Pathobiome 2018

18th-20th March 2018 –

Palais des congrés

Ajaccio, France

Abstract book

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Welcome message

It is a real pleasure to welcome you for this second International Symposium on the “Pathobiome”.

The thread of this meeting is the pathology at the omics era.

Understanding whether and how microorganisms help or harm has always been a major challenge since the discovery of microbes and of the threats they constitute for humans and their domesticated animals and plants. Large-scale sequencing has revolutionized microbiology by giving us access to a wealth of previously unsuspected microorganisms. It has made us microbiologists revisit the pathology paradigm, inherited from Pasteur and Koch, on “one pathogen-one disease”, to fully embrace Haeckel and Darwin theories in ecology and evolution. We now look at the pathology as a dysfunctioning of a whole ecosystem, which size can be as small as an individual, e.g. a human, an animal, a plant, or as large as specific environments, e.g. soils, fields, prairies, forests or lakes.

This meeting series thus emerged from the convergence of microbiology classical disciplines including virology, bacteriology or parasitology and second, it integrated mathematic modelling, statistics and computer sciences. The challenges ahead are enormous to overcome scientific and technological issues and we need to develop artificial intelligence approach to support our own “natural” intelligence! It is with a bit of uncertainty and angst but foremost with enthusiasm that we feel this is the way we have to “reinvent” microbiology, with an “EcoHealth” approach, as we know now that human health largely depends on the health of its environment.

The aim of this meeting is to seed and share new ideas and results, and challenge our minds with new concepts. We have invited speakers renowned in their fields to share their experience and their feelings; these interactions will be of particular interest for young scientists. Beyond getting to know each other and exchanging ideas, we hope that the informal times during this meeting will set up new collaborations and reinforce existing ones.

Changes in Science also have an impact on its interactions with Society. The increased sophistication and power of our understanding and methods raise hopes and fears. Both sometimes originate from insufficient or biased media coverage. It is, thus, also our mission to explain “our science” to the public, avoiding hype and triumphalism, but pointing out scientific advances and their potential positive or negative societal impact. As part of this meeting, we have on Sunday evening a specific general audience conference in French on the “Human microbiome and the pathology” given by Joel Doré, a pioneer in the field. With this conference, and the ensuing question time, we hope to promote interactions with a large audience of people interested in scientific issues and methods, and how they can meet societal concerns.

Finally, it is also a special pleasure to welcome you to Corsica, a wonderful environment and a frontline laboratory as an island, for the survey of microbial ecosystems confronted to global changes and the intensification of trade. Scientists are particularly welcome here!

Pascale Serror & Mylène Ogliastro,
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Scientific Program

Sunday, March 18, 2018

16:00 – 17:00 Registration of Attendees

17:00 - 19:00 Welcoming and introduction

Science-Society interaction — Public lecture in French on the role of the microbiota in infectious diseases.

Joël Doré (INRA, Jouy en Josas, France)

Monday, March 19, 2018

08:30-08:45 Welcoming introduction

08:45-09:30 Opening lecture — Anna-Liisa LAINE (Helsinki University, Finland)

Within host pathogen diversity - causes and consequences.

Session 1: From pathogens to pathobiome

9:30-10:30 Keynote lecture — Muriel Vayssier-Taussat (INRA, France)

From tick borne pathogens to tick borne pathobiome.

10:30-11:30 Posters & Coffee Break

11:30 -12:30 Oral presentations

James Doonan (Bangor University, UK)
Distinguishing pathogen from symbiont in the acute oak decline pathobiome.

Kari Saikkonen (Natural Resources Institute Finland, Finland)
The outlook for plant pathobiome research.

Emilie Lejal (INRA, Maisons-Alfort, France)
Dynamics and co-occurrences of *Ixodes ricinus*-borne pathogens.

12:45-14:00 Lunch break

Session 2: From the host to pathobiome

14:00-15:00 Keynote lecture — Brett Finlay (University of British Columbia, Canada)

The role of the microbiome in enteric infections.

15:00-16:00 Oral presentations

Shaynoor Dramsi (Institut Pasteur, France)
Competitive advantage of *Streptococcus gallolyticus* in colon cancer microbiota

Pascale Serror (INRA, Jouy-en-Josas, France)
Lactobacillus paracasei CNCM I-3689 reduces vancomycin-resistant *Enterococcus* persistence and promotes *Bacteroidetes* resilience following antibiotic challenge.

Philippe Velge (INRA, Nouzilly, France)
Salmonella carrier state in chicken: role of gut microbiota in the Super-shedder and Resistant phenotypes.

16:00-17:00 Posters & Coffee Break

17:00-18:30 Oral presentations — Nicolas Barnich (Université Clermont Auvergne, Inserm, INRA, France)

Adaptation of the pathobiont adherent-invasive *E. coli* to gut environment in the context of Crohn's disease.

Yannick Tremblay (Institut Pasteur, Paris, France)
Role of deoxycholate in induction of *Clostridium difficile* biofilm formation.

Biet Franck (INRA, Nouzilly, France)
Mycobacterium avium subsp. *paratuberculosis* hosted by free-living amoebae .

Derek Lundberg (Max Planck Institute for Developmental Biology, Germany)
Are strong natural ACD6 alleles in *A. thaliana* microbial paranoia?

Tuesday, March 20, 2018

08:30-09:15 Opening lecture — Stéphane Hacquard (Max Planck Institute for Plant Breeding, Germany)
Microbial interactions and community assembly at the root-soil interface.

Session 3: The pathobiome as a driver of pathogen adaptation and diversification

09:15-10:15 Keynote lecture — Fabrice Roux (INRA/CNRS, Toulouse, France)
A genomic map of local adaptation to microbiota and potential pathobiota in *Arabidopsis thaliana.*

10:15-11:15 Posters & Coffee Break

11:15-12:35 Oral presentations — Talia Karasov (Max Planck Institute, Germany)
Long-term maintenance and expansions of dozens of pathogenic *Pseudomonas* strains within *A. thaliana* populations.

Tania V. Fort (INRA, Pessac, France)
Vertical transmission of microbiota is influenced by maternal and environmental effects in sessile oak (*Quercus petraea*).

Philippe Roumagnac (CIRAD, France)
Geometagenomics illuminates the impact of agriculture on the distribution and prevalence of plant viruses at the ecosystem scale.

Rosanna Wright (University of York, UK)
The structure and genetics of cross-resistance evolution in bacteria-phage interactions.

12:45-14:00 Lunch break

Session 4: Statistical and Mathematical modeling to unravel interactions

14:00-15:00 Keynote lecture — Otso Ovaskainen (University of Helsinki, Finland)

How to make more out of community data? A conceptual framework and its implementation as models and software.

15:00-16:00 Oral presentations — Julien Chiquet (AgroParitech/INRA, Paris, France)
Network Reconstruction from Ecological Count Data with a Sparse Poisson Lognormal Model.

Arnaud Cougoul (INRA, Theix, France)

Limits of statistical detection of microbial associations from metagenomic data.

PAUVERT Charlie (INRA, Bordeaux, France)
Cropping system shapes foliar fungal networks of grapevine.

16:00-16:30 Young scientific awards for oral and poster presentations.

Conférence Grand public
Sunday 18 March 2018, Palais des congrès, Ajaccio.

La symbiose Homme-microbiote dans la santé et la maladie.

Joël Doré. Micalis & Metagenopolis, INRA, Jouy-en-Josas, France.

L'Homme est une symbiose qui se met en place dès la naissance entre des cellules, tissus et organes humains d'une part et un cortège de microorganismes d'autre part. Ils interagissent étroitement et l'harmonie de cette relation est un élément majeur du maintien en bonne santé. Chaque personne porte ainsi 100.000 milliards de bactéries dont la population la plus dense est localisée dans l'intestin. Ce microbiote contribue à la digestion, produit des vitamines et des composés et molécules signales d'intérêt pour l'hôte, protège de la prolifération de microorganismes environnementaux et éduque et stimule le système immunitaire. Il influence également le comportement via l'axe intestin-cerveau.

Des avancées techniques majeures ont permis de dépasser l'impossibilité de cultiver de nombreux symbiontes du microbiote dominant. Le dernier pas a consisté à caractériser le microbiote par une approche de métagénomique, donnant accès à l'intégralité des gènes du microbiote dominant, et ainsi à son potentiel fonctionnel global. Ces travaux ont permis de décrire 10 millions de gènes et de mettre en évidence les 3 grandes structures écologiques du microbiote : les entérotypes. Ils ont également conduit à documenter une altération de la composition du microbiote dans l'ensemble des grandes pathologies de société dont l'incidence n'a cessé de croitre depuis les années 1950. Ces maladies chroniques non-transmissibles - inflammatoires, métaboliques, allergiques, dégénératives, et neurologiques (neurodégénératives et psychiatriques) - présentent toute une altération de composition du microbiote intestinal dominant mais également toujours une déviation de paramètres de physiologie qui traduisent une altération de la symbiose Homme-microbes. De façon récurrente, ce sont une perte de diversité (de richesse en gène notamment), la diminution des proportions de certaines bactéries protectrices du microbiote normal et l'augmentation de pathobiontes qui sont observées du côté du microbiote. En parallèle du côté de l'hôte les grandes constantes sont une perméabilité intestinale accrue, un tonus inflammatoire élevé et une situation de stress oxydant local et systémique. Il est probable que ces signaux d'aggravation respectifs conduisent à un état pathologique qui s'auto-entretient en un cercle vicieux dans nombre de contextes de pathologies chroniques ; une situation que la médecine actuelle, médecine de l'organe centrée sur les symptomes, n'est pas préparée à traiter dans sa globalité.

Plusieurs facteurs environnementaux ont pu contribuer à cette altération de la symbiose Homme-microbes en quelques générations. La transition nutritionnelle qui a conduit à passer de plus de 60 à moins de 20 grammes de fibres en moyenne par jour, avec un apport accru de sucres simples et de graisses et protéines animales, aura eu un impact fort sur la diversité globale et la composition du microbiote. Les pratiques et l'environnement entourant la naissance ont également beaucoup changé allant jusqu'à interdire la transmission verticale normale du microbiote de la mère au nouveau-né, comme le fait le recours aux antibiotiques en période périnatale, ou encore la césarienne à laquelle certaines régions du monde ont recours pour 9 naissances sur 10 aujourd'hui. L'exposition à de nombreux xénobiotiques alimentaires et environnementaux est enfin un facteur d'agression des muqueuses et de sollicitation du système immunitaire qui s'est accru même s'il reste encore peu documenté. Le constat actuel est alarmant avec la perspective d'ici 2025 d'environ une personne sur 4 touchée par l'une des pathologies chroniques listées ci-dessus. Cela vaut aussi pour les maladies infectieuses dans la mesure ou les agressions imposées de façon répétée à nos microbiotes conduisent à la fragilisation ou la perte de cette fonction clé qu'est la barrière contre la prolifération de bactéries environnementales, notamment pathogènes. L'un des exemples extrêmes bien connu est l'infection à *Clostridium difficile* induite à l'hôpital par un traitement antibiotique. Cette observation souligne l'urgence de prendre en compte la symbiose Homme-microbes parmi les éléments de physiopathologie potentiels dans un contexte d'augmentation de l'incidence des maladies que la génétique ne peut en aucun cas expliquer.

L'analyse métagénomique du microbiote peut fournir des informations importantes pour le clinicien. Les travaux récents ont montré que la richesse en gènes du microbiote dominant, signature de sa

diversité, est un marqueur de santé. Une faible richesse en gènes du microbiote est associée à un phénotype dégradé dans les maladies métaboliques et prédit également une moins bonne réponse à une intervention nutritionnelle. Dans les maladies inflammatoires, elle est associée à une plus grande fréquence des phases aigües de la maladie. Dans les maladies hépatiques, la perte de richesse est associée à la sévérité de la condition du patient. La faible richesse est aussi associée à une moindre robustesse et accompagne souvent les processus infectieux, y compris lorsqu'elle est iatrogénique, induite par une démarche thérapeutique. Il devient dès lors assez clair qu'un monitoring dans le temps permettrait de prédire le risque et la vitesse d'aggravation de nombreuses pathologies.

Si l'on a aujourd'hui, par nos changements conscients de modes de vie, d'habitudes alimentaires et de pratiques cliniques, fragilisé la symbiose Homme-microbes au point de faire la part belle à de nombreuses pathologies, il n'en reste pas moins que la compréhension au plan mécanistique la rupture de symbiose peut être la clé de la restauration d'un contexte protecteur. Comprendre la relation homme-microbes c'est en effet se donner les moyens d'agir par l'apport de petites molécules ciblant le dialogue bactéries-cellules, ou des souches bactériennes symbiontes protectrices, voire de cocktails ou d'écosystèmes complets comme le propose aujourd'hui le transfert de microbiote allogénique ou autologue.

Pour conclure, il est légitime de reconnaitre que beaucoup reste à faire pour documenter au plan mécanistique la rupture de symbiose Homme-microbes dans les troubles psychiatriques pour en faire un levier potentiel d'accompagnement des approches thérapeutiques existantes.

