing bacteria. Reunion Island, located in the Southwest Indian Ocean and well connected to Europe, Southern Africa, and the Indian subcontinent, is both a recipient and potential hub for the spread of ESBL and carbapenemase-producing pathogens. In this study, hospital wastewater effluents from Reunion Island were analysed to assess microbial communities and their resistomes. Five metagenomic samples were collected and sequenced using PacBio long-read sequencing. Assembly and binning tools were compared, using metaMDBG or metaFlye for assembly and PacBio's HiFi-MAG-Pipeline for binning, to determine their effect on the number and quality of metagenome-assembled genome (MAGs) obtained. A total of 374 and 295 high-quality MAGs (completeness ≥90% and contamination ≤5%) were obtained for the metaMDBG and metaFlye approach, respectively. MAGs representative of 118 species from 93 genera were obtained, with a predominance of *Pseudomonas*, *Flavobacterium*, and *Aeromonas*. Resistome analysis revealed the presence of ESBLs such as *bla*GES-1, *bla*GES-5, *bla*VEB-9, *bla*SHV-106, and various carbapenemases including *bla*OXA-10, *bla*OXA-17, *bla*OXA-347, *bla*OXA-4, *bla*OXA-912, *bla*NDM-1, and *bla*VIM-4. This work supports early monitoring efforts to identify potential hospital outbreaks and their sources within Reunion Island to improve infection control.

P015

Impact of biofilms and quorum sensing on the emergence of heteroresistance to last-resort antibiotics

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Abstract

Antibiotic heteroresistance presents a growing public health concern since the phenotype is associated with treatment failure and is hard to detect using conventional diagnostic testing. In this study, we characterized heteroresistance to last-resort antibiotics in *Klebsiella oxytoca*, an opportunistic pathogen associated with hospital-acquired infections alongside exploring the role of biofilms and quorum sensing in enhancing this phenotype.

Heteroresistance was characterized in six isolates (clinical and environmental) using population analysis profiling (PAP). Using biofilm and antibiotic stress assays, the frequency of resistant subpopulations was compared between planktonic-phase and biofilm-derived bacteria in the presence of polymyxin B, colistin and meropenem at high concentrations (32-512 µg/mL). Pre-formed biofilms were treated with antibiotics, followed by viability assessment and live/dead imaging using confocal laser-scanning microscopy. Using *Vibrio harveyi* BB170 bioluminescence assays, we characterized the presence and levels of A1-2 quorum-sensing molecules in all six isolates.

The isolates were confirmed to exhibit heteroresistance, indicated by the 8-fold difference between the MICs (32-64 µg/mL) and the maximum non-inhibitory concentration (2 µg/mL). Four out of the 6 isolates were strong biofilm formers. They demonstrated increased resistance to all the antibiotics, as evident in an increase in their MICs compared to the planktonic phase bacteria. Supernatants of all six isolates induced luminescence in *V. harveyi*, indicating active production of AI-2 quorum-sensing molecules, with fold-induction analysis showing variable levels among the isolates.

These findings support evidence that biofilms enhance antibiotic heteroresistance and highlight the need for targeted approaches to prevent persistent infections in clinical settings.

P016

Biotechnological Characterisation and Genetic Analysis of Plant-Associated Lactic Acid Bacteria

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Abstract

The global market for plant-based (PB) milk alternatives continues to expand with sales predicted to reach $52bn (USD) by 2032. Within the European market, sales of PB cheese and yogurt analogues are rapidly increasing. Similar to traditional dairy-based fermented products, PB cheese and yogurt analogues may be produced via the fermentation of PB milk alternative substrates by Lactic Acid Bacteria (LAB) including *Streptococcus thermophilus*, species of *Lactococci* and various members of the *Lactobacillaceae*. However, many of these industrialised species of LAB display significant evolutionary regression, owing to niche adaptation within the rich milk environment. As such, many dairy-adapted strains display biotechnological redundancy when applied to complex PB milk alternative substrates. Contrastingly, "wild" LAB have retained a larger repertoire of metabolic bio-machinery which allows for proliferation on differing substrates.

In this study we screened a variety of phyllosphere samples for the presence of viable LAB which harbour potential for the fermentation of PB milk alternatives. Following speciation via 16s rRNA sequencing, isolates were evaluated for their biotechnological potential including carbohydrate utilisation patterns, protease activity, EPS production, antibiotic resistance, phage susceptibility and in the case of *Lactococci* – cell wall polysaccharide type. Eighteen isolates – 9 *Lactococci,* 6 *Leuconostoc*, 2 *Pediococci* and 1 *Lactiplantibacillus* – were further subject to whole genome sequencing and detailed bio-informatic analysis, with genotype to phenotype matching being employed to support experimental findings.

Here, we present the outcomes of our combined analysis and highlight the biotechnological potential of plant-associated LAB as novel starter cultures for the PB fermentation industry.

P017

Integrated genome-metabolome profiling unveils *Trichoderma* sp. strain AM6 as a key modulator of terpenoid metabolism in tea (*Camellia sinensis* L.)

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Abstract

Tea (*Camellia sinensis* L.) is highly valued for its terpenoids and flavonoids, which significantly contributes to its flavor and health benefits. However, the microbial influence, particularly *Trichoderma* spp. and other beneficial bacterial consortia, on tea's terpenoid metabolism remains underexplored. In this study, we characterized a tea root-associated novel, indigenous *Trichoderma* sp. strain AM6 throu integrated genome and metabolome profiling. The strain possesses a 39.91 Mbp genome size with 98.8% assembly completeness and a fully assembled mitochondrial genome. Metabolic pathway analysis from the genomic insights revealed that AM6 exclusively utilizes the mevalonate pathway for terpenoid biosynthesis, unlike tea plants, which employ both mevalonate (cytosolic) and MEP/DOXP (plastidial) pathways. Untargeted LC-ESI-MS/MS identified a total of 11,841 secondary metabolites, including a rich diversity of monoterpenoids, diterpenoids, a sesquiterpenoids. Phylogenomic analysis confirmed AM6 as a distinct *Trichoderma* species with unique adaptations. GC-MS profiling showed enhanced volatile terpenoid production in tea plants inoculated with either AM6 alone or in combination with a synthetic microbial consortium. Transcriptome analysis of tea roots revealed upregulation of HMGCR and downregulation of DXS transcripts, suggesting microbe-induced metabolic shift towards the mevalonate pathway. Our findings unraveled the potential of *Trichoderma* sp. AM6 as a key modulator of tea terpenoid metabolism, offering promising applications for improving tea quality through targeted microbiome management.

P018

Oligopeptide-modified chimeric oligonucleotides as targeted therapeutics for the ESKAPE pathogens

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Abstract

Antimicrobial resistance (AMR) is a global issue projected to cause 10 million deaths per year by 2050. Thus, there is a need for novel and innovative therapies that can overcome the resistance developed by bacterial pathogens. Antisense oligonucleotides (ASOs) are RNA therapeutics that can hybridise to a target gene and prevent it from being translated. I aim to develop ASOs that target resistance genes of the ESKAPE pathogens, to resensitize them to existing antibiotics.

Target genes are selected by conducting bioinformatics analyses on selected strains, and ASOs are designed to be complementary to the genes of interest. Delivery into bacterial cells is facilitated by conjugating the ASOs to a cell penetrating peptide (CPP), using click chemistry. CPPs are synthesised using solid-phase peptide synthesis and purified by HPLC. Fluorescently labelled CPPs are utilised to evaluate their uptake into different ESKAPE pathogen species using super resolution microscopy and flow cytometry.

A list of target genes conserved in multiple strains of each species was obtained for each of the ESKAPE pathogens and further *in silico* analyses are ongoing to assess their suitability as ASO targets. Assessment of the CPP pVEC's entry into different species of the ESKAPE pathogens revealed that uptake varies between different species. Thus, pVEC is an ideal delivery molecule for some ESKAPE pathogens, but not others. Several CPPs are being considered as alternative delivery molecules in addition to pVEC. This interdisciplinary project is establishing chemical biology and molecular microbiology methodologies to enable the re-purposing of antibiotics for ESKAPE pathogens.

P019

Phage susceptibility of an Irish *Listeria monocytogenes* strain collection to environmentally and commercially sourced phages.

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Abstract

Listeria monocytogenes is a foodborne pathogen, often associated with ready to eat (RTE) foods, which can lead to food recalls and potentially deadly outbreaks of disease in risk groups which include pregnant women, newborns, the elderly and immunocompromised. Lytic bacteriophages have been evaluated as additional antimicrobial aids to bolster current interventions offered to the food industry, due to their inherent antibacterial properties and ability to propagate. We compared the susceptibility of a diverse collection of Irish-relevant *Listeria monocytogenes* strains from environmental, food and clinical sources to phages P61 and P53, isolated from grass silage, and a commercially available phage product. Using spot and efficiency-of-plaquing assays 164 strains showed varying susceptibility to the environmental and commercial phages. For phage P53, 60% of strains were susceptible, 22% were insensitive and 18% were not susceptible. For phage P60, 46% of strains were susceptible, 27% were found insensitive and 27 showed no susceptibility. The phages were capable of infecting broad range of *L. monocytogenes* strains without obvious correlation to serogroup, clonal complex or source type. Analysis of the efficacy of the commercial phage product is currently underway. This study demonstrates the importance of screening phages across a diverse strain collection to identify insensitive or resistant strains capable of escaping infection. This has important implications for the development of effective phage cocktails.

P020

Chemical messages mediating a global virulence switch in ESKAPE pathogen behaviour

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Abstract

Cell-to-cell communication in microbial systems is known for its vital role in cellular signalling and gene expression. Microbial functions associated with bacterial virulence, pathogenicity, host-microbe interaction, and biofilm development are mediated by cellular systems. The extent to which chemical diversity it tolerated in natural signalling systems in poorly understood, and newly discovered ligand-receptor combinations are changing how core functions such as quorum sensing are viewed. Furthermore, interference in these signalling systems can modulate microbial virulence and pathogenicity, and microbial infection caused by drug-resistant pathogens. Pyrones have recently been identified as LuxR agonists, while plant-derived phytochemicals such as coumarins have been found to shape microbiome dynamics and pathogen behaviours from a broad spectrum of ecosystems. Here we uncovered a new role for pyrone and coumarin signals in suppressing key chronic phenotype behaviours in *Pseudomonas aeruginosa* and other ESKAPEE pathogens, as well as in the fungal opportunist *Aspergillus fumigatus*. Proteomic analysis revealed strong suppression of WspR (cyclic di-GMP), consistent with the observed biofilm and twitching suppression. We also uncovered an important 'hydroxylation-bias' favouring coumarin and umbelliferone (7-OH) in the specific competitive inhibition of the Pseudomonas Quinolone Signal (PQS), associated with reduced activity of a PqsR translational fusion and suppression of pyocyanin production. Conversely, esculetin (6,7-OH) was most effective at Acyl Homoserine Lactone (AHL) QS biosensor inhibition. As the very real threat posed by antimicrobial resistance persists, these data support a role for chemical messages in delivering an ecological solution to dysbiosis in the host-microbe interaction.

P021

Genomic and phenotypic changes associated with the acquisition of fluoroquinolone resistance in *Campylobacter jejuni*

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Abstract

Campylobacter jejuni is the leading cause of bacterial gastroenteritis worldwide, with fluoroquinolone resistance posing a persistent challenge despite regulatory interventions. Resistance in *C. jejuni* is associated with mutations in the *gyrA* gene which lead to a relaxation of DNA supercoiling. This is known to impact several key phenotypes, including motility, biofilm formation, and virulence, with important implications for transmission and severity of disease. In this study, two laboratory and two recently isolated *C. jejuni* strains were repeatedly passaged on increasing concentrations of ciprofloxacin, yielding isolates with low (up to 10µg/mL), medium (up to 40µg/mL), and high levels (>40µg/mL) of ciprofloxacin resistance. PCR confirmed the presence of the T86I mutation in the *gyrA* gene of these resistant isolates, and motility testing via the soft agar method revealed strain-dependent differences between susceptible and resistant isolates. Fluorescence microscopy was used to assess viability and structure of biofilms under microaerobic and aerobic conditions, with changes in biofilm morphology observed in resistant isolates. Additionally, assays were conducted to evaluate changes in protein secretion and aerotolerance, and chloroquine gel electrophoresis was used to compare plasmid supercoiling levels between susceptible and resistant isolates. Isolates were whole genome sequenced to identify shared SNPs associated with the phenotypic shift that accompanies fluoroquinolone resistance. These results build on previous observations demonstrating that the acquisition of resistance-conferring mutations in *gyrA* leads to changes in phenotypes with implications for *C. jejuni* transmission and virulence.

P023

The development of novel freeze-dried bacteriophage formulations that can be used for the treatment of a range of bacterial infections.

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Abstract

Antimicrobials, formerly hailed as “miracle drugs”, are the cornerstone of modern medicine and indispensable in treating and preventing infections in humans, animals and plants. However, due to the rise of antimicrobial resistance (AMR), infections that were once easily treatable with first-line antimicrobials are becoming increasingly difficult to manage, posing a major global health threat. Bacteriophage therapy is one of the most promising alternative treatments for bacterial infections. With the resurgence in confidence in phage therapy, the formulation of bacteriophage products for therapeutic application is necessary for commercial circulation. However, bacteriophages often lose activity during formulation and storage, especially in suspension. This study investigates lyophilisation, or freeze-drying, as a method to preserve phage viability in a solid state for long-term storage at ambient temperatures. The study also intends to test the effects of different cryoprotectants on long-term phage stability and activity. Preliminary results suggest that, even though adding a cryoprotectant such as 10% w/v sucrose to bacteriophage stocks before being freeze-dried did not improve phage viability, it was necessary to improve product stability and prevent cake collapse, serve as a bulking agent and improve the product’s physical appearance. The aim is to incorporate the solid formulations into a range of pharmaceutical products such as rod-type inserts for vaginal rings, wound dressings, and hydrogels.

P024

Utilising an in vitro gut model to investigate drug-microbiome interactions

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Abstract

Inter-individual variability in treatment response poses a significant challenge in patient care. The gut microbiome, which varies widely among individuals, affects drug responses. This study aimed to explore how the variable response medicine donepezil impacts the gut microbiome of a healthy volunteer using a reproducible in vitro gut model.

Method: A batch fermentation gut model was optimized to replicate the distal colon environment, considering pH, temperature, and anaerobic conditions. Its functionality was validated through experiments with an untreated negative control, a positive antibiotic control to induce dysbiosis, and donepezil, a variable-response drug. Samples were collected at t = 0, 8, 24, and 48 hours for DNA extraction and 16S rRNA V4 sequencing. Additionally, the growth of gut microbiota isolates was assessed with and without donepezil.

Results: Sequencing results revealed the presence of a diverse microbiome in the simulated models, dominated by the phyla Firmicutes and Bacteroidetes. Notably, a significant change in the microbiome was observed between t=0 and t=8 h, before stabilisation at the 24 h timepoint. Significant changes in beta diversity were observed in microbiome populations by 24 h. The growth of certain gut bacteria isolates was decreased by donepezil.

