



MICROBIOLOGY DAY

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27TH OF MAY 2025

BOOKLET

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AND

Natacha JANISZEWSKI, creator of the logo of this event

THANKS TO



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MICROBIOLOGY DAY PROGRAM

- 8.30 am **Ouverture des portes**

- 9.00-9.05 am

Introduction

Alexia Damour

- 9.05-10.30 am **Assemblée Générale MicroBio-NA**

- Introduction: Remerciements, Intervention de M. Philippe Nauche,
Présentation du réseau Microbio-NA
- Présentation des actions de communication
- Présentation du PSGAR MIE
- Présentation des avancées des 3 groupes de travail du Réseau
- Conclusion et ouverture.
- Temps de questions

- 10.30 am-11 am **Coffee break**

- 11 am-12.35 am **Physiology of pathogenic microorganisms**

Chairperson: Eloise Bertiaux and Karine Frénal

- 11.00 am: **Philippe Bastin, Invited Speaker, Paris**

Cutting it too short: an intriguing interplay between flagellum and cell growth in trypanosomes

- 11.45 am: **Maxime Lefranc, Bordeaux**

Emergence of Wickerhamomyces anomalus invasive infections in injecting drug users

- 12.00 am: **Antoine Loquet, Bordeaux**

New solid-state NMR approaches to decipher the molecular organization of fungal and yeast cell wall

- 12.15 am: **Damien Achard, CEVA**

Impact of 150 mg/kg dosage of paromomycin on gut microbiota in healthy calves

- 12.35 am-1.40 pm **Lunch break**

- 1.40 pm – Intervention pour congrès SFM

- 1.45 pm-3.00 pm **Host-pathogen interaction**

Chairperson: Alexia Damour and Jean-Christophe Meunier

- 1.45 pm: **Gaetan Ligat, Invited speaker, Toulouse**

Molecular biology of host-HCMV interactions in brain tumors and therapeutic innovations

- 2.30 pm: **Camille Lefevre, Limoges**

Polymorphism study and characterization of functional domains of the human cytomegalovirus portal protein pUL104

- 2.45 pm: **Cynthia Cavillon, Poitiers**

Impact of physioxia and role of HIFs in skin infection by Usutu

- 3.00 pm – 4.15 pm **Poster session & coffee break**

- 4.15 pm–5.20 pm **Environmental microbiology**

Chairperson: Hélène Agogué and Sophie Nolivos

- 4.15 pm: **Émilie Sevin, TOOPI Organics**

Expanding the frontiers of urine upcycling : LACTIPI PLUS® a new product by TOOPI Organics

- 4.35 pm: **Abiola Saheed Akinwale, Pau**

Pharmaceuticals in waste water treatment plants: microbial diversity and biodegradation under pharmaceuticals pressures

- 4.50 pm: **Ana Luzia Lacerda, La Rochelle**

The plastisphere: a comprehensive description of geographic and temporal patterns across the Mediterranean Sea and the Atlantic Ocean

- 5.05 pm: **François Maclot, Bordeaux**

Deciphering the complex ecology of plant and mycoviruses in wild grasses by analysing the virome in individual plants

- 5.20–5.30 pm **Awards and concluding remarks**

Chairperson: **Alexia Damour and Karine Frénal**





PHYSIOLOGY OF PATHOGENIC MICROORGANISMS



Philippe Bastin, Invited speaker

Trypanosome Cell Biology Unit, Institut Pasteur, Paris

Cutting it too short: an intriguing interplay between flagellum and cell growth in trypanosomes

Trypanosomes are flagellated parasitic protists responsible for various infectious diseases such as sleeping sickness or Chagas disease. Their flagellum is essential for cell motility and division, but also for morphogenesis and attachment to tissues in their insect vector. Flagellum length is modulated to adapt to the different environments they encounter during their life cycle. In this presentation, we will report investigations in *Trypanosoma brucei* to monitor but also to alter flagellum growth, with various consequences on motility and morphogenesis. Surprisingly, the picture is not black and white, revealing an unexpected interplay between flagellum elongation and cell growth.

Maxime Lefranc

Microbiologie Fondamentale et Pathogénicité – MFP – UMR 5234 – CNRS – Université de Bordeaux

Emergence of *Wickerhamomyces anomalus* invasive infections in injecting drug users. M. Lefranc, M. Desnos-Ollivier, P. Baudino, K. Boukris-Sitbon, C. Plaisant, F. Dalle, F. Lanternier, S. Imbert

Wickerhamomyces anomalus (*Candida pelliculosa*) is a yeast found in various environmental niches and biotechnological processes. It is barely involved in human infections, even if some reports suggest its epidemic potential, particularly in premature infants. Recently we faced an increased number of *W. anomalus* invasive infections in our institution. Therefore, we decided to investigate them from a nationwide perspective. We analyzed retrospectively consecutive *W. anomalus* invasive infections reported to the French National Reference Center for Mycoses and Antifungals between 2013 and October 2024. Moreover, we studied their genetic diversity by short-tandem repeat (STR)-based genotyping, including a broad range of *W. anomalus* invasive and non-invasive clinical isolates.

Regarding centers that participated exhaustively in the surveillance during the whole study period, 43 *W. anomalus* invasive infections (37 fungemia, 4 osteoarticular and 2 eye infections) were recorded from 34 patients, by 16 French tertiary care centers, including 9 patients only in ours. Surprisingly, 24 patients (70.1%) were injecting drug users (IDUs). Moreover, we found a significant increase in the mean annual number of *W. anomalus* infections between 2013–2020 (2 cases/year) and 2021–2024 (4.5 cases/year) ($p=0.02$). This increase was mainly driven by the emergence of IDU-related infections, as 14 of them (58.3%) occurred after 2021. STR genotyping was performed on 48 *W. anomalus* isolates

and showed a great genetic diversity among isolates involved in IDU-related infections. Nevertheless, we identified identical genotypes shared by several patients and a cluster of close genotypes involving 4 IDUs from 3 different centers, suggesting a common infection source. Furthermore, for one IDU with fungemia, we isolated concomitantly a *W. anomalus* from the cotton used to filter the drug before injection and kept by the patient. Interestingly, these 2 isolates shared the same genotype, highlighting the increased risk associated with poor drug use practices.

This nationwide retrospective study highlights the recent emergence of *W. anomalus* invasive infections in IDUs, which seems to be associated with common exogenous sources and/or poor drug use practices. Nevertheless, prospective studies and environmental samples are needed to identify potential sources and prevent further infections.

Antoine Loquet

Chimie et Biologie des Membranes et des Nano-objets
CBMN – UMR 5248 – CNRS – Bordeaux INP – BSA

New solid-state NMR approaches to decipher the molecular organization of fungal and yeast cell wall

Most microbes possess a "cell surface", i.e. a critical surface architecture which can have several purposes: protection against the external environment, a mimicry strategy to evade host defenses, or a pathogenic molecular weapon to damage host cell membranes. Molecular understanding of the microbial cell envelope remains one of the most active research fields in microbiology, because many essential processes related to the survival of the microbe and its ability to infect host cells are taking place at the cell surface and are driven by surface components. The ability to study such cell surfaces at a molecular level is of prime interest, to understand how pathogenic and parasitic microbes can survive, proliferate and carry out their harmful action. Current state-of-the-art analytical approaches to study cell surface have two main drawbacks: (i) they are destructive or rely on the extraction of weakly-bound molecules, requiring chemical or biochemical methods prior to the analysis. (ii) Because each component is quantified in isolation, the global organization and interplay between components is missing. Here we present our recent advances using solid-state NMR spectroscopy to investigate the molecular organization of fungal, bacterial and yeast cell surface at atomic resolution: the architecture of *Aspergillus fumigatus* cell wall during its conidial morphotype transition (1) and the molecular distinction between cell wall and capsular polysaccharides in *Cryptococcus neoformans*.

(1) Solid-state NMR molecular snapshots of *Aspergillus fumigatus* cell wall architecture during a conidial morphotype transition. Lamon G, Lends A, Valsecchi I, Wong SSW, Duprè V, Lafont F, Tolchard J, Schmitt C, Mallet A, Grélard A, Morvan E, Dufourc EJ, Habenstein B, Guijarro JL, Aimanianda V, Loquet A. PNAS 2023

(2) Molecular Distinction of Cell Wall and Capsular Polysaccharides in Encapsulated Pathogens by In Situ Magic-Angle Spinning NMR Techniques. Lends A, Lamon G, Delcourte I, Sturny-Leclerc A, Grélard A, Morvan E, Abdul-Shukoor MB, Berbon M, Vallet A, Habenstein B, Dufourc EJ, Schanda P, Aimanianda V, Loquet A. J Am Chem Soc. 2025



Impact of 150 mg/kg dosage of paromomycin on gut microbiota in healthy calves

Pascal Butty¹, Anne Trotell¹, Damien Achard¹, Y. Jaquemet², L. Hernandez², J. Le Guennec³, P-Y. Moalic³, F. M'Zali⁴

¹ Ceva Santé Animale, Libourne, France, ² Pigase, 227 chemin de Sapeins, 01480 Chaleins, France, ³ Bio Chêne Vert Finalab, 4 rue Théodore Botrel, 22600 Loudéac, France, ⁴ Aquitaine Microbiologie / university of Bordeaux, Bâtiment Bordeaux Biologie Santé, 2 rue Dr Hoffmann Martinot, Bordeaux, France

Objective

This study aimed to evaluate the effect of oral paromomycin administration over five days on resistance development in the intestinal commensal microbiota of healthy calves.

Materials and Methods

Twenty-five healthy calves aged 12 to 21 days, originating from seven French farms, were included in the study. They were housed collectively, fed milk replacers twice daily, and randomized into a treatment group (Gabbrovet Multi®, Ceva Santé Animale, 150 mg/kg daily for five days; n=23) or a control group without treatment (n=2). Daily monitoring included fecal consistency, depression scores, and appetite (evaluated on a 0-2 scale). Fecal samples were collected at four time points: before treatment (Day-1), during treatment (Day+4), and after treatment (Day+20, Day+36). Samples were immediately frozen at -80°C then transferred to the microbiology lab for isolation and microbiological analysis of commensal Escherichia coli strains. For each fecal sample, 20 purified and randomly selected E. coli colonies were selected.

To manage the high number of strains, related strains were grouped using mass spectrometry (MaldiTof Biotyper Compass Explorer software). Minimum inhibitory concentrations (MICs) for paromomycin were determined using a customized microdilution method (UMIC), along with aminoglycoside antibiograms following CLSI guidelines. Resistance evolution was tracked by comparing strain data at different time points relative to Day-1, using the CA-SFM kanamycin breakpoint for Enterobacteriaceae.

Results

A total of 1,780 E. coli strains were isolated and analyzed. On Day-1, commensal E. coli populations comprised 69% susceptible and 31% resistant clones. Treatment with paromomycin did not induce resistance in previously susceptible E. coli clones. Instead, the treatment eliminated a substantial proportion of susceptible E. coli, resulting in a temporary predominance of preexisting resistant clones within the intestinal microbiota. This disruption was short-lived, as susceptible clones began to reappear post-treatment. Indeed, by Day+36, susceptible E. coli accounted for 40% of the population, compared to 7% on Day+20, 10% on Day+4.

Conclusions

No evidence of resistance acquisition in commensal E. coli was observed during this study. MIC values for individual clones remained stable across sampling points. The paromomycin treatment effectively disrupted the microbiota by favoring preexisting resistant clones, but the flora gradually reverted to its initial composition after treatment cessation. The high bactericidal dosage used in this study likely played a role in preventing resistance development. However, caution is advised for lower dosages (e.g., prophylactic regimen), which are known to increase the likelihood of resistance emergence.