Pour en savoir plus …

Blottière HM, deVos WM, Ehrlich SD, Doré J. Human intestinal metagenomics – state of the art and future. Curr Opin Microbiol 2013, in press. [Review]

Le Chatelier E, Nielsen T, Qin J, Prifti E, Hildebrand F, Falony G, Almeida M, Arumugam M, Batto JM, Kennedy S, Leonard P, Li J, Burgdorf K, Grarup N, Jørgensen T, Brandslund I, Nielsen HB, Juncker AS, Bertalan M, Levenez F, Pons N, Rasmussen S, Sunagawa S, Tap J, Tims S, Zoetendal EG, Brunak S, Clément K, Doré J, Kleerebezem M, Kristiansen K, Renault P, Sicheritz-Ponten T, de Vos WM, Zucker JD, Raes J, Hansen T, MetaHIT consortium, Bork P, Wang J, Ehrlich SD, Pedersen O. Richness of human gut microbiome correlates with metabolic markers. Nature. 2013, 500:541-546.

Lepage P, Leclerc M, Joossens M, Mondot S, Blottière HM, Raes J, Ehrlich SD, Doré J. A metagenomic insight into our gut's microbiome. Gut. 2013 Jan;62(1):146-58.

Li J, Jia H, Cai X, ... Pedersen O, Doré J, Ehrlich SD, MetaHIT Consortium, Bork P, Wang J. An integrated reference gene catalog of the human gut microbiome. Nature Biotech. 32:834-841. 2014

Larraufie P, de Wouters P, Potocki-Veronese G, Blottière HM, Doré J. 2015. Functional metagenomics to decipher food–microbe–host crosstalk. Proceedings of the Nutrition Society. 74:1–4.

Dore, J and Blottiere, H. 2015. The influence of diet on the gut microbiota and its consequences for health. Curr Opin Biotechnol.. 32:195-199

Blottiere, H and Dore, J. 2016. Impact des nouveaux outils de métagénomiques sur notre connaissance du microbiote intestinal et de son rôle en santé humaine. Médecine Science. 32: 944-51

Oral presentations

Opening lecture, day 1

K1. Within-host pathogen diversity – causes and consequences.

Anna-Lisa Laine
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Pathogens are prevalent across all ecosystems and they may have strong negative effects on their hosts. Hence, there is a pressing need to understand risks of infection and how these evolve. To date, host-pathogen interactions have been largely viewed within the one host-one parasite framework although in reality the same host may be attacked by a myriad of pathogenic microbes. As molecular tools have become increasingly available for the study of parasites, we now know that a single host individual can support a highly diverse pathogen community. However, remarkably little is known about the factors that determine which pathogens co-occur within the same host individual and how they interact.

In my talk I will give a brief overview of what are the current opportunities and gaps in knowledge in understanding the causes and consequences of variable within-host pathogen communities. I will then present 1) A case study of strain diversity of *Podosphaera plantaginis* infecting the same host. Our analysis of 1363 pathogen populations for two consecutive years revealed coinfection – a prerequisite of sex - to be spatially variable but more common than expected by chance. We identify hotspots for recombination and a direct ecological benefit of outcrossing. Jointly these results confirm that outcrossing has direct epidemiological consequences as well as a major impact on pathogen genetic diversity, thereby promoting facultative sex in pathogens. 2) A case study of spatially variable virus communities infecting a highly fragmented host population network. Experimentally we have assessed the relative roles of the host and environment in determining the structure of virus communities.

Session 1: From pathogens to pathobiome

Keynote lecture

K2. From Tick-Borne Pathogens to Pathobiome.

Muriel Vayssier-Taussat,
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Ticks, as vectors of several notorious zoonotic pathogens, represent an important and increasing threat for human and animal health in Europe. Until recently, research laboratories were used to be specialized on a limited number of tick-borne pathogens. Within the last few years, the rapid development of Next Generation Sequencing and various high throughput methods has facilitated complete screening of pathogens within their hosts, discovery of new pathogens or the detection of unexpected ones. As expected, those approaches have allowed detection without *a priori* established tick-borne pathogens, such as the *Borrelia*, *Anaplasma*, *Coxiella*, *Francisella* or *Rickettsia* genus. Those genera, belong to both pathogenic bacteria for vertebrates and/or endosymbionts. The genetic proximity of some tick-borne pathogens to mutualistic symbionts hosted by ticks is indeed evident when studying phylogenies of several bacterial genera. Consequently, ticks represent a compelling yet challenging system in which to study microbiomes and microbial interactions, and to investigate the composition, functional, and ecological implications of bacterial communities. Appreciation of these complex systems is expanding our understanding of tick-borne pathogens, leading us to evolve a more integrated view that embraces the 'pathobiome'; the pathogenic agent integrated within its abiotic and biotic environments. Deciphering the relationships within pathobiome will garner invaluable information, which may aid in the future development of vector-borne pathogen transmission control strategies.

O1. Distinguishing pathogen from symbiont in the acute oak decline pathobiome.

Doonan J[1]*, Denman S[2], Pachebat JA[3], McDonald JE[1]

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The United Kingdom's native oak are under serious threat from Acute Oak Decline (AOD). Stem tissue necrosis is a primary symptom of AOD and several novel bacteria are associated with necrotic lesions. Two members of the lesion pathobiome, *Brenneria goodwinii* and *Gibbsiella quercinecans*, have been identified as causative agents of necrosis. However, additional bacteria including *Lonsdalea quercina* ssp. *britannica* and *Rahnella* spp. have been detected in the lesion microbiome, but their role in the decline is unclear. Consequently, information on the genome-encoded virulence factors that mediate lesion formation is critical to understand the role of these bacteria as pathogens or symbionts in the AOD lesion microbiota. Here, the whole genomes of lesion microbiota were sequenced, annotated and compared against canonical bacterial phytopathogens and symbionts. Genome-wide virulence factor clustering demonstrated that all studied members of the AOD lesion microbiota possessed genes associated with phytopathogens. However, the genome of *B. goodwinii* was characteristic of a necrogenic phytopathogen, validating ecological studies that implicate it as the key causal agent of AOD lesions. We demonstrate that in combination with ecological data, whole genome sequencing provides key in complex pathosystems.

Key words: Acute Oak Decline (AOD), *Brenneria goodwinii*, *Gibbsiella quercinecans*, necrosis, pathobiome, phytopathogens

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O2. The outlook for plant pathobiome research.

K. Saikkonen*

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Recent advances in tools and technologies in molecular biology have broadened capabilities to study diversity and functions of pathobiomes. Utilizing these new tools and technologies allows exploring the whole plant microbiome, and "the omics" approaches examining e.g. how genomics, proteomics and metabolomics translate into structures and functions involved in the interactions among plant and its microbial partners. Partly resultant contemporaneous insight has been the hologenome theory of evolution. However, human perspective and conventional disciplines of life sciences might constrict and/or distract understanding the nature of microbe-plant interactions. Here, I discuss the spectrum of plant-microbe emphasizing that these interactions are labile in ecological and evolutionary time. Consequently, the very same microbes are often "labelled" as pathogens, parasites and endophytes and sometimes even named differently. I propose that diverse assemblages of plant associated microbes form a holobiont, an extended phenotype and the target of phenotypic selection. Then, I contend that pathogen–plant interactions follow similar evolutionary and ecological processes as other host–mutualist or host–parasite interactions, and therefore need not to be treated differently. Taking into account this spectrum and complexity of interactions, plant microbiome provides a fertile ground for ecologists and evolutionary biologists interested in multi-trophic interactions, evolution of life histories, co-evolutionary processes and speciation.

References

Saikkonen K., S.H. Faeth, M.L. Helander And T.J. Sullivan (1998) Fungal endophytes: a continuum of interactions with host plants. Annu. Rev. Ecol. Syst. 29: 319-343.
Saikkonen K., P. Wäli, M. Helander And S.H. Faeth (2004) Evolution of endophyte-plant symbioses. Trends Plant Sci. 9: 275-280.
Wäli P. P., P.R. Wäli, K. Saikkonen and J. Tuomi (2013) Is the pathogenic ergot fungus a conditional defensive mutualist for its host grass? PLoS ONE 8(7): e69249.

Keywords: holobiont, pathogens, parasites, endophytes, multi-trophic interactions, evolution of life histories, co-evolution

O3. Temporal variability and ecological patterns of *Ixodes ricinus*-borne pathogens over three consecutive years.

Lejal E.*, Pollet T. and Vayssier-Taussat M.

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Ixodes ricinus is the predominant tick species in Europe and is recognized as the primary European vector of bacterial diseases in humans. This tick species carries a large variety of pathogens, including *Borrelia burgdorferi* sensu lato, the causative agent of Lyme borreliosis, and is frequently co-infected (Moutailler et al., 2016). The recent pathobiome concept (Ryan et al., 2013 ; Vayssier-Taussat et al., 2014, 2015) leads us to adapt our point of view and consider pathogens in their abiotic and biotic environment while studying them. While more information is now available on *I. ricinus*-borne pathogens, little is known about their temporal dynamics. Using a high-throughput screening method (microfluidic PCRs), we characterized the *I. ricinus*-borne pathogen dynamics through three consecutive years and determined if this dynamic follows recurrent seasonal patterns. We led an ecological survey on the temporal dynamics of *I. ricinus*-borne pathogens. About 1000 ticks (nymphs and adults) were collected monthly for 3 years in the Senart forest (south of Paris, France). After individual crushing and DNA extraction, ticks were analysed using the microfluidic PCRs approach, allowing to assess in each tick, the presence and prevalence of 40 selected pathogens. Over the one thousand ticks tested from April 2014 to Mai 2017, 29.7% were positive for at least one tested micro-organism and 4.5% were co-infected with at least two pathogens. Four different genera were mainly identified: *Borrelia* spp, *Anaplasma* spp, *Rickettsia* spp, and *Babesia* spp. High variations of pathogen infection rate were observed from one month to another, with a minimum of 14.3% in October 2014 and a maximum of 50.9 % in June 2014. However, no clear temporal patterns (i.e. seasonality) were detected and no yearly recurrence was observed whatever the taxonomic scale. These innovative results are the first step before studying potential links between *I. ricinus* microbiome (16S rRNA gene sequencing) and pathogen dynamics using networks analysis.

References : Moutailler S, Valiente Moro C, Vaumourin E, Michelet L, Tran FH, Devillers E, et al. (2016) Co-infection of Ticks: The Rule Rather Than the Exception. PLoS Negl Trop Dis. 10(3): e0004539. Vayssier-Taussat M, Kazimirova M, Hubalek Z, Hornok S, Farkas R, Cosson JF, et al. (2015) Emerging horizons for tick-borne pathogens: from the 'one pathogen–one disease' vision to the pathobiome paradigm. Future Microbiol. 10(12): 2033–2043. Vayssier-Taussat M, Albina E, Citti C, Cosson JF, Jacques MA et al. (2014) Shifting the paradigm from pathogens to pathobiome: new concepts in the light of meta-omics. Front Cell Infect Microbiol. 4:29. Ryan E. (2013) The Intestinal Pathobiome: Its Reality and Consequences Among Infants and Young Children in Resource-Limited Settings. The Journal of Infectious Diseases, 208 (11): 1732–1733.

Keywords: Tick-borne pathogens - *Ixodes ricinus* - Pathogen dynamics - Seasonality

Session 2: From hosts to pathobiome

Keynote lecture

K3. The role of the microbiome in enteric infections.

B. Brett Finlay
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It is now well established that the microbiota have a profound influence on the outcome of enteric infections, including those caused by *Salmonella* and pathogenic *Escherichia coli*. However, the mechanisms behind these effects are still poorly understood. We have begun to closely examine the interactions between microbiota and the enteric pathogens *Salmonella* and *Citrobacter rodentium* (as a model for EPEC and EHEC). We have found significant cross talk between the pathogens, the microbiota and their metabolites, and host responses, including effects on pathogen virulence systems, affecting gene expression. This has led to the identification of genes that are known to impact virulence, yet their mechanisms are undefined. Collectively, by better understanding the crosstalk between microbiota and enteric pathogens will not only increase our understanding of pathogenesis, but provide potential future methods to develop novel preventatives.

O4. Competitive advantage of *Streptococcus gallolyticus* in colon cancer microbiota.

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Colonization by *Streptococcus gallolyticus* subsp. *gallolyticus* (SGG) is strongly associated with the occurrence of colorectal cancer (CRC), although the factors leading to its successful colonization are unknown. Here we elucidate crucial steps that explain how CRC favors SGG colonization. Using mice genetically prone to CRC, we show that SGG colonization is 1,000-fold higher in tumor-bearing mice than in normal mice. This selective advantage occurs at the expense of resident intestinal enterococci. A novel SGG-specific locus encoding a bacteriocin (gallocin) is shown to kill enterococci *in vitro*. Importantly, bile salts strongly enhance this bacteriocin activity *in vivo*, thus leading to greater SGG colonization. Constitutive activation of the Wnt pathway, among the earliest signaling alterations in CRC, increases luminal bile acid production and favors colonization by SGG. We conclude that CRC-specific conditions promoted SGG colonization of the gut by replacing commensal enterococci in their niche.

Keywords: *S. bovis* biotype I, colon, bacteriocin, colorectal cancer, APC/Notch

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O5. *Lactobacillus paracasei* CNCM I-3689 reduces vancomycin-resistant *Enterococcus* persistence and Bacteroidetes resilience following antibiotic challenge.