Conclusion: The gut model employed showed considerable promise as a simulated gut environment for the anaerobic growth and maintenance of gut microbiota. Further optimisation of the model, including timing of drug addition, will improve the robustness of this system as a model for studying the effects of variable response drugs on the gut microbiome.

P025

Assessing the proteolytic ability of food microbes as a potential tool to improve digestibility of plant-based protein

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Abstract

Global growth in plant-based diets highlights potential nutritional challenges, particularly concerning plant protein quality and digestibility. Fermentation offers a promising approach to improve the nutritional, functional, and organoleptic properties of plant proteins. The initial research goals of the "UP-LIFT" project are to screen a biobank of food-grade microorganisms for their ability to hydrolyse plant-based proteins isolated from crops including potato, fava bean, and pea. Screening to date has been carried out with microbial strains on fava bean protein isolated at Teagasc, and a commercially available pea protein isolate. Protein suspensions (5% w/v) in H_2O were inoculated with overnight cultures of candidate microorganisms and incubated for 72 hours. Changes in microbial load (CFU's) and pH were observed, and final fermentation products were solubilised in 1% SDS for protein analysis via BCA assay and SDS-PAGE. Strains of *Yarrowia lipolytica*, *Bacillus subtilis*, and *Bacillus licheniformis* have been demonstrated to break down pea and fava bean protein, based on changes in their protein composition visualised in SDS-PAGE, and >1.0 $\log_{10}$ increases of microbial load in the fermentations. A plate-based proteolytic assay based on pea protein isolate, currently under development aligns with SDS-PAGE results obtained post-fermentation utilising the same starting inoculum and comparable fermentation conditions. The initial findings of this research are promising, showing potential of mass screening of strains able to process plant proteins. Future work will focus on utilising screened strains in optimised bioprocesses with the aim of improving nutritional and functional properties of plant protein for novel ingredient development.

P026

Microbiome and Resistome Responses to Flooding in Agricultural Soils.

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Abstract

Background:

Antimicrobial resistance (AMR) and climate change are two escalating threats to global health. Climate-driven increases in flooding events have been observed worldwide, with significant implications for environmental health. The environment can harbour antimicrobial resistance genes (ARGs), which may spread among microbial communities through horizontal gene transfer.

Methods:

8 flooded and 8 non-flooded soil samples were compared using a combined approach of shotgun metagenomic sequencing and high-throughput qPCR array targeting ARGs. Metagenomic data was used to determine taxonomic composition and resistance profile, while qPCR array was used to detect 96 specific ARGs. Additionally, preliminary binning for metagenome-assembled genome (MAG) recovery is underway, with the goal of linking assembled genomes to ARGs identified through metagenomic analysis.

Results:

Metagenomic data revealed that flooded soils showed differences in community composition, including increased relative abundance of facultative anaerobes such as *Anaeromyxobacter* and *Rhodomicrobium*. In contrast, bacterial genera such as *Bradyrhizobium* and *Kribella* were decreased in flooded soil. A general decrease in ARG abundance was observed in flooded soils. While metagenomic data from non-flooded soils showed a reduction in only a few genes, qPCR array results revealed a wider decrease, including genes such as *qepA, aad*, and *vanA.* Ongoing MAG generation will potentially reveal high-quality bins that may be linked to ARG-carrying taxa.

Conclusion:

These results highlight the potential impact of flooding on microbial community composition and ARG distribution in soil.

P027

Isolation and identification of antibiotic resistant bacteria and genes from soil. The effect of animal manure use as a fertiliser on antimicrobial resistance in an Irish beef and sheep farm.

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Abstract

Introduction:

Antimicrobial resistance (AMR) is a public health issue affecting many areas including agriculture. In farms, animal manure can contain antibiotic resistant genes, antibiotic resistant bacteria and undegraded antibiotics. Due to the use of animal manure as a fertiliser and its deposition on soil, there are fears that AMR could spread through the food chain from animals to the environment. Therefore, the impact of animal manure spreading on AMR needs to be investigated.

This study aims to; take soil and animal manure samples before and after animal manure spreading; extract DNA and characterise the taxonomic and AMR gene profile using shotgun sequencing and isolate resistant bacteria to investigate multidrug resistance (MDR) and resistance mechanisms.

Methods:

Animal manure samples were taken before spreading and soil samples taken before, 2 weeks, 1, 2 and 3 months after manure spreading. Bacterial DNA was extracted from soil and manure and sent for shotgun sequencing.

Antibiotic resistant bacteria were isolated by diluting soil samples in phosphate buffered saline solution and inoculating directly onto antibiotic media.

Results:

36 colonies resistant to ampicillin, ertapenem, ciprofloxacin, colistin, chloramphenicol, trimethoprim, amikacin and tetracycline were isolated so far and are being tested for multidrug resistance. These results highlight the presence and dissemination of MDR on farms.

Conclusion:

This work investigates the effects of animal manure use as a fertiliser on the spread of AMR on farms. Shotgun sequencing data will reveal the impact of animal manure spreading on the microbial taxonomic and AMR gene profile of soil in agricultural fields.

P028

Rapid Formation of Aerobic Granules as a Source of Fast-Aggregating Bacteria for Cell-to-Cell Immobilization in Bioaugmentation for Nutrient Removal from Wastewater

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Abstract

Background:
Aerobic granulation involves a carrier-free, self-immobilisation process where microbial biomass forms dense aggregates. These granules may serve as a promising source of fast-aggregating bacteria that can act as natural immobilizing partners for pollutant-degrading microbes in bioaugmentation strategies targeting nutrient removal from wastewater.

Methods:
Three sequencing batch reactors (SBRs) were operated under high hydraulic selection pressure and elevated organic loading rates to induce rapid aerobic granule formation. Aerobic granules and sludge development were monitored over 8 days through morphological observations and total suspended solids (TSS) measurements.

Results:
Granules with diameters between 1- 4 mm were observed within 30 hours of reactor operation, indicating rapid microbial aggregation. Biomass accumulation increased steadily after the initial washout, with reactor R2 reaching a settled biomass height of 29 cm by day 7. However, sustained granulation was not achieved beyond 48 hours. By day 8, all reactors showed granule deterioration and filamentous overgrowth, attributed to high organic loading and short cycle times. These findings highlight the limitations of operational conditions that prioritise rapid granulation but compromise long-term stability.

Conclusion:
Early-stage granule formation under stressed operational conditions provides a viable route for isolating fast-aggregating bacteria. These bacteria will be isolated and evaluated for their potential to act as biological immobilization supports via cell-to-cell aggregation for co-culturing with functional nutrient-removing bacteria. This approach could enhance bioaugmentation by improving microbial retention, synergy, and reactor stability in wastewater treatment applications

P030

A molecular dissection of glyphosate metabolism by *Sinomonas atrocyanea* ATCC13752

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Abstract

Glyphosate (N-(phosphonomethyl)-glycine), a broad spectrum organophosphonate herbicide, is the most widely used herbicide globally. Despite its benefit to agriculture, widespread usage has been linked with adverse ecological and health complications. Whilst abiotic strategies can address these ecotoxicological consequences, biodegradation offers an alternative strategy. Surprisingly, however, fundamental questions remain unanswered about its metabolism by microorganisms.

Sinomonas atrocyanea ATCC13752, is a biphasic Gram positive bacterium deposited in culture collection prior to the invention of glyphosate. Pipke and Amrhein (1988) reported its ability to degrade glyphosate as sole phosphorus source, producing AMPA (Aminomethylphosphonic acid) as a primary metabolite, before its subsequent mineralisation. Though dominant in soil, this degradation pathway, is rarely found in axenic cultures. We now seek to use a range of strategies to dissect *S.atrocyanea*'s ability to metabolise glyphosate in order to understand the evolutionary origin of this trait within the soil microbiome.

In agreement with Pipke and Amrhein, *S.atrocyanea* grew on glyphosate and KH2PO4 as sole phosphorus sources. Unexpectedly no growth was observed on AMPA or other organophosphonates even when multiple media supplementations were utilised. Further investigation, confirmed by DAPI staining, revealed that S.atrocyanea accumulated intracellular polyphosphate, and was tolerant to glyphosate up to a concentration of 50 mM.

Surprisingly bioinformatic analysis reveals the genome sequence of *S.atrocyanea* ATCC13752 contains no known phosphonate metabolizing genes. A proteomic workflow has been created that will allow us, for the first time to understand more about not only glyphosate degradation, but also phosphorus metabolism more widely, in *S. atrocyanea* ATCC13752.

P032

Identification of a MarR-Family Regulator of Ectoine Biosynthesis in M. kenyense AMO1 Through Integrative Genomics and Motif Discovery

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Abstract

Ectoine is a high-value compatible solute widely used in pharmaceuticals, cosmetics, and biotechnology due to its protective properties under osmotic and thermal stress. Methanotrophs, bacteria that utilise methane as their sole carbon and energy source, are emerging as sustainable platforms for ectoine biosynthesis. *Methylomicrobium kenyense* strain AMO1 is particularly promising, producing ectoine at lower salinities (3–5%), offering industrial advantages such as reduced bioreactor corrosion and operational costs. Hence, elucidating the regulatory mechanisms governing ectoine biosynthesis in this organism is essential to optimise its metabolic potential and improve ectoine production efficiency. To uncover regulatory elements, we performed hybrid whole-genome sequencing of *M. kenyense* AMO1, assembling the genome into four contigs (N50: 4.4 Mb). Canonical ectoine biosynthesis genes (ectABC) and their promoter regions were identified. BPROM analysis revealed a σ^{70}-dependent promoter upstream of *ectA*, suggesting transcription as a single operon. MEME Suite identified a conserved 20-bp motif in this promoter region, also found near a MarR-family transcriptional regulator (GIBCMODD_02113). Protein annotation using InterProScan confirmed this regulator's domain profile, indicating its potential role in ectoine biosynthesis modulation. This integrative genomic approach identifies a MarR-family regulator as central to ectoine biosynthesis regulation, laying a foundation for future experimental validation and biotechnological optimisation.

P033

Genomic Analysis of Multiple Vancomycin-Resistant and Vancomycin-Susceptible Enterococcus faecium isolates Recovered Simultaneously from Individual Hospitalised Patients Reveals Universal Within-Host Diversity.

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Abstract

Vancomycin-resistant *Enterococcus faecium* (VREfm) is a major hospital-associated pathogen with a high prevalence in Ireland. Clinical microbiology laboratories routinely investigate one VREfm isolate per patient for surveillance, which fails to account for potential multiple colonising strains.

Whole-genome sequences of up to 100 *E. faecium* isolates (VREfm and vancomycin-susceptible *E. faecium* (VSEfm)) per rectal swab from known VREfm-positive patients in a large Irish hospital were determined using the MiSeq platform (Illumina). Isolate diversity was investigated by core-genome multilocus sequence typing (cgMLST) using Seqsphere+ v8.5.1 (Ridom GmbH), based on 1423 genes. cgMLST clusters were determined using single-linkage clustering and a pairwise allelic difference threshold of ≤20.

To date, *E. faecium* isolates from three patients have been sequenced. cgMLST analysis revealed extensive within-host diversity, with ≥ two Sequence Types (STs) identified in each patient. ST80 predominated: Patient 1, (36/46, 78.2%); Patient 2, (69/72, 95.8%); Patient 3, (13/14, 92.8%). Multiple complex types (CTs) were identified among ST80s in all patients: Patient 1, 36 ST80 isolates comprised CT9300 (n=8, 22.2%) and CT2933 (n=28, 77.7%); Patient 2, 69 isolates comprised CT6738 (n=52, 78.8%) and CT9300 (n=17, 24.6%); Patient 3, 13 isolates comprised CT6988 (n=9, 69.2%) and CT2933 (n=4, 30.7%). In all three patients, > 51% isolates were VREfm.

The results of this study to date indicate potential universal within-host *E. faecium* diversity in VREfm-positive patients in an Irish hospital, raising concerns over reliance on investigating one isolates per patient for VREfm surveillance. A further 700 isolates from seven other patients are being investigated to confirm these findings.

P034

Concentration-Dependent Effects of Furanone C-30 as a positive control on Quorum Sensing and Motility Phenotypes in *C. violaceum* and *P. aeruginosa*

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Abstract

Background:
Quorum sensing (QS) is a cell-to-cell communication mechanism in bacteria that regulates gene expression and group behaviours. Synthetic furanone-C30 is widely used as a QS inhibitor and positive control in bacterial phenotypic assays; however, its effectiveness across various QS assays and bacterial species remains inconsistently validated. This research investigates how furanone-C30 affects two well-established QS model organisms, *Chromobacterium violaceum* ATCC 12472 and *Pseudomonas aeruginosa* PAO1.

Methods:

Growth curves were established by culturing *C. violaceum and P. aeruginosa* in Luria Bertani broth separately for 24hurs, with furanone-C30 concentrations ranging from 10-100 µg/mL. After determination of non-toxic concentrations (without growth inhibition), QS-related phenotypic assays were performed, violacein production in *C. violaceum* and swimming and swarming motility assays in *P. aeruginosa*.

Results:

At 10 µg/mL, furanone-C30 did not significantly affect bacterial growth over 24 hours in both organisms, validating this concentration for subsequent assays. Higher concentrations caused significant growth inhibition and cell death. Furanone-C30, at 10 µg/mL, effectively reduced violacein production and inhibited swimming motility, confirming its role as a robust positive control in these QS assays. However, it failed to inhibit swarming motility, indicating assay-specific limitations. Molecular analysis results indicated the need for further investigation on other genes involved in the QS mechanism inhibition.

Conclusion:

Our findings emphasize the need to use non-cytotoxic concentrations of furanone-C30 to distinguish true QS inhibition from growth effects. Although effective in some assays, its variable performance highlights the importance of assay-specific validation and routine growth curve assessment in QS studies.

P036

A Unified 72-Hour Protocol for Rapid Pathogen Detection in Environmental and Food Samples

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Abstract

Outbreaks and product recalls linked to fruits and vegetables contaminated with foodborne pathogens have highlighted the vulnerability of fresh produce to environmental contamination. These risks can appear in open-field production and throughout the food production chain, from cultivation to post-harvest handling. As part of the SafeGreen project, which aims to strengthen pathogen monitoring in fruit and vegetable production systems, this work focuses on developing a time-efficient and reliable method for detecting pathogens across diverse environmental contexts. This study presents the adaptation of a unified protocol for the collection, processing, and analysis of multiple sample types, including water, soil, environmental surfaces, and fresh produce. The protocol focuses on detecting pathogens of major public health relevance: pathogenic *Escherichia coli*, *Listeria monocytogenes*, and *Salmonella enterica*. The entire process is structured to be completed within 72 hours, from sample collection to preliminary results. It incorporates tailored sampling methods depending on the sample type, followed by cold-chain transport, selective enrichment and rapid molecular detection using Multiplex PCR. Special attention was given to optimizing incubation temperatures, antibiotic concentrations, and collection protocols to minimize microbial viability loss and ensure sample representativeness under varied environmental conditions. By unifying these steps, this protocol facilitates more efficient and reliable environmental surveillance and supports epidemiological studies requiring timely data. Its adaptability across sample types makes it particularly useful for monitoring contamination sources in agricultural settings, water bodies, and food production environments. Ultimately, this contributes to improved public health response strategies by providing information within a significantly reduced timeframe.