HOST-PATHOGEN INTERACTIONS



Gaëtan LIGAT, Invited speaker

Virus and brain cancer, Infinity Institute, Toulouse

Molecular biology of host-HCMV interactions in brain tumors and therapeutic innovations

Out of all brain cancers, glioblastoma multiforme (GBM) is the most common and most aggressive malignant primary brain tumor. Despite considerable research efforts, treatment options for GBM remain limited. Strong evidence has established that up to 15% of human cancers are directly caused by viruses. For example, hepatitis B virus has been shown to be responsible for the development of liver cancer. Herpesvirus such as Epstein-Barr and Kaposi's sarcoma virus have also been implicated in the development of various cancers such as lymphoid malignancies and epithelial cancers. In addition, other viruses such as Human cytomegalovirus (HCMV), are strongly suspected of having a link with GBM since the seminal work of Charles Cobbs, who discovered the presence of the virus in a large proportion of brain tumors. Indeed, although HCMV affects 50–80% of the worldwide population, it is, strikingly, found in more than 95% of malignant gliomas. Recent studies have shown for the first time that HCMV leads to the carcinogenesis of human mammary cells and induces glioblastoma upon infection of human astrocytes.

Interestingly, many studies have suggested that anti-HCMV therapy can restrain GBM progression *in vitro* and *in vivo*. Valganciclovir has demonstrated a promising survival benefit in both newly diagnosed and recurrent GBM. Furthermore, the hypothesis of an oncogenic role of HCMV raises hope for the development of a vaccine that could prevent the occurrence of various cancers such as GBM, either directly caused by HCMV and/or for which HCMV is an aggravating factor. Thus, HCMV offers a starting point for the identification of new GBM drivers, together with an alternative target for the development of new therapeutic strategies by targeting host factors or viral proteins.



Polymorphism study and characterization of functional domains of the human cytomegalovirus portal protein pUL104

Camille Lefèvre, Claire Gourin, Gaël Champier, Sébastien Hantz

Human cytomegalovirus (HCMV) is an opportunistic pathogen infecting a large proportion of the world's population. Usual antiviral treatments such as ganciclovir target viral polymerase. They are responsible for toxicity and the emergence of resistance mutations, restricting their use. Therefore, it seems necessary to develop inhibitors of other steps of the viral replication cycle. Letermovir inhibits the encapsidation of the viral genome within neoformed capsids, a step performed by the terminase complex (pUL56-pUL89-pUL51). However, its use has already led to the emergence of resistance mutations in all three subunits of the complex (1).

The pUL104 portal protein, which assembles into a dodecamer to allow viral DNA to enter neoformed capsids, interacts with the terminase complex (2). Consequently, pUL104 could also be a target of letermovir. Moreover, no functional domain has been described within this HCMV protein to date. Through Sanger sequencing of pUL104 under letermovir-induced selection pressure in clinical strains, we identified a mutation substituting an alanine by a serine at position 115, potentially involved in resistance. Furthermore, structural modeling of pUL104 revealed the presence of a potential leucine-zipper domain (position 261 to 282, allowing protein-protein interaction), a nuclear localization sequence (3) (NLS ; position 625 to 635, allowing protein to enter the nucleus) and a monomer interaction domain (position 440 to 459, allowing dodecamerization of pUL104).

These domains were mutated in BAC-HCMV by "en passant" mutagenesis ; the four leucines of the leucine-zipper domain were substituted by alanine, while the NLS and the monomer interaction domain were deleted. Viral replication was studied after transfection of recombinant BAC-HCMV into human fibroblasts of the MRC-5 line. To study the NLS and the monomer interaction domain, the UL104 gene mutated at these domains was cloned in fusion with m-Cherry or GFP fluorescent proteins within the pCI-neo vector. The impact of the mutations on the nuclear localization of pUL104 was observed by confocal fluorescence microscopy. Data from our microscopy analyses, antiviral assays and replicative capacity tests have already demonstrated the importance of these functional domains for viral replication.

Thus, this study is essential to deepen our understanding of the mechanism of action of letermovir and associated resistance mechanisms, as well as the functioning of the portal protein pUL104, essential for the encapsidation step and a potential new antiviral target.

References

- (1) Chou and Watanabe, 2024
- (2) Dittmer et al., 2005
- (3) Gaël Champier, 2006

Impact of physioxia and role of HIFs in skin infection by Usutu

Cynthia Cavillon, Sonia Lacourt, Nicolas Lévéque, Hamid-Reza Rezvani, Charles Bodet, Magali Garcia

Usutu virus (USUV) is an emerging arbovirus belonging to the Orthoflavivirus genus that is actively circulating in Europe and recently in Nouvelle Aquitaine. Its transmission to humans, particularly following inoculation into the skin during a mosquito's blood meal, can lead to influenza-like illness and meningoencephalitis.

The skin is divided into 3 successive layers from the deepest to the most superficial: the hypodermis, the dermis and the epidermis. The dermis is made up of fibroblasts, immune cells and blood vessels. The epidermis, composed mainly of keratinocytes, is not vascularised. Oxygenation occurs via the passive diffusion of oxygen carried in the vessels of the dermis. The oxygenation rate (ppO_2) of the epidermis varies from 8% in cells close to the dermis to 1% in cells further away.

We previously demonstrated that keratinocytes are permissive to USUV infection leading to an innate immune response at a ppO_2 of 20%. However, data highlights the impact of ppO_2 on the immune response and viral replication, notably via the Hypoxia Inducible Factor (HIF)-1 α or HIF-2 α , transcription factors whose expression is regulated according to the level of cellular oxygenation.

Given the role of HIFs in cell physiology and the different skin ppO_2 compared to conventional culture, the objectives are to determine the impact of skin physioxia and the role of HIFs during USUV skin cell infection. Viral replication, the immune and metabolic responses were characterized in control keratinocytes and keratinocytes underexpressing HIF-1 α or HIF-2 α .

First stimulated with DMOG, a hypoximimetic molecule preventing the degradation of HIFs under a ppO_2 of 20%, a decrease in viral replication was noticed in cells underexpressing HIF-1 α compared to control cells as well as a significantly increased antiviral response, both at basal level and during infection. On the contrary, viral replication and antiviral response were similar in cells underexpressing HIF-2 α to control cells.

Then, cells were infected under a ppO_2 of 1% in a hypoxia chamber, showing a decrease in viral replication in cells underexpressing HIF-1 α compared to control cells. In addition, cells cultured at 1% O₂ expressed more antiviral response genes during infection than those cultured at 20% O₂.

Taken together, these data suggest a role for HIF-1 α during USUV infection of keratinocytes.





ENVIRONMENTAL MICROBIOLOGY



Emilie SEVIN

Responsable R&D, TOOPI Organics, Loupiac de La Réole

Expanding the frontiers of urine upcycling : LACTIPI PLUS® a new product by TOOPI Organics

Claire Peyruchat, Kenza Boubekri, Hany Abdo, Emilie Sevin

TOOPI Organics is the first start up that commercialises urine-based and fermented biostimulants.

To address agro-ecological transition and save drinking water resources, TOOPI Organics developed a waterless urine collecting system. Then, the stabilised collected urine is upcycled as fermentation media to allow Plant Growth Promoting Bacteria (PGPB) proliferation in industrial bioreactors. In a circular approach of the nutrients cycle, the fermented urine-based biostimulants are applied on various field crops and in arboriculture to improve vegetative growth of plants.

In 2024, TOOPI Organics launched a new microbial product: LACTIPI PLUS®, based on *Lacticaseibacillus paracasei* fermentation on optimised urine-based media. Thus, TOOPI Organics acquired a scale up line, from 1L to 5kL bioreactors, to achieve its industrial process development. Nowadays, this product is manufactured on a 5kL-industrial bioreactor and distributed in France and Belgium.

With more than 20 fields trials, this product proved its agronomic efficiency. Indeed, TOOPI Organics demonstrated plant nutrients absorption efficiency and crop biomass increase for grapevine, pines, olive trees in response to LACTIPI PLUS® application. Moreover, when applied in partial or total substitution of mineral fertilisers, trials on different crops tend to indicate similar agronomic yields.

Abiola Saheed AKINWALE

Institut des Sciences Analytiques et de Physico-Chimie pour l'Environnement et les Matériaux - IPREM - UMR 5254 - CNRS, Université de Pau et des Pays de l'Adour

Pharmaceuticals in waste water treatment plants: microbial diversity and biodegradation under pharmaceuticals pressures

Abiola Saheed Akinwale, Bahia Khalfaoui Hassani, Claire Gassie, Marie Morère, Mathilde Monperrus and Rémy Guyoneaud

Pharmaceuticals are a major group of emerging pollutants primarily used in human and veterinary medicine to prevent or treat diseases and maintain quality of life. The detection of pharmaceuticals in several environmental compartments, such as wastewater treatment plants (WWTPs), has become a global scientific and regulatory concern. Conventional wastewater treatment processes are unable to remove many pharmaceuticals because of the mixture's complexity and their low biodegradability

(Voravich et al., 2023). Many studies have investigated the potential of microbial degradation of pharmaceuticals (Narayanan et al., 2023), because of their ecofriendly and economic advantages over other conventional treatment approaches. In our study, we characterized and isolated microbial communities (enrichments) and strains from two WWTPs under two enrichment conditions (in the presence and absence of carbon source) and ketoprofen and oxazepam exposure at 1 ppm and 1000 ppm. Differences in microbial community composition were obtained between pollutant enrichments primarily due to the impact of individual pharmaceutical compounds and the availability of additional nutrient sources irrespective of the WWTP. Also, we have noticed concentration-dependent trends in microbial associations under high and low concentrations of ketoprofen and oxazepam. High dominance of a few bacteria genera was recorded in the 1000 ppm ketoprofen and oxazepam enrichments, while more diverse bacteria genera dominated the enrichments at 1 ppm concentration of the compounds. Moreover, most of the isolated strains in all conditions are phylogenetically closer to strains that have been reported for degradation of different organic pollutants. Overall, this study will contribute to a more comprehensive understanding of the dynamics of microbial community structure associated with pharmaceutical compound degradation using indigenous microbial diversities in the aerobic sludge of full-scale WWTPs and give future clues to obtain efficient strains/consortia for their biodegradation.

REFERENCES

- Ganthavee, V., and Trzcinski, A. (2023). Removal of pharmaceutically active compounds from wastewater using adsorption coupled with electrochemical oxidation technology: A critical review, *Journal of Industrial and Engineering Chemistry*, Volume 126, Pages 20–35, ISSN 1226-086X, <https://doi.org/10.1016/j.jiec.2023.06.003>.
- Narayanan, M., Kandasamy, S., Lee, J., & Barathi, S. (2023). Microbial degradation and transformation of PPCPs in aquatic environment: A review. *Heliyon*, 9. <https://doi.org/10.1016/j.heliyon.2023.e18426>.

Ana Luzia Lacerda

Littoral ENvironnement et Sociétés – LIENSS – UMR 7266 – CNRS, Université de La Rochelle

The plastisphere: a comprehensive description of geographic and temporal patterns across the Mediterranean Sea and the Atlantic Ocean

A.L. Lacerda^{1,2}, R. Casotti³, J.F. Briand⁴, V. Lenoble⁵, S.M. Lorenzo⁶, F. Kessler⁷, A. Barre⁴, C.M.M. Pérez⁶, V.F. González⁶, J.M.A. Garda⁶, C. Murano³, V. Donnarumma³, E. Oreste⁷, H. Joyce⁸, C. Hannon⁸, R. Nash⁸, F. Orange⁹, J. Frias⁸, M.L. Pedrotti².