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Among intestinal pathobionts, enterococci, in particular vancomycin-resistant enterococci (VRE), are a leading cause in critically ill, elderly and immunocompromised patients of health-care associated and community-acquired infections with life threatening issues. Promoting intestinal resistance against enterococcal overgrowth and carriage after antibiotic treatments could reduce the risk of VRE infections. Our objective was to investigate the effects of two *Lactobacillus* strains on the colonisation and persistence of a VRE strain. We used an intestinal colonisation mouse model based on an antibiotic-induced microbiota dysbiosis to mimic enterococci overgrowth and VRE persistence. Each *Lactobacillus* spp. was administered daily to mice starting one week before antibiotic treatment until two weeks after antibiotic and VRE inoculation. For each trial, we monitored transient colonisation and persistence of VRE and indigenous enterococci, and we characterized the fecal

microbiota longitudinally by 16S rRNA gene sequencing. At endpoint, we analysed ileal and colonic response on a selection of host genes by transcriptomic analysis. Of the two strains, *Lactobacillus paracasei* CNCM I-3689 improved VRE-shedding. Resilience of mainly Bacteroidetes associated with a modulation of host factors leads us to propose an indirect anti-VRE effect related to recovery of members of the phylum Bacteroidetes. This work supports non-antibiotic strategies against opportunistic enterococci after antibiotic-induced dysbiosis, and opens research avenues to identify keystone species of anti-VRE effect.

Keywords: Vancomycin-resistant *Enterococcus* (VRE), colonization resistance, gut microbiota, antibiotics.

O6. *Salmonella* carrier state in chicken: role of gut microbiota in the super-shedder and resistant phenotypes.

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Salmonella are enteric bacteria recognized as an important economic and public health problem throughout the world. Depending on serovar and host, *Salmonella* infection can range from a life-threatening systemic infection to an asymptomatic infection. *Salmonella*-carrier animals are a serious food safety issue, because high-level shedding infected individuals may contaminate human through consumption of infected meat. However, despite the importance of *Salmonella* persistence as a reservoir of disease, little is known concerning the mechanisms at play in farm animals. The development of a new infection model in isolator allowed us to show that in absence of animal reinfections and cross contaminations between chicks, *S. Enteritidis* induces a heterogeneity of fecal excretion and caecal colonization contrary to what is observed in flocks. This heterogeneity of infection corresponds to extreme phenotypes including (1) Super-shedder individuals, functioning as a reservoir for the pathogen and constantly disseminating *Salmonella* (2) Resistant chicks. Multiple well-known factors influence the dynamics of *Salmonella* in its hosts, including features of host (health, age, immune system status, host genetics, diet) or pathogen (virulence factors, exposure dosage). Our data showed that modulation of gut microbiota before infection modulates *Salmonella* colonization. Consistent with this idea, the transfer of gut microbiota, collected before infection from individuals that have later developed the high-shedding syndrome yielded to the development of the Super-shedder phenotype 4 days post infection. In line with this experiment, the analysis of faecal microbiota composition before and after infection revealed significant differences among Super-Shedder and Resistant chicks. Moreover, presence of some gut bacteria before infection correlated to the Resistant phenotype. Taken as a whole, these results suggest that *Salmonella* colonization is inhibited and/or promoted by a subset of microbes naturally found in varying abundances within the gut microbiota. Such microorganisms may naturally be missing or scarce before infection in the chicks that later develop the Super-shedder phenotype. This work opens new avenues to identify gut bacteria able to prevent *Salmonella* colonization and to define new strategies to control *Salmonella* infection.

Keywords: *Salmonella*, carrier state, gut microbiota, Super-shedder, chicken.

O7. Adaptation of the pathobionte adherent-invasive *Escherichia coli* to gut environment in the context of Crohn's disease.

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Crohn's Disease (CD) is a chronic inflammatory disease of the gastrointestinal tract involving the dynamic interaction of host genetics, the microbiome and inflammatory responses. Ileal mucosa of CD patients is often abnormally colonized by adherent-invasive *Escherichia coli* (AIEC). AIEC are considered to be pathobionts, because they colonize and promote inflammatory diseases due to the adaptive evolution of their genome in a susceptible host. AIEC virulence factor expression is required for optimal gut colonization capacity. Interestingly, differential flagellin regulation was observed between commensal *E. coli* (HS) and AIEC (LF82) strains: flagellum expression by AIEC bacteria, in contrast to that of commensal E. coli, is enhanced under intestinal conditions (bile acids and mucins). Flagella are involved in AIEC ability to cross mucus layer *in vitro* and *in vivo*, conferring a selective advantage in penetrating mucus layer and reaching epithelial surface. Global RNA sequencing of AIEC strain LF82 show an explosive effect of bile acids with a dysregulation of about 40% of the genome, with a global upregulation of genes involved in degradation and downregulation of those implicated in several biosynthesis. Ethanolamine utilization bestows a competitive advantage on AIEC strains, which are capable of degrading it. Activation of metabolic pathways that interplay together to provide an energy benefit to AIEC. Finally, AIEC utilize serine metabolism pathway to gain an edge over competing *E. coli* strains in the inflamed gut. Meanwhile, amino acid metabolism has minimal effect on their competitive fitness in the healthy gut. The availability of luminal serine used for the competition of *E. coli* is largely dependent on dietary intake; whereby the inflammation-induced blooms of AIEC are significantly blunted when serine are removed from the diet. These results improve the understanding of the global regulatory network under environmental stress and provide potential features to prevent AIEC colonization of patients with CD.

Keywords: adherent-invasive E. coli, gut adaptation, intestinal colonization and inflammation

O8. Role of deoxycholate in induction of *Clostridium difficile* biofilm formation.

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Clostridium difficile (CD) is a gram-positive spore forming bacterium, strictly anaerobic that is the most common cause of nosocomial diarrhoea. We assumed that biofilm formation might play a role in the adaptation of CD to gut colonization and/or relapse of the CD infection. Recent studies indicate that bile acids have different effects on the ability of CD to colonize GI tract. Indeed, both cholate (CA) and deoxycholate (DCA), a secondary bile acid metabolized from CA, stimulates CD spores germination. However, DCA is toxic to vegetative CD cells. Therefore, in healthy microbiota, CD vegetative cells are exposed to the toxic effect of DCA in the colon. In contrast, during dysbiosis, transformation of CA to DCA is prevented and the CA level remains high leading to spore germination and expansion of CD in the colon. We provide experimental evidence that DCA strongly stimulates CD biofilm formation on abiotic surface. The characterization of the matrix indicated that eDNA and

proteins are required for the DCA-induced biofilm while polysaccharides are not incorporated. In addition, global regulators such as CodY, CcpA and SigL involved in the metabolic regulation are essential for the biofilm formation in response to DCA. Finally, we showed that the adaption to long-term exposure to DCA repress production of toxins and spores directly via DCA or by the DCA-induced biofilm. Altogether these results suggest that DCA is an important signal in the infectious life cycle of CD that allow persistence of CD in the GI tract when a normal microbiota is restored, increasing the relapse's risk. In agreement when we tested the ability of CD to form mixed biofilm with *Clostridium scindens* (CS), a commensal bacteria metabolizing CA into DCA, we showed that dual-species cultures of CD with CS lead to an increase in biofilm formation and cell survival.

Keywords: *Clostridium difficile*, Bile salts, gut dysbiosis, Biofilm

O9. Environmental *Mycobacterium avium* subsp. *paratuberculosis* hosted by free-living amoebae.

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Mycobacterium avium subsp. *paratuberculosis* is responsible for *paratuberculosis* in animals. This disease, leading to an inflammation of the gastrointestinal tract, has a high impact on animal health and an important economic burden. The environmental life cycle of *Mycobacterium avium* subsp. *paratuberculosis* is poorly understood and several studies suggest that free-living amoebae might be a potential environmental host. Free-living amoebae are protozoa found in water and soil that are described as reservoirs of pathogenic and non-pathogenic bacteria in the environment. Indeed, bacteria able to survive within these amoebae would survive phagocytosis from immune cells. In this study, we assessed the *in vitro* interactions between several strains of *Mycobacterium avium* subsp. *paratuberculosis* and *Acanthamoeba castellanii*. The results indicate that strains including fields isolates representing the major genetic lineages were able to grow within the amoeba and that they can survive for several days within their host. To explore the presence of *Mycobacterium avium* subsp. *paratuberculosis* in environmental amoebae, we sampled water from farms positive for *paratuberculosis*. A *Mycobacterium avium* subsp. *paratuberculosis* strain was detected within an environmental amoeba identified as related to the poorly described *Rosculus* genus. The bacterial strain was genotyped, showing that it was similar to previous infectious strains isolated from cattle. In conclusion, we described that various *Mycobacterium avium* subsp. *paratuberculosis* strains were able to grow within amoebae and that these bacteria could be found on farm within amoebae isolated from the cattle environment. It validates that infected amoebae might be a reservoir and vector for the transmission of *Mycobacterium avium* subsp. *paratuberculosis*.

Keywords: *Paratuberculosis* Amoebae Reservoir, Vector Environment

O10. Are strong natural ACD6 alleles in *A. thaliana* microbial paranoia?

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Accelerated cell death 6 (ACD6) is a transmembrane protein widely conserved in *A. thaliana* that functions in a positive feedback loop with the defense hormone salicylic acid (SA). It helps to control expression of several immune receptors critical in detecting microbe-associated molecular patterns (MAMPs) (1). Wild plant populations maintain variation in this gene (2). In particular, some accessions including Est-1 have a hyperactive allele that, in lab conditions, results in necrosis in older leaves, overall higher SA levels, stunted growth, and more robust immunity to common pathogens (3). Other accessions such as Col-0 have milder alleles that do not impart these phenotypes. I planted outdoors in Germany and Sweden (where CRISPR/Cas9 mutants have a clear non-GMO legal status) several plant genotypes originating from Est-1 and Col-0 parents that have sequence variation only at the ACD6 locus, allowing for a direct test of the influence of ACD6 on microbiome assembly. The plants germinated in wild soil and overwintered exposed to the elements. I also grew the same lines in the institute greenhouse in wild soil. I processed rosettes from eight to twelve independent plants per location per genotype and will do so again for the current season, ultimately resulting in a dataset of nearly 300 plants; from each plant analysis includes 16S and ITS amplicon sequencing, metagenome sequencing, plant transcriptome sequencing, and plant hormone measurements including SA. I will present data from the 2016-2017 season. This work provides insight into the maintenance and selection of a gene for which a variety of alleles persist in wild stands.

References

(1) Zhang Z, Shrestha J, Tateda C. et al., Molecular Plant 7 (8): 1365–83 (2014).
(2) Świadek M, Proost S, Sieh D et al. The New Phytologist 213 (2): 900–915 (2016).
(3) Todesco M, Balasubramanian S, Hu T, et al. Nature 465 (7298): 632–36 (2010).

Keywords: microbiome, ACD6, phyllosphere, metagenome, field experiment

Opening lecture, day 2

K4. Microbial interactions and community assembly at the root-soil interface.

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The roots of healthy plants host a rich diversity of microbes that evolved independently within distinct kingdoms of life. However, the relevance of intra- and inter-kingdom microbe-microbe interactions for community assembly and host-microbiota balance is poorly understood. First, we examined the role of interbacterial antagonistic interactions in root microbiota establishment utilizing a phylogenetically-diverse culture collection of the *Arabidopsis thaliana* root microbiota. A large-scale screen for antimicrobial-mediated interbacterial interactions showed that the majority of strains exhibited unique inhibitory or sensitivity patterns and that antibiosis of root-associated commensals might be greater than that of soil bacteria during root microbiota establishment. We then selected 13 highly competitive (HC) and 13 highly sensitive (HS) bacterial strains for community perturbation experiments in a gnotobiotic plant system and showed that HC strains are key community members that sculpt bacterial assemblages along the soil-root continuum. Second, we profiled bacterial, fungal and oomycetal communities established in the roots of *Arabidopsis* populations and found disproportionate negative associations between root-associated prokaryotic and eukaryotic taxa (>90%). We established corresponding microbial culture collections, which comprise more than 50% of the most abundant root-associated microbes detected by culture-independent community sequencing. Re-colonization of germ-free Arabidopsis with mono- and multi-kingdom synthetic microbial consortia revealed the profound influence of the bacterial root microbiota on fungal and oomycetal community structure and diversity, leading to increased plant growth and survival. Deconvolution of 2,862 binary bacterial-fungal interactions *in vitro*, combined with perturbation experiments *in planta*, indicates that the protecting activity conferred by the bacterial root microbiota is a redundant trait that confers robust maintenance of host-microbial homeostasis.

Session 3: The pathobiome as a driver of pathogen adaptation and diversification

Keynote lecture

K5. A genomic map of local adaptation to microbiota and potential pathobiota in *Arabidopsis thaliana*.

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A current challenge in microbial pathogenesis is to identify biological control agents that may prevent and/or limit host invasion by microbial pathogens. *In natura,* hosts are often infected by multiple pathogens. However, most of the current studies have been performed under laboratory controlled conditions and by taking into account the interaction between a single commensal species and a single pathogenic species. The next step is therefore to explore the relationships between host-microbial communities (microbiota) and microbial members with potential pathogenic behavior (pathobiota) in a realistic ecological context. In the present study, we investigated such relationships within root and leaf associated bacterial communities of 163 ecologically contrasted *Arabidopsis thaliana* populations sampled across two seasons in South-West of France. In agreement with the theory of the invasion paradox, we observed a significant humped-back relationship between microbiota and pathobiota α-diversity that was robust between both seasons and plant organs. For the microbiota, we observed a strong dynamics of seasonal community succession in most populations. Accordingly, the potential pathobiota composition was explained by combinations of microbiota OTUs that were season specific. This result suggests that the potential biomarkers controlling pathogen's invasion are highly dynamic. Finally, we conducted a Genome Environment Association analysis using more than 1.4 million SNPs to finely map genomic regions associated with bacterial community descriptors. We detected neat peaks of association related to the presence/absence of co-occurring OTUs. In addition, the identification of QTLs for diversity and composition of bacterial communities highlighted the benefit of exploring diffuse biotic interactions. A substantial fraction of SNPs related to bacterial community descriptors were significantly enriched in the extreme tail of a genome scan of adaptive spatial differentiation, indicating that the identified candidate genes have been shaped by natural selection.