P037

Comparative analysis of virulent and resistant *Listeria monocytogenes* phenotypic and genotypic characteristics to identify novel genetic markers of food persistence.

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Abstract

Background -

Listeria monocytogenes is a Gram-positive, rod-shaped, intracellular, foodborne pathogen with robust stress response mechanisms. It is important to study acid resistance since weak acids play an important role in food preservation, along with encountering weak acids in hosts' innate barrier in the stomach.

Methods –

A collection of 224 *L. monocytogenes* strains collected over the years by our lab together with collaborators, will be used in this study. Phenotypic characteristics were tested by an agar-based assay to investigate their growth and survival properties in presence of a range of organic acid food preservatives and food sanitizers. High-quality images of all the plates were scanned every 24 h and pixel intensity were measured using ImageJ.

Results –

Range of sensitivity against acetic and lactic acid was observed. Lineage II strains (n=102) had significantly poorer growth after 72 h on BHI agar with 30mM acetic acid at pH 5.8 compared to lineage I strains (n= 94; $p < 0.05$). This difference was even larger when only clinical isolates were compared between two lineages. Lineage I clinical strains were more resistant to acetic acid than lineage II strains ($p < 0.01$). In contrast, lineage II strains were significantly more resistant to lactic acid (25mM at pH 5.8) than lineage I strains ($p < 0.01$) based on colony growth recorded at 72h.

Conclusion –

Wide range of sensitivity was detected in the whole strain collection. Genome sequence analysis between closely related strains that differ phenotypically with respect to acetate and lactate resistance are underway and would be presented.

P038

Modulation of *Listeria monocytogenes* adhesion and biofilm formation by catecholamines (epinephrine and norepinephrine)

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Abstract

Listeria monocytogenes is a Gram-positive bacterium pathogen responsible for severe infections such as septicemia and meningitis in immunocompromised patient and in pregnant women. While catecholamines such as epinephrine and norepinephrine are known to influence human physiology and the behaviour of many Gram-negative pathogens, their impact on Gram-positive pathogens including Listeria monocytogenes is not well known.

*In our study, th*e effect of catecholamines on the physiology of *Listeria monocytogenes* was evaluated using *in vitro* assays. Growth kinetics have been done in iron-deficient media with or without these molecules. Their impact on biofilm formation was assessed using crystal violet staining and confocal microscopy. Autoaggregation was estimated by optical density measurements and scanning electron microscopy. Finally, bacterial adhesion to intestinal Caco-2/TC7 cells was quantified on BHI plates after 1h30 of infection.

We showed that epinephrine and norepinephrine enhanced the growth of *Listeria monocytogenes* in an iron-deficient culture medium. An increase of autoaggregation, biofilm formation and adhesion to intestinal cells was also observed. These results suggest that catecholamines can modulate the behaviour of *Listeria monocytogenes*. Mechanisms involved remain to be explored.

P040

Proteomic Analysis of the Response on Candida albicans to a Novel Silver Based Antifungal Agent – Ag(PPO)2

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Abstract

Candida albicans is an opportunistic human pathogenic yeast which is capable of causing superficial and systemic disease. Candidosis affects many immunocompromised individuals, and the acquisition of resistance is compromising therapy. Current antifungal therapies can display a high level of cytotoxicity and there is an urgent need for the development of novel therapeutics. There is a historical precedence for the use of silver as an antimicrobial agent, more recently there has been interest silver (I) complexes containing phenanthroline (phen) derived ligands. Based on this we have synthesised a suite of silver (I) *bis*(phenoxazine) complexes and investigated their antifungal activity. The lead compound is a *bis*(propylphenoxazine) silver(I) complex also known as $Ag(PPO)_2$, which has shown strong antifungal activity with low toxicity. A quantitative label-free proteomic approach was employed to ascertain a possible mechanism of action for this complex. This analysis revealed potential disruption to the fungal cell wall and to cellular respiration. The cell wall alterations were confirmed using scanning electron microscopy. The adherent capabilities of the yeast to human tissue were also investigate and a reduction is adherence was observed.

P041

Genomic characterisation of Salmonella Agama from badgers and livestock abortions

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Abstract

Salmonella species are of major public health significance, capable of causing major food-borne outbreaks and disease incidents in various domestic animals. Clinical signs in domestic animals include septicemia, enteritis, respiratory infections and arthritis. Furthermore, salmonellosis has previously been described as a cause of abortions in domestic animals.

The Agri-Food and Biosciences Institute Veterinary Sciences Division (AFBI VSD), based in Northern Ireland (NI), previously assessed a proportion of recent ruminant abortion cases in sheep and cattle hosts and revealed they were due to Salmonella species - specifically serovar Salmonella Agama.

In this study, we use whole genome sequencing (WGS) and downstream bioinformatics to genetically characterise locally derived S. Agama isolates using in silico serotyping, multi locus sequence typing (MLST), phylogenetic analysis, and pangenome analysis tools. Furthermore, we provide a comparison between NI (n=12), mainland UK (GB; n=64), and Republic of Ireland (ROI; n=1) S. Agama isolates to assess local genetic diversity against the wider landscape.

P042

Can We Mitigate Antimicrobial Resistance in Pig Manure for Sustainable Agriculture?

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Abstract

Animal slurry is a cost-effective, and environmentally beneficial fertiliser that can reduce the agricultural carbon footprint. However, microbial studies have linked slurry application to increased levels of pathogenic bacteria such as *Escherichia coli,* and antimicrobial resistance genes (ARGs). This is a concern from a One Health perspective due to the potential transmission of pathogens and ARGs to humans, animals and the environment.

Previous metagenomic and high-throughput qPCR analyses from our laboratory identified that extended storage, composting and anaerobic digestion effectively reduces pathogens, ARGs and mobile resistance elements in pig slurry. In this work, I focus on culturable *E. coli* isolated from these samples. Antimicrobial susceptibility testing was performed on *E. coli* from stored (n = 211) and composted (n = 61) manure. Whole genome sequencing (Illumina short-read) was performed, and genomes were screened for multilocus sequence types, serotypes, antimicrobial and biocide resistance, virulence factors and plasmids.

Preliminary data from early treatment timepoints shows diversity in *E. coli.* A high prevalence of co-resistance against ampicillin, tetracycline, sulphonamide and trimethoprim was observed, encoded by a variety of resistance genes that are likely plasmid-encoded. Characterisation of isolates from later treatment stages is ongoing and will provide further insights into the efficacy of pre-treatment on reducing *E. coli*-associated risks.

P043

Hi-C Proximity Ligation Reveals Microbial Genome Structure and Host–Mobile Element Associations in the Bovine Rumen

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Abstract

The rumen microbiome plays a central role in host digestion and microbial ecosystem function, yet its genome-level structure and associated mobile genetic elements (MGEs) remain difficult to resolve due to its high complexity and density. One approach to overcome this challenge is to use library preparation methods that employ proximity ligation, allowing co-location of DNA sequences originating from the same cell. In this study, we applied Hi-C proximity ligation-based metagenomics to characterize the rumen microbiomes of twelve dairy cows fed one of four distinct diets: Grass, Grass-Clover, Grass-Clover-Plantain, and Grass-Concentrate. This strategy enabled the recovery of 4,194 microbial metagenome-assembled genomes (MAGs), 18,771 viral sequences, and 1,307 plasmids. Many viral and plasmid sequences were successfully linked to their microbial hosts using Hi-C contact data. Among the viral sequences, integrated prophages formed a notable subset, revealing widespread phage–host associations within the rumen ecosystem. Antimicrobial resistance genes were detected in both chromosomal and mobile genetic contexts. Functional annotations revealed a conserved repertoire of fermentative, fibre-degrading, and nitrogen-transforming pathways, with widespread distribution across taxonomically diverse genomes. Carbohydrate-active enzymes were dominated by glycoside hydrolase families, reflecting the fibre-rich nature of the rumen environment. These findings demonstrate the power of Hi-C sequencing to resolve genome structure and mobile element associations in complex microbiomes. The resulting genome-resolved framework offers valuable insights into rumen microbial ecology and sets the foundation for future work on functional dynamics, microbial interactions, and gene transfer in host-associated environments.

P044

The effect of wood-derived xylooligosaccharides on utilization and gut colonisation potential in human- and bovine-associated Levilactobacillus brevis

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Abstract

In Ireland, 11% of the total land area is covered by forest, and around 8.2 million tonnes of wood waste are produced annually from construction, demolition, and deforestation. Wood waste can provide a sustainable source of wood-derived xylan and xylooligosaccharides (XOS), proposed as next-generation prebiotics for human health and animal feed supplements. However, whether human- and bovine-associated commensals demonstrate different preferences and utilisation of these prebiotics and impact on gut colonisation potential to enhance gut health is unknown. In this study, four lactobacilli species, human-associated (Lacticaseibacillus rhamnosus, Lacticaseibacillus paracasei) and human- and bovine-associated (Lactobacillus acidophilus, Levilactobacillus brevis), were investigated for their utilisation mechanisms of xylans and XOS of varying lengths. XOS improved the growth of lactobacilli strains by 5-30%, particularly by mixing different concentrations and lengths of XOS (cocktail mix, CTM). Degradation profiles of xylan and XOS CTM indicated that bacterial utilisation mechanisms were not restricted by host species but were strain-specific, and L. brevis xylanase was produced within 2 h of supplementation. Xylan and XOS also impacted on commensal ability to form biofilms which affects colonisation in the gut. This research provides key insights into the different utilisation mechanisms in human and bovine digestion. Therefore, tailored combinations of valorised xylan and XOS of different lengths could be targeted for different outcomes in animal and human health.

P047

Investigating the Importance of *M. avium* and Alveolar Macrophage Iron Handling in the Context of Chronic Lung Disease

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Abstract

Nontuberculous mycobacteria (NTM) comprise a diverse group of environmental mycobacteria, distinct from *Mycobacterium tuberculosis (M. tuberculosis)* and *M. leprae*. These opportunistic pathogens can cause chronic respiratory infections, especially in individuals with underlying lung diseases, such as COPD, bronchiectasis, cystic fibrosis and asthma. A characteristic feature of these chronic lung conditions is iron dysregulation in the airways, with alveolar macrophages (AMs) accumulating excess intracellular iron—creating a niche that may favour intracellular pathogens, such as *M. avium*, one of the most common causative agents of NTM lung infection.

This study tests the hypothesis that *M. avium* exploits macrophage iron stores to enhance its intracellular replication. Using murine foetal liver-derived AMs (FLAM) and primary human AMs as *in vitro* models for infection, we have shown iron supports enhanced bacterial replication. Treatment with the iron chelator 2,2'-bipyridyl significantly reduces *M. avium* intramacrophage replication, in line with previous reports that iron deprivation limits *M. avium* intracellular replication.

Under defined iron-limited and iron-replete conditions, the iron-associated genes ***bfr*** and ***irtA***, which are homologous to well-characterised iron storage and uptake genes in *M. tuberculosis*, exhibit temporally dynamic and iron-responsive regulation, indicating coordinated bacterial strategies to balance iron acquisition and storage, according to environmental availability.

This work advances our understanding of how altered lung iron metabolism contributes to NTM pathogenesis. These findings underscore the critical role of accessible intracellular iron pools in supporting *M. avium* persistence, and highlight targeting bacterial iron storage and acquisition as promising adjunctive therapeutic approaches.

P049

AMicrobioM: Establishment of a Culture Collection of Seaweed Associated Microbes

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Abstract

The marine environment, specifically the underexplored seaweed-associated microbiome (SAM), holds great potential as a natural reservoir for novel sustainable bioactive compounds. Indeed, seaweed lives in proximity with microbes that are known to produce a variety of bioactive compounds potentially relevant to human and crop health, as well as other biotechnological applications.

To isolate and characterise this resource, seaweed samples (from the West and East coast of Ireland) were rinsed in sterile artificial seawater, and the tissue disrupted in a ball mill. The resulting suspension was diluted 1:10 in artificial seawater and plated on 8 different culture media (including a unique medium containing 0.1X R2A and 1g/L of sterile ground seaweed biomass) and 2 temperatures (8 and 18°C) to maximise the recovery of different bacterial species.

Our isolation methodology yielded a broad spectrum of cultivable bacteria, and ~1,500 isolates were selected for the collection. We found a higher number of recoverable colonies originating from the medium supplemented with host seaweed biomass, indicating that many bacteria likely use their host as a carbon source and therefore require its addition in the medium. Preliminary analysis of 163 isolates revealed the presence of 65 unique species and 7 potentially novel species, highlighting the undiscovered potential of this marine niche.

This work establishes a valuable culture collection of seaweed-associated microbes. All isolates, sequence data, and metadata will be deposited in an open-access repository. The AMicrobioM collection is open for academic collaborations to uncover novel bioactives and enzymes of biotechnological relevance.

P050

AMicrobioM: Exploring Seaweed Associated Microbiomes as a Source of Bioactive Plant Biostimulants

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Abstract

Climate change presents a growing threat to global food systems, requiring the development of sustainable agricultural solutions. One promising avenue lies in bioactive compounds produced by marine bacteria associated with seaweeds. These compounds, from the seaweed-associated microbiome (SAM) include potential crop biostimulants such as growth promoting factors (plant phytohormones and analogues), defence elicitors, and antimicrobials effective on crop pathogens.

Thirteen macroalgal species were collected from the east (Dublin) and west (Galway) coasts of Ireland. Seaweed samples were barcoded via Sanger sequencing, and culturable bacteria were isolated under varied media and growth conditions. 16S rRNA sequencing were used for bacterial identification. In parallel, 16S metabarcoding is being used to characterise microbial diversity and composition in seaweed samples.

We isolated ~ 1,500 bacterial colonies from our summer sampling campaign. 163 colonies have been sequenced to date, identifying 65 species, including *Pseudoalteromonas, Cobetia, Polaribacter*, *Flavobacterium, Algibacter, Halomonas,* and *Cognaticolwellia.* Several isolates showed <97% sequence identity with NCBI references, suggesting the presence of potentially novel species. Notably, *Pseudalkalibacillus hwajinpoensis* was identified and is of interest due to its known inhibitory activity against *Gibberella fujikuroi*, a fungal rice pathogen.

Future work will focus on screening extracts from these isolates to identify bioactive fractions that 1) promote plant germination and growth, 2) trigger plant defence responses and 3) suppress known crop pathogens. The most promising extracts will be fractionated and chemically characterised using HPLC, MS/MS, and NMR. Purified compounds will be evaluated in crop pot trials to assess their effects on growth and disease resistance.