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Plastics at sea are a global ecological threat, not only as debris but also as a substrate for microbial communities, known as the ‘plastisphere’. The factors shaping these communities remain uncertain; however, variations between virgin and recycled polymers, combined with environmental differences, may influence both early and mature plastisphere development. As part of the JPI-Oceans project ‘Integrated approach to the fate of microplastics towards healthy marine ecosystems (MicroplastiX)’, this study provides the most extensive and geographically consistent analysis of the marine plastisphere to date. Three polymers – virgin low-density polyethylene (LDPE), recycled polypropylene (PP-PC), and the biopolymer polylactic acid (PLA) – were incubated in situ across six locations in Europe and South America. Using standardized protocols and DNA metabarcoding, we assessed how biogeography, environmental variables, polymer type, and exposure time influence prokaryotic and eukaryotic groups colonizing plastics. Results show that biogeography and seasonal factors shape plastisphere communities



more than polymer type, with distinct differences between Mediterranean and Atlantic sites. Over 70 prokaryotic taxonomic orders and 50 eukaryotic groups were identified, including potential plastic degraders (*Oleibacter*, *Alcanivorax* spp) and pathogens (*Pseudomonas*, *Candida* spp), which were present in multiple sites. Understanding the factors shaping plastisphere communities is essential for assessing species dispersal, biodegradation potential of plastics at sea, and the public health risks that should be considered when developing global strategies to mitigate ocean plastic pollution.

François Maclot

Biologie du Fruit et Pathologie – BFP – UMR 1332 – INRAE – Université de Bordeaux

Deciphering the complex ecology of plant and mycoviruses in wild grasses by analysing the virome in individual plants

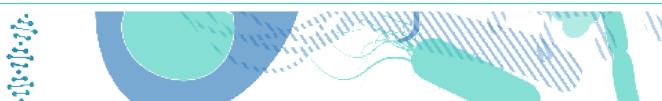
François Maclot, Armelle Marais, Thierry Candresse, Sébastien Massart

Before the domestication of plants, plant viruses were co-evolving with wild plants growing in mixed species communities, thereby potentially resulting in complex interactions (antagonism, commensalism, mutualism). The development of agriculture deeply modified ecosystems, altering the dynamics of virus-plant pathosystems and accelerating the rate of virus evolution and emergence. High-throughput sequencing technologies have now enabled more comprehensive studies of viromes at different scales, from individual plants to entire ecosystems, offering insights into virus ecology within agro-ecological landscapes. Recent virome studies revealed diversified and largely unknown viral communities in natural ecosystems, with high rates of co-infection and a high abundance of persistent or cryptic viruses. However, many studies focused on plant pools, showing the viral richness but missing important ecological information such as viral prevalence, co-infection and spatial distribution of virus infection. In this context, we conducted a study in the Natural Park “Burdinale-Mehaigne” (Belgium) to explore virus diversity and ecology in individual plants and pooled samples of two grass species, *Lolium perenne* and *Poa trivialis*, from pastures and high biological value grasslands.

Using a virion-associated nucleic acids (VANA) metagenomic approach on 143 individual plants, the study found for both host species a higher virus prevalence (79%) in pastures, dominated by phytoparaviruses, whereas grasslands showed lower virus prevalence (48%), with a predominance of mycoviruses. High prevalence of co-infected plants was observed but with low virus accumulation. For yellow dwarf viruses (B/CYDVs), comparison of sequenced genomes within and between plants suggested a high genetic diversity, and a novel BYDV-like species was identified in *Poa trivialis*. Additionally, *Lolium perenne* was identified as a key virus reservoir, hosting up to 25 different virus species compared *Poa trivialis* infected by up to 16 viruses. These findings underscore the significant role of wild plant communities as virus reservoirs, influencing virus ecology across both natural and agricultural landscapes.



POSTER



	Last name	First name	Title
1	ACHARD	Damien	Impact of 150 mg/kg dosage of paromomycin on gut microbiota in healthy calves
2	ANDREU PAZOS	Marina	Identification of Key Bacteria involved in the transmission of antibiotic resistance using a One Health approach
3	ARNOULD	Kathyanna	Characterisation of TbSeipin : a key protein involved in lipid droplet formation in <i>Trypanosoma brucei</i>
4	ARVY	Nathalie	Lipid Droplets: Beyond Mere Energy Storage, Key Players in Viral Propagation
5	AUTIN	Michel	Interplay between HIV-1 integration and transcription
6	BARROUILHET	Sophie	First evidence of a mercury resistance mechanism in an anaerobic bacterium: impact on mercury accumulation and methylation
7	BENOIT	Nathanaël	A ubiquitin-like protein controls assembly of a bacterial Type VIIb secretion system
8	BOUREIMA ABDOU	Bachir	Transfer of Integrative and Conjugative Elements and Chromosomal DNA in <i>Mycoplasma hominis</i> under Cell Culture Conditions
9	BRUNET	Thibault	Étude des résistances au céfiderocol de bactérie GRAM-négatives d'origines cliniques
10	CASCIANI	Silvia	Hydrogen at depth. An investigation into life limits in underground reservoirs.
11	DENIMAL	Marina	Cytométrie en flux et étalement : Deux approches complémentaires pour mieux comprendre l'effet des antibiotiques
12	FAUTRAS	Yoann	Study of the aerobic and anaerobic sedimentary bacteriobiota
13	FIÉVET	Valentin	Investigating species compositional and metabolomic dynamics in a <i>Fusarium</i> synthetic community under abiotic pressures
14	GOUDENECHÉ	Pierre	Exploring the Role of Maternal Nutrition and Gut Microbiota in Neuroprotection Against Neonatal Hypoxia-Ischemia. Study in Ratss
15	GRAU	Henri	Améliorer l'action de la colistine face à <i>Klebsiella pneumoniae</i> multirésistante via des nanoparticules chargées en farnesol
16	JACQUET	Chloé	Fidaxomicin, a potential new drug for combinatorial therapies against <i>Mycobacterium abscessus</i>
17	JARRY	Fabien	Antibiogramme rapide par SdFFF à partir de flacons d'hémocultures positifs à <i>Escherichia coli</i>
18	JEHANNE	Quentin	Recurrent <i>C. jejuni</i> and <i>C. coli</i> infections: the contribution of genomics to the characterization of two cases

19	JESSU	Amélie	Pathophysiology of Mpox virus skin infection.
20	KALLISSERI PARAMBIL	Anagha	NMR SPECTROSCOPY TO STUDY PROTEIN ASSEMBLIES ON BACTERIAL MEMBRANE
21	KHOURY	Joy	Co-occurrence between Fusarium and bacterial microbiota in wheat grains
22	LACERDA	Ana Luzia	Projet EMERG : Exosome microbien et Risque sanitaire – Intérêt d'une Gestion One Health des enjeux liés aux grippes zoonotique
23	LAVIELLE	Sarah	Investigating the role of Gut Microbiota in Glioblastoma development
24	MOUVILLE	Clémence	Phage MDAΦ entry mechanisms shape pilin variant populations of <i>Neisseria meningitidis</i>
25	NAVARRO	Marine	Fungal microRNAs: key players in interaction
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1 – Damien Achard, DVM (Doctor in Veterinary Medicine)

Ruminant Innovation Leader, CEVA

Impact of 150 mg/kg dosage of paromomycin on gut microbiota in healthy calves

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Objective

This study aimed to evaluate the effect of oral paromomycin administration over five days on resistance development in the intestinal commensal microbiota of healthy calves.

Materials and Methods

Twenty-five healthy calves aged 12 to 21 days, originating from seven French farms, were included in the study. They were housed collectively, fed milk replacers twice daily, and randomized into a treatment group (Gabbrovet Multi®, Ceva Santé Animale, 150 mg/kg daily for five days; n=23) or a control group without treatment (n=2). Daily monitoring included fecal consistency, depression scores, and appetite (evaluated on a 0-2 scale). Fecal samples were collected at four time points: before treatment (Day-1), during treatment (Day+4), and after treatment (Day+20, Day+36). Samples were immediately frozen at -80°C then transferred to the microbiology lab for isolation and microbiological analysis of commensal Escherichia coli strains. For each fecal sample, 20 purified and randomly selected E. coli colonies were selected.

To manage the high number of strains, related strains were grouped using mass spectrometry (MaldiTof Biotype Compass Explorer software). Minimum inhibitory concentrations (MICs) for paromomycin were determined using a customized microdilution method (UMIC), along with aminoglycoside antibiograms following CLSI guidelines. Resistance evolution was tracked by comparing strain data at different time points relative to Day-1, using the CA-SFM kanamycin breakpoint for Enterobacteriaceae.

Results

A total of 1,780 E. coli strains were isolated and analyzed. On Day-1, commensal E. coli populations comprised 69% susceptible and 31% resistant clones. Treatment with paromomycin did not induce resistance in previously susceptible E. coli clones. Instead, the treatment eliminated a substantial proportion of susceptible E. coli, resulting in a temporary predominance of preexisting resistant clones within the intestinal microbiota. This disruption was short-lived, as susceptible clones began to reappear post-treatment. Indeed, by Day+36, susceptible E. coli accounted for 40% of the population, compared to 7% on Day+20, 10% on Day+4.

Conclusions

No evidence of resistance acquisition in commensal E. coli was observed during this study. MIC values for individual clones remained stable across sampling points. The paromomycin treatment effectively disrupted the microbiota by favoring preexisting resistant clones, but the flora gradually reverted to its initial composition after treatment cessation. The high bactericidal dosage used in this study likely played a role in preventing resistance development. However, caution is advised for lower dosages (e.g., prophylactic regimen), which are known to increase the likelihood of resistance emergence.

2 - Marina ANDREU PAZOS

*Anti-Infectieux : supports moléculaires des résistances et innovations thérapeutiques
- RESINFIT - UMR 1092 – INSERM, Université de Limoges*

Identification of Key Bacteria involved in the transmission of antibiotic resistance using a One Health approach

Marina Andreu Pazos, Thibault Stalder

Antibiotic resistance (AR) has become a global health threat. Many of the AR genes found in today's pathogens originate from environmental bacteria and are often carried on mobile genetic elements like plasmids, which can be transferred between bacteria by horizontal gene transfer. Determining how AR genes spread within and across microbiomes and ecosystems is crucial to understand AR emergence and spread. Previous studies have shown that some bacteria carry more AR genes and plasmids than others in the same community. In this study, we hypothesize that some bacteria disproportionately contribute to the spread of these genes across ecosystems, acting as keystones in the transmission of AR.

To address this hypothesis, we use a One Health approach, analyzing the IMG/PR database that gathers plasmid sequences from human, animal and environmental sources. To identify which bacteria are more frequently associated with AR plasmids and more widely spread between humans, animals and environments, we use a network approach that connects plasmids to their hosts and habitats. Preliminary analysis of the database, which includes 29120 plasmids derived from metagenomes, revealed three major classes: Clostridia, Gammaproteobacteria, and Betaproteobacteria, and Bacilli. Notably, the top bacterial genera most connected to AR plasmids and widely spread across ecosystems include *Escherichia* spp., *Staphylococcus* spp., and *Salmonella* spp., all of which have several plasmids associated with at least one AR gene. Future work will incorporate the PLSDB database.

3 - Kathyanna ARNOULD

Microbiologie Fondamentale et Pathogénicité - MFP - UMR 5234 – CNRS, Université de Bordeaux

Characterisation of TbSeipin: a key protein involved in lipid droplet formation in *Trypanosoma brucei*

Kathyanna Arnould, Perrine Hervé, Emmanuel Tetaud, Frédéric Bringaud, Loïc Rivière

Lipid droplets (LDs) are conserved organelles found in all eukaryotes, primarily involved in lipid storage. However, their role during key parasitic stages of *Trypanosoma brucei* remains poorly understood. In this study, we identified TbSeipin, a key protein involved in lipid droplet formation in *T. brucei*. Using a CRISPR/Cas9 genome editing technology recently optimized in our team, we performed endogenous tagging and a knockout (KO) strategy to investigate TbSeipin's localization and function. Comprehensive analyses – including omics approaches, imaging, and fluorescence assays – revealed that TbSeipin regulates LD size and growth. Heterologous complementation in *Saccharomyces cerevisiae* confirmed its conserved function with its yeast counterparts. Unexpectedly, TbSeipin knockout also altered cell cycle progression, which differs from findings in other organisms. This suggests that TbSeipin may have additional roles in *T. brucei* beyond lipid metabolism, potentially influencing cell cycle progression through direct or indirect mechanisms that are not yet fully understood.