O11. Long-term maintenance and expansions of dozens of pathogenic *Pseudomonas* strains within *A. thaliana* populations.

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Crop disease outbreaks are commonly associated with the clonal expansion of single pathogenic lineages, until new genetic resistances are deployed by plant breeders. Whether similar boom-and-bust scenarios hold for natural plant pathogens is mostly unknown. To address this question, we are studying the bacterial genus, *Pseudomonas*, known to contain important plant pathogens, and its colonization of a wild host, *Arabidopsis thaliana*. We initially carried out a multi-year 16S rDNA survey of *Pseudomonas* in *A. thaliana* leaves at hundred-km scales. The most common lineage, defined as a 16S rDNA operational taxonomic unit (OTU), corresponded to a pathogenic clade present in all sites. It was the most successful plant colonizer and was significantly correlated with total microbial load. To determine whether this OTU ubiquity indicated the clonal expansion of a single *Pseudomonas* lineage, or whether the 16S rDNA similarity masked greater underlying diversity, we cultured and sequenced 1,524 *Pseudomonas* genomes from more than 130 *A. thaliana* individuals. We found that this persistent OTU was composed of dozens of pathogenic lineages that differ in gene content and disease phenotype. This clade diverged more than 100,000 years ago, prior to *A. thaliana* recolonization after the last glacial maximum. Single lineages reached high frequencies within plants, but different lineages dominated across plants. The abundance of this OTU, along with the maintenance of diversity in this pathogen lineage, suggests not only that genetic innovations underlying the success of the lineage are rooted deeply in the OTU phylogeny, but also that in contrast to crop systems, no single strain overtook *A. thaliana* populations. An important question for the future is how much of this is explained by host genetic diversity, and how much by an environment more complex than that in agricultural systems.

Keywords: Pathogen diversity, clonal expansion, wild vs. agricultural populations, from metagenomics to strains

O12. Vertical transmission of microbiota is influenced by maternal and environmental effects in sessile oak (*Quercus petraea*)

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Seeds facilitate transmission of microorganisms from one generation to another. These microorganisms can have beneficial, neutral or detrimental effects on plant fitness. Conversely, the dynamics and evolution of seed-borne microorganisms, including pathogens, depend on seed dispersal, seedling survival and microbial interactions within the seed. These ecological and

evolutionary relationships between the plant host, seed-borne pathogens and seed-associated microbiota have been overlooked so far. Here, we analyzed for the first time the microbial turnover in seeds of a forest tree species (*Quercus petraea*), using a hierarchical sampling design including three spatial scales (seed micro-environment, mother tree and forest site). A total of 125 acorns, collected either in the canopy or on the ground, were analyzed. The microbiota in the microenvironment of each seed (formed by twigs and leaves, or litter and soil, respectively) was also characterized. We focused on the fungal component of the microbial community as *Q. petraea* often hosts seed-borne fungal pathogens (*Ciboria* sp.). We combined a metabarcoding approach with quantitative PCR to characterize the richness, composition and abundance of the fungal community. Our results revealed significant effects of the mother tree and forest site on the fungal community of the acorns that are in the canopy. Interestingly, the maternal effects disappear when acorns fall on the ground as seed-borne fungal taxa are largely replaced by soil-borne taxa. Future analyses using Hierarchical Modelling of Species Communities (HMSC) will enable us to highlight the associations between seed-borne pathogens and other microorganisms. We will discuss the implications of our results on local adaptation processes.

Keywords: seed microbiota, maternal effect, fungal community, forest pathogen, metabarcoding, Quercus.

O13. Geometagenomics illuminates the impact of agriculture on the distribution and prevalence of plant viruses at the ecosystem scale.

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Disease emergence events regularly result from human activities such as agriculture, which frequently brings large populations of genetically uniform hosts into contact with potential pathogens. Although viruses cause nearly 50% of emerging plant diseases, there is little systematic information about virus distribution across agro-ecological interfaces and large gaps in

understanding of virus diversity in nature. Here we applied a novel landscape-scale geometagenomics approach to examine relationships between agricultural land use and distributions of plant-associated viruses in two Mediterranean-climate biodiversity hotspots (Western Cape region of South Africa and Rhône river delta region of France). In total, we analysed 1725 geo-referenced plant samples collected over two years from 4.5 km x 4.5 km grids spanning farmlands and adjacent uncultivated vegetation. We found substantial virus prevalence (25.8–35.9%) in all ecosystems, but prevalence and identified family-level virus diversity were greatest in cultivated areas, with some virus families displaying strong agricultural associations. Our survey revealed 94 previously unknown virus species, primarily from uncultivated plants. This is the first effort to systematically evaluate plant-associated viromes across broad agro-ecological interfaces. Our findings indicate that agriculture substantially influences plant virus distributions and highlight the extent of current ignorance about the diversity and roles of viruses in nature.

Keywords: Spatial metagenomics, plant-associated viruses, Mediterranean agroecosystems

O14. The structure and genetics of cross-resistance evolution in bacteria-phage interactions.

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Antagonistic coevolution between bacteria and phage, resulting in reciprocal selection for improved resistance and infectivity, highly influences the structure of microbial communities. However, the impact of specific resistance mutations is dependent on the structure and degree of cross resistance between phage strains. We selected spontaneous resistance mutants of *Pseudomonas aeruginosa* against 27 phage strains individually to determine the structure, genetic basis and cost of cross-resistance. Whilst the magnitude of cross-resistance provided by mutations selected against different phage strains varied greatly, this was not limited by associated fitness costs, and demonstrates that broad resistance ranges can be selected even in the absence of multiple phage strains. Using network analysis to characterise the structure of cross-resistance within the phage collection, we identified two distinct phage groups, with high within- but low between-group cross-resistance. Examining resistance mutations, which provide only within-group cross-resistance revealed a distinct molecular target for each group; resistance mutations targeted type IV pilus within Group 1 and lipopolysaccharide biosynthesis within Group 2. More generalist resistance, providing between-group cross-resistance, was provided by mutations affecting *rpoN*, and alternative sigma factor, which controls expression of many lifestyle-associated traits including motility, biofilm formation and quorum sensing. Further, we showed that cross-resistance could predict the efficacy of phage cocktails, both in terms of the rate of resistance evolution and ability to suppress bacterial growth. These results suggest that cross-resistance is likely to be a common property of bacteria-phage communities, and that the structure of cross-resistance could strongly influence their structure and stability. Additionally, understanding these processes could greatly improve the treatment of bacterial infections, for example, limiting cross-resistance could drastically improve the rational design of phage therapy cocktails.

Keywords: Phage therapy, cross-resistance, experimental evolution

Session 4: Mathematical modeling to unravel interactions

Keynote lecture

K6. How to make more out of community data? A conceptual framework and its implementation as models and software.

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A central aim community ecology is to understand the processes that determine the assembly and dynamics of species assemblages at different spatiotemporal scales. To facilitate the integration between conceptual and statistical approaches in community ecology, we have developed Hierarchical Modelling of Species Communities (HMSC) as a general, flexible framework for modern analysis of community data. HMSC belongs to the class of joint species distribution models, and it makes it possible to derive simultaneously species- and community level inference from data on species occurrences, environmental covariates, species traits, and phylogenetic relationships. HMSC applies to a wide variety of study designs, including hierarchical data, spatial data, temporal data, and spatio-temporal data. I describe the general HMSC framework and discuss its applicability with case studies that include both macro-organisms and microbiota.

O15. Network Reconstruction from Ecological Count Data with a Sparse Poisson Lognormal Model.

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Many microbes associate with higher eukaryotes and impact their host vitality. Understanding how host-associated microbiomes are assembled and maintained is a crucial first step in the path to enhancing microbial communities for the host benefit. In particular, uncovering direct interactions among communities members is necessary to identify keystone species and systemic taxa that are critical in preserving the structure of ecological communities. The ease and relatively low cost with which sequence data can now be collected on microbiomes using either marker genes (16S rRNA, 18S rRNA, ITS, etc.) or whole genome sequencing also means that there is a growing demand for robust methods that can uncover interactions while being well suited to peculiarities of composition communities inferred from DNA sequences. The most important peculiarities are that i) species abundances are measured as counts (number of reads) and ii) those abundances are relative rather than absolute. The question of finding direct interactions can be formulated as a problem of network inference - or network reconstruction - for which the Gaussian setting provides a canonical way. In a Gaussian world, direct interaction between two species can be faithfully modeled as partial correlation between those taxa so that there is a clear path from statistical dependencies to ecological interactions. Unfortunately, the Gaussian assumption does not apply to count data typically produced by 16S surveys. To circumvent this limitation, state of the art model-based approaches for the reconstruction of ecological networks (SparCC, REBACCA, SPIEC-EASI, etc) all use a two-step strategy. In the first step, counts are either directly transformed to pseudo Gaussian observations using simple transforms, or converted to proportions first which are themselves transformed to pseudo Gaussian observations. The second step applies a (partial) correlation-based approach from the abundant literature of Gaussian graphical models on the pseudo-Gaussian observations from the first step. We adopt a different stance in this work by recoursing on a latent model where we directly model counts using Poisson distributions that are conditional to latent (hidden) Gaussian correlated variables. Thus, in this model (known as a Poisson lognormal), the dependency structure is completely captured by the latent layer. The hidden variables are sometimes referred to as abundance basis but are not a simple transformation of the counts. This modeling can be easily extended to account for the effects of known covariates or the presence of offset terms. This latter effect typically enables us to adapt to varying library sizes in NGS experiments and to reconstruct a single network for microbes sequenced using different marker genes. To perform network inference, we add some sparsity inducing constraints on the inverse covariance matrix of the latent Gaussian vector to select only the most important interaction between species. Unlike the usual Gaussian setting, the penalized likelihood is generally not tractable in this framework. We resort instead to a variational approximation for parameter inference and solve the corresponding optimization problem by alternating a gradient descent on the variational parameters and a graphical-Lasso step on the covariance matrix. We show that the sparse Poisson Lognormal approach has better performance than the existing methods on simulation. We then illustrate our approach on several datasets from microbial ecology. In particular, we consider a dataset set studying the interactions between the pathogen *Erysiphe alphitoides* and other collected bacterial and fungal species in oak trees. We show how accounting for sequencing depth via offsets and integrating external covariates in the model (which was never done in the existing literature to our knowledge) drastically changes the topology of the latent network between species. This analysis demonstrates the centrality of *Erysiphe alphitoides* in the latent network.

Keywords: Network Inference, Count data, Multivariate Analysis, Variational Inference

O16. Limits of statistical detection of microbial associations from

metagenomic data.

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Pathogens and the pathobiome they interact with can be characterized by 16S rRNA sequencing that reveal the microbial composition of a studied environment. The impact of microbial interactions on the properties of microbiota is a topic of key interest in disease ecology. Network analysis is often employed to characterise potential microbial interactions (Layeghifard et al., 2016). It typically requires identifying pairwise statistical associations between the occurrence or abundance of bacterial operational taxonomic units (OTUs) (Faust and Raes, 2012).Microbiota contain hundreds of OTUs, most of which are rare, including pathogens. This feature of community structure can lead to methodological difficulties to detect associations: simulations have shown that methods for detecting pairwise associations between OTUs (which presumably reflect interactions) yield problematic results (Weiss et al., 2016). Rare OTUs are commonly removed restrictively in an empirical filtering step resulting in a loss of information (Friedman and Alm, 2012; Kurtz et al., 2015). We explored the statistical testability of such associations given occurrence and read abundance data. The goal was to understand the impact of OTU rarity on the testability of correlation coefficients. We found that a large proportion of pairwise associations, especially negative associations, cannot be reliably tested. Investigations of microbial agents for biological control purposes are therefore restrained. Consequently, identifying testable associations could serve as an objective method for trimming datasets (in lieu of current empirical approaches). Our threshold is constructed for presence-absence data and read count data and it depends on number of samples and on OTU prevalence.

References

Faust K, Raes J. (2012). Microbial interactions: from networks to models. Nat Rev Microbiol 10: 538–550.
Friedman J, Alm EJ. (2012). Inferring Correlation Networks from Genomic Survey Data. PLoS Comput Biol 8: 1–11.
Kurtz ZD, Müller CL, Miraldi ER, Littman DR, Blaser MJ, Bonneau RA. (2015). Sparse and Compositionally Robust Inference of Microbial Ecological Networks Von Mering C (ed). PLOS Comput Biol 11: e1004226.
Layeghifard M, Hwang DM, Guttman DS. (2016). Disentangling Interactions in the Microbiome: A Network Perspective. Trends Microbiol xx: 1–12.
Weiss S, Van Treuren W, Lozupone C, Faust K, Friedman J, Deng Y, et al. (2016). Correlation detection strategies in microbial data sets vary widely in sensitivity and precision. Isme J 10: 1669–1681.