P051

Sustainable bioplastic production by *Cupriavidus necator* using waste cooking oil

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Abstract

Polyhydroxybutyrate (PHB), a biodegradable polymer offers a promising friendly alternative to petroleum-based plastic. *Cupriavidus necator* is a well-known PHB producer that accumulate about 80% of PHB of its DCW. However, the high-cost of carbon sources remains a major barrier to commercial-scale production. Waste cooking oil (WCO) being rich in carbon and abundantly available, represents a low-cost substrate for PHB biosynthesis. In this study, *C. necator* was used to convert WCO to PHB, and process optimization was performed by examining the effects of phosphorus limitation, WCO concentration (2-10%), nitrogen source (ammonium sulfate, sodium nitrate, ammonium nitrate, ammonium chloride, peptone, urea, and yeast extract), and incubation time (24-120 h) on PHB production. Additionally a simplified mineral free production medium containing only WCO and yeast extract was tested for economic viability. Results demonstrated that *C. nectar* could efficiently metabolize WCO, achieving a PHB accumulation of 22.88% under initial conditions. Optimization revealed that the best conditions for PHB production were 0.04% of phosphorus, 2% WCO, yeast extract as a nitrogen source, and 72 - 96 h of incubation. Under these conditions, maximum PHB concentration reached 6.75 g/L, biomass 7.42 g/L, and maximum PHB accumulation 93% the highest reported for this strain. However, the cost reduction trial using a mineral free medium resulted in lower accumulation (74.9%). of, respectively. Characterization analysis of the produced PHB showed similar properties to Sigma standard PHB. Overall, the findings confirm WCO as an effective and economical substrate for PHB production, with potential for industrial-scale application following further optimization.

P052

Exploring the role of Lipoxins as novel therapeutics for chronic wound infections

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Abstract

Chronic wound infections are characterized by a dysregulated healing process and are often exacerbated by the presence of biofilm-forming microorganisms. Persistent inflammation is a hallmark of these wounds, making the modification of the host inflammatory response an attractive therapeutic strategy. Lipoxins, a class of Specialized Pro-Resolving Mediators (SPMs), are endogenously produced to help resolve inflammation. While endogenous Lipoxin A4 (LxA4) is known to improve wound healing and has been shown to affect *P. aeruginosa* biofilm formation, its direct effects on various bacteria are not fully understood.

In this study, we investigated the direct effect of LxA4 on biofilm formation of *P. aeruginosa* and *Staphylococcus aureus* (MSSA and MRSA) strains. Biofilm biomass was quantified using crystal violet staining, and cell viability was assessed via resazurin assays.

Our experiments showed that LxA4 had little to no direct effect on biofilm formation in any of the tested strains. However, treatment with LxA4 (1-10nM) was found to increase *P. aeruginosa* cell viability within a biofilm. Future work will investigate if this increased viability affects the response of *P. aeruginosa* to antibiotic treatment and will also test the effect of LxA4 on *S. aureus* biofilm cell viability.

Beyond this, we will use a keratinocyte scratch assay to explore the therapeutic effects of lipoxin in a host-pathogen system, testing its impact on wound healing in the presence or absence of bacterial cell-free supernatant.

P055

The role of post-translational modifications in protecting bacteriocins during gut transit.

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Abstract

Bacteriocins are ribosomallly- synthesised antimicrobial peptides typically classified as peptides with or without post-translational modifications (PTMs). Bacteriocins have different levels of PTMs and if they are to be used as potential orally delivered therapeutics information on their stability in the gastrointestinal (GI) environment is essential. This is also important where bacteriocins are to be used as food biopreservatives where their breakdown during digestion might be desirable as to not impact the microbiome. We have previously shown that nisin can survive transit through the pig intestine fully intact where it causes changes in the gut microbiota. We selected five bacteriocins to represent a range of diverse levels of PTMs: Capidermicin (leaderless), Pediocin PA-1 (pediocin-like), Leucocyclocin C (circular), Thuricin CD (sactipeptide) and Nisin A (lanthipeptide). These were chosen to conduct a comparative analysis of their stability under conditions mimicking the GI tract. Each bacteriocin was analysed for the presence of potential enzyme cleavage sites (pepsin, trypsin, chymotrypsin and proteinase K) and the resultant post-cleavage fragment masses were calculated. Each bacteriocin was then synthesized or purified before being subjected to enzyme treatment. Aliquots taken at various time points were tested against an indicator strain by well diffusion assay to assess activity followed by matrix-assisted laser desorption/ionization time of flight mass spectrometry to identify fragment/peptide masses. Bacteriocins were also exposed to an array of pH and temperature conditions to assess other aspects of stability. These studies confirmed that the protease sensitivity of the bacteriocins was related to the complexity of their structures.

P056

The Overlooked Threat: Campylobacteriosis Burden, Resistance Patterns, and Molecular Insights in India

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Abstract

Background: Campylobacter is a leading global cause of gastroenteritis, yet remains underdiagnosed in India due to limited surveillance and diagnostics.

Methods: We aimed to study the burden, AMR, and genomic epidemiology of campylobacteriosis in a large multicentric study across North India where 1,127 human diarrhoeal samples and 906 animal samples were tested using culture, real-time PCR, and whole genome sequencing.

Results : Out of 1,127 human diarrhoeal samples, 2.04% were culture-positive, while 9.67% were RTPCR-positive (*C. coli* 49.5%, *C. jejuni* 43.1%, mixed 7.4%). Among 906 animal samples, 24.7% yielded Campylobacter isolates—most from poultry (34.3%) and pigs (30.3%). Species distribution included *C. coli* (59.4%), *C. jejuni* (38.4%), and *C. hyointestinalis* (2.2%), the latter reported for the first time in India. Notably, *C. jejuni* was absent in pig samples. Meat accounted for 27.2% of animal isolates.

Antimicrobial resistance was high: ciprofloxacin (96.9%), tetracycline (50.8%), azithromycin (23.1%), and gentamicin (13.9%), with *C. jejuni* showing higher resistance. Phylogenetic analysis showed 70% of poultry, 20% of goat, and 10% of pig isolates clustered with human strains. Thirteen sequence types (STs) were identified, grouped into CC828 and CC21. Indian animal isolates closely clustered with Vietnamese human and Japanese pig strains, highlighting regional zoonotic transmission and genetic overlap.

Conclusions: Campylobacter is a significantly underrecognized cause of diarrhoea in India. Its highly plastic genome enables rapid adaptation and widespread dissemination. Our findings suggest the circulation of shared, resistant lineages across Asia—likely driven by food chain transmission—highlighting an urgent need for integrated One Health surveillance.

P057

S. aureus survival in the bloodstream modulated by Tca proteins.

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Abstract

Staphylococcus aureus is the leading cause of mortality from sepsis, with over 300,000 deaths in 2019. The bloodstream is a heavily protected niche, where *S. aureus* must withstand many host antimicrobials to establish an infection and cause sepsis. TcaR, a predicted MarR-family transcriptional regulator, has previously been identified to regulate several virulence genes, associating it with adhesion, surface protein expression, and resistance to heat and oxidative stress. As part of the teicoplanin associated locus (tca), TcaR was hypothesised to also regulate *tcaA*, which remodels the cell wall to adapt to the bloodstream environment. Our investigations demonstrate that mutation of *tcaR* confers increased resistance to cell envelope targeting antimicrobial peptides and fatty acids, phenocopying results obtained in a *tcaA* null strain. Contradictory to a Δ*tcaA* strain, loss of *tcaR* does not alter wall teichoic acid abundance in the cell envelope. In addition, a *tcaR* null strain demonstrates increased resistance to hydrogen peroxide mediated killing, whilst loss of *tcaA* does not influence this phenotype. Together these overlapping and disparate phenotypes indicate that TcaR mediated phenotypes are not solely due to regulation of tcaA, with this regulator impacting alternative pathways which facilitate survival in the presence of serum and blood derived antimicrobials. Current investigations are into survival against neutrophils and within whole blood, to further confirm relevance clinically. Additionally, qRT-PCR is underway to establish conditions TcaR is active, and if it regulates key genes such as *tcaA*.

P064

β-Lactams reduce toxicity in Staphylococcus aureus in a teichoic acid-dependent manner

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Abstract

Staphylococcus aureus infections place a significant burden on public health, representing the leading cause of mortality from bacteraemia globally. Toxin production is a key feature of staphylococcal pathogenicity, contributing to extensive tissue damage and driving the onset of sepsis. There is a growing interest in deploying existing antibiotics as strategies to reduce bacterial toxicity. Previous research using β-lactams, a staple treatment option, to reduce staphylococcal toxicity, however, has produced conflicting results regarding their effect on toxin production. Here, we aim to ascertain the effect of β-lactams on toxicity in both reference and clinical strains, comprehensively determine the mechanism responsible, and establish whether this effect is strain-dependent. We show that exposure of *S. aureus* to a range of sub-inhibitory concentrations of various β-lactams targeting each penicillin-binding protein significantly decreased bacterial toxicity as measured by lysis of the HL-60 neutrophil cell line. Analysis of secreted proteins revealed this decreased toxicity was specifically due to decreased secretion of phenol-soluble modulin (PSM) toxins, with the secretion of other toxins not affected. Focusing specifically on the β-lactam cefazolin, we found that PSMs were present inside treated cells, indicating that they were being retained upon exposure. We used isogenic mutant sets to identify both wall- and lipoteichoic acids as crucial in mediating this inhibition of PSM secretion. In conclusion, sub-inhibitory concentrations of β-lactams inhibit staphylococcal toxicity in a teichoic acid-dependent manner, and therefore part of their success as antimicrobials may be attributed to this anti-virulence activity.

P065

Risk mapping Environmental Antimicrobial Resistance in Ireland

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Abstract

Antimicrobial resistance (AMR) is a growing global concern, recognised as a One Health issue due to its ability to spread across species and borders. In Ireland, the absence of comprehensive environmental AMR surveillance and limited data on its transmission to humans and animals make it difficult to assess potential health risks. Additionally, the complexity of analysing publicly available data on AMR drivers within a specific area further restricts efforts to predict where and when AMR health impacts may occur.

We present an integrated approach using Geographic Information Systems (GIS) and machine learning (ML) to analyse the spatial and temporal drivers of environmental AMR. The results are organised into thematic map layers for weighted overlay analysis, and identify AMR risk hotspots across Ireland. Our methodology integrates publicly available data from sectors including agriculture, healthcare, wastewater, and climate. We employ tools such as Google Earth Engine and apply ML techniques such as self-organising maps for clustering, classification, risk assessment, and prediction.

We place particular emphasis on bathing and groundwater sources as key pathways for AMR transmission. This focus is especially relevant in Ireland, where private wells are widely used for both human and animal consumption, and where water-based recreation is an important part of inland and coastal life. Initial results have identified AMR risk hotspots associated with elevated levels of E. coli and enterococci at several bathing locations. We hope our findings will support policy development and help guide surveillance and prevention strategies aimed at curbing the emergence and spread of AMR.

P066

Feeding the Gut from the Mouth: Exploring the Oral-Gut Axis of the Human Mycobiome

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Abstract

The human body, and in particular, the oral cavity and gut, are host to diverse populations of fungi that have been poorly characterised compared to the bacterial fraction of the human microbiome. Moreover, the potential relationship between these spatially separated fungal communities remains largely unexplored, despite its possible implications for health, disease diagnostics, and therapeutic strategies. This study aimed to investigate the diversity and relationship between fungal species inhabiting the oral cavity and gut to assess whether the same species and strains exist across these sites within individuals. Stool and saliva samples from 40 healthy individuals were cultured, enumerated, and subjected to chromogenic differentiation based on colony color and was confirmed by ITS sequencing. Fungi were detected in both stool, (1.45 $\pm$ 0.86) x 10^3CFU/g, and saliva samples, (0.20 $\pm$ 0.22) x 10^3CFU/mL, in 25% (n=10) of individuals, while 35% (n=14) had fungi only in stool, (0.58 $\pm$ 0.86) x 10^3CFU/g, and 2.5% (n=1) only in saliva, 0.08 x 10^3CFU/mL. Among the matched oral and gut samples, *Candida parapsilosis* was the most frequently identified species in both sample types (40%; n=4), followed by *Candida albicans* (10%; n=1). These findings highlight a potential oral-gut axis in the human mycobiome and identify key fungal species for further investigation. Further genotypic and phenotypic characterization of these isolates is ongoing to determine whether the gut mycobiota is seeded from the oral cavity, offering new insights into fungal transmission, colonisation dynamics and disease risk in healthy human populations.

P068

Comparative Thermal Inactivation of Salmonella and Listeria monocytogenes in Bovine Manure and Slurry

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Abstract

Zoonotic pathogens such as *Salmonella* and *Listeria monocytogenes* pose significant public health risks when present in livestock waste used for land application. Both bovine manure and slurry are commonly applied as organic fertilizers, yet they differ in physical and chemical characteristics that in turn may influence pathogen survival. This study aimed to compare the thermal inactivation kinetics of *Salmonella* and *L. monocytogenes* in these matrices at various temperatures (55, 62.5 and 70°C), and to assess the impact of single verses multi-species inoculation on pathogen survival.

The pathogens were inoculated individually and as a mixed species cocktail into bovine manure and slurry, followed by heat treatment in a precisely controlled water bath. Results showed a strong temperature dependence for bacterial inactivation. Complete inactivation occurred within 1-2 minutes at 70°C and 62.5°C. In contrast, at 55°C, both species demonstrated extended survival, with *Salmonella* detectable up to 30 minutes in manure and up to 15 minutes in slurry. The matrix played a significant role, with pathogens surviving longer in manure than in slurry. Notably, mixed-species inoculation resulted in slower inactivation compared to individual inoculations. For example, *L. monocytogenes* was detectable until 2.5 minutes in manure when co-inoculated with *Salmonella*, but only until 2 minutes when inoculated alone, suggesting potential microbial interactions that enhance thermal tolerance.

These findings underscore the importance of tailoring thermal treatment protocols to specific waste types and highlight the need to consider the inoculation strategy when evaluating pathogen reduction.

P069

Beyond in vitro: matrix-specific growth of *Listeria monocytogenes* on ready-to-eat spinach.

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Abstract

Listeria monocytogenes can grow on refrigerated ready-to-eat foods, causing listeriosis, a severe, potentially fatal illness for immunocompromised individuals. While cold-adapted strains are known to up-regulate cold-shock proteins, enhancing survival under low-temperature conditions in vitro, their performance on real food matrices, particularly leafy greens, remains under-explored.

To address this gap, we investigated growth dynamics of cold-adapted *L. monocytogenes* strains on spinach during simulated post-processing contamination. Strains from the DetCon consortium with contrasting growth patterns at 4 °C and 10 °C on agar were compared with a reference isolate. Unlike previous protocols using pre-culturing at 30 °C before cold exposure, strains were directly inoculated into 7 °C medium, reflecting realistic cold-chain conditions, before use in growth trials. Spinach from three production system: laboratory-grown (Trumpet), supermarket-sourced, and organically farmed (Erbette), was inoculated with 100 CFU/g of cold-adapted *L. monocytogenes* and stored at 7 °C following EU Reference Laboratory challenge protocols.

Unexpectedly, a strain previously classified as “slow-growing” showed greater growth than a “fast-growing” strain on organic spinach. In contrast, on supermarket spinach, another “fast-growing” strain exhibited higher growth, while growth on lab-grown spinach remained consistent. In vitro assays at 7 °C revealed similar growth patterns, though one strain was less cold-adapted.