4 – Nathalie ARVY

Biologie du Fruit et Pathologie – BFP – UMR 1332 – INRAE – Université de Bordeaux

Lipid Droplets: Beyond Mere Energy Storage, Key Players in Viral Propagation

N. Arvy, M. Batsale, L. Jambou, L. Sofer, V. Simon, C. Quinteau, N. Doner, R. Mullen, D. Coulon, C. Brehelin, S. German-Retana

Lipid droplets (LDs) are universal dynamic organelles, consisting of a core of neutral lipids surrounded by a phospholipid monolayer associated with enzymes and structural proteins. Far beyond mere energy reservoirs, they play a key role in cellular homeostasis by regulating lipid metabolism, energy storage, and even signal transduction. Their involvement in infectious processes has already been demonstrated during fungal and bacterial attacks in plants, but their role in virus-plant interactions, as already reported in humans, has only recently been explored (Dai et al., 2024; Wang et al., 2024).

In this study, we present the first comprehensive analysis of LDs accumulation and their recruitment to viral replication compartments (VRCs) during infection by turnip mosaic virus (TuMV, a member of the Potyvirus genus) in both *Arabidopsis thaliana* and *Nicotiana benthamiana*, using confocal microscopy. We observed a significant accumulation of neutral lipids in infected leaves, confirming that TuMV infection induces LDs biogenesis in *N. benthamiana*. Furthermore, transmission electron microscopy imaging revealed that these newly formed LDs are located near TuMV-induced VRCs.

We specifically focused on two protein families essential for LD biosynthesis and stability: SEIPINs and LIPID DROPLET ASSOCIATED PROTEINS (LDAPs), the latter being also involved in lipase recruitment. TuMV propagation was significantly reduced in *Arabidopsis* *ldap* and *seipin* mutants, while it was enhanced in plants overexpressing these genes, both in inoculated and systemic leaves. Our data suggest that SEIPIN and LDAP proteins play a role in both viral movement and replication, making them key factors in LDs recruitment during viral infection in plants. Collectively, our results highlight a pro-viral function of LDs in TuMV-infected plants.

5 – Michel Autin

Microbiologie Fondamentale et Pathogénicité – MFP – UMR 5234 – CNRS, Université de Bordeaux

Interplay between HIV-1 integration and transcription

Autin M., Bertinetti C.*¹, Zgadzay Y., Lapailleurie D., Munier-Lehmann H., Bonomi M., Ruff M., Lesbats P. and Parissi V*

«Host chromatin invasion by retroviruses relies on their stable integration into the chromosomes of infected cells. Integration is catalyzed into chromatin by the integrase protein (IN) within the Strand Transfer Complex (STC). Completion of the integration process requires then the disassembly of the STC, allowing DNA repair at the insertion site and subsequent expression of viral genes. This post-integration disassembly step remains poorly known.

IN protein carries additional functions including binding to the genomic viral RNA, especially to the TAR region, thereby assisting in virion assembly. Mutations in the IN carboxyl-terminal domain known to bind RNA affect both IN stability at the integration site and transcription of viral genes (1). Furthermore, recent findings demonstrate that the binding of IN to TAR RNA induces structural changes in both IN and TAR in addition to enhance Tat binding to RNA potentially optimizing the viral transactivation (2).

Based on these data, we proposed that the binding of TAR to IN may occur after integration, leading to STC destabilization and promoting its dissociation from the nucleosomal site. To investigate this hypothesis, we performed in vitro dissociation experiments of STC reconstituted onto human

nucleosomes. RNA fragments were found to efficiently dissociate the STC, whereas DNA structures appeared to be significantly less effective. This dissociation was further confirmed using Biolayer Interferometry. Among all the nucleic acid structures tested, the TAR RNA element exhibited the most potent dissociation properties. Biochemical characterization of the nucleoprotein complexes released after RNA-induced dissociation revealed that the IN and LEDGF/p75 was specifically dissociated from the nucleosomal integrated product, while preserving chromatin structure. These results suggest that RNA, and more specifically the TAR element, produced immediately after integration, could contribute to STC dissociation, facilitate the transactivation process, and the subsequent post-integration events. The initial results obtained in cells are consistent with and support our working hypothesis. They could demonstrate, for the first time, an interaction at the integration site between integrase and newly transcribed TAR RNA.

Our work highlights a novel functional interplay between HIV-1 integration and transcription, which requires precise coordination to ensure the optimal invasion of host chromatin by the virus. Ongoing pharmacological approaches targeting this process, as well as cellular analyses of this process will also be discussed (MS under preparation).

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2. Rocchi,C. et al., *Int. J. Mol. Sci.* 2022»

6 - Sophie BARROUILHET

Institut des Sciences Analytiques et de Physico-Chimie pour l'Environnement et les Matériaux - IPREM - UMR 5254 - CNRS, Université de Pau et des Pays de l'Adour

First evidence of a mercury resistance mechanism in an anaerobic bacterium: impact on mercury accumulation and methylation

BARROUILHET S, MONPERRUS M, GASSIE C, LE BARS M, LE GOHALEN A, DOLLA A, KHALFAOUI B, GUYONEAUD R, ISAURE M, GONI URRIZA M

Mercury (Hg) is a persistent pollutant leading to environmental and health issues. While Hg resistance mechanisms are well characterized in aerobic microorganisms, they remain unidentified in anaerobes. Since some anaerobic bacteria are able to transform Hg(II) into methylmercury (MeHg), a potent neurotoxin, deciphering Hg resistance mechanisms is necessary to understand how anaerobes deal with Hg toxicity. Previous differential transcriptomic analysis on the anaerobic bacterium *Pseudodesulfovibrio hydrargyri* BerOc1 identified a cluster of genes overexpressed at 0.5 µM of Hg(II). They display sequence homologies with both efflux and metal resistant systems. This study aims to determine the role of this cluster in Hg resistance and methylation. Single BerOc1 mutant strain, deleted of the efflux system, and double mutant strain, deleted of both the efflux and metal resistant systems, were generated to conduct a comparative phenotypic analysis with the wild-type (wt). Sensitivity to Hg(II) and to MeHg was monitored by following bacterial growth, while MeHg production and intracellular Hg(II) concentrations were measured by GC-ICP MS on cells exposed to different Hg(II) concentrations (from 0.0005 to 50 µM). Below 0.5 µM of Hg(II), no differences were observed for bacterial growth and intracellular Hg(II) concentration between wt and mutant strains. However, above 0.5 µM, sensitivity to Hg(II) increased for mutant strains with a higher impact for the double mutant. Therefore, 0.5 µM of Hg(II) was determined as the threshold concentration inducing resistance in the wt strain. Moreover, above 0.5 µM, single mutant strain accumulated 2 to 10 times more Hg(II), whereas double mutant accumulated 2 to 200 times less Hg(II) compared to wt depending on Hg(II) concentrations. Our results support a two-level resistance mechanism: a first resistance (low level) induced by a Hg-scavenging (metal resistant) and a second resistance (higher lever) induced by the efflux system. Interestingly, MeHg production decreased in both mutant strains, even in the single mutant that showed higher intracellular Hg(II) contents. The accumulated Hg(II) was not available for methylation. This study highlights, for the first time, a mechanism of Hg resistance in anaerobic bacteria. This finding is a key step toward understanding the MeHg production and Hg(II) dissemination in environmental ecosystems.



7 - Nathanaël BENOIT

Microbiologie Fondamentale et Pathogénicité - MFP - UMR 5234 - CNRS, Université de Bordeaux

A ubiquitin-like protein controls assembly of a bacterial Type VIIb secretion system

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Bacteria inhabit densely populated environments where they compete for nutrients, resources, and space. To gain an advantage, bacteria have developed secretion systems that release effector proteins into their surroundings or directly into other cells. Type VII secretion systems (T7SS) are crucial bacterial nanomachines that mediate interbacterial competition and host pathogen interactions in Gram-positive bacteria. The T7SSb secretion system, present in Firmicutes such as *Bacillus subtilis* and *Staphylococcus aureus*, plays a crucial role in this bacterial competition, process in which an attacker bacterium kills a prey bacterium. The T7SSb system regulates bacterial populations found in biofilms and microbiota, particularly during infection. In *Bacillus subtilis*, the T7SSb is encoded by the yuk operon, which includes genes encoding membrane components, cytosolic proteins, and secreted toxins. These toxins, such as RNases, inhibit the growth of target cells that lack the corresponding antitoxin. Despite their importance, the structural basis for assembly and substrate transport in T7SSb, a widely distributed T7SS variant, remains poorly understood.

New data obtained in the laboratory show the first cryo-EM structure of the T7SSb core complex from *Bacillus subtilis*, revealing how a ubiquitin-like protein, YukD, coordinates assembly of the secretion machinery. YukD forms extensive interactions with the central channel component YukB and promotes its association with the pseudokinase YukC, creating a stable building block for channel assembly. Using microscopy and competition assays, we demonstrate that YukD is essential for proper T7SSb complex formation and bacterial killing. Our findings reveal how bacteria have adapted a ubiquitin-like protein as a structural regulator for assembling a large secretion complex.

8 - Bachir BOUREIMA ABDOU

Microbiologie Fondamentale et Pathogénicité - MFP - UMR 5234 - CNRS, Université de Bordeaux

Transfer of Integrative and Conjugative Elements and Chromosomal DNA in *Mycoplasma hominis* under Cell Culture Conditions.

B. Boureima-Abdou, Nadège Henin, C. Le-Roy, J. Guiraud, V. Dubois, E. Baranowski, C. Bébéar and S. Pereyre

Mobile genetic elements drive bacterial evolution by promoting genetic plasticity and adaptation. Among them, *Mycoplasma* Integrative and Conjugative Elements (MICEs) challenge the notion that mycoplasmas evolved solely through genome reduction. Despite their importance, MICEs remain poorly characterized, particularly in *Mycoplasma hominis*, a commensal and opportunistic pathogen of the human urogenital tract.

To investigate the conjugative properties of *M. hominis* ICE, we conducted mating experiments under cell culture condition. HeLa cells were co-incubated for seven days with a donor strain carrying the tetracycline resistance gene inserted in an intergenic region of the ICE to track the ICE and a recipient strain harboring a different resistance marker (to gentamicin or ofloxacin). Potential transconjugants were selected on agar

plates containing both antibiotics.

Whole genome sequencing and SNP polymorphism analyses revealed two distinct horizontal gene transfer (HGT) mechanisms. While the ICE was transferred from the donor to the recipient strain, a Mycoplasma Chromosomal Transfer (MCT) occurred in the opposite direction, from the recipient to the donor cell, independently of ICE mobilization and involved the exchange of large chromosomal regions.

These findings reveal an efficient HGT system in *M. hominis*, enhancing genome plasticity and adaptation, with potential implications for antibiotic resistance and mycoplasma evolution.