Keywords: microbial ecology, community structure, association network, data filtering, correlation bounds

O11. Cropping system shapes foliar fungal networks of grapevine.

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Microbial networks support many ecosystem services, including disease regulation. A current challenge is to reconstruct these networks from environmental DNA, and to assess their potential adaptation and resilience to environmental disturbances. Here, we investigated the impact of the cropping system on foliar fungal networks of grapevine. Fungal association networks were reconstructed from metabarcoding data in three organic plots and three conventional plots, after

having selected the best bioinformatic pipeline using a fungal mock community (about 200 fungal strains) as standard. A total of 120 networks were inferred for each plot by varying the network construction parameters in SparCC. Whole-network and node-level topological properties were computed and compared between cropping systems. The dissimilarity between networks was calculated and partitioned into the dissimilarity due to both turnovers of associations and species. We showed that foliar fungal α-diversity was significantly higher in organic plots at the time of sampling, and that the community composition significantly differed between cropping systems. *Erysiphe necator*, a major foliar fungal pathogen of grapevine, was significantly less abundant in organic plots than in conventional plots, and its centrality in the association networks was higher. The dissimilarity of networks between cropping systems was mainly due to changes in associations rather than changes in species composition and this result was robust to variations in network construction parameters. This latter finding highlights the importance of monitoring changes in microbial association networks, in addition to changes in microbial community α-diversity and composition.

References

Friedman, J and EJ. Alm. 2012. “Inferring Correlation Networks from Genomic Survey Data.” PLoS Computational Biology 8 (9): e1002687.

Poisot, T et al. 2012. “The Dissimilarity of Species Interaction Networks.” Ecology Letters 15 (12):1353–61.

Keywords: network inference, microbial networks, fungal pathogen, metabarcoding, phyllosphere, grapevine, organic farming

Poster presentations

P1. Relationship between rice genetic diversity and microbiota composition in traditional Yuanyang rice terraces of China.

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Rice has been annually cultivated for more than 1400 years in the Yuanyang terraces (YYT) of Yunnan, China. Interestingly, the level of diseases observed in this region has always remained low and, consequently, the YYT rice production has never been constrained by a heavy disease burden. A recent study has shown that the arsenal of resistance genes overall born by the rice traditional varieties that are grown in YYT prevents the dissemination of the highly virulent fungal pathogen *Magnaporthe oryzae* (Liao et al 2016). While the use of more than 40 traditional rice varieties has been maintained for centuries in YYT, but the use of a few modern improved varieties has been drastically increased the last few years. This significant change in the overall level of diversity of the rice varieties growing in YYT may affect the long-term sustainable rice protection of this region. Our overarching hypothesis is that the simplification of the varietal landscape is driving a modification of the YYT rice microbiota, which may by extension have an impact on the YYT plant pathogen dynamics, and further on the pathogen emergence or non-emergence. In this study, we have focused our sampling design on a YYT village where both traditional and modern varieties have been equally cultivated for 3-4 years. We have sampled 9 fields cultivated with traditional rice varieties and 9 fields cultivated with improved rice varieties. The 180 collected plants (10 plants X 18 fields) were genotyped after partial genome sequencing (genotyping-by-sequencing approach, GBS). The GBS library was established using the ApekI restriction enzyme, we genotyped the panel of samples with up to 5000 informative SNP markers. This GBS study has confirmed the high degree of diversity of YYT traditional rice varieties in comparison to the low level of diversity of the modern improved varieties that are increasingly introduced in the YYT region. We have further characterized the roots and the stem microbiota using the virion-associated nucleic acid metagenomics-based approach to characterize the virus communities and metabarcoding methods for the bacteria and fungi communities. Besides highlighting the characterization of the microbiota of each rice variety, we will present preliminary results on the relationship between the YYT host genetic diversity and the rice microbiota composition.

References

Liao, J., Huang, H., Meusnier, I., Adreit, H., Ducasse, A., Bonnot, F., Pan, L., He, X., Kroj, T., Fournier, E., Tharreau, D., Gladieux, P., and Morel, J.B. 2016. Pathogen effectors and plant immunity determine specialization of the blast fungus to rice subspecies. eLife 5: e19377.

Keywords: rice, genetic, diversity, microbiota, China

P2. Liver microbiome of *Peromyscus leucopus*, a key reservoir host species for emerging infectious diseases in North America.

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Microbiome studies generally focus on the gut microbiome, which is composed of a large proportion of commensal bacteria. Here we propose a first analysis of the liver microbiome using next generation sequencing as a tool to detect potentially pathogenic strains. We used *Peromyscus leucopus*, the main reservoir host species of Lyme disease in eastern North America, as a model and sequenced V5-V6 regions of the 16S gene from 18 populations in southern Quebec (Canada). The *Lactobacillus* genus was found to dominate the liver microbiome. We also detected a large proportion of individuals infected by *Bartonella vinsonii arupensis*, a human pathogenic bacteria responsible for endocarditis, as well as *Borrelia burgdorferi*, the pathogen responsible for Lyme disease in North America. We then compared the microbiomes among two *P. leucopus* genetic clusters occurring on either side of the St. Lawrence River, and did not detect any effect of the host genotype on their liver microbiome assemblage. Finally, we report, for the first time, the presence of *B. burgdorferi* in a small mammal host from the northern side of the St. Lawrence River, in support of models that have predicted the northern spread of Lyme disease in Canada.

Keywords: 16S, NGS, *Peromyscus leucopus*, Microbiome, *Borrelia*, *Bartonella*

P3. Characterization of the rice microbiota: development and contribution of the 'culturomics' approach.

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Plant microbiota that colonizes different compartments of the plant host plays a key-role in nutrient availability, growth promotion and plant health. Understanding and managing plant microbiota could allow promoting beneficial microbial communities and reducing the impacts of detrimental microbes. While metabarcoding analyses targeting different regions of the 16S rRNA have recently gained popularity to describe large microbial communities of different ecosystems, these culture-independent surveys may have presented a depth bias and failed in detecting microbial populations with low concentrations. The diversification of culture conditions together with a high throughput identification by mass spectrometry system (MALDI-TOF), i.e. a culturomics approach, can greatly increase the number of detected species (Lagier et al., 2015). Microbial culturomics has been shown to reveal new bacterial repertoires non covered by metabarcoding sequencing in the human gut microbiome study (Lagier et al., 2015). In some cases, culture techniques detected even more bacterial species. In addition, a better understanding of the ecology of the plant-associated microbiota and the interactions between members of the community may require the use of synthetic microbial communities. Using rice as a plant model we aim at describing the spatio-temporal dynamics of rice endophyte microbiota. The 16S rRNA metabarcoding approach will be combined with culturomics approaches in order to compare both molecular and 'cultivable' diversity and evaluate their potential complementarity. Our objectives are to (i) establish a culture collection, (ii) develop a MALDI-TOF database for routine identification of bacterial isolates and (iii) compare the metabarcoding approach and the culturomics approach in terms of characterization of microbial community diversity. We isolated more than 2000 bacterial colonies from superficially rice disinfected tissues collected in two rice fields of Camargue in the Provence region of France. Colonies were first identified by a sanger sequencing of the complete 16S rRNA gene. These colonies were also identified by mass spectrometry and the score identification obtained from the current MALDI-TOF database revealed gaps in identifying plant-associated bacteria. One challenge will be to enrich the database with data from plants to make it valuable in plant microbiota analyses. We will further compare the community diversity as described by the metabarcoding approach and the 'cultivable'

fraction.

References
Lagier, J.C., Hugon, P., Khelaifia, S., Fournier, P.E., La Scola, B., and Raoult, D. 2015. The rebirth of culture in microbiology through the example of culturomics to study human gut microbiota. Clin. MIicrobiol. Rev. 28(1): 237-264.

Keywords: Mass spectrometry, Endophytes

P4. Genomic insights into the pathogenicity of *Rickettsiella* spp., intracellular bacteria of arthropods.

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Gammaproteobacteria of the genus *Rickettsiella*, closely related to *Legionella* and *Coxiella*, are well known as pathogenic bacteria in many arthropods (Bouchon et al. 2011). In woodlice, while *Rickettsiella* was primarily only described as a virulent agent (Bouchon et al., 2011; Cordaux et al., 2007), we showed that some strains may be non-pathogenic (Dittmer et al., 2016; Bouchon et al. 2016). Non-pathogenic strains were also reported in aphids where they act as mutualists conferring benefits to their hosts by protecting against predators (Tsuchida et al. 2010) or a fungal pathogen (Lukasik et al. 2012). *Rickettsiella* was recently found highly abundant in ticks with no evidence of virulence (Duron et al. 2016). However, a strain initially identified as *Diplorickettsia massiliensis* (Mediannikow et al. 2010) but nested in the *Rickettsiella* genus (Leclerque & Kleespies 2012), was later recognized as a human pathogen (Subramanian et al. 2012). *Rickettsiella* therefore constitutes a particularly interesting group to study the evolutionary emergence of pathogenicity. Unfortunately, there are only a very few genomic data for *Rickettsiella*: only one whole and annotated genome of *R. isopodorum* from the woodlouse *Trachelipus rathkei* has been recently published (Wang & Chandler 2016), whereas two draft genomes were available: *R. grylli* isolated from an unidentified woodlouse (GenBank AAQJ00000000) and *D. massiliensis* from the tick *Ixodes ricinus* (Mathew et al. 2012). Interestingly several genomic islands have been identified in *R. isopodorum* but absent in *R. grylli*. By NGS metagenomics approaches, we completed these data with five new genomes of *Rickettsiella* from distinct species of woodlice. Phylogenomics showed that these genomes were closely related to *R. grylli* and *R. isopodorum* but distantly related from *D. massiliensis*, all belonging to a well-defined *Rickettsiella* genus. Comparative genomics allowed us to identify T4SS secretion systems and a high number of putative virulence factors including eukaryote-like domain-containing proteins.

Keywords: Pathogenicity, *Rickettsiella*, Intracellular, Bacteria, Arthropods

P5. Host-pathogen-commensal interactions in fish.

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The aquaculture industry is a fast growing food sector that faces important challenges. Among these,

sanitary issues are the most important obstacles to the development of a sustainable aquaculture, since fish farmers suffers major economic losses due to infectious diseases and insufficient pathogen control (FAO reports 2014 (1) and 2016 (2)). To cope with bacterial diseases, farmers essentially rely on the recurrent use of antibiotics incompatible with the development of a sustainable aquaculture. The recognition that animal-associated commensal microbiota play a crucial role in protecting their hosts against invading pathogens through a process called colonization resistance suggests that a better understanding of the host-pathogen-commensal interactions could have important outcomes for innovative control of infectious diseases. Although still in its infancy, microbial ecology of host-pathogen interactions has significantly progressed in the last years, and many studies investigating bacterial pathogenesis conceptually as an "ecological problem" are being undertaken. Such ecosystem-based studies should ultimately provide important information for the development or improvement of alternative control strategies such as the use of improved diets, probiotics and prebiotics, and new models and modeling approaches have emerged to answer these questions of both fundamental and medical relevance. However, these models essentially used terrestrial animals (mostly mammalian) so far (3). The Infection et Immunité des Poissons team (INRA), the Genetics of Biofilms Unit (Institut Pasteur) and the MARBEC Unit (IFREMER) are combining their expertise in microbiology, bacterial ecology and fish infectiology in order to address host-pathogen-commensal interactions issues using relevant fish species (i.e., rainbow trout and sea bass) for the aquaculture industry. A special focus will be made on the family *Flavobacteriaceae* that encompasses a variety of devastating fish pathogens (4) as well as environmental species. Our work should provide a better understanding of the microbiota associated with the fish ecosystems (e.g., gills, skin mucus, intestine), including the pathogenic species.

References
1: FAO (2014). The State of World Fisheries and Aquaculture 2014.
2: FAO (2016). The State of World Fisheries and Aquaculture 2016.
3: Colston TJ & Jackson CR. Microbiome evolution along divergent branches of the vertebrate tree of life: what is known and unknown. Mol Ecol. 2016, 25(16):3776-800.
4: Loch TP & Faisal M. Emerging flavobacterial infections in fish: A review. J Adv Res. 2015, 6(3):283-300.

Keywords: Fish, Pathogen, Flavobacteria, Virulence, Microbiota

P6. N-actetyl-glucosamine influences the biofilm formation of *Escherichia coli*.

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The mucous layer is a physical barrier that limits the contact between bacteria and host epithelial cells. There are growing evidences that microbiota-produced metabolites can also be specifically sensed by pathogens as signals to induce or repress virulence genes (2,7,9). We show that the mucin sugars N-acetyl-glucosamine (NAG) and sialic acid can reduce biofilm formation of commensal and pathogenic *Escherichia coli* strains. *E. coli* catabolism of NAG and sialic acid leads to the production intracellular NAG-6-P that will inactivate the regulator NagC (3). NagC is a known repressor of its own operon, but was also shown to activate the expression of several genes in *E. coli* (1,3,5,6). We demonstrated that the inactivation of the regulatory protein NagC, by addition of NAG or by mutation, reduced the biofilm formation of adherent and invasive *E. coli* (AIEC) strain LF82 and enterohemorrhagic *E. coli* (EHEC) strain EDL933 under static conditions. Moreover, no additional repression was observed upon the addition of NAG in the Δ*nagC* mutant. Thus, NagC could be a positive regulator of biofilm formation

in *E.coli*. Interestingly, real-time monitoring of biofilm formation of LF82 biofilm using microfluidic system (8) showed that *nagC* mutation impairs the early process of biofilm promotion of LF82. Thus, NAG sensor NagC is involved in the early steps of biofilm formation of AIEC strain LF82 under both static and dynamic conditions. Its implication is partly due to the activation of type 1 fimbriae (2,6). By affecting the concentration of free NAG available in the digestive tract, gut bacterial species expressing N-acetylglucosaminidase (4) might therefore influence *E. coli* biofilm formation through a modulation of NagC activity.