These findings highlight the need to understand cold adaptation mechanisms and strain-specific behaviour interacts with food matrices. This study underscores the importance of considering strain phenotype, food matrix, and environmental adaptation in risk assessments to improve predictive models and mitigation strategies for RTE produce under cold-chain conditions.

P071

Identification of Immunogenic Protein Biomarkers of *Mycobacterium bovis* for Next-Generation Bovine Tuberculosis Diagnostics

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Abstract

Mycobacterium bovis, the causative agent of bovine tuberculosis (bTB) and a member of the *Mycobacterium tuberculosis* complex, is a serious concern to cattle, wildlife, and public health due to its wide host range and potential for zoonotic transmission. Conventional diagnostic techniques, such as sputum microscopy and culture-based assays, have a significant drawbacks such as long processing times, low sensitivity, and the requirement for specialized lab space. To address these challenges, the present study aims to identify multiple specific protein biomarkers of *M. bovis* with high immunogenic potential for integration into an electrochemical biosensor that would offer rapid, sensitive, and field-deployable diagnostic capabilities. Initially, potential antigens were selected based on the existing literature and known associations with *M. bovis* pathogenicity. These antigens were further evaluated using multiple bioinformatics databases and tools to assess key parameters such as antigenicity, immunogenicity, allergenicity, toxicity, and subcellular localization. Through comprehensive *in silico* analysis and rigorous screening, eight candidate protein biomarkers were shortlisted. These proteins demonstrated strong antigenic and immunogenic profiles, were non-toxic and non-allergenic, and were predicted to be surface-exposed or secretory, making them suitable for biosensor applications. The ultimate objective of this work is to incorporate these biomarkers into a next-generation electrochemical biosensor, thereby enabling early and accurate detection of bTB infections. This approach has the potential to significantly enhance disease surveillance, limit transmission, and reduce economic losses in the livestock industry, particularly in low-resource settings with limited access to advanced veterinary diagnostic facilities.

Keywords: Bovine Tuberculosis, Biomarker, Electrochemical biosensor, *Mycobacterium bovis,* Antigen screening

P072

Investigating how *Staphylococcus aureus* manipulates neutrophil metabolism to facilitate intracellular survival

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Abstract

Background

Staphylococcus aureus is an opportunistic bacterium and major global cause of death, compounded by antibiotic resistance and immune evasion. It can survive within host cells, including neutrophils, permitting dissemination and potentially altering neutrophil metabolism and function. We aim to investigate how *S. aureus* alters neutrophil metabolism to facilitate intracellular survival.

Methods

HL60 neutrophil-like cells and primary human neutrophils from healthy volunteers were infected with *S. aureus* isolates exhibiting altered phagocytic uptake and intracellular persistence, alongside parental wild type (WT) JE2. Mutations included: *mprF*::tn, *spA/sbi*::tn, *agr*::tn, and in enzymes of carbon and nitrogen metabolism. Phagocytosis and intracellular survival were assessed using gentamycin protection assays and quantified by enumeration on TSA. Neutrophil metabolism was assessed via SCENITH flow cytometry and pharmaceutical inhibition of key metabolic pathways.

Results

Compared to WT, phagocytosis of *spA/sbi:*::tn increased, and intracellular survival of *mprF*::tn was reduced at 6- and 24-hours post-infection. SCENITH revealed increased glucose and mitochondrial dependence in *mprF*::tn-infected cells, while WT displayed lower dependence. Intracellular survival increased in *mprF*::tn and WT following inhibition of glycolysis, fatty acid transport, and oxidative phosphorylation after 6 hours, and in WT after 24 hours, suggesting transient metabolic shifts. Inhibition of glycolysis and oxidative phosphorylation together increased intracellular survival in both strains, only WT maintained this effect indicating greater adaptability.

Conclusion

Intracellularly persistent *S. aureus* demonstrate greater adaptability to metabolic changes during infection. Understanding how *S. aureus* manipulates these pathways to survive within neutrophils may reveal novel therapeutic targets.

P073

Large-scale *in silico* analysis of polyhydroxyalkanoate synthases in *Pseudomonas*: diverse enzymes for sustainable bioplastics production

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Abstract

Pseudomonas is one of the leading bacterial producers of polyhydroxyalkanoates (PHAs), a sustainable alternative to petroleum-based plastics. PHA synthases (PhaCs) are the key enzymes for their production, but their diversity, distribution, and structure across the genus remain poorly understood.

We constructed and analysed a database of 859 *Pseudomonas* genomes from 186 species using *in silico* methods. PhaC-encoding genes were identified through multiple sequence alignments and classified into the four reported PhaC classes. Amino acid sequences of the identified enzymes were extracted for phylogenetic inference and 3D structure prediction, among other comparative analyses.

Our results show that 738 strains encode two class II PhaCs, consistent with the canonical gene cluster reported for PHA synthesis in *Pseudomonas*. However, six of these strains had a third and one a fourth class II PhaCs. Additionally, 38 genomes encoded one class I PhaC, and two encoded a second. Despite belonging to the same class, many PhaCs displayed substantive sequence variation. Based on phylogenetic inferences, we proposed new subclasses — and even a novel class. Structural modelling, however, revealed a highly conserved 3D structure across the analysed PhaCs.

Genomic context analysis showed that several *phaC* genes were located within putative genomic islands, phages, or plasmids, supporting their acquisition *via* multiple horizontal transfer routes and highlighting their dissemination potential.

To our knowledge, this is the first large-scale study of PhaCs revealing their remarkable diversity across the *Pseudomonas* genus. Further studies are needed to understand their enzymatic properties to produce PHAs with varied monomers and competitive with petroleum-based plastics.

P074

Patterns of antimicrobial resistance in pathogens across One Health in Ireland - An In-silico Approach

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Abstract

Background: Antimicrobial resistance (AMR) is a growing global concern, threatening the effectiveness of treatments for infectious diseases. To support early detection and response efforts, we used an *in-silico* approach to assess AMR patterns in pathogens isolated from humans, animals, and the environment in Ireland.

Methods: In this study, we analyzed 11,577 pathogen isolates from the NCBI Pathogen Detection system, representing 47 clinically important species, including *Escherichia coli*, *Salmonella enterica*, *Staphylococcus aureus*, and others. These isolates were from diverse sources -humans, animals, and environmental samples. Using an *in-silico* approach, we analyzed isolates for resistance genes, plasmids, and virulence factors to build a comprehensive pathogen profile.

Results: Our analysis revealed significant resistance across multiple antibiotic classes. *Klebsiella pneumoniae* showed the highest number of resistant isolates (n=197), followed by *E. coli* and *Shigella* (n=183), and *Salmonella enterica* (n=159). Plasmid detection was highest in *E. coli* and *Shigella* (22.4%), followed by *Salmonella enterica* (15.9%) and *K. pneumoniae* (14.8%). Similarly, *E. coli* and *Shigella* carried the most virulence genes (41.29%), with notable contributions from *S. aureus*, *Bacillus cereus*, and *Yersinia enterocolitica*. An increased resistance pattern was observed in isolates collected during the COVID-19 pandemic, likely linked to high antimicrobial usage.

Conclusion: Rapid and accurate detection of emerging AMR determinants among pathogens is a significant aspect of public health surveillance that helps to combat the spread of AMR efficiently. The present study provides baseline data to implement control measures that can be taken in the hotspots.

Keywords: antimicrobial resistance, in-silico analysis, surveillance, Ireland, one health

P075

Evaluating recombinant expression of phage endolysins in Escherichia coli.

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Abstract

Bacteriophages produce a potent class of lysis-inducing enzymes, referred to as endolysins, that have shown potential as antibacterial agents, including as decontamination agents in food processing environments. Endolysins exhibit species- and strain-specific activity, making them highly valuable antimicrobials. However, producing them at commercially related scales is fraught with issues such as poor solubility and host toxicity during recombinant expression in *Escherichia coli*. This study investigates the impact of expression strain selection and incubation temperature on the recombinant production of two endolysins, MATN from *Staphylococcus aureus* phage (C.A. Boncompain, unpublished data) and T7L, a lysozyme from *E. coli* phage T7. Using small-scale expression experiments and SDS-PAGE densitometry analysis, we assessed protein yield, solubility and inducibility of the endolysins. In the *E. coli* strain Rosetta pLysSRARE, expression at lower temperatures (18–24°C) improved the solubility, inducibility and compared to *E. coli* strain KRX which produced higher yields at higher temperatures, (30-37°C), albeit with reduced inducibility. It was also found that neither strain selection nor temperature could improve the solubility of MATN, warranting further experimentation identifying the domains contributing to the inherent insolubility. Our findings demonstrate the importance of selecting an appropriate host strain and optimising expression conditions for efficient endolysin production, as criteria such as inducibility are important when producing proteins that are potentially toxic to the host strain

P076

Tracking Antibiotic Resistance in Slurry-Exposed Soil for Monitoring of Antimicrobial Resistome Changes

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Abstract

Background. The continued rise on antimicrobial resistance (AMR) is a significant, multifaceted threat to human society. Many AMR genes have their origins and reside in soil environments. The interactions of these environments with farmers, plant and animal agricultural products creates the possibility for AMR transmission to humans.

Slurry spreading is a common agricultural practice with high potential to transmit resistant microorganisms from farm animals to both farmers and the soil. This work aims to evaluate the effect of slurry spreading on the soil antimicrobial resistome.

Methods. Isolates were collected at three different time points: before and two and four weeks after slurry spreading. Isolates were screened for resistance to Amikacin, Ampicillin, Aztreonam, Cefepime, Chloramphenicol, Ciprofloxacin, Colistin, Ertapenem, Tetracycline, and Trimethoprim, by plating onto media with antibiotics. The isolates' antibiotic resistance profile was characterised via antibiotic disc diffusion. DNA isolation was prepared for 16S identification as well as metagenomic shotgun sequencing (bulk soil samples).

Results. Resistance to all antibiotics was obtained at all timepoints. Number of resistant isolates recovered after slurry spreading was double that recovered prior to spreading. At four weeks, the number decreased slightly but was still higher than prior to spreading. Beta-lactam resistance, including Aztreonam, Ampicillin, and Cefepime were common at all time points, with some Ertapenem resistance obtained.

Conclusion. This study is part of the multidisciplinary Resist AMR group, and it highlights how AMR in the soil and certain agricultural practices, such as slurry spreading, can impact he overall dissemination of AMR in the farm environment.

P077

Integrated Constructed Wetlands for One Health AMR Surveillance Across Diverse Wastewater Sources.

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Abstract

Antimicrobial resistance (AMR) is acknowledged as one of the greatest challenges to global health, with estimates that by 2050, AMR will cause 10 million deaths annually unless action is taken. While efforts to fight AMR have traditionally focused on clinical and veterinary settings, but increasing attention is now being given to the role of the environment. This project specifically focuses on Integrated Constructed Wetlands (ICWs) which are shallow, free surface-water wetlands, densely vegetated with Irish plant species to treat through-flowing waters. This study examined the presence of antimicrobial resistant organisms (AROs) in influents and effluents from seven ICWs across Ireland. Microbial parameters assessed include total viable counts, *E. coli*, enterococci, and AROs including ESBL-producing, fluoroquinolone- and carbapenemase-resistant Enterobacterales, *Pseudomonas aeruginosa*, and *Acinetobacter baumannii*. Culture-based methods and MALDI-TOF were used for bacterial isolation and identification.*E. coli* was the predominant species identified, and fewer isolates were recovered from effluents. The antimicrobial susceptibility profile for 16 antimicrobials was determined by disk diffusion for a bank of 380 *E. coli* isolates obtained from both influent and effluent in different ICWs. The profiles were compared for *E. coli* isolates obtained from both antimicrobial-containing selective media (n=240) and from non-antimicrobials media (n=140). Isolates from antimicrobial-containing media higher levels of resistance than those from non-antimicrobial containing media, indicating the importance of appropriate media . These findings support the use of *E. coli* as an effective AMR indicator and demonstrate the utilisation potential of ICWs as a sustainable strategy for mitigating environmental AMR using a One Health approach.

P078

Copper and zinc from slurry applications shape grassland microbial community structure

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Abstract

Soil microbes are crucial in maintaining soil health and enhancing agronomic productivity. This study investigates long-term cow and pig slurry applications on soil microbial community structures, soil chemistry, and herbage yield in a ~50-year Long-Term Slurry (LTS) experiment on a permanent grassland located in County Down, Northern Ireland. Illumina and PacBio rRNA sequencing (analysing 16S and ITS genera) were used to investigate soil samples from the experiment's treatments to depths of 15cm. Slurry type and application rate were found to alter the soil microbial community. In comparison to the non-amended control treatment and inorganic fertiliser treatment, higher slurry rates had the greatest log2 fold change of relative microbial abundance, alongside increasing soil pH and total carbon. Soil pH, copper and zinc concentrations were found to be significant key drivers of soil microbial community shifts (permutation test, $P<0.001$). More specifically, copper primarily influenced 16S and zinc was the greatest influence on ITS genera. CAP analysis revealed that relative abundance changes of 16S genera diverged depending on slurry type and rate, with ITS genera only distinguished by rates of slurry.

P080

Distribution of Antimicrobial Resistance Genes *blaTEM, blaCTX-M, blaVIM,* and *blaKPC* in Irish Farm Soils.

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Abstract

Background

The β-Lactam class of antimicrobials are used to treat bacterial infections in human and veterinary medicine. As with other classes of antimicrobials, resistance to β-lactams has been reported. Development of plasmid mediated resistance is of particular concern as it allows for the horizontal spread of β-lactam resistance genes, such as *blaTEM, blaCTX-M, blaVIM,* and *blaKPC*, between bacteria.

Soils support a complex ecosystem of micro-organisms. Human activity, such as farming or improper/overuse of antimicrobials, contaminates soils with antimicrobials and forces an evolutionary shift in bacteria towards developing or selecting for antimicrobial resistance.

Methods

DNA was extracted from 3000 soil samples obtained from Irish farms. Sample DNA presence was confirmed using 16S qPCR. The presence of the *blaTEM, blaCTX-M, blaVIM,* and *blaKPC* antimicrobial resistance genes was assessed, again by qPCR. The data obtained was then used to create a heat map of antimicrobial resistance gene distribution in Irish farm soils across Ireland.

Results/Discussion

The study highlighted the frequency and distribution of several β-lactam class of antimicrobial resistance genes in Irish farm soils.

Conclusion

The data compiled can be used to inform farmers and policy makers of the distribution of antimicrobial resistance genes across Irish farms. The creation of the heat map can be used as a useful visual aid when interacting with the public, farmers and veterinarians.

P081

Discriminating viable and dead cells in complex microbial communities: combining PMA treatment, Nanopore sequencing, and development of a novel bioinformatics tool

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Abstract

BackgroundDNA from dead cells can overestimate diversity and bias microbiome analyses, obscuring the true composition of viable communities. Propidium monoazide (PMA) selectively inhibits DNA from non-viable cells, enabling more accurate profiling of microorganisms in the environment.