9 - Thibault BRUNET

Pharmacology of Antimicrobial Agents and antibioResistance - PHAR2 - U1070 - INSERM, Université de Poitiers

Etude des résistances au céfiderocol de bactéries GRAM-négatives d'origines cliniques Thibault Brunet, Sandrine Marchand, Frédéric Tewes

«Le céfiderocol est une céphalosporine conçue pour traiter les infections graves causées par des bactéries Gram-négatives multirésistantes. Il est utilisé comme dernier recours dans des infections telles que les infections urinaires compliquées et les pneumonies associées à l'hôpital. Sa structure unique, qui inclut un sidérophore en plus du noyau β -lactame, lui permet d'exploiter les mécanismes de transport du fer pour pénétrer l'espace périplasmique des bactéries. Toutefois, des résistances cliniques ont déjà été observées, notamment contre *Acinetobacter baumannii*, mettant en lumière les limites de son efficacité 1. Cette étude visait à étudier la sensibilité des bactéries GRAM-négatives lors d'un traitement au céfiderocol. Nous avons d'abord déterminé les CMI du céfiderocol pour des souches cliniques d'*Acinetobacter baumannii* et de *Klebsiella pneumoniae* en milieu déplété en fer. Ensuite, les tests de CMI séquentielle ont évalué l'apparition de résistances sous exposition à des concentrations suboptimales pendant 7 jours. Pour une souche résistante d'*A. baumannii* OXA-24, OXA-65, TEM-1B, la CMI du céfiderocol a augmenté de 16 mg/L à 2048 mg/L après cinq passages. Pour une souche sensible, la CMI du céfiderocol a augmenté de 0,25 mg/L à 256 mg/L après 7 jours. Ces résultats montrent que, malgré des CMI inférieures au seuil de résistance fixé à 2 μ g/mL par l'EUCAST, l'émergence de résistances significatives a été observée en seulement quelques jours. Dans la suite de l'étude, la stabilité de ces résistances au céfiderocol sera évaluée par des passages successifs en milieu déplété en fer, et les mutations potentielles seront identifiées par séquençage des génomes bactériens à l'aide de la technologie Nanopore.

1.Karakonstantis, S., Rousaki, M. & Kritsotakis, E. I. Cefiderocol: Systematic Review of Mechanisms of Resistance, Heteroresistance and In Vivo Emergence of Resistance. *Antibiotics* 11, 723 (2022).»

10 - Silvia CASCIANI

Institut de chimie de la matière condensée de Bordeaux - CMCB - UMR 5026 – CNRS, Université de Bordeaux

Hydrogen at depth. An investigation into life limits in underground reservoirs

Silvia Casciani, Gabriella Gallo, Martina Cascone, Donato Giovannelli, Samuel Marre, Anaïs Cario

Hydrogen plays a key energetic role as a keystone molecule bridging diverse scientific domains—from astrobiology and biotechnology to the biogeochemistry of extreme environments. It is a fundamental electron donor and primary energy currency in microbial metabolisms, particularly within anaerobic environments where it drives and connects a wide array of microbial processes in geological settings.



Cytométrie en flux et étalement : Deux approches complémentaires pour mieux comprendre l'effet des antibiotiquesMarina Denimal¹, Jérémie Moreau¹, Sandrine Marchand^{1,2}, Julien Buyck¹, Jonathan Clarhaut^{1,2}

Introduction : *Pseudomonas aeruginosa* est un pathogène opportuniste classé comme prioritaire par l'OMS en raison de sa forte implication dans les infections nosocomiales et de l'émergence croissante de résistances, nécessitant de nouveaux antibiotiques en urgence. (1–3) L'étude de l'activité des antibiotiques repose souvent sur des courbes de bactéricidie (TKC), mais cette méthode n'est pas toujours adaptée à l'analyse de profils particuliers comme les persisters (4,5) ou à des mélanges de souches. Dans ce contexte, la cytométrie en flux offre de nombreux avantages, notamment grâce au marquage direct des souches par des protéines fluorescentes (ex : GFP) ou à l'utilisation de sondes fluorescentes ciblant des fonctions cellulaires comme le métabolisme. L'utilisation de la cytométrie en flux permettrait une analyse quantitative rapide de l'effet bactéricide d'antibiotiques.

Objectif : L'objectif de ce travail est de comparer l'évaluation de l'activité antibiotique à l'aide de méthodes conventionnelles à celle obtenue par cytométrie en flux couplée à l'imagerie.

Méthode : Des TKC ont été réalisées afin d'évaluer la dynamique de croissance de *Pseudomonas aeruginosa* PAO1-eGFP exposée à différentes concentrations de ceftazidime, ceftazidime/avibactam (avibactam à 4 mg/L), tobramycine et méropénème. Des prélèvements ont été réalisés à différents temps d'incubation (0, 2, 4, 6, 24 et 30 h). La répétabilité et la reproductibilité de l'approche ont été évaluées à l'aide de tests en triplicat. La méthode conventionnelle reposait sur l'ensemencement de 50 µL de l'échantillon à la suite de dilution en série, sur gélose au charbon actif, suivie d'un comptage des colonies après incubation à 37°C sur la nuit. Pour l'approche cytométrique, 50 µL de chaque échantillon a été marqué avec le colorant membranaire SYTOX™ Blue, puis analysé sur un cytomètre Attune® CytPix (Thermo Fisher Scientific). Pour étudier le marquage des événements, deux lasers ont été utilisés. Un laser bleu (488 nm) permettant la mesure de l'émission de la GFP dans le canal BL1 (530/30 nm). Un laser violet (405 nm) permettant la mesure de l'émission du SYTOX™ Blue dans le canal VL1 (440/50 nm).

Résultats : Les CMI des différents antibiotiques de notre souche PAO1-eGFP étaient comparables à la souche PAO1 de référence: ceftazidime (CMI = 2 mg/L), ceftazidime/avibactam (CMI = 2 mg/L), tobramycine (CMI = 0,25 mg/L), et le méropénème (CMI = 0,5 mg/L). Les courbes de bactéricidie obtenues par la méthode conventionnelle révèlent des profils similaires pour l'ensemble des antibiotiques testés : une diminution marquée de la population bactérienne à des concentrations supérieures à la CMI, une réduction suivie d'une reprise de croissance après 6 heures d'exposition à la CMI, et une croissance comparable au témoin pour les concentrations inférieures. En revanche, l'utilisation de la cytométrie a permis de distinguer des comportements différents dépendant de l'antibiotique utilisé. Après exposition à la ceftazidime seule ou en combinaison avec l'avibactam, deux populations ont été observées : l'une de taille normale, l'autre allongée, suggérant un blocage de la division cellulaire. Seules les bactéries GFP(+)/Sytox(-) corrélaient avec les CFU/mL, les GFP(+)/Sytox(+) étant non cultivables. Avec la tobramycine, bien que les CFU/mL deviennent nulles à forte concentration dès 2 h post-exposition, des cellules GFP(+) étaient encore détectées, suggérant une viabilité transitoire non traduite en croissance. Concernant le méropénème, certaines bactéries à paroi altérée GFP(+)/Sytox(+) restaient cultivables à 1×CMI, tandis qu'à 8×CMI, des formes sphéroïdes atypiques étaient détectées, sans croissance en CFU/mL.

Conclusion : La cytométrie en flux permet de mettre en évidence l'impact morphologique de l'antibiotique sur *Pseudomonas aeruginosa* mais permet aussi une analyse quantitative rapide de l'effet bactéricide d'antibiotiques au cours du temps. Cependant une bactéricidie totale qui pouvait être observée avec plusieurs antibiotiques à fortes concentrations par l'approche traditionnelle en comptage sur gélose n'était pas observée en cytométrie, les mécanismes expliquant ces différences sont actuellement en cours d'étude.

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13 - Yoann FAUTRAS

Chimie et Biologie des Membranes et des Nano-objets - CBMN - UMR 5248 - CNRS - Bx INP - BSA, Bordeaux INP

Study of the aerobic and anaerobic sedimentary bacteriobiota

FAUTRAS Yoann, JULIENNE Sarah, EXODIA Consortium, BOURILLOT Raphaël, CARIO Anaïs, VILAIN Sébastien

«The thesis is part of a larger project aiming to analyze the Extracellular Polymeric Substances (EPS) contents of sediments from modern and ancient estuaries (ANR "EXODIA"). EXODIA aims to better define the composition and function of EPS in modern and ancient estuaries, and their interaction with clay and metals through diagenesis. The aim of the thesis is to isolate, characterize and identify the culturable microorganisms potentially involved in the estuarine sediment EPS cycle (anaerobic and aerobic bacteria), in particular strains able to produce high quantities of EPS and/or able to degrade EPS. A maximum of microorganisms from the most representative sediment horizons on 6-m deep cores will be isolated by a culturomics strategy relying on the use of 16 media and 2 temperatures of incubation as well as enrichment strategies to focus on EPS degrading and biofilm-forming strains. After isolation and constitution of a strain's library, each strain will be grown as planktonic and sessile pure culture and their ability to produce EPS (mucoid character), to form biofilms and to produce EPS-modifying enzymes (DNases, proteases, hydrolases, lyases) will be assayed. Antibiosis and antibiofilm activities of isolates against bacterial reference strains in biofilm research (*Escherichia coli* and *Pseudomonas aeruginosa*) will be carried out. All the information relative to the isolates will be listed in a publicly available database. The identification of isolates will be performed in first intention by mass spectrometry. In parallel, genomic identification of the microbiome in sediment samples will be performed. This will allow us to estimate if our isolates are representative of the global community.»

14 - Valentin FIÉVET

Mycologie et Sécurité Alimentaire - MycSA - UR 1264 - INRAE, Université de Bordeaux

Investigating species compositional and metabolomic dynamics in a *Fusarium* synthetic community under abiotic pressures

Valentin Fiévet, Laetitia Pinson-Gadais, Stéphane Bernillon, Louis Carles, Florence Richard-Forget

Fusarium species are causative agents of *Fusarium* head blight (FHB), a devastating fungal disease affecting cereal crops worldwide. FHB causes yield losses and grain contamination with mycotoxins (type A and B trichothecenes, zearalenone, enniatins, beauvericin), posing significant health and food safety concerns. Several *Fusarium* species occupying the same ecological niche during infection likely interact, modulating FHB outcomes, including symptoms and mycotoxin contamination. However, most studies have focused on single *Fusarium* species, particularly *F. graminearum*, considered as the main causal agent of FHB, which has proven to be insufficient for a comprehensive

understanding of the disease.

This study aims to investigate the species compositional and metabolomic dynamics of a synthetic community called Meta-Fusarium. This community was built with one strain of each of the seven major FHB species encountered in Europe (*F. graminearum*, *F. culmorum*, *F. poae*, *F. avenaceum*, *F. sporotrichioides*, *F. langsethiae*, *F. tricinctum*) and exposed *in vitro* to different abiotic pressures. Changes in temperature and oxidative stress, key abiotic constraints that Fusarium species must cope with during plant infection, were considered. Our data indicated that, regardless of the culture conditions, *F. culmorum* followed by *F. graminearum* and to a lesser extent *F. poae*, were the predominant species. Species composition was shown to be affected by abiotic factors, *F. culmorum* being favoured by a decrease in temperature while *F. graminearum* was more abundant under oxidative stress. Similarly, metabolomic profiles and mycotoxin accumulation by the Meta-Fusarium were influenced by abiotic conditions. While Type B trichothecenes were the dominant mycotoxins, whatever the culture conditions, the 15-ADON mycotoxin produced by *F. graminearum*, was quantified in higher amounts under oxidative stress.

These results provide the first assessment of behaviour of a complex Fusarium synthetic community, moving towards a mechanistic understanding of Fusarium interactions and their impact on disease outcomes.