References: 1-Barnhart, M.M., Lynem, J., and Chapman, M.R. (2006). GlcNAc-6P levels modulate the expression of Curli fibers by *Escherichia coli*. J Bacteriol 188(14), 5212-5219. 2-Barnich, N., Boudeau, J., Claret, L., and Darfeuille-Michaud, A. (2003). Regulatory and functional co-operation of flagella and type 1 pili in adhesive and invasive abilities of AIEC strain LF82 isolated from a patient with Crohn's disease. Mol Microbiol 48(3), 781-794.3-El Qaidi, S., Allemand, F., Oberto, J., and Plumbridge, J. (2009). Repression of galP, the galactose transporter in *Escherichia coli*, requires the specific regulator of N-acetylglucosamine metabolism. Mol Microbiol 71(1), 146-157. 4-Derrien M, van Passel MW, van de Bovenkamp JH, Schipper RG, de Vos WM, Dekker J. (2010). Mucin-bacterial interactions in the human oral cavity and digestive tract. Gut Microbes. 1, 254-268. 5-Le Bihan, G., Sicard, J.F., Garneau, P., Bernalier-Donadille, A., Gobert, A.P., Garrivier, A., et al. (2017). The NAG Sensor NagC Regulates LEE Gene Expression and Contributes to Gut Colonization by *Escherichia coli* O157:H7. Front Cell Infect Microbiol 7, 134. 6-Sohanpal, B.K., El-Labany, S., Lahooti, M., Plumbridge, J.A., and Blomfield, I.C. (2004). Integrated regulatory responses of fimB to N-acetylneuraminic (sialic) acid and GlcNAc in *Escherichia coli* K-12. Proc Natl Acad Sci U S A 101(46), 16322-16327. 7-Sicard, J.F., Le Bihan, G., Vogeleer, P., Jacques, M., and Harel, J. (2017). Interactions of Intestinal Bacteria with Components of the Intestinal Mucus. Front Cell Infect Microbiol 7, 387. 8-Tremblay, Y.D., Vogeleer, P., Jacques, M., and Harel, J. (2015). High-throughput microfluidic method to study biofilm formation and host-pathogen interactions in pathogenic *Escherichia coli*. Appl Environ Microbiol 81(8), 2827-2840. 9-Vogt, S.L., Pena-Diaz, J., and Finlay, B.B. (2015). Chemical communication in the gut: Effects of microbiota-generated metabolites on gastrointestinal bacterial pathogens. Anaerobe 34, 106-115.

Keywords: *Escherichia coli*, biofilms, mucus, N-acetyl-glucosamine

P7. Airborne dispersion of Leptospirosis in a meat processing plant.

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Introduction: Leptospirosis is the most common occupational zoonosis in New Zealand, with the highest incidence observed in meat processing workers and farm workers. New Zealand has a high incidence of human infection relative to other temperate developed countries, and the organism is widespread in livestock. Serological testing has confirmed infection in livestock presenting to abattoirs and in meat workers. The objective of this study was to determine whether leptospires were present in bioaerosols within the abattoir.
Methods: Ambient air samples (n=18) were collected in an abattoir from ovine and bovine processing areas, using a SASS 3100 high volume sampler located adjacent to workers performing exsanguination (halal sticking), pelt removal, evisceration, a splitting saw (bovine only) and boning or meat cutting. Nucleic acid (DNA) in the bioaerosol samples was amplified using multiple displacement amplification (MDA) for metagenomic analysis, but the material was also tested for specific pathogenic species including *L. interrogans* sv *Pomona* and *L. borgpetersenii* sv Hardjobovis by quantitative PCR. The original (unamplified) DNA samples were also tested.

Result: Leptospires were detected in 11 of the (MDA) samples from both ovine and bovine processing areas at the splitting saw, evisceration, exsanguination and pelt removal. There was no evidence of leptospires in samples taken in the boning or meat cutting areas, or in the five blanks taken. Two of the original DNA samples, both from the ovine pelt removal area, also tested positive for leptospires. **Discussion:** This is the first study to show that leptospires can be detected in a bioaerosol within an abattoir, suggesting a possible route of transmission to meat workers. The organism was detected at locations adjacent to slaughter, pelt removal and evisceration, with the strongest evidence near ovine pelt removal. This distribution directly mirrors the pattern of risk shown in serological testing of meat workers.

Keywords: Leptospirosis, bioaerosol, slaughterhouse

P8. Toward the definition of *Aedes albopictus* and *Aedes koreicus* pathobioma from an area of recent invasion in northern Italy.

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Aedes mosquitoes are considered highly successful invasive species globally. They are also vectors of several viruses of medical importance. As other species, they host a community of bacteria in their midgut, which can play an important role in affecting their fitness, physiology, food digestion, metabolism, immunity, adaptation to new environmental conditions including the capacity to transmit pathogens. Using a metagenomic approach we characterized the microbial community of two mosquito species of medical relevance: *Aedes albopictus* and *Aedes koreicus*. Using high-throughput sequencing we analysed the v3-v4 hypervariable region of 16s rRNA of the midgut from 30 non-fed *Ae. albopictus* females and 30 non-fed *Ae. koreicus* females from the Province of Trento. Alpha and beta diversity indices were used to assess the diversity and richness of bacterial communities in both mosquito species and the differences among countries. The two species showed a large core microbiota, including 75.98% of the identified Operational Taxonomic Units, largely composed by species of the genus *Pseudomonas*, suggesting a common developmental environment. Notably Wolbachia, an intracellular endosymbiont of mosquitoes known to modulate their ability to transmit many pathogens, was present in *Ae. albopictus* (0.1%) but not in *Ae. koreicus*, while Asaia spp. was found mainly in *Ae. koreicus* (14.42%) and in very low proportions in *Ae. albopictus* (0.07%). In conclusion, assessing the composition and diversity of invasive mosquito species gut microbiota provides the basis for the development of further research studies aimed at characterizing the effect of environmental conditions on vectorial capacity and therefore the actual disease hazard within a new habitat.

Keywords: *Aedes albopictus*, *Aedes koreicus*, pathobioma, northern Italy.

P9. The transcriptional factor sigma H as a virulence regulator in *Corynebacterium pseudotuberculosis*.

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Corynebacterium pseudotuberculosis is the etiological agent of Caseous Lymphadenitis (CLA), a disease that affects small ruminants and presents great zoonotic potential [1]. This bacterium persists up to eight months in soil and survives in adverse conditions outside and inside the host, where it replicates intracellularly [2]. Many studies on the pathogenesis of a range of infectious agents have dedicated attention to proteins that participate in gene modulation and may play a role in adaptation for survival under harsh environmental conditions. One example involves the RNA polymerase sigma subunits, which are transcriptional factors known for the activation of specific gene sets in response to external stimuli [3]. Among the sigma factors, the factor Sigma H (σH) stands out as an important driver of virulence and stress response in *Mycobacterium tuberculosis*, a pathogen that is closely related to *C. pseudotuberculosis* [4]. In this work we construct a mutant strain of *C. pseudotuberculosis* for the sigma factor H and evaluate the differentially expressed proteins in the mutant compared to the wild-type strain. Our results suggest that the factor σH participates in the metabolism of nucleic acids and other proteins related to the homeostasis of transcriptional and translational events. Also, σH showed to be important for full persistence of *C. pseudotuberculosis* in BALB/c mice, as recovery of mutant strain from spleen and mesenteric lymph nodes is diminished in comparison with wild-type bacteria after 72 hours of experimental infection. This work represents an important step towards a better comprehension of the regulatory mechanisms through which *C. pseudotuberculosis* establishes infection and the development of a live-attenuated vaccine against CLA.

Funding: Coordination for the Improvement of Higher Education Personnel (CAPES) and the National Council for Scientific and Technological Development (CNPq).

References: [1] Baird, G.J. and Fontaine, M.C. *Corynebacterium pseudotuberculosis* and its role in ovine caseous lymphadenitis. J Comp Pathol 137:179-210, 2007. [2] Dorella, F.A.; Pacheco, L.G.C.; Oliveira, S.C.; Miyoshi, A.; Azevedo, V. *Corynebacterium pseudotuberculosis*: microbiology, biochemical properties, pathogenesis and molecular studies of virulence. Vet Res. 37:201-218, 2006.[3] Souza, B.M.; Castro, T.L.P.; Carvalho, R.D.O.; Seyffert, N.; Silva, A.; Miyoshi, A.; Azevedo, V. σECF factors of gram-positive bacteria: A focus on *Bacillus subtilis* and the CMNR group. Virulence. 5:1-14, 2014.[4] Manganelli, R.; Voskuil, M.I.; Schoolnik, G.K.; Dubnau, E.; Gomez, M.; Smith, I. Role of the extracytoplasmic-function sigma factor sigma(H) in *Mycobacterium tuberculosis* global gene expression. Mol microbiol. 45:365-74, 2002.

Keywords: *Corynebacterium pseudotuberculosis*, factor sigma H, virulence

P10. Pathogen communities in wild plant populations at the agro-ecological interface.

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Pathogen communities in wild plant populations at the agro-ecological interface. The species community in which the host is embedded is expected to affect the pathogen community that it is infected by and vice versa. This may be especially pronounced in agro-ecological landscapes where species diversity between wild and cultivated area changes drastically. However, there is lack of empirical data on how agricultural land use and the local plant community affect how pathogen communities are formed and their evolutionary potential. We investigated spatial variation and impact of agricultural practices on five recently discovered virus species in 28 *Plantago lanceolata* populations across the Åland Islands, SW Finland. We collected samples and data on plant species diversity, soil characteristics as well as population size and connectivity. The data were collected at three spatial scales: in the immediate proximity of the host plant, within host populations and at the

metapopulation scale to be able to estimate the spatial scale in which the processes operate. We screened viral communities in 280 plant individuals and sequenced partial virus isolates from the agricultural and non-agricultural populations. We identified factors affecting virus prevalence and diversity across different geographical scales. We show that population connectivity, size and soil nutrient levels are the main drivers for variation in virus infection within populations. The viruses investigated were common in all vegetation types but virus diversity was higher in agricultural populations than in non-agricultural populations. When we looked at the immediate plant community near the host plant we observed lower virus diversity with high plant species diversity in non-agricultural populations where as in agricultural populations this relationship changed. We also found that different isolates were common in agricultural and non-agricultural populations. These results shed light on how pathogen communities are formed and suggest that agricultural practices may have a profound impact on processes governing pathogen epidemiology at population and local scales.

Keywords: pathogens plants communities

P11. Prevalence, Diversity and Transmission of *Culex pipiens* densovirus.

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Densoviruses (*Parvoviridae*) are small, non-enveloped, single-stranded DNA viruses. They infect many species of arthropods, especially insects, like mosquitoes. While most of the mosquito densoviruses are closely related, *Culex pipiens* densovirus (CpDNV) totally differs from other mosquitoes densoviruses known so far. With their ambisense genome organisation and bigger genome size, they indeed represent a new type of mosquito densovirus. CpDNV have been isolated from laboratory *Cx. pipiens* mosquitoes 18 years ago. However, their prevalence in nature and the outcome of their interactions with (i) their host and (ii) the rest of their host's microbiome remained unexplored. A singularity of their hosts, *Cx. pipiens* mosquitoes, is that they are always found infected with five genetically distinct groups of endosymbiotic bacteria *Wolbachia* (wPip). Being vertically and maternally transmitted, *Wolbachia* manipulate their hosts' reproduction to ensure their spread within the host population. This fascinating endosymbiont has also been shown to protect several arthropod hosts against viral infection. In this context, we first explored the prevalence of this particular mosquito densovirus, CpDNV, in natural populations of *Cx. pipiens* mosquitoes. Our first results showed a high prevalence of CpDNV all around the world suggesting their importance on the evolution and the ecology of this host-endosymbiont system. We are currently studying their diversity and their transmission modes, to better understand both how they persist in nature and how they interact with their host (*Cx. pipiens*) - symbiont (*Wolbachia*) system.

Keywords: *Culex pipiens*, mosquito, densovirus, *Wolbachia*, host-symbiont-pathogen interaction

P12. From salines and serpentines: adaptive evolution of *Brassicaceae* and their microbiome.

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There is emerging realisation that every plant is endowed with its own adaptable microbiome. A growing literature shows plant health, growth, and biomass production are influenced by microbiomes. In *Arabidopsis thaliana*, for example, fungal endophyte communities vary with host genotype and show strong interactions between microbial species. Interaction between host plant and microbiome has already been successfully used by application of selected bacteria or fungal strains on crops to enhance stress resilience to drought in grapevine, generally promoting plant growth and stress resistance in wheat and increase yield in nutrient stressed barley cultivars. This suggests the potential for engineering beneficial microbial communities. We recently observed dramatic within-species, population-level differences in resilience to extreme saline and serpentine environments in several tractable *Brassicaceae* species. We hypothesise that differences in plant microbiomes may influence the remarkable resilience contrasts we observe. In particular, a subset of *Arabidopsis arenosa* populations grow in drought prone serpentine sites while a subset of *Brassica fructiculosa* populations are locally adapted to high salt conditions, whereas other populations in both species are not adapted to these challenges. Thus, we are constructing microbiome atlases to investigate whether microbiome communities may facilitate adaption to extreme environments. Additionally, because serpentine and high saline soil populations have a common characteristic of being very drought prone, we will test for shared drought specific patterns of community change between the plant and microbial genomes. We focus on populations that are adapted to high saline soils and populations adapted to serpentine soils, as both present stringent challenges that are directly relevant to growing worldwide agricultural need.