MethodsWater kefir was used as a model and prepared under aerobic and anaerobic fermentation (48 h each), followed by thermal treatments at room temperature (control), 45 °C, 60 °C, or 95 °C for 5 min. At each temperature, samples were subjected to complete or partial heat-killing and treatment with and without PMA (25 µM). This factorial design yielded 168 samples. DNA was extracted using the QIAamp PowerFecal Pro Kit and sequenced using full-length 16S rRNA gene Nanopore sequencing. Bioinformatic analyses included taxonomic classification, alpha and beta diversity metrics, statistical testing and preliminary development of a novel computational tool.

ResultsPMA significantly altered microbial community composition, with stronger effects observed in heat-treated samples. Lactobacillaceae abundance decreased by 20–35% following PMA treatment, while Acetobacteraceae and Leuconostocaceae increased, indicating viability for these taxa. Shannon diversity decreased by 0.4–0.8 units in PMA-treated groups. The largest shifts were detected in 95 °C complete heat-kill samples, whereas partial heat-kill at 60 °C retained a more diverse viable fraction.

ConclusionPMA treatment enhances the resolution of viable microbiome profiling in complex communities. Thermal processing strongly influences viability patterns. This study establishes a framework for accurately assessing microbial viability and introduces a complementary bioinformatics approach to predict the contributions of dead cells, with wider applications in microbial ecology and microbiome research.

P083

Addressing the Global Threat of Antimicrobial Resistance in Ireland: A Comprehensive One Health Approach Integrating Agriculture, Human Health, and EnvironmentAuthors:

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Abstract

The rise of antimicrobial resistance (AMR) poses a significant threat to global health, food security, and the environment. This multidisciplinary project, involving four PhD projects and experts from plant science, microbiology, genetics, AMR research, computer science, bioengineering, and sociology, aims to study environmental and human resistomes in agricultural settings, while analysing practices and policies to recommend institutional reforms.

We will use a multidisciplinary design to investigate and characterise the microbiome and resistome across environmental, plant, animal, and human systems on farms. The research involves identifying the persistence of AMR genes in the environment and analysing risk of transfer to humans. To further develop actionable insights, we will identify transmission risks for farmers *via* the air and farm environment during agricultural practices and subsequently utilise *ex-vivo* organoid platforms to understand the susceptibility of the microbiome to the resistome present on farms. Data collection will include soil and plant sampling, microbiome sequencing, and resistome profiling.

We will conduct a mixed-methods evaluation of agricultural practices and policies governing antimicrobial use in Ireland through document analysis and engagement with farmers and policy actors.

Findings from each project will contribute to a holistic understanding of AMR transmission. The combined results will be disseminated to key stakeholders and guide evidence-based recommendations for improved practices and policy interventions to reduce AMR exposure and transmission risks. Addressing AMR requires a comprehensive One Health approach. By utilising multidisciplinary expertise this project seeks to provide actionable solutions to the AMR challenge and inform sustainable policy reforms.

P084

Synergistic Effects of Plasma-Activated Water and Gentamicin Against *E. coli* Biofilms

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Abstract

Intravesical gentamicin is used to prevent urinary tract infections (UTIs) and urological complications following renal transplantation. As antimicrobial resistance increases, alternative strategies for biofilm eradication are needed. Plasma-activated water (PAW)—water treated using cold atmospheric plasma—has demonstrated antimicrobial and biofilm-penetrating properties. This study investigates whether gentamicin remains active in PAW and whether the combination can synergistically reduce the minimum biofilm eradication concentration (MBEC) of *E. coli* biofilms.

Since *E. coli* is responsible for ~75% of UTIs, reference strains were treated with PAW, gentamicin, and gentamicin reconstituted in PAW. Minimum inhibitory concentrations (MICs) were determined to assess if gentamicin retains its antimicrobial activity in PAW. *E. coli* was then challenged with these treatments in both planktonic form and as biofilms grown on silicone discs to evaluate bacterial eradication and whether gentamicin in PAW more effectively disrupts biofilms than gentamicin alone.

Planktonic *E. coli* was eradicated using PAW alone within 30 minutes, and *E. coli* biofilms were eradicated within 60 minutes. MIC testing confirmed that gentamicin remained active in PAW, showing values comparable to gentamicin in water. This was also observed with other uropathogens tested, including *K. pneumoniae*, *P. mirabilis*, and *E. faecalis*. Notably, gentamicin reconstituted in PAW increased biofilm susceptibility, reducing the MBEC compared to gentamicin alone. Furthermore, this effect was achieved with only 30 minutes of treatment, versus the overnight exposure typically required for MBECs.

Gentamicin retained its antimicrobial activity in PAW and the combination resulted in a lowering of the MBEC required for silicone-based *E. coli* biofilms.

P085

Virological Correlates of Molecular Detection of Avian Influenza Viruses in Water Samples from the Island of Ireland

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Abstract

Emerging infections continue to exert a major burden on human and animal health yet are inherently unpredictable. Avian influenza (AI) poses a risk to wildlife, livestock and humans. While current approaches to monitor AI in the wild have limitations such as low sensitivity and bias towards severe cases, additional methods, such as environmental surveillance, could mitigate these. Furthermore, virus shed into the environment plays a direct role in causing outbreaks. We have recently used human wastewater testing to identify and sequence both human and avian influenza viruses together. Additionally, we have adapted this technique to characterisation of AI in natural wild bird habitats. However, how molecular detection correlates with risk is unknown, particularly regarding estimates for infectivity of environmental viruses. Determination of the environmental drivers of AI infectivity may allow greater prediction and control of incursions. Therefore, we established a system to measure the impact of environmental matrix taken from local waterbodies on influenza A virus (IAV) infectivity *in vitro* and how this relates to PCR detection and sequencing. Remarkably, a lab isolate of IAV (mammalian H1N1 'WSN') retained its infectivity in liquid suspension. However, changes in incubation time, temperature and nature of the matrix influenced its fate; longer incubations, supra-natural temperatures and lower pH reduced infectivity. Work is ongoing to make this model more complex to allow a forward screen of Ireland's waterbodies to identify hotspots – and correlates – of potential viral persistence to enhance regional preparedness against AI.

P086

Investigating the Impact of Chemical Pollution on Viral Infection and Innate Immunity

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Abstract

Emerging zoonotic diseases like influenza A viruses (IAVs) or coronaviruses increasingly reflect complex interactions between the pathogen, their human and non-human hosts and ecological disruptions such as pollution. Indeed, both humans, livestock and wildlife can be exposed to relatively high levels of pollutants, such as lead (Pb). However, the impact of such chemical pollutants on viruses remains relatively underexplored, particularly at a molecular and cellular level. Therefore, we investigated the hypothesis that exposure to Pb exacerbates the risk of zoonotic viral spillover, via altering virus replication dynamics, and innate immune response. A cell culture model of lead-induced changes in host physiology and viral infection was established in human and animal (dog) cell lines with IAV infection. Furthermore, bioactive human and dog interferon (IFNs) were produced and characterised. Preliminary data confirms a cytotoxic effect of Pb on mammalian cells, providing a foundation for subsequent viral infection assays. Established *in vitro* data will be integrated with ongoing monitoring of wild mammal biology and pathogen prevalence, focusing on Eurasian otters (*Lutra lutra*), a sentinel species (closely related to dogs in the order *Carnivora*) for environmental contamination and one involved in zoonotic viral infections. By integrating cellular and ecological data, this interdisciplinary research addresses key gaps on chemical pollutants in disease emergence, which could inform initiatives to limit the impact of zoonosis.

P087

Validation of a catchment contamination risk map (CCRM) for Shiga toxin-producing E. coli (STEC) presence in groundwater wells in Ireland

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Abstract

Unregulated private groundwater sources are widely used for drinking water in Ireland and represent an important transmission route for waterborne pathogens including Shiga-toxin producing Escherichia coli (STEC). This study aimed to develop and validate a tool (catchment contamination risk map, CCRM) to predict groundwater microbial contamination risk in Irish hydrogeological sub-catchments.

The CCRM is based on a multi-criterion risk mapping approach which integrates georeferenced data on human STEC enteritis, potential STEC sources, environmental pathways and receptors. Three catchments (extreme, high and moderate risk categories) were selected for validation field studies. Groundwater wells (n=15-21 per catchment, matched by infrastructural characteristics to include an equal proportion of types) were assessed for E. coli fortnightly (MPN/100 mL, Colilert-18®). One litre of E. coli positive samples was filtered (0.45 µM), filters enriched in mTSB, and Shiga toxin (stx1 and stx2) detection performed on DNA extracts by PCR.

To July 2025, 27 of 93 samples (29%) were E. coli positive, with 14/31 (45%), 7/28 (25%) and 6/34 (18%) in the extreme, high and moderate risk catchment, respectively. There was a significant difference (p=0.03; Fisher's exact test) between extreme and moderate catchments. The stx1/2 detection rate in E. coli positive samples was 37% (10/27), with 43% in extreme (6/14) and high-risk (3/7) catchments, and 17% (1/6) in the moderate. Detection of stx2 (90%) far exceeded stx1 (50%).

Hitherto, findings support the CCRM being as or more useful than currently used groundwater vulnerability classifications. Continued validation is planned over the peak human infection season (to October 2025).

P088

Evolutionary Divergence Between Ligand and Receptor Drives Functional Differences in Mucosal Immunity

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Abstract

Respiratory viruses such as influenza A virus (IAV), coronaviruses, and paramyxoviruses continue to pose major One Health threats. The type III interferon lambda (IFNλs) cytokines constitute one of the first responding immune defences at barriers tissues like the lung. Although primarily studied for their antiviral activity, IFNλs play crucial roles in bacterial, fungal, and helminth infections. IFNλs are a good target for the development of prophylactic measures to control epidemics and pandemics. Like other immune gene families, IFNλs exhibit a high degree of variability but the functional consequences of such genetic differences remain poorly defined and understood. In an antiviral screen of mammalian IFNλs we identified several proteins with species-specific activity. However, the mechanism behind these differences is unknown. Here, we hypothesise that IFN-receptor co-evolution controls species restriction in antiviral activity. To investigate this, we combine computational, molecular and infection assays to determine the mechanistic basis of antiviral potency. Our work has identified potential genetic and structural markers for IFN-receptor incompatibility which are guiding functional experiments. Preliminary work suggests that simple chimeras between functional and non-functional IFNs are unlikely to yield bioactive proteins, necessitating a targeted approach at the receptor interface. Understanding how subtle sequence variation alters IFNλ function will provide insight into cytokine evolution and may inform the development of more effective, One-Health antiviral interventions.

P089

Endolysins for Microbiome Editing - LysH1 is a Novel Enterococcus faecium Phage Endolysin with Activity Against Ruminococcus gnavus in a Simplified Human Gut Consortium.

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Abstract

The human microbiome plays an important role in human health and disease. In recent years, a number of studies have associated changes in the composition of the gut microbiome with inflammatory bowel disease (IBD). One microbe that has been implicated in IBD is Ruminococcus gnavus (aka Mediterraneibacter gnavus). Transient blooms of R. gnavus in the human gut correlate with increased inflammatory symptoms in patients. Interventions that could deplete R. gnavus without causing significant collateral damage could prove useful as a possible therapy. Endolysins are phage-derived peptidoglycan hydrolases that could be promising in this context. We identified, cloned, and expressed a novel Enterococcus faecium bacteriophage lysin, LysH1. Structural analysis of this endolysin revealed a two-domain structure composed of a catalytic domain and a previously uncharacterised domain. We constructed a green fluorescent protein hybrid fusion that allowed us to functionally characterise this unknown domain as a cell wall binding domain. This is the first instance of experimental characterisation of the cell wall binding domain of an E. faecium endolysin. Testing full-length LysH1 against a panel of strains revealed that it has cross-species lytic activity against R. gnavus. Due to the strong lytic activity of LysH1 against R. gnavus, its potential as a novel therapeutic was assessed in a simplified human intestinal microbiota (SIHUMI) model. LysH1 was shown to target R. gnavus in this defined population, without impacting the other members of the consortium. This suggests that LysH1 could be a therapeutic candidate for the depletion of R. gnavus in patients with IBD.

P090

Characterising the Airborne Microbiome and Antimicrobial Resistance in Irish Farm Environments

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Abstract

Antimicrobial resistance (AMR) is a pressing global health threat, with agricultural antibiotic use significantly contributing to the spread of resistant bacteria and antibiotic resistance genes (ARGs) in the environment. While multiple pathways of AMR dissemination have been studied, the role of aerosols as potential vectors remains largely unexplored. The 2021 Environmental Protection Agency (EPA) gap analysis identified air and aerosols as the least investigated transmission routes of antimicrobial-resistant organisms (AROs) and ARGs. This research aims to bridge that gap by characterising the aerosol microbiome in Irish farm environments using both culture-based and culture-independent approaches. Livestock operations, including activities such as animal handling and manure management, generate substantial bioaerosols, potentially exposing farm workers to airborne resistant bacteria and fungi.

A total of n=12 aerosol samples were collected, using the AirPrep™ Cub Sampler-ACD210, during high-exposure activities on a single farm in Ireland. The aerosol microbiome was characterised by sequencing on a NextSeq™ 2000 P4 XLEAP-SBS™ run and by culture-based techniques. This study offers new insights into the role of aerosols as vectors of AMR in agricultural settings and highlights the occupational health risks for farm workers. Data generated will feed into the larger ResistAMR programme tackling AMR from a One Health perspective.

P091

A unique case study of an inter-kingdom emphysematous pyelonephritis infection in a renal transplant patient.

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Abstract

Emphysematous pyelonephritis (EPN) is a rare, life-threatening kidney infection, characterised by the production of intra-parenchymal gas, most commonly caused by *Escherichia coli* in immunocompromised individuals. This case study presents a complex case of EPN in a renal transplant recipient at Belfast City Hospital, with an inter-kingdom co-infection involving bacteria and fungi across multiple medical devices.

A nephrostomy tube, ureteric stent, and Yeates drain were obtained from the same renal transplant patient. The stent and drain were colonised by *Candida parapsilosis* and extended-spectrum beta-lactamase (ESBL) producing *E. coli*, whereas the nephrostomy tube was colonised *by Nakaseomyces glabratus* and ESBL-producing *E. coli*.

All *E. coli* isolates were non-biofilm formers and exhibited multi-drug resistance, including against several antibiotics commonly used in UTI management. The *E. coli* isolated from the nephrostomy tube also exhibited resistance to trimethoprim (MIC = 1024 mg/L), unlike those isolated from the other medical devices, which were sensitive (MIC = 0.125 mg/L). All fungal isolates formed biofilms, resulting in increased antifungal tolerance versus those in planktonic form.

Kidney transplant patients who develop EPN have poor diagnostic outcomes, often resulting in graft loss and mortality. In this case, the allograft was lost due to damage from EPN, but the patient survived. To date, there are no published case reports which document EPN in a renal transplant involving multi-kingdom biofilm infection. This case study highlights the potential for multiple colonisation events of MDR organisms at different medical devices in EPN and the diagnostic and therapeutic challenges posed as a result.