15 – Pierre GOUDENECHE

UMR5536, Université de Bordeaux

Exploring the Role of Maternal Nutrition and Gut Microbiota in Neuroprotection Against Neonatal Hypoxia-Ischemia. Study in Ratss

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Neonatal hypoxia-ischemia (NHI) remains a leading cause of perinatal mortality and long-term neurological impairment. While therapeutic hypothermia is currently the only approved treatment, it fails to protect nearly 50% of treated newborns, highlighting the urgent need for alternative neuroprotective strategies. Our team has previously demonstrated that maternal dietary supplementation with resveratrol (RSV), a polyphenol derived from grapes, offers neuroprotection in a rat model of NHI, characterized by reduced brain lesion volumes and preserved sensorimotor functions. To enhance the bioavailability and translational potential of this strategy, we evaluated two complementary approaches: (i) maternal supplementation with piceatannol, a hydroxylated analogue of RSV with improved pharmacokinetic properties, and (ii) a polyphenolic cocktail (RSV + ϵ -viniferin + pterostilbene), combining compounds with distinct mechanisms of action and half-lives to maximize synergy. Both interventions exhibited greater neuroprotective effects than RSV alone. Given the potential role of the gut-brain axis, we investigated whether these effects might be mediated through modulation of the neonatal gut microbiota. To test this hypothesis, we conducted fecal microbiota transplantation (FMT) from pregnant dams supplemented with the polyphenolic cocktail to pups born to non-supplemented dams, from postnatal day 0 (P0) to day 7 (P7), the day of NHI induction. Preliminary results indicated that microbiota modulation via FMT significantly reduced brain lesion volumes in the short term. Furthermore, behavioral assessment revealed that NHI-pups receiving FMT from cocktail-supplemented dams maintained better behavioral abilities compared to untreated NHI control. These findings suggest that part of the neuroprotective effects of maternal polyphenol supplementation may be mediated through gut microbiota modulation.

16 - Henri GRAU

Pharmacology of Antimicrobial Agents and antibioResistance - PHAR2 - U1070 - INSERM

Améliorer l'action de la colistine face à *Klebsiella pneumoniae* multirésistante via des nanoparticules chargées en farnesol

Henri Grau, Farris Daffa Imtiyaz, Sandrine Marchand, Rémy Bonnin, Laurent Dortet, Julien M. Buyck, Frédéric Tewes

Nos travaux précédents ont montré que des nanoparticules lipidiques chargées en farnésol (F-LNPs) renforcent l'efficacité de la colistine (CST), réduisant sa CMI jusqu'à 64 fois contre *Acinetobacter baumannii* et *Escherichia coli*. Cette étude étend cette approche à *Klebsiella pneumoniae*, en explorant les mécanismes d'action de la combinaison CST + F-LNPs et son impact sur la résistance.

Des CMI ont été déterminées sur 18 isolats cliniques de *K. pneumoniae*, traités par la CST seule ou en association avec des F-LNPs. Trois isolats, présentant des modifications de leur LPS (pEtN, L-Ara4N ou les deux), ont été sélectionnés pour comprendre le mécanisme d'action de l'association CST + LNPs. Onze passages successifs ont été réalisés afin de décrire l'induction de résistance, accompagnées d'une analyse du potentiel zéta au passage 11. Une étude de la perméabilité membranaire a été réalisée à l'aide de l'iodure de propidium pour la membrane interne (IM) et de la nitrocéfine pour la membrane externe (OM). Enfin, l'efficacité de la combinaison a été testée *in vivo* dans un modèle d'infection *Galleria mellonella*.

Les F-LNPs ont permis de réduire la CMI de la CST de 8 à 1024 fois sur les 18 isolats. Sur les trois souches sélectionnées, la combinaison CST + F-LNPs a empêché le développement de résistance, et donc l'augmentation de la CMI au fil des passages, contrairement à CST seule, dont la CMI est passée de 4 à ≥ 1024 mg/L. Cette élévation s'accompagnait d'une variation du potentiel zéta, passant de $-25,5 \pm 3,1$ mV à $-14,4 \pm 2,9$ mV, alors qu'il restait stable autour de $-21,2 \pm 1,9$ mV en présence de CST + F-LNPs. Les tests de perméabilité ont montré une augmentation significative de la perméabilité de l'IM et de l'OM avec les F-LNPs, qui était renforcé avec la CST. *In vivo*, la combinaison CST + F-LNPs a permis une survie de 43 % des larves à 7 jours, contre 0 % pour la CST seule à la même dose.

Nos résultats *in vitro* démontrent que les LNPs améliore l'efficacité de la CST contre des souches de *K. pneumoniae* résistantes à la colistine, en perturbant davantage les membranes bactériennes et en limitant l'émergence de la résistance. Les résultats *in vivo* montrent une augmentation significative de la survie lors d'infections par des souches résistantes.

17 - Chloé JACQUET

Pharmacology of Antimicrobial Agents and antibioResistance - PHAR2 - U1070 - INSERM, Université de Poitiers

Fidaxomicin, a potential new drug for combinatorial therapies against *Mycobacterium abscessus*

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Mycobacterium abscessus (Mabs), an opportunistic pathogen, is associated with severe pulmonary outcomes in susceptible population like cystic fibrosis patients. Treatment requires prolonged multidrug regimens (12–18 months) with cure rates below 30%, largely due to intrinsic resistance mechanisms and limited effective



antimicrobials. Recently, fidaxomicin—a macrocyclic antibiotic approved for *Clostridium difficile* infections—was identified as a potential candidate against Mabs⁽¹⁾. In this study, we assessed the antimicrobial activity of fidaxomicin against Mabs and explore its interaction with six commonly used antibiotics. The minimal inhibitory concentration (MIC) of fidaxomicin was determined against *M. abscessus* ATCC 19977T reference strain using the broth microdilution method in Middlebrook 7H09 medium (10% OADC and 0.5% glycerol) (n=4). Results were read after 3 days of incubation at 35 ± 2 °C. Synergistic activity between fidaxomicin and azithromycin, amikacin, apramycin, imipenem, linezolid, or tigecycline was evaluated using the checkerboard method (n=3). The efficacy of each combination was quantified by calculating the Fractional Inhibitory Concentration Index (FICI), with synergy defined as minimal FICI ≤ 0.5. The MIC of fidaxomicin was 8–16 mg/L. Fidaxomicin combined with azithromycin showed a synergistic effect (minimal FICI= 0.375) as well as with imipenem and tigecycline (minimal FICI of 0.375 and 0.5 respectively). No synergistic interaction was found with combination of fidaxomicin with either amikacin, apramycin or linezolid (minimal FICI > 0.5).

The synergistic association of fidaxomicin with azithromycin, imipenem or tigecycline is highlighting the potential of fidaxomicin as a valuable component of combination therapy.

¹Sun, Q. et al. *J. Med. Microbiol.* 71, (2022).

18 – Fabien JARRY

*Anti-Infectieux: supports moléculaires des résistances et innovations thérapeutiques – RESINFIT
– UMR 1092 – INSERM, Université de Limoges*

Antibiogramme rapide par SdFFF à partir de flacons d'hémocultures positifs à *Escherichia coli*

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Le sepsis est l'une des principales causes de mortalité dans le monde. Des études ont montré que l'initiation précoce d'une antibiothérapie adaptée est fortement corrélée à un meilleur pronostic vital des patients atteints de bactériémie. Cependant, les méthodes conventionnelles d'antibiogramme nécessitent 16 à 24 heures afin de fournir des résultats à partir de flacons d'hémocultures positifs. Il est donc important d'obtenir les résultats de l'antibiogramme le plus rapidement possible.

L'objectif de cette étude est d'évaluer les performances de la technologie de Fractionnement par couplage Flux-Force de Sémination (SdFFF) et sa capacité à fournir un antibiogramme rapide en moins de 3 heures, directement à partir de flacons d'hémocultures positifs.

La SdFFF, méthode apparentée à la chromatographie liquide, est dépourvue de phase stationnaire et met en jeu un couple flux de phase mobile et champ multi-gravitationnel qui permet de séparer les bactéries en fonction de leurs caractéristiques physico-chimiques telles que leur taille, leur forme et leur densité. Pour déterminer les performances de la SdFFF, des hémocultures enrichies de 100 isolats de *Escherichia coli* ont été utilisées. Une suspension calibrée d'hémocultures positives a été incubée seule ou

en présence de 5 antibiotiques différents d'intérêt clinique (Amoxicilline, Trimethoprime-Sulfamethoxazole, Céfotaxime, Ciprofloxacine et Gentamicine) à des concentrations correspondant aux concentrations critiques selon le référentiel CA-SFM/EUCAST 2024 pendant 2 heures à 37°C. Après centrifugation, les suspensions bactériennes sont injectées dans la machine de SdFFF. L'étude des signaux d'élution entre les échantillons non traités et les mêmes échantillons traités et la comparaison avec la méthode de référence par micro-dilution en milieu liquide (BMD) ont permis de mettre en place une stratégie d'interprétation afin de classer les bactéries comme sensibles ou résistantes à chaque antibiotique. Enfin, afin de valider notre approche, nous avons travaillé en conditions réelles à partir de 40 flacons d'hémocultures positifs à E. coli issus de patients hospitalisés au CHU de Limoges selon la même méthodologie.

Pour les 100 hémocultures enrichies d'isolats de E. coli, les résultats montrent un accord de catégorie (CA = 99,8%), une sensibilité (Se = 99,5%) et une spécificité (Sp = 100%) élevées : aucune erreur majeure (ME, souches classées comme résistantes par la méthode SdFFF et sensibles par la méthode BMD) et 1 erreur très majeure (VME, souches classées comme sensibles par la méthode SdFFF et résistantes par la méthode BMD). Ces résultats sont conformes aux recommandations du CLSI (CA \geq 90%) et de la norme ISO 20776-2:2021 (Se, Sp \geq 95%). Parmi les 40 hémocultures positives à E. coli du CHU de Limoges, 37 (92,5%) ont permis d'obtenir un signal interprétable après 2 heures d'incubation à 37°C. Les résultats obtenus étaient conformes à ceux obtenus précédemment (CA, Se, Sp = 100%).

Ces résultats préliminaires indiquent que la SdFFF permet la réalisation d'un antibiogramme rapide directement à partir d'hémocultures positives à E. coli dans un délai inférieur à 3 heures. Des essais sur un plus grand nombre d'isolats, d'espèces bactériennes et de familles d'antibiotiques permettront de tester la robustesse et l'universalité de la technologie SdFFF.

19 - Quentin JEHANNE

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Université de Bordeaux*

Recurrent *C. jejuni* and *C. coli* infections: the contribution of genomics to the characterization of two cases

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Campylobacter species are the most common cause of bacterial gastro-enteritis and, in 1% of cases, can be responsible for bacteraemia. We compare here two clinical cases of common variable immunodeficiency (CVID) patients with Campylobacters-associated diarrhoea and repeated bacteraemia over a period of almost 10 years. The main goal of this study was to analyse and compare the genome of each isolated strain for both cases during follow-up. For patient «B», 18 isolates of *C. coli* (BC) and 17 isolates of *C. jejuni* (BJ) were sampled between 2014 and 2024. For patient «T», 10 isolates of *C. coli* (TC) were sampled between 2019 and 2024. Next-generation sequencing was used to confirm the

species and perform molecular antimicrobial resistance identification, MLST typing, core-genome MLST, core-genome SNP (cgSNP) analysis and source attributions. For patient B, all 10 *C. coli* belong to the same CC and ST and were attributed to the chicken reservoir. However, the 5 *C. jejuni* (attributed to the ruminant reservoir) showed variable CC and ST. We identified two clusters from 2014 and one from 2022–2023. The cgSNP analysis also displayed three clusters of *C. jejuni* and confirmed that each *C. coli* most likely came from the same initial strain. Additionally, a 16S rDNA mutation in position A1358G associated to gentamicin resistance was found among *C. coli* from 2022. Recent *C. coli* were also resistant to ertapenem which is associated to an amino-acids duplication within the PorA protein sequence. For patient T, each *C. coli* has the same CC, ST and porcine origin. Isolates from 2023 displayed an A1358G 16S rDNA mutation and one isolate a duplication in PorA sequence. Retrospective analyses of antimicrobial therapy for both patients showed an association between the administration of antibiotics and the appearance of resistance markers, suggesting an in vivo selection pressure. Here, we described two cases of recurrent *C. coli* and *C. jejuni* infections among CVID patients, with strains able to escape to successive antimicrobial therapies with specific DNA mutations. These molecular events have never been described before among *Campylobacter* species.