Keywords: *Brassica fructiculosa*, *Arabisdopsis arenosa*, salt stress, serpentine adaption, microbiome

P13. The impact of genotype and warming on the leaf fungal patho- and microbiome.

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The leaf surface of plants is a habitat to many fungal species. The leaf surface fungal species communities can be structured by both abiotic and biotic factors, such as host genetic identity and elevated temperatures. However, only few manipulative experiments were conducted to assess the impact of these two factors on above-ground microbial communities. We set up a warming experiment to investigate how foliar fungal pathogens and endophytes are affected by elevated temperatures and host genetic identity. In this study, we particularly focused on: 1) the impact of host genetic identity and warming on the resistance of the host tree to a specialist pathogen, the powdery mildew *Erysiphe alphitoides*, and 2) the impact of host genetic identity and warming on the composition of foliar fungal communities of *Quercus robur*. Preliminary results show that powdery mildew was more abundant in the non-heating treatment, but the effect also differed among the host genetic identities. This suggests that effects of temperatures on host-pathogen systems may be mediated by the interaction of environment and the genetic identity of the host, and may therefor lead to changes in host-pathogen interactions.
Keywords: Endophytes, pathogens, species interactions, fungal-climate interactions, host genotype

P14. A library preparation optimized for RNA virus metagenomics allows

sensitive detection of an arbovirus in wild-caught vectors.

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The study of viral communities has recently been boosted by the use of the so-called high-throughput sequencing (HTS) technologies. Coupled with random amplification of nucleic acids, HTS allows the identification of viral species present in a given sample without a-priori knowledge on their identity but their resemblance to known viruses. The huge interest of this approach for virus ecology and diagnostics has triggered several technical improvements in most steps required in virus metagenomics, from nucleic-acid extraction to bioinformatics analysis of sequencing results. Surprisingly, there is yet a key step that has received little attention, that of library preparation. Here, we describe a library preparation for exploring viral diversity in a large number of samples with the use of high-throughput sequencing. This method uses custom adaptors that are PCR ligated, allowing low cost, high multiplexing and fully exploitable read lengths. After validation of our method with artificial viral communities, we have tested it with two samples set from wild-caught arthropod vectors showing that our approach allows to identify an arbovirus in highly-degraded samples with a relatively high sensitivity.

Keywords: Metagenomics; arbovirus; arthropod vectors.

P15. Conserved virome diversity and structure in the mosquito vector *Culex pipiens*.

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Recent epidemics caused by different mosquito-borne viruses underline the viral diversity associated to mosquitoes. However, beyond human viral pathogens, we know little on other viruses of the

mosquito virome. This unexplored diversity probably influences vector competence and other aspects of the mosquito biology, as shown for the mosquito bacteriome. We have analysed the virome of *Culex pipiens*, a mosquito vector of important arboviruses like Rift Valley fever virus or West Nile fever virus. To this end, we have coupled a metagenomics approach with a large sampling campaign involving different countries and habitats around the Mediterranean basin. Our results show for the first time conserved patterns in virus diversity among mosquito populations, as well as specificities probably linked to different environmental conditions. The discovery of a ubiquitous group of viruses strongly supports the existence of a core virome in *Culex pipiens* that is likely to influence mosquito physiology.

Keywords: virome, arbovirus, mosquito, vector

P16. Epichloë endophyte effects on leaf blotch pathogen (*Rhynchosporium* sp.) of tall fescue vary among grass origin and environmental conditions.

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Systemic Epichloë endophytes are common fungal symbionts of many cool-season grasses. They are known for their capability of increasing host plant tolerance against biotic and abiotic stressors, including grass pathogens. However, results on endophyte-mediated disease resistance have been ambiguous, and the underlying mechanisms of disease resistance remain unknown. We studied how Epichloë endophytes affect naturally occurring leaf blotch (*Rhynchosporium* sp.) pathogen infections of wild and cultivated tall fescues (*Schedonorus phoenix*). Endophyte-infected and uninfected tall fescues were grown in a common garden experiment in southern Finland for eight growing seasons. The experimental plants were subjected to nutrient and water treatments. Our preliminary results show that the effects of endophytes on leaf blotch infection incidences varied among plant origins under different environmental conditions. Overall, American cultivars had lower pathogen infections than the Nordic wild grasses. American manipulatively endophyte-free plants had considerably higher pathogen levels compared to their endophytic counterparts under some treatments. Endophytic wild plants from Åland, Finland, exhibited higher leaf blotch incidences than naturally or manipulatively endophyte-free plants, whereas with plants from Gotland, Sweden, the case was opposite. These results indicate that Epichloë endophytes may either suppress or increase pathogen *Rhynchosporium* infections in tall fescues, depending on grass origin and environmental conditions.

Keywords: Leaf blotch, *Rhynchosporium*, *Epichloë*, Fungal endophytes, Grasses

P17. Immune response in relation to bovine mammary gland microbiome during transition to once-a-day milking.

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Bovine mastitis is an inflammatory disease of the mammary gland, often due to bacterial infections. We have recently established a link between bovine teat microbiome and history of animal with regard to mastitis, suggesting a link between teat microbiota and health. In an attempt to further explore this

relation, we investigated response to transition from twice to once-a-day (OAD) milking, which can be seen as a disturbance of the mammary gland ecosystem. Such practice is generally regarded as risky due to a possible inflammation. We thus questioned whether response to OAD-milking was related to bovine teat microbiome. The bovine teat microbiome was investigated in 31 quarters just prior to transition to OAD milking. All quarters were free of inflammation and infection at sampling time but displayed different response to transition to OAD milking: some quarters developed mastitis (G1), while others did not show any infection during OAD milking period (G2 to G5). Nevertheless, groups G2 to G5 exhibited different immune response. G2 did not show any inflammation all along the OAD-milking experiment. G3 displayed inflammation at D14, as revealed by IL8 secretion in milk, a pro-inflammatory cytokine. Finally, G4 and G5 exhibited early inflammation and this inflammation still occurred at D14 for G5. Taxonomic profiles were determined at D0 revealing a higher alpha-diversity in G4 and G5 compared to G2. Clustering of the quarters based on their bacterial composition made it possible to separate G1 and G2 (Cluster 1) from G4 and G5 (Cluster 2). Discriminant analysis of taxonomic profiles between these clusters revealed several taxonomic differences. This study clearly underlines the link between teat microbiome and immune response in the bovine mammary gland. This work is part of the RUMINFLAME project funded by the INRA MetaProgramme GISA.

Keywords: bovine mammary gland microbiota, mastitis, immune response, once-a-day milking

P18. Virome NGS analysis of pests and pathogens for plant protection.

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Despite the role of viruses as an important challenge for society as cause of several diseases in humans, animals and plants, recent research has evidenced a more general “ecological” role of viruses as regulators of microbial communities, from the biogeochemical recycling cycles of the oceans, to altering mutualistic symbiotic relationships in different systems. Viruses are also a major driver of microbial evolution and bear a formidable potential as biotechnological tools including gene therapy, vaccine production and targeted pathogen removal. The H2020 VIROPLANT project (call SFS17) will address the role of the “virome” as a sub-component of the microbiome, with the aim of applying NGS technology and empirical biological experiments to develop new environmentally friendly virus-based control strategies to manage bacterial, fungal and insect vector-transmitted plant diseases (phytoplasmas and viruses) as well as insect pests. So far, plant protection through viruses has relied on limited successful model systems; moreover, viruses infecting bacteria (phages) are also state of the art tools with potential to cope with antibiotic resistant bacterial strains in human and veterinary medicine. VIROPLANT will pursue the use of viruses to increase the arsenal for the control of plant diseases caused by the most important biotic stresses. Protocols of risk assessment will be implemented for virome-based model control strategies. A business plan for representative categories of virome-based control strategies will be included. Stakeholders and SMEs will be involved in bringing the most promising products to the market. Interdisciplinary approaches involving sociological and communication expertise will specifically address the issue of how to convey to the general public the complex beneficial aspects that viruses can bring to plant biotic stress control, overcoming possible societal reluctance towards the use of virus-related new technologies. The Consortium involves 11 research organizations with complementary expertises from eight EU Countries, four SMEs and two stakeholders.

P19. Characterization of LLS: a new bacteriocin from *Listeria* epidemic strains.

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Listeria monocytogenes (Lm) is a Gram-positive bacterial pathogen responsible for food-borne infections in humans. Lm strains preferentially associated to human listeriosis outbreaks display a pathogenicity island that encodes for listeriolysin S (LLS)1, a bacteriocin active *in vitro* against Lm LLS-negative strains, *Lactococcus lactis* and *Staphylococcus aureus*[2-4]. *In vivo*, the increased virulence of Lm LLS-positive strains is associated to modulation of the host intestinal microbiota and colonization of the host intestinal tract in orally-infected animal models[2-4]. As adaptation to the gastro-intestinal tract seems a critical feature of epidemic listeriosis, we intend to explore this phase of listeriosis by investigating: (a) the molecular mechanisms that characterize the bactericidal activity of LLS in target bacterial species, (b) the mechanisms involved in the LLS expression in the gut. To understand the mechanism of action of LLS on target bacteria, the LLS was fused to different tags with the aim to purify the molecule and perform microscopy studies. We confirmed that LLS is located on the membrane of LLS producer cells and that LLS activity induces cell membrane irreversible permeabilization on target bacteria and reversible cell membrane permeabilization on producer bacteria. Our *in vitro* and microfluidics preliminary results suggest that the LLS bactericidal activity is mediated through a contact dependent mechanism. Furthermore, to understand the regulation of LLS expression in the gut we will perform transcriptomics of Lm present in the intestine of germ free mice. Finally, a high throughput screening of gut analogue compounds will be performed to find the activation signal(s) of LLS.

References

1. Cotter, P. D. et al. Listeriolysin S, a Novel Peptide Haemolysin Associated with a Subset of Lineage I *Listeria monocytogenes*. PLoS Pathog. 4, e1000144 (2008).
2. Quereda, J. J. et al. Bacteriocin from epidemic *Listeria* strains alters the host intestinal microbiota to favor infection. Proc. Natl. Acad. Sci. 113, 5706–5711 (2016).
3. Quereda, J. J. et al. Listeriolysin S Is a Streptolysin S-Like Virulence Factor That Targets Exclusively Prokaryotic Cells *In Vivo*. mBio 8, e00259-17 (2017).
4. Quereda, J. J., Meza-Torres, J., Cossart, P. & Pizarro-Cerdá, J. Listeriolysin S: A bacteriocin from epidemic *Listeria monocytogenes* strains that targets the gut microbiota. Gut Microbes 1–8 (2017).

Keywords: Listeriolysin S, bacteriocin, virulence factor, gut microbiota, membrane permeabilization

P20. New insights into cow holobiont in relation to health.

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Infectious diseases have been traditionally considered as the result of the bipartite interaction between a given pathogen and its host. Recent advances in high-throughput sequencing technology have uncovered the complexity of the various microbiomes associated with the host and symbiotic microbiota have emerged as a main player of the infectious process. In cattle, major efforts have been devoted to

the characterization of the microbiome associated to different anatomical sites in relation to animal performance and health. However, these have mainly focused on the comparison of microbiomes of healthy versus diseased animals. Issues that remained to be addressed include the role in disease development of the microbiome associated to the affected organ as well as the impact of microbiome located at remote body sites. An increasing amount of studies mainly conducted in human have shed light on the capacity of a microbiome to affect both local and distant sites within a same host. Here, we propose to explore the structure, diversity and dynamics of microbiomes associated to 4 anatomical sites in cows before and after calving. The interdependence of these microbiome in relation to animal health and genetics will also be investigated. For this purpose, over one thousand samples were collected from 45 primiparous prim' Holstein cows selected from two divergent lineages with high or low milk somatic cell counts that are respectively more or less susceptible to intra-mammary infections. Sampling were performed at 4 time points and from 4 anatomic sites: nasal, genital, buccal (as a proxy for rumen), and foremilk (as a proxy for internal teat microbiome). Here, results will focus on defining and analyzing the structure of the microbiomes at one-month post-partum. Overall, these data should contribute to providing a holistic view of bovine microbiomes in relation to health. This work is part of the MICROCOSM project funded by the INRA MetaProgramme MEM.

Keywords: bovine microbiome, health, holobiont, host genetics

P21. Host genotype in explaining within-host virus communities in a transplant experiment.

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Individuals are often simultaneously infected by multiple pathogenic microbes. However, host-pathogen research has traditionally been conducted in the "single host—single pathogen" framework, and the role of host resistance in shaping pathogen communities remains largely unexplored. To test whether host resistance is the key determinant of within-host pathogen communities, we performed a field transplant experiment where we placed healthy replicates of multiple cloned *Plantago lanceolata* individuals to wild *P. lanceolata* populations in the Åland Islands to acquire natural virus infections. We sampled these experimental plants multiple times over the growing season to detect five common viruses with specific PCR-primers, and to track temporal changes in the within-host virus communities. We found differences in virus infections among genotypes and plants kept in different populations. Our results suggest that host genotype is important but does not alone determine the composition of pathogen communities.