P094

A novel pipeline combining bioinformatic approaches to horizontal gene transfer detection and pathogenicity in prokaryotes

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Abstract

Recent publications highlight the central role of horizontal gene transfer (HGT) in the shaping prokaryote evolution. HGT allows bacteria to rapidly adapt to new environments, while contributing to the spread of antimicrobial resistance and virulence genes. Antimicrobial resistant bacteria are a critical global health issue, therefore it is important to understand HGT to mitigate the threat posed by antimicrobial resistance. A range of computational HGT detection tools have been developed which analyse different criteria to detect HGT events, combining these approaches has been found to improve significantly the quality of predictions. Here, publicly available Salmonella Typhi and Salmonella Typhimurium short reads were downloaded from GenBank. The raw Illumina reads were trimmed using fastp with tail cutting and deduplication enabled. The processed reads were assembled with SPAdes using the 'careful' parameter. CheckM2 was used to assess the resulting assembly quality; genomes with >70% completeness and <5% contamination were considered for downstream analysis. Abricate was used to identify acquired AMR and virulence genes as well as the presence of plasmids. HGT events between genera were identified using MetaCHIP, while Gubbins was used to detect HGT within lineage groups. This workflow will then be used to assess the potential association between HGT events and the presence of AMR genes, virulence genes and plasmids. Future work aims to generate an approach to HGT detection which utilises complementary methods simultaneously, to capture a more comprehensive scope of HGT events within a species or lineage, while also detecting inter-species gene transfer.

P096

Relationship between soil microbial VOCs and nitrogen cycling communities : impact on N_2O emissions

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Abstract

The widespread use of nitrogen fertilisers in agriculture is one of the leading factors behind the rising greenhouse gas (GHG) soil emissions. Despite considerable research effort, N_2O emissions from soil microbial processes remain challenging to predict. However, one overlooked aspect affecting soil N_2O emissions is nitrification inhibition caused by microbial volatile organic compounds (VOCs), which are ubiquitous in soil.

This study aims to determine the impacts of soil VOCs on nitrogen pools, N_2O emissions and microbial communities by manipulating VOC levels through reciprocal oxic-anoxic incubations of soil microcosms. Our working hypothesis is that under oxic conditions, soil VOCs levels remain low while NH_4^+ is converted to NO_3^- by nitrifiers (AOA, AOB). Inducing anoxic conditions should allow a build-up of VOCs and the conversion of NO_3^- to N_2O by denitrifiers. We expect that oxic incubation following anoxic conditions will show reduced conversion of NH_4^+ to NO_3^- due to nitrification inhibition caused by higher VOC levels.

N_2O and VOCs emissions will be analysed respectively with a GC-ECD, and thermal desorption GC-MS. Simultaneously, geochemical analyses will be performed to follow nitrogen conversions (NH_4^+, NO_3^-). Microbial community shifts will be tracked by 16S rRNA gene sequencing and qPCR will be used to quantify the abundance of 11 nitrogen cycle-related genes (e.g., amoA, nosZ). The results will reveal key information about how soil VOCs mediate N cycling and enable future studies on the effect of nitrification-inhibiting VOCs on soil microbial gene expression and their potential to predict N_2O emissions.

P097

Benchmarking K-mer Classifiers for Trace-Level Pathogen Detection in Long-Read Soil Metagenomes

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Abstract

Surveillance of antimicrobial resistance (AMR) in environmental reservoirs such as soil requires accurate detection of low-abundance pathogens. However, the reliable identification of organisms like Klebsiella pneumoniae from long-read metagenomic data remains a significant challenge. Rigorous benchmarking is needed to establish the true detection and quantification limits of bioinformatic classifiers.

Methods: We conducted an in silico spike-in experiment to evaluate the Kraken2/Bracken pipeline. Using Badread, Oxford Nanopore (ONT) reads were simulated from a K. pneumoniae chromosome under three quality profiles (Q10, Q14, Q20). These were spiked into a public soil metagenome at relative abundances from 1% to 0.001%, creating 12 test conditions. Detection rates and Spearman correlations between expected and observed abundances were calculated.

Results: The pipeline detected the target chromosome in all 12 spike-in conditions, with a limit of detection below 0.001%. At 0.01% abundance with Q20 reads, 1,021 reads were correctly assigned. Quantification was highly accurate, with Spearman's rho = 1.0 ($p = 0.083$) across all quality profiles. Higher read quality consistently increased recovery, with Q20 reads yielding the greatest sensitivity at trace abundances.

Conclusion: Kraken2/Bracken demonstrates high sensitivity and quantitative reliability for low-abundance pathogen detection in complex soil metagenomes. This benchmarking framework supports confident application of K-mer-based tools in AMR surveillance and pathogen tracking, even when targets occur at trace levels.

P098

Soil microbial community structure and soil carbon dynamics in fields and hedgerows of Northern Irish Grasslands

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Abstract

It is estimated that globally grasslands store 236 giga tons of carbon (C) in soil belowground, playing an important role in global climate change mitigation. This is particularly important for Northern Ireland (NI) where grasslands cover ~95% of farmland. As part of Soil Nutrient Health Scheme (SNHS) project we selected 250 grasslands across NI, representing wider active grassland management practices across NI, and are measuring soil C stocks, available nutrients, carbon lability, age of carbon via radio carbon dating and microbial community structure (MCS) via phospholipid fatty acid analyses up to 30 cm depth, extending to 1 m depth later this year (2025), sampling both within the farm and nearby hedgerows. Our preliminary results show that grassland MCS is dominated by Gram-negative bacteria, and it significantly varies with depth, and between fields and hedgerows. Relative abundance of Gram-negative bacteria increased with depth while that of Gram-positive and fungi decreased with depth. Soil MCS is also significantly affected by season and field/hedgerow topography. Similarly, C and N content in hedgerow soils are greater than nearby farm soils, showing significant interactions with topography and soil microbial community structure. Our preliminary results show significant increase in C and N stocks from the year 2004 to 2024, indicating C sequestration in NI farms over the last two decades. Currently we are exploring the relationship between soil microbial community structure and carbon dynamics in these grasslands. Findings from our research will provide important insights into grassland carbon sequestration and sustainable land management for NI.

P099

NPower: A Step Towards Solving Europe's Nutrient Excess Crisis

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Abstract

Nitrogen (N) and phosphorous (P) are essential nutrients for food production, however, the accumulation of excess N and P in air, soil, and water leads to severe environmental damage and biodiversity loss. The EU-funded NPower project aims to demonstrate technologies and best practices to recover these nutrients, restoring natural balances. Sustainable nutrient management at scale requires collaboration between scientists, engineers, technologists, policy makers, primary producers, and other key actors to make connections and bridge gaps across multiple sectors. To achieve this, NPower aims to build strong regional networks that promote circular value chains to benefit our society and economy.

Microbiologists represent a key contingent providing vital insights and expertise in the transition towards a circular nutrient economy. The nutrient recovery technologies being developed at NPower demonstration sites in Spain include the use of bacterial cocktails for the treatment of pig manure, polyphosphate-accumulating organisms and denitrifying bacteria for nutrient recycling, and the production of fertilisers and biogas from anaerobic digestion processes, while testing the safety and microbial diversity of recovered fertilisers.

The NPower Irish Cluster together with Regional Clusters in Spain, Belgium, and Finland serve as knowledge hubs to evaluate the applicability and transferability of solutions to each region. This approach will facilitate the future implementation of nutrient management solutions across Europe's diverse landscapes, addressing national and regional realities. The Irish Regional Cluster is open to microbiologists interested in shaping best management practices and policy recommendations through co-creation workshops with diverse actors from researchers and practitioners to industry and government representatives.

P100

Role of *Candidatus* Moeniiplasma glomeromycotorum endobacteria in the interaction between arbuscular mycorrhizal fungi and crop plants

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Abstract

Arbuscular mycorrhizal fungi (AMF) colonize plant roots and play an important role in plant nutrition (i.e. phosphorus and nitrogen uptake). This nutritional symbiosis can be beneficial for plant growth, yield, and productivity which has created a research momentum on their possible use in agriculture as an alternative to fertilizers. Despite AMF being the most studied mycorrhizal group, few studies focus on AMF endobacteria. *Candidatus* Moeniiplasma glomeromycotorum (*Ca*Mg) is the most prevalent endobacteria of AMF and its function remains unknown. We hypothesize that *Ca*Mg might not only influence AMF biology but also the association between AMF and host plants. The aim of this project is to explore the role of *Ca*Mg in the AMF symbiosis with barley (*Hordeum vulgare*) and alfalfa (*Medicago sativa*), two important crop plants in Ireland.

Greenhouse experiments will be carried by inoculating barley and alfalfa seedlings with AMF in the presence and absence of *Ca*Mg. Throughout the experiment image-based phenotyping will be used to assess the health status of plants and after harvesting, classical metrics will be assessed (i.e. shoot and root biomass, root system architecture, tissue mineral content, root colonization and sporulation success) to compare the different treatments. Additionally, plant cellular responses to *Ca*Mg presence will be measured by analysis of differential gene expression, proteomic and metabolomic changes between treatments.

This will be important to establish the role of *Ca*Mg on its host and to broaden the research perspective of plant-microbe interactions as potential solution for more sustainable agriculture practices.

P101

Understanding the metabolic cross talk to enable sustained nasal colonisation by Staphylococcus aureus

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Abstract

Staphylococcus aureus asymptomatically colonises the anterior nares of approximately 30% of healthy individuals, acting as a reservoir for subsequent infections. Despite extensive research, the specific metabolic mechanisms enabling persistent nasal colonisation remain incompletely understood.

To elucidate the metabolic pathways and adaptive strategies utilised by *S. aureus* during nasal colonisation and identify key factors distinguishing successful colonisers to better develop therapeutics for decolonisation.

Transposon mutagenesis of the USA 300 derivative JE2 *S. aureus* strain was performed to generate mutants of genes in key carbon and nitrogen metabolic pathways. Mutant strains were evaluated in vitro for growth kinetics and metabolite utilisation patterns in intracellular survival using the gentamicin protection assay. In vivo colonisation fitness was assessed using a murine model of nasal colonisation.

S. aureus can survive within macrophages and nasal epithelial cells. In order to do this, *S. aureus* must adapt to environments with differing nutritional compositions and selection pressures. I have shown that it utilises pyruvate and the citric acid (TCA) cycle metabolism to survive intracellularly in human macrophages. Nitrogen metabolism, as well as TCA cycle metabolism have roles in successful colonisation of a murine model, although only the requirement for TCA cycle intermediates has been shown in a nasal epithelial cell line cultured in nutrient rich media.

S. aureus nasal colonisation depends on sophisticated metabolic adaptations that exploit host and site-specific nutrient sources while modifying the local microenvironment. In the future, these findings may provide new targets for anti-colonisation strategies and enhance our understanding of host-pathogen metabolic interactions.

P102

A Pilot Study to Assess Biofilm Formation capabilities of bacterial Isolates from Burn Wound patients attending the National Burns Unit, St. James's Hospital

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Abstract

Burn injuries continue to remain a clinical burden in high-income, developed countries due to their complexity, associated co-morbidities and intense resource requirement for treatment. Biofilms are important targets in burn treatment to decrease the staggering mortality rates in this patient cohort, of which 42-65% can be attributed to infection. Previous audit in the unit found key pathogens isolated from burn wounds included S*taphylococcus aureus* (35.5%), other coagulase negative *staphylococci* (21.4%), and *pseudomonas* species (8%). In patients requiring prolonged admission over 30 days, there was a trend of these key pathogens isolated throughout hospital stay. Infection had a statistically significant correlation with mortality in the unit ($P<0.01$). Early Identification of patients colonised with biofilm forming, antimicrobial resistant strains of bacteria impact appropriate and effective antimicrobial stewardship.

Pathogenic bacteria were collected from tissue swab samples from patients admitted to the National Burns Unit in St James's hospital over a 3-month period. Bacteria were identified employing culture-based techniques and MALDI-ToF mass spectrometry. Biofilm forming capabilities of bacterial isolates was measured using the crystal violet biofilm assay.

A diverse variety of bacteria were isolated from these wound swabs, namely key pathogens found in audit. All isolates tested were capable of forming biofilms. Furthermore, the biofilm formation capabilities of isolates collected from individual patients over time varied widely.

This pilot study has revealed the high level of biofilm forming bacteria in wound infections. Biofilms are inherently resistant to antibiotics and these results could influence burn wound care in the future.

P103

Investigating the use of plasma activated hydrogels against biofilms

Bryan Sim, Thomas Thompson, Brendan Gilmore

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Abstract

Biofilms are complex extracellular matrices that various bacteria and pathogens form which adhere to surfaces. Unlike planktonic cells, biofilm-associated bacteria exhibit better resistance to antibiotics and other environmental stresses. This poses a problem in healthcare settings, such as contamination of medical devices and repeated infections of patients.

Staphylococcus aureus biofilms were grown for 48 hours on different material surfaces and rinsed with PBS, including 24-well plates, titanium discs and porcine skin models. Various hydrogel formulations were developed, which are then used to coat the materials with grown biofilms. Both hydrogel and material are then treated with cold atmospheric plasma to form a thin hydrogel film across the surface of the material, over the biofilms. TSB were then added along with Resazurin assay and incubated to quantify the activity of the bacteria post plasma treatment. The remaining culture was serially diluted, plated onto TSA, incubated overnight, and number of colonies formed were counted. Hydrogels were also loaded with different concentrations of antibiotics to test for combined effectiveness against biofilms. Different plasma treatment times were used to check if the duration of treatment affects the extent of damage done to the biofilm.

Our results have shown that plasma treating titanium discs coated with hydrogels has managed to reduce bacterial counts present on the titanium discs, regardless if hydrogel is loaded with antibiotics or not.

P104

"Innovative TB Detection: A High-Performance qPCR-based assay for diverse clinical samples"

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Abstract

Tuberculosis (TB) remains a major global health challenge, particularly in resource-limited settings. We developed an optimised quantitative PCR (qPCR) assay to enhance TB detection sensitivity. The assay was evaluated using 101 inactivated sputum samples from patients in South Africa, in collaboration with Stellenbosch University. Among 51 Xpert MTB/RIF Ultra-positive samples, our test detected 50, yielding a sensitivity of 98.04%. Of the 50 Xpert-negative samples, 41 were not detected, resulting in a specificity of 82.00%. ROC curve analysis showed an AUC of 0.98, with optimised cutoffs achieving 96.08% sensitivity and 92.0% specificity.

To assess the assay's versatility, we conducted a pilot study using inactivated faecal swabs, a sample type easier to collect—especially in children or individuals unable to produce sputum, such as those with HIV co-infection. We collected 56 faecal swabs from patients at a respiratory clinic in Bristol. Of 21 patients with active TB (9 pulmonary, 12 extrapulmonary), 6 were detected—all pulmonary TB. The 3 undetected pulmonary cases were microscopy-negative, suggesting low bacterial load.