20 – Amélie JESSU

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Pathophysiology of Mpox virus skin infection

Amélie Jessu, Emilie Murigneux, Inas Daoudi, Mustapha Si-Tahar, Nicolas Lévéque, Magali Garcia, Charles Bodet

«Mpox virus (MPXV) is the etiological agent of mpox, a zoonotic disease responsible for a global outbreak in 2022. The transmission primarily occurred through direct skin to skin contact but other routes, such as respiratory secretions, may also contribute to its dissemination. Currently, few data are available on the interactions between human skin cells and MPXV as well as on the inflammatory and antiviral immune responses induced. Our work aimed to characterize the permissiveness of primary human keratinocytes and fibroblasts to MPXV infection. Both cell types were infected for 72 hours, and DNA and RNA were extracted. Viral replication was assessed by qPCR and infectious viral titers were determined using the TCID₅₀ assay. The expression of inflammatory and antiviral targets was studied using RT-qPCR analyzes. Our results demonstrate that MPXV is capable of replicating in both human keratinocytes and fibroblasts from 24 hours post-infection (hpi) and viral DNA levels increased up to 72 hpi. In fibroblasts, the production of infectious viral particles was detected in the extracellular media at 72 hpi. At 48 hpi, MPXV infection resulted in an inflammatory response, with upregulation of cytokines and chemokines such as CXCL3 and CXCL8 in keratinocytes and fibroblasts. The antiviral response appeared to differ between the cell types, with upregulation of interferon-stimulated genes such as IFIT2, IFIT3, and ISG15 in fibroblasts, while these genes were downregulated in keratinocytes. Taken together, our results suggest that response to MPXV cutaneous infection varies depending on the resident skin cell type. Thus, the use of more complex models, such as ex vivo human tissue explants reflecting the architecture of the skin, would help to better understand the host response to MPXV infection.»

21 – Anagha KALLISSEI PARAMBIL

Institut Européen de Chimie et Biologie - IECB - UAR 3033 - CNRS - US001 - INSERM

NMR SPECTROSCOPY TO STUDY PROTEIN ASSEMBLIES ON BACTERIAL MEMBRANE

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Keywords: Antimicrobial resistance, ZipA protein, NMR Spectroscopy

Antimicrobial resistance in bacteria is an escalating public health threat, making it crucial to identify new therapeutic targets [1]. Understanding the atomic mechanisms behind protein-membrane interactions in bacteria offers a promising strategy to discover novel drug targets. Our research focuses on the ZipA protein, which plays a vital role in bacterial cell division [2], controlling the initial molecular steps during set-up of the division site in certain bacterial pathogens such as *Escherichia coli*. Using both solution and solid-state Nuclear Magnetic Resonance (NMR) spectroscopy, we investigate the molecular structure of ZipA in a membrane-mimicking environment. By targeting the protein structure in a native-like setting, this study aims to uncover insights into the structures and the interactions with the membrane at the atomic level, providing crucial information for the development of new antimicrobial agents.

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DOI:10.5772/intechopen.101671

[2] J Lin, Yu-pin, 2019, 9, 18712.

22 – Joy KHOURY

Mycologie et Sécurité Alimentaire – MycSA – UR 1264 – INRAE, Université de Bordeaux

Co-occurrence between Fusarium and bacterial microbiota in wheat grains

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«Fusarium Head Blight (FHB) represents a significant threat to wheat production worldwide, causing substantial yield losses and grain quality deterioration through mycotoxin contamination. While the influence of climatic conditions, agronomic practices, and host genotype on FHB incidence and *Fusarium* species composition is well-established, the role of wheat microbiota in modulating *Fusarium* infection remains poorly understood. This study aims to determine whether and how wheat-associated bacteria influence the species composition of *Fusarium* in wheat grains. Bacterial diversity analysis has been conducted by using 16S rRNA gene sequencing data from over 500 wheat grain samples collected across diverse French agricultural regions.

Influence of abiotic parameters such as wheat variety, agricultural practices, climatic conditions and mycotoxin contamination levels on bacterial diversity and community composition is currently being evaluated. Co-occurrence network analysis will integrate bacterial and *Fusarium* diversity data from the same samples to identify significant positive and negative associations between specific taxa and mycotoxin production. These associations will be tested in vitro with co-cultures of bacteria (single strain and synthetic communities) and *Fusarium*. The results of the interaction will be evaluated in vitro by measuring bacterial and fungal growth (via qPCR) and mycotoxin production (via HPLC-DAD and LC-MS).

This study will provide critical insights into the ecological interactions between wheat-associated bacteria and *Fusarium* pathogens, potentially leading to innovative biological strategies for FHB management and mycotoxin mitigation in wheat production systems.»



23 – Ana Luzia LACERDA

Littoral ENvironnement et Sociétés – LIENSs – UMR 7266 – CNRS, Université de La Rochelle

Projet EMERG : Exosome microbien et Risque sanitaire – Intérêt d'une Gestion One Health des enjeux liés aux grippes zoonotique

LACERDA Ana Luzia; PAOLETTI Mathieu; CRAVO-LAUREAU Cristiana; DEFAYE Baptiste; DELHAES Laurence; AGOGUÉ Hélène

Dans le cadre des Programmes Scientifiques de Grande Ambition Régionale – Maladies Infectieuses Émergentes et Risques Sanitaires (PSGAR-MIE), le projet EMERG vise à étudier l'évolution de l'exosome microbien, incluant les bactéries, archées, champignons et micro-eucaryotes, dans les zones côtières et marines de Nouvelle-Aquitaine. L'objectif principal est de caractériser la diversité et la dynamique des communautés microbiennes dans ces environnements afin de mieux comprendre les liens entre les conditions écologiques, les interactions microbiennes et l'émergence des virus de la grippe aviaire. Les prélèvements sur le terrain ont commencé en mars 2025 et sont réalisés sur six sites représentatifs : Yves, La Palmyre, Audenge, Moliets, Chizé et l'observatoire marin SOMLIT au large de La Rochelle. Les échantillons sont collectés dans trois compartiments environnementaux – eau, sédiment et air – selon une stratégie de suivi saisonnier sur une période d'un an. L'analyse des communautés microbiennes repose sur des techniques de métabarcoding d'ADN environnemental à haut débit, ciblant des marqueurs génétiques spécifiques : le gène 16S rRNA pour les bactéries et les archées, le gène 18S rRNA pour les micro-eucaryotes, et la région ITS2 pour les champignons. Les profils de diversité microbienne seront croisés avec les paramètres environnementaux mesurés sur site (température, pH, salinité, oxygène dissous), ainsi qu'avec les conditions météorologiques (p. ex. vent, précipitations). Une attention particulière sera portée à l'identification des configurations microbiennes et des conditions environnementales susceptibles de favoriser la présence et la persistance des virus de la grippe aviaire dans la région Nouvelle-Aquitaine. Les exposomes des sites à faible probabilité d'infection seront comparés à ceux des zones où des cas confirmés ont été observés, dans le but d'identifier des signatures microbiennes, abiotiques et biotiques, spécifiques aux contextes d'épidémie. Les données seront analysées à l'aide d'outils biostatistiques et d'analyses de réseaux écologiques afin d'explorer les schémas d'interaction microbienne et d'identifier des indicateurs écologiques robustes. Ces bioindicateurs pourraient contribuer à la surveillance environnementale et aider à anticiper les conditions favorables à l'émergence d'épisodes de grippe aviaire. Ce projet s'inscrit dans une démarche interdisciplinaire et intégrative 'Une Seule Santé', portée par une collaboration entre l'Université de La Rochelle, l'Université de Pau et des Pays de l'Adour, et le Centre Hospitalier Universitaire (CHU) de Bordeaux.

24 – Sarah LAVIELLE

Institut de Biochimie et Génétique Cellulaires – IBGC- UMR 5095 – CNRS, Université de Bordeaux

Investigating the role of Gut Microbiota in Glioblastoma development

Sarah Lavielle, Manon Lemaitre, Doriane Bomont, Thomas Daubon, Océane Martin

Glioblastoma is the most common and aggressive brain tumor in adults, characterized by a poor prognosis, with a median survival of approximately 14 to 15 months and frequent therapeutic failure. To improve patient survival and quality of life, it is essential to identify the factors contributing to the initiation, progression, and therapeutic resistance of this cancer. In this study, we focus on the gut-brain axis, specifically by exploring the potential

role of the bacterial microbiota, in glioblastoma progression. The bacterial microbiota refers to the diverse community of bacteria that colonizes the gut, which has been shown to play a crucial role in several brain pathologies, including Parkinson's disease and Alzheimer's disease. More recently, emerging studies have suggested that the bacterial microbiota may influence glioblastoma, but the underlying mechanisms remain unclear. This project has two main objectives :

1. Evaluate the effect of bacterial microbiota depletion on glioblastoma progression.
2. Investigate the mechanisms by which gut bacteria influence glioblastoma, by studying tumor immune system modulation in vivo and the impact of gut-derived metabolites on glioblastoma stem cells in vitro.

To achieve this, C57BL/6 mice were implanted or not with murine glioblastoma stem cells (mGB2) and treated or not with antibiotics to deplete their gut microbiota. Tumor growth was monitored by bioluminescence imaging. Macrophages and T-cells infiltration into the tumor was analyzed using RNAscope multiplex staining. Finally, the effect of two metabolites (serotonin and butyrate), which are either produced or regulated by the gut microbiota, was tested in vitro on murine (mGB2) and patient-derived (P3) glioblastoma stem cells to assess their impact on cellular invasion and proliferation.

The results showed reduced tumor progression in microbiota-depleted mice. Additionally, antibiotic-treated mice exhibited a significant decrease of immune cell infiltration at the tumor site. In vitro, glioblastoma cells treated with serotonin proliferated more but exhibited reduced invasion.

In contrast, butyrate significantly decreased proliferation but increased invasion. These findings suggest a role of bacterial microbiota on glioblastoma progression. Bacteria may stimulate the recruitment of immune cells into the tumor, benefiting its growth. The tumor could also exploit certain bacterial metabolites to promote its development.

This project aims to contribute to a better understanding of the gut-brain axis in glioblastoma and may help identify novel therapeutic strategies targeting bacterial microbiota to improve treatment outcomes for patients with this aggressive cancer.

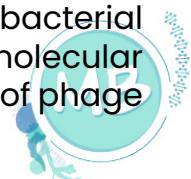
25 – Clémence MOUVILLE

Institut Européen de Chimie et Biologie – IECB – UAR 3033 – CNRS – US001 – INSERM, Université de Bordeaux

Phage MDA Φ entry mechanisms shape pilin variant populations of *Neisseria meningitidis*

Clémence Mouville, Antoine Brizard, Morgane Wuckelt, Mélanie Montabord, Julie Meyer, Mathieu Coureuil, Emmanuelle Bille

«*Neisseria meningitidis* is a commensal bacterium of human nasopharyngeal mucosa that can cross the nasopharyngeal barrier and spreads in the bloodstream. A filamentous phage, designated MDA Φ for Meningococcal Disease Associated, has been associated with invasive meningococcal diseases in young adults. This phage can infect meningococci using their type IV pili (TFP) and hijacks the TFP secretin to be extruded without damaging the host. MDA Φ seems to increase the occurrence of diseases by increasing bacterial colonization at the site-of-entry. In this work, our aim was to understand the molecular mechanism by which MDA Φ infects *N. meningitidis*. We focused on the first step of phage



interaction with bacteria. Investigations with deletion mutants of genes involved in the TFP machinery have shown that phage entry requires a functional and retractable TFP. This result is consistent with the literature on Ff or CTXΦ phages that interact directly with the pili tip. However, we found no evidence for the interaction of MDAv with the meningococcal TFP tip. We therefore focused on the possible interaction between the TFP fiber and the phage capsid. As PilE is subject to antigenic variation, we have identified variants of PilE that prevent entry of MDAΦ. Infection then takes place in a limited subset of bacteria expressing specific pilE sequences.