Importantly, the use of inactivated samples enables testing in Containment Level 2 laboratories, significantly expanding access to molecular diagnostics in settings without high-containment facilities. This proof-of-concept study highlights the assay's potential to improve TB diagnostics, particularly for pulmonary TB. Further investigation is needed to optimise detection in extrapulmonary TB, including exploration of additional sample types

P105

Exploring the combined effects of gentamicin and plasma-activated water against uropathogenic biofilms

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Abstract

The rising levels of antimicrobial resistance call for new antibacterial and antibiofilm treatment options. A possible approach is the use of plasma-activated water (PAW), which has been shown to have antibacterial properties and can improve antimicrobial susceptibility by disrupting biofilms. In this project we explore the possible synergy between PAW and the aminoglycoside gentamicin against uropathogenic biofilms.

In this study, *E. coli* biofilms were grown on silicone discs, as *E. coli* is the most common pathogen associated with urinary tract infections. Both planktonic and biofilm forms of *E. coli* reference strains were exposed to deionised water, PAW, gentamicin and gentamicin reconstituted in PAW to perform kill curves. Minimum inhibitory concentrations (MIC) and minimum biofilm eradication concentrations (MBEC) were obtained to evaluate whether gentamicin can retain antimicrobial activity in PAW, and if their combined effects reduce the MBEC of uropathogenic biofilms.

Treatment of planktonic cells with PAW produced complete eradication within 30 minutes, while *E. coli* biofilms were eradicated within 1 hour. Gentamicin was able to retain its antimicrobial properties when reconstituted in PAW, exhibiting a similar MIC to gentamicin alone. Furthermore, the combined effects of gentamicin and PAW can lower the MBEC of uropathogenic biofilms, presenting a promising approach for tackling antimicrobial resistance.

P107

Inactivation of *Listeria monocytogenes* using UVC-LED and Far-UVC Technologies

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Abstract

Listeria monocytogenes provides significant challenges for food producers in Ireland, in particular, those producing 'Ready-To-Eat' (RTE) foods. UVC radiation with traditional low-pressure (LP) lamps is approved for surface decontamination and in the processing of some foods. As a result of a global effort to reduce mercury emissions, there is a push to consider alternative lamps such as UVC-LED and Far-UVC to replace LP-mercury containing lamps. In addition to being mercury free, Far-UVC has significantly reduced health and safety concerns with regards to human exposure. Assessment of a strain collection of *L. monocytogenes* isolates from diverse sources in Ireland has highlighted the intra-species phenotypic heterogeneities in tolerances to certain stresses. This study aimed to assess the antilisterial efficacy to UVC using UVC-LED and Far-UVC technologies and to screen the collection to identify strains with higher tolerance. Strains were individually cultured in TSB to stationary phase and inoculated in 5mL of PBS at a cell concentration of approximately 6 log CFU/mL. Strains were separately treated with UVC-LED (265nm) and Far-UVC (222nm) at UV doses targeting roughly 2-4 log reductions. Treated strains were recovered and enumerated on non-selective agar (TSA). Log reductions were calculated for each strain and UVC treatment combination. Results revealed significant differences in sensitivities to UVC radiation across the strain collection using both lamps. This phenotypic data highlights the need for consideration of strain-level variability when implementing control measures for *L. monocytogenes* and the need for a representative set of isolates when assessing the efficacy of risk reduction measures.

P108

Novel Phosphorus Utilisation Pathways in the archaea Haloferax volcanii DS2.

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Abstract

Phosphorus is an essential but often limited nutrient, particularly in extreme environments such as hypersaline lakes. While bacterial phosphorus acquisition is well understood, archaeal mechanisms remain poorly characterised. *Haloferax volcanii* DS2, a model halophilic archaeon, offers an opportunity to investigate these understudied pathways.

A combination of comparative genomics and laboratory experiments was used to assess the phosphorus metabolism of *H. volcanii*. BLAST analysis identified homologues of known bacterial phosphorus metabolism genes. Concurrently, growth experiments tested the organism's ability to utilise various complex phosphorus sources including adenosine monophosphate (AMP), polyphosphate (PolyP), phytate, and phosphonoacetate.

The results showed that *H. volcanii* can utilise AMP and PolyP as effectively as inorganic phosphate, demonstrated by comparable increases in protein concentration and optical density. Free phosphate accumulation in these cultures indicated active enzymatic breakdown of complex sources. Notably, many known bacterial phosphate metabolism genes were absent or misannotated in the genome, implying the presence of novel, archaeal-specific mechanisms. A focused growth study with AMP revealed a delayed but enhanced growth phase, suggesting a physiological adaptation period.

These findings indicate that *H. volcanii* employs distinct strategies for phosphorus acquisition, potentially involving unknown enzymes. The study highlights the need to explore archaeal nutrient cycling and provides a foundation for biotechnological applications in phosphorus-limited environments such as sustainable agriculture and marine ecosystem management.

P109

Meta-Hort: Investigating Agronomic and Postharvest Influences on the Microbiome of Horticultural Produce

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Abstract

Meta-Hort is an applied research project exploring how agronomic and postharvest practices influence the microbiome of horticultural produce. The current focus is on strawberries and button mushrooms—two high-value, perishable crops with known microbial complexity and wide consumption. These crops serve as ideal models to investigate how factors such as ripeness at harvest, storage duration, and growth media impact microbial community structure and stability.

The project integrates shotgun metagenomic sequencing and traditional culturing methods to characterize microbial diversity across different production scenarios. Protocols for DNA extraction from complex plant and fungal matrices have been optimized, alongside host DNA depletion strategies. Bioinformatic workflows—employing tools such as Kraken2, GTDB, and HUMAnN3—are used for taxonomic and functional profiling. In parallel, viable microbial isolates are being recovered for downstream probiotic potential screening.

By comparing locally grown and imported production models, Meta-Hort aims to determine how cultivation and supply chain practices modulate food-associated microbiomes. Future work will extend to ex vivo colon fermentation models to assess microbiome-host interactions relevant to gut health.

This research supports the development of microbiome-informed horticultural strategies and contributes to sustainable, health-promoting food systems.

P112

Identification of the Antimicrobial Potential of Methylglyoxal Present in Natural Honey Commercialized in the City of Cali

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Abstract

Antimicrobial resistance is a critical global health challenge, demanding sustainable and innovative therapeutic alternatives. Honey, a natural product with bioactive properties, is a potential antimicrobial agent due to its physicochemical characteristics, including methylglyoxal (MGO), a reactive compound with documented bactericidal effects.

This study evaluated the antimicrobial potential and MGO content of two natural honeys (liquid and creamy) commercialized in Cali, Colombia. NMR spectra confirmed the presence of MGO in low concentrations (quantification in progress). Antimicrobial activity was assessed using the well-diffusion assay against *Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 9027, and *Candida albicans* ATCC 10231.

Preliminary results showed inhibition of *E. coli* with halos of 0.4–0.5 cm for honeys supplemented with 0.07% MGO, comparable to the 0.5 cm halo from the positive control. Creamy honey alone produced a 0.5 cm inhibition halo, consistent with NMR findings. No inhibition was observed against *C. albicans* at 0.07% MGO; higher concentrations (0.08%–1.5%) are being tested to determine the minimum inhibitory concentration. Evaluation with *P. aeruginosa* and naturally resistant strains is ongoing.

These findings suggest that the type of honey and MGO content, likely influenced by floral origin, modulate antimicrobial activity. This study, **"Identification of the Antimicrobial Potential of Methylglyoxal Present in Natural Honey Commercialized in Cali, Colombia,"** is part of the project **"Synergistic Evaluation of Natural Extracts for the Enhancement of Honey Bioactive Properties,"** highlighting the ecological and biomedical relevance of Colombian honey for AMR mitigation and the valorization of local apiculture.

Keywords: methylglyoxal, honey, antimicrobial resistance, well-diffusion assay, NMR

P113

Potassium iodide enhances the antimicrobial activity of plasma-activated water.

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Abstract

Background: Plasma-activated water (PAW) generates reactive oxygen and nitrogen species (ROS/RNS) in acidic aqueous conditions, offering a promising, chemical-free antimicrobial approach. The addition of potassium iodide (KI) may enhance this effect by producing reactive iodine species (RIS) such as hypoiodous acid (HIO).

Methods: Spark-discharge PAW was generated and supplemented with low concentrations of KI (10–100 µM). Chemical analysis measured changes in H_2O_2 and NO_3^- levels, while antimicrobial activity was assessed against planktonic cells of Escherichia coli, Listeria monocytogenes, and Salmonella enterica, as well as biofilms. Scavenger assays identified key ROS/RNS contributing to the antimicrobial mechanism. Stability of PAW-KI was monitored over 14 days at 4 °C.

Results: KI addition led to a dose-dependent increase in H_2O_2 concentration, from ~1.2 mM (PAW alone) to ~1.8 mM at 30 µM KI, and a ~17% increase in NO_3^-. PAW + KI achieved complete inactivation of E. coli and L. monocytogenes within 3 minutes, while PAW alone required over 10 minutes; S. enterica showed partial resistance. Biofilm eradication was also significantly enhanced with KI. Equivalent H_2O_2 + KI solutions failed to match the rapid killing, suggesting in situ RIS generation plays a crucial role. Scavenger assays confirmed the involvement of H_2O_2 and singlet oxygen, while ozone and superoxide were dispensable. PAW-KI remained chemically and microbiologically stable for at least 14 days at 4 °C.

Conclusion:

KI-enhanced PAW generates RIS in situ, enabling rapid, broad-spectrum antimicrobial action and biofilm eradication. This residue-free, stable, and scalable system offers a sustainable disinfection platform for food safety and clinical use.

P114

Characterisation of two Proteus phage endolysins with different metal cofactor requirements

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Abstract

Proteus mirabilis is a Gram-negative uropathogenic bacterial species responsible for many catheter-associated urinary tract infections (CAUTIs). Due to the growing rates of antimicrobial resistance among CAUTI pathogens, novel antimicrobial solutions are urgently needed. Bacteriophage endolysins are an emerging class of antimicrobial agents, but no endolysins from *P. mirabilis* phages have been reported to date. Here we describe two new *Proteus* phage endolysins, LysPM1 and LysPM2, containing Peptidase_M15_3 and CHAP domains, respectively. We experimentally confirmed that both enzymes have antibacterial activity against several *Proteus* spp. members when used in combination with outer membrane permeabilizers, including EDTA, citric acid, chloroform, as well as against frozen bacteria. Both enzymes have pH optimum around 7-8 and are thermally stable up to 60°C. Both endolysins completely lost the lytic activity upon treatment with EDTA, suggesting that divalent cations participate in the catalytic mechanism; the addition of Ca^{2+} and Zn^{2+} to LysPM1, and Ca^{2+} and Mn^{2+} to LysPM2 partially restored the activity. No detectable effect of NaCl concentration was observed in the range of concentrations tested.

Our study confirms LysPM1 and LysPM2 as first experimentally characterised *Proteus* phage endolysins. These findings could guide further development of phage-derived lytic enzymes for treatment of *Proteus* infections.

P115

The Application of Bioengineering strategies to evaluate the potential of Nisin in Microbiome Editing

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Abstract

Bacteriocins are antimicrobial peptides produced by some bacteria that can have a narrow or broad spectrum of activity against other bacteria. Their modes of action are varied and complex, targeting key cellular processes and structures to kill or inhibit growth. The specificity of bacteriocins to certain microbes limits the evolutionary pressure of resistance development. It is this specificity that significantly differentiates bacteriocins from broad-spectrum conventional antibiotics. Nisin, the most studied bacteriocin, has a long history of use as a food preservative and it is becoming increasingly apparent that it shows promise as a clinical alternative to antibiotics due to its effectiveness against drug-resistant pathogens. However, its use in clinical settings is limited owing to its sensitivity to proteolytic destruction in the upper gastro-intestinal tract and broad activity spectrum. Bioengineering strategies present promising avenues for overcoming the limitations of nisin, thereby enhancing its application as a potential precision therapeutic. This study aims to apply these experimental approaches to produce peptide derivatives with combinations of desirable properties with a view to precise sculpting of the human microbiota with capacity to positively impact human health.

P116

Evaluation of bile salt hydrolase inhibitor efficacy for modulating host bile profile and physiology using a chicken model system.

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Abstract

Gut microbial enzymes, bile salt hydrolases (BSHs) are the gateway enzymes for bile acid (BA) modification in the gut. This activity is a promising target for developing innovative non-antibiotic growth promoters to enhance animal production and health. Compelling evidence has shown that inhibition of BSH activity should enhance weight gain by altering the BA pool, host signalling and lipid metabolism. We recently identified a panel of promising BSH inhibitors. Here, we address the potential of them as alternative, effective, non-antibiotic feed additives, for commercial application, to promote animal growth using a chicken model. In this study, the in vivo efficacy of three BSH inhibitors (caffeic acid phenethylester, riboflavin, carnosic acid) were evaluated. 7-day old chicks (10 birds/group) were either untreated or they received one of the specific BSH inhibitors (25 mg/kg body weight) via oral gavage for 17 days. The chicks in treatment groups consistently displayed higher body weight gain than the untreated chicks. Metabolomic analysis demonstrated that BSH inhibitor treatment led to significant changes in both circulating and intestinal BA signatures in support of blunted intestinal BSH activity. Consistent with this finding, liver and intestinal tissue RNA-Seq analysis showed that carnosic acid treatment significantly altered expression of genes involved in lipid and bile acid metabolism. Taken together, this study validates microbial BSH activity inhibition as an alternative target and strategy to antibiotic treatment for animal growth promotion.

Keywords: Bile salt hydrolase (BSH), Bile acid, Non-antibiotic growth promoter, Chicken model, Inhibitors, Liquid Chromatography coupled to Tandem Mass Spectrometry (LC-MS/MS), Metabolomics, Transcriptomics, Host-microbiota interaction.

P117

Microbial and Metabolic Mediators of Host–Microbe Dialogue in Early Life

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Abstract

Gut-resident bacteria produce a diverse array of metabolites, either directly synthesized or derived from dietary and host substrates. These compounds influence gut health, maintain homeostasis, and enable communication between the gut and distant organs. Critically, these biochemical interactions shift with microbial dynamics across life stages, forming unique species and strain level metabolic fingerprints. Yet, the common ground lies in their function—the metabolites they produce or transform.

Among early colonisers, Bacteroideales (notably *Bacteroides*) and Proteobacteria (including *E. coli*) are abundant but remain under characterised in terms of their contributions to the host–microbe interactions. We hypothesised that these groups offer essential, diet-responsive metabolic functions that support initial colonisation and foundational gut development.

To investigate this, infant stool samples were collected at one week, one month, and three months of age. Bacterial isolates were tracked longitudinally and identified through whole-genome sequencing to capture species- and strain-level variation. Complementary metabolomic and phenotypic analyses revealed diet-modulated production of immunomodulatory lipids and bile acids in vitro.

Longitudinal tracking confirmed that key strains persisted across the first three months of life, indicating those that are transient, tourist and those that form a stable foundation.

These findings position early colonisers as metabolically active, significant, and responsive to dietary input. Specific strains may hold potential as probiotics to support both early-life development and long-term health.

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