Next, we have shown that purified TFP and MDAΦ form bundles together. Additional analysis of the charged amino acids of TFP and those of MDAΦ capsid coat supported our hypothesis. Finally, we showed that T4P with positive electrostatic potential favour phage infection and allow strong bacterial adhesion to endothelial cells and vice versa. Together, our data support a new model of interaction between filamentous phages and type IV pili that could take part in the selection of pathogenic strains of *N. meningitidis*.»

26 – Marine NAVARRO

Mycologie et Sécurité Alimentaire – MycSA – UR 1264 – INRAE, Université de Bordeaux

Fungal microRNAs: key players in interaction

Marine Navarro, Fabien Dumetz, Nadia Ponts.

The Fusarium genus comprises over 80 phytopathogenic species responsible of Fusarium Head Blight, or FHB, a devastating cereal disease worldwide. FHB causes significant economic losses and causes health problems due to the production of mycotoxins by fungi, thermostable molecules dangerous to humans and animals, if ingested. FHB results from the interaction of several Fusarium species, including up to sixteen different species. Recent studies have shown that the infection stage and mycotoxin production are influenced by intra-microbial interactions mediated by molecular dialogue. We hypothesized that this molecular dialogue is mediated by microRNAs (miRNAs). MiRNAs are small non-coding RNAs, of 18–25 nucleotides, that induce post-transcriptional gene silencing. The abundance and nature of miRNAs are influenced by various factors such as stress, developmental stages, and microbial interactions. The aim of our research is to explore miRNAs of Fusaria during interactions.

To achieve this, confrontation tests between *Fusarium graminearum* and other Fusaria were set up to investigate interaction zones. Morphological modifications were initially observed using confocal microscopy, which raises questions about the mechanisms behind these morphological changes. Multi-omics approach was realized, including comparative transcriptomic analyses to identify miRNAs and their potential targets during Fusaria interactions. In addition, metabolomic analyses using HPLC-MS complemented this approach to detect signature metabolites of the interaction.

These results will give us keys to the diagnosis and prevention of FHB and mycotoxin accumulation in the plant system.

27 – Garance NOELLIER

Institut des Sciences Analytiques et de Physico-Chimie pour l'Environnement et les Matériaux – IPREM – UMR 5254 – CNRS, Université de Pau et des Pays de l'Adour

Microbial Assemblages and Metal Pollution Impacts in Mediterranean Wetlands

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Mediterranean wetlands face increasing pressures from metal(loid) pollution and environmental change, threatening their ecological balance and biodiversity. This study uses 16S and 18S rRNA metabarcoding to explore how microbial communities—bacteria and microeukaryotes—respond to varying metal concentrations and physicochemical conditions in three contrasting wetland sites. The results reveal that bacterial communities are highly sensitive to environmental gradients, particularly metal pollution, with specific taxa showing strong positive or negative correlations with metal levels. In contrast, eukaryotic assemblages appear more stable, suggesting a broader tolerance to environmental stress. Overall, microbial community structure reflects site-specific environmental pressures, highlighting the value of microorganisms as sensitive indicators of wetland health. These findings offer important insights into the ecological impacts of pollution and support the integration of microbial bioindicators into wetland monitoring and conservation strategies across the Mediterranean region.

28 – Arthur PETIT

Chimie et Biologie des Membranes et des Nano-objets – CBMN – UMR 5248 – CNRS – Bx INP – BSA, Université de Bordeaux

Production and purification of endolysins as a novel biocontrol agent in winemaking

Arthur Petit, Cas Mosterd, Amandine Guinoiseau, Claire Le Marrec, Agnès Hocquellet

«Endolysins are enzymes encoded by bacteriophages that specifically target bacterial cell walls, offering a promising alternative to conventional antimicrobial treatments. Their high specificity and effectiveness make them attractive candidates for various biotechnological applications, including food safety and preservation. In winemaking, where lactic acid bacteria (LAB) can cause spoilage or unwanted fermentations, endolysins could serve as effective biocontrol agents. Our study aimed to produce and purify recombinant phage endolysins from wine LAB in *Escherichia coli*. We cloned gene constructs into inducible expression vectors, and then transformed them into *E. coli* strains optimized for protein production. Expression was induced in small-scale cultures, and cells were lysed by sonication. Soluble and insoluble protein fractions were analyzed by SDS-PAGE and Western blot. Although protein expression was detected by Western blot using anti-His antibodies, the expression levels appeared low. These preliminary results lay the foundation for further optimization of expression conditions and purification strategies, with the goal of developing endolysins as biocontrol agents in the winemaking process.»

30 – Axel PITARD

Littoral ENVironnement et Sociétés – LIENSS – UMR 7266 – CNRS, Université de La Rochelle

Des molécules d'origine végétale : solutions naturelles pour lutter contre les maladies infectieuses ?

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«Ce projet de recherche vise à explorer le potentiel d'extraits végétaux en tant qu'agents anti-infectieux, avec pour principal objectif d'identifier des composés bioactifs capables de lutter contre des microorganismes pathogènes. Ce projet s'inscrit dans le cadre du Laboratoire Commun PHYTOMAR'INNOV qui débute en partenariat entre le Laboratoire LIENSS (UMR 7266 CNRS – La Rochelle Université) et la société Valbiotis (France).



Les résultats obtenus jusqu'à présent montrent une activité significative, en particulier contre des bactéries coques Gram positives multirésistantes aux antibiotiques. Ces résultats mettent en lumière le rôle prometteur des extraits végétaux comme sources potentielles de nouveaux traitements, notamment en réponse à la résistance aux antibiotiques des bactéries pathogènes. En intégrant des approches analytiques modernes pour identifier les composés actifs, ce projet ouvre des perspectives pour le développement de solutions naturelles et durables adaptées aux besoins médicaux et vétérinaires actuels pour lutter contre des maladies infectieuses.»

31 – Lucie ROUZOULENS

*Anti-Infectieux: supports moléculaires des résistances et innovations thérapeutiques - RESINFIT
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Development of a pseudotyped retroviral particle system for Human Cytomegalovirus (HCMVpp)

Lucie Rouzoulens, Sophie Alain and Jean-christophe Meunier.

Human cytomegalovirus (HCMV) infection is generally asymptomatic in healthy individuals, but the virus will persist for life in most adults. Reactivation is often observed in immunocompromised patients and can lead to serious organ damages and graft rejection for transplanted patients. Also, infection of the fetus is the major cause of congenital birth defect. Antivirals targeting HCMV are available but their effectiveness is largely partial and serious side effects are common. Thus, it is essential to develop safe, innovative and more robust therapeutic strategies. In this project, we aim at deciphering precisely the entry step mechanisms in order to identify pertinent viral envelop protein therapeutic targets. However, studies aiming at characterizing the viral entry strategy using cell culture systems were limited so far by the sizeable genomic complexity of HCMV, and the unusually high number of envelope protein displayed on this virus. To overcome this "protein background noise", we want to develop a simplified "pseudo-virus" system for HCMV. The main interest of this "retroviral pseudoparticles system" is that envelope proteins expressed on the surface of the infectious particles can be chosen at will. Then, the role of envelope proteins can be precisely deciphered whether expressed alone or in combinations. After validation of this HCMVpp system, we will construct retroviral particles carrying envelope proteins of different viral strains and recombinants. We will then perform a comparative analysis of viral entry and resistance of these particles. In order to better understand the mother-to-infant HCMV transmission mechanisms, we will also identify "differentiation epitopes" on envelope proteins. The influence of these epitopes on viral entry and neutralization will be examined using the HCMVpp system. The HCMVpp system we will develop may be highly useful for the international scientific community for implementing new innovative therapeutic strategies.

32 –Lisa SCILLIA

Microbiologie Fondamentale et Pathogénicité - MFP - UMR 5234 – CNRS, Université de Bordeaux

Modified Antisense Oligonucleotides : a new advance against antibioresistance ?

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Antibioresistance is a major global public health concern. Its human impact was assessed at 1.27 million deaths worldwide in 2019, and the latest estimates predict

up to 39 million deaths by 2050, or approximately 2.4 million annually (Kariuki S., *The Lancet*, 2024). Combating antibiotic resistance therefore requires coordinated and urgent action. In its 2024 priority pathogens list, the World Health Organization (WHO) categorized Enterobacteriaceae such as *Escherichia coli* producing extended-spectrum β -lactamases (ESBLs)—notably those resistant to third-generation cephalosporins (3GCs)—as critical for Research & Development.

One promising strategy to counter antibiotic resistance involves improving existing antibiotics. In this context, the ARNA and MFP laboratories (Bordeaux University) have developed an approach using antisense oligonucleotides (ASOs) complementary to the blaCTX-M-15 mRNA, with the aim of inhibiting the translation of CTX-M-15, the most prevalent ESBL in Europe.

The experimental model used is an *E. coli* TcK12 strain expressing CTX-M-15, which confers resistance to ceftriaxone (CFX), one of the most widely prescribed 3GCs. The addition of a lipid moiety (LASO) at the 5' end of the ASO led to a 26-fold reduction in the minimum inhibitory concentration (MIC) of CFX (MICCFX) for both the TcK12 strain and a clinical isolate.

Western blot analyses ($n=2$) quantifying CTX-M-15 protein levels in TcK12 demonstrated a 4-fold inhibition in the presence of LASO alone, and a 2-fold inhibition when LASO was combined with CFX. However, the specificity and reproducibility of these results remain to be fully validated.

In the event that non-specific or non-reproducible effects are confirmed, alternative mechanisms responsible for the observed reduction in MICCFX will need to be explored in order to substantiate the therapeutic potential of LASOs as a strategy to combat antimicrobial resistance.

33 – Sephora THERESINE-AUGUSTINE

Microbiologie Fondamentale et Pathogénicité – MFP – UMR 5234 – CNRS, Université de Bordeaux

Optimization of a gut-on-chip for the study of the pathogenicity of *Candida* yeasts.

Sephora THERESINE-AUGUSTINE, Fernanda LOPEZ-GARCIA, Karine DEMENTHON.

Commensal *Candida* yeasts, can become pathogenic and resistant to treatments when the host immune system is compromised, leading to severe infections. Current biological models fail to accurately replicate the complexity of the human body. Organs-on-chips (OOCs) are an innovative alternative to model organ functionality and recapitulate some of their physiological or pathological features in-vitro. Building on previous results demonstrating the successful differentiation of caco-2 cells into a functional intestinal epithelium layer, we propose to enhance the currently developed gut-on-a-chip by vascularizing it and making it immunocompetent. Our aim is to get closer to a physiologically realistic microenvironment to study intestinal *Candida* infections. In this project, we managed to obtain a differentiated endothelial monolayer whose cells align themselves in the direction of flow. We showed that circulating THP-1 immune cells are able to migrate between the microfluidic channels in response to inflammation triggered by *C. albicans* infection. It was also possible to observe yeast being phagocytized by THP-1 cells. These findings demonstrate the potential of the gut-on-a-chip as a powerful tool for studying the dynamics of *Candida* infections and immune responses.





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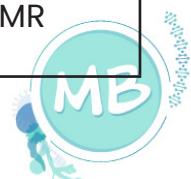
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