Keywords: *Plantago lanceolata*, host resistance, pathogen community, coinfection, plant virus, transplant experiment

P22. Why such a contrast between the low prevalence of zoonotic pathogens in questing ticks and the high prevalence in their vertebrate reservoir hosts?

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In a context of emerging or re-emerging tick-borne diseases, many studies have measured the prevalence of zoonotic agents in hosts and vectors, using cross-sectional protocols at different spatial scales. Surprisingly, a frequent contrast occurs between a high prevalence in the vertebrate hosts and a low prevalence in questing ticks. Furthermore, investigations on the species of the genera *Anaplasma*, *Babesia* and *Borrelia* have shown that several taxa co-circulate and are maintained in vertebrate host populations. Indeed, the co-infection of the individual vertebrate host by several taxa and genetic variants is the rule, rather than the exception. Based on a review of recent papers on ticks and tick-borne diseases, we hypothesize that high and repeated exposure to ticks under natural conditions causes the frequent re-infection of vertebrates with a diversity of tick-transmitted taxa, leading to a high overall prevalence of infections and co-infections. However, the immunity induced by this frequent exposure could keep the intensity of co-infections by different strains of the same taxa at a low level within individual hosts, which in turn could lead to a low frequency of acquisition by the ticks during blood-meals. Furthermore, in order to complete their life-cycle, the tick-transmitted organisms need also to resist the tick innate immunity, to compete with its microbiote and to be able to colonize the salivary glands of the vector. Facing these successive selective pressures, the maintenance of the infections in the host populations can be achieved only if ticks are abundant, to compensate for a low prevalence in tick populations. We argue that a meta-community approach taking into account the functional traits of the different pathogen taxa and their intra-taxa diversity is required to fully understand the dynamic interplay between the actors of the pathosystem and the potential emergence of pathogenic strains.

Keywords: ticks, tick-borne infections, meta-communities, diversity, reservoir

P23. The soil microbiota diversity influences the transcriptome of the telluric protist responsible for the clubroot.

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Plasmodiophora brassicae (Pb) is an obligate biotrophic pathogenic protist responsible for the clubroot, a root gall disease of *Brassicaceae* species. As main ways of disease control (crop rotations and cultivation of tolerant varieties) have shown their limits, there is a need to design alternative and durable methods based on ecological concepts. Interactions between plants and their microbiota could contribute to the emergence of innovative plant protection strategies. In this context of complex interactions [plant genotypes X soil microbiota X pathogen], the general hypotheses are that microbial diversity in the soil can modify Pb performances directly or indirectly by increasing plant defense, and that plant and microbiota function expressions are connected to organize plant physiological responses against the pathogen. Here, the main objective is to determine which molecular mechanisms of Pb are affected by modifications of soil microbial communities with a dual-RNAseq transcriptomics approach. For this, a time-course experiment was conducted to study interactions between Pb, two *B. napus* genotypes, and three soils harboring different microbiota diversities (High, Medium, Low). These soils were obtained by microbial diversity manipulation experiments, which consisted to inoculate sterilized

initial native soil by suspensions/dilutions coming from the same initial soil. First, the soils, characterized with bacterial 16S and fungal 18S markers, displayed, as expected, different levels of richness and diversity, particularly between High and Low soils. Secondly, the study showed that the soil microbial diversity levels had an impact on disease development (symptom levels and pathogen quantity). Finally, the Pb transcriptional patterns were also modulated by these microbial diversities, and these modulations were dependent of the kinetic time and interestingly of the host genotype plant. The functional analysis of gene expressions allowed highlighting main Pb genes families that were modulated. To go further in the interactions' characterization, the plant transcriptome will also be studied.

Keywords: Plant pathogen ; transcriptome ; soil microbiota

P24. Interactions between the wild plant host *Solanum dulcamara* and *Phytophthora infestans*, the causal agent of potato late blight.

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Late blight in potato, caused by infection with the oomycete *Phytophthora infestans*, leads to large economic losses. Introduction of new resistant potato varieties is countered by rapid evolution of new pathogenicity factors in *P. infestans*. Increased insights into plant-pathogen interactions in natural systems can aid our understanding of epidemiology of late blight as well as the development of more environmentally friendly and durable plant defence. We have studied interactions between *P. infestans* and the wild potato relative *Solanum dulcamara*. *S. dulcamara* shows variable resistance to *P. infestans* infection in the lab and in an experimental garden. Moreover, we found that susceptible plants can be tolerant of *P. infestans* infection, which led to overcompensation in growth for lower levels of infection, suggesting a potential benefit of the presence of *P. infestans*. Despite finding susceptible genotypes in our experiments, we have rarely detected infected leaves in wild populations. However, soil sampling of the rhizosphere of *S. dulcamara* showed occurrence of *P. infestans* in winter, spring and summer samples, indicating overwintering in the rhizosphere. Preliminary results showed that isolates from the rhizosphere had no negative effect on *S. dulcamara* roots, but were more pathogenic than a lab strain on potato leaves. Our results does not suggest strong pathogenic effects of *P. infestans* on leaves of the wild host *S. dulcamara*, but indicates that in particular the rhizosphere of *S. dulcamara* could influence epidemiology of potato late blight.

Keywords: *Solanum dulcamara*, *Phytophthora infestans*, Potato late blight epidemiology, Plant tolerance, Pathogen off-season survival

P25. Do soil microbial communities diversity modified plant - bioagressor interactions?

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The consequences of decline in biodiversity for ecosystem processes and functioning have long been of considerable interest. The significance of biodiversity loss in microbial communities is challenged by the concept of functional redundancy. Since different species can have the same function in ecosystems, functional redundancy predicts that the loss of species does not necessarily alter ecosystem functioning because of their replacement by other species for maintaining processes. Therefore, it is classically assumed that 'the more, the better' in terms of biodiversity levels, however species-rich microbial communities potentially led to high competition and antagonistic interactions that may reduce community functioning, and may have consequences on plant growth and health or soil biological barrier effect against pathogens. We investigate the consequence of *Brassica napus* - microbiome diversity interaction on different bioagressors fitness following a life-history traits approach. Microbial diversity was manipulated through a removal-recolonization experiment. The impact of the removal experiment on the microbial diversity was evaluated using high-throughput sequencing. *Brassica napus* were cultivated in these soils and confronted to three different bioagressors: the protist *Plasmodiophora brassicae*, the nematode *Heterodera schachtii* and the cabbage root fly *Delia radicum*. The effect of soil microbial communities on each bioagressor were evaluated by measuring different life history traits, which are a reliable estimate of bioagressor feeding success and expected fitness of the system. The result obtained showed a reduction of disease index or bioagressors fitness in the less diverse communities. These results were discussed in regard of plant metabolites and community ecology.

Keywords: Microbial diversity - *Brasica napus* - pathognes - bioagressors - fitness - life history traits

P26. Metabolic processes mainly impacted by fixed coregenome variants during adaptations of *Salmonella* serovars to mammalian- and avian-hosts.

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With regard to the fact that many of the bacterial genomic studies exploring evolution processes of the host adaptation focus on the accessory genome describing the gains and losses of genes, we developed a new approach focusing on the fixed coregenome variants in order to describe the modification of metabolic processes during the colonization of new habitats by *Salmonella* serovars. More precisely, we recently published (Felten et al. 2017) bioinformatic tools allowing (i) variant calling analysis and robust phylogenetic inference based on SNPs and recombination events, (ii) detection of fixed coregenome variants distinguishing homoplastic and non-homoplastic SNPs and InDels, and (iii) gene-ontology enrichment analyses to decrypt metabolic processes involved in adaptation of *Salmonella enterica* subsp. *enterica* to mammalian- (S. Dublin), multi- (*S. Enteritidis*), and avian- (S. Pullorum and S. Gallinarum) hosts. Confirming that the monophyletic clade S. Dublin diverged from the polyphyletic clade *S. Enteritidis*, which includes the divergent clades *S. Pullorum* and *S. Gallinarum*, the 'VARCall' workflow produced highly confident variants for downstream robust phylogenetic inference (i). Supporting the phylogenetic reconstruction, the scripts 'phyloFixedVar' and 'FixedVar' detected intragenic and non-homoplastic fixed variants (ii). Based on those synonymous and non-synonymous fixed variants, the scripts 'GetGOxML' and 'EveryGO' identified representative metabolic pathways linked to host adaptation using the first gene-ontology enrichment analysis based on fixed coregenome variants (iii). The new coregenome approach that we propose, links detection of fixed SNPs and InDels with regards to inferred phylogenetic clades and gene-ontology enrichment analysis in order to decrypt the evolution of metabolic processes at the coregenome scale during the adaption of *Salmonella* serovars Dublin (i.e. mammalian-hosts), Enteritidis (i.e. multi-hosts), Pullorum (i.e. avian-hosts) and Gallinarum (i.e. avian-hosts). All these polyvalent Bioinformatic tools can be applied on other bacterial genus without additional developments.

References. Felten A., Vila Nova M., Durimel K., Guillier L., Mistou M.Y., Radomski N. (2017) First gene-ontology enrichment analysis based on bacterial coregenome variants: insights into adaptations of *Salmonella* serovars to mammalian- and avian-hosts. BMC Microbiology 17(1):222.

Keywords: Bacterial fixed variants; Bacterial genomics; Gene-ontology enrichment analysis

P27. Experimental evolution of a *Bacillus thuringiensis acrystalliferous* strain in *Galleria mellonella*.

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The continuous exposition of a pathogenic bacterium in a host during a serial passage experiment (SPE) may facilitate the apparition and posterior fixation of mutations that favour its growth and multiplication in the host environment1. These changes, can be traced during an SPE by whole genome sequencing of the evolved variants 2, 3. Here we describe the results obtained for a SPE using a *Bacillus thuringiensis* streptomycin resistant crystal minus strain (Bt407 Cry$^-$)4 using *Galleria mellonella* larvae as a host system. Our objective was to describe the history of the events, which arose during the evolution of the pathogen in one of its natural hosts and explain phenotypic variations based on genotypic differences. The parental strain was entirely re-sequenced by Illumina NextSeq system prior to the study in the host. An infection protocol was established where *G. mellonela* larvae were force-fed with spores of the Bt407 Cry$^-$ strain, and spores collected from dead larvae where used to re-infect new *G. mellonella* individuals in a next passage. The experiment lasted for 20 passages and with 9 lines in parallel. The genetic changes, which happened during the experimental evolution, were monitored by whole genome sequencing of the evolved populations at passages 5, 10, 15 and 20. In parallel, the SPE evolved strains were tested for virulence to *G. mellonella*, persistence (bacterial load in the cadavers) and resistance to antimicrobial peptides (lag times and growth rates in presence or absence of nisin, an antimicrobial peptide used as a food preservative) and compared with the initial parental bacteria and with populations evolved *in vitro*. The fitness of the evolved strains was also measured by mixing the same amount of the SPE evolved and ancestral lineages and measuring the outcome of competition by estimating the rate of change in their respective frequencies over the course of the competition trial.

References: 1. Ebert, D. Experimental evolution of parasites. Science 282, 1432–5 (1998).2. Koskiniemi, S. et al. Pathoadaptive mutations in *Salmonella enterica* isolated after serial passage in mice. PLoS One 8, e70147 (2013).3. Masri L, Branca A, Sheppard AE, Papkou A, Laehnemann D, Guenther PS, et al. Host–Pathogen Coevolution: The Selective Advantage of *Bacillus thuringiensis* Virulence and Its Cry Toxin Genes. PLoS Biol13(6): e1002169. (2015)4. Sheppard, A. et al. Complete Genome Sequence of *Bacillus thuringiensis* Strain 407 Cry-. Genome Announc. 1, e00158-12 (2013).

Keywords: Experimental evolution, gut ecosystem, whole genome sequencing, *Bacillus thuringiensis*

P28. From pathobiome to socio-pathosystems: questioning disease management practices in Corsica.

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Shifting from pathogen to pathobiome paradigm raises questions on infectious disease management strategies. Disease-based strategies can be antagonist to reach a level of animal health in animal farming systems. According to our results in Corsica, bovine tuberculosis, echinococcosis, trichinosis, hepatitis E virus and Aujeszky virus are pathogens found together in the same pig herds but these pathogens fall under various modes of regulations, based on the use of different tools.
For example, we show co-infection between Aujeszky and hepatitis E virus as an indicator of animal infectious interaction between domestic pigs and wild boars, which pattern is different according to herd management practices. Whereas Aujeszky virus is supposed to be the object of drastic management measures, HEV management does not exists. Else, whereas bovine tuberculosis and trichinosis are detected through systemic controls in slaughterhouses, bovine tuberculosis is subject to specific epidemiological surveys and prophylaxis on cattle but not on pigs. *Echinococcosis* and *trichinosis* raise questions on slaughterhouse geographical control at the scale of the territory, and the identification of various HEV strains highlight different infectious pathways.
Moreover, implementing a pathogen management measure can affect an equilibrium at the levels of the animal and the herds as well as at the level of the territory. The notion of socio-pathosystem provides a framework to capture multi-pathogen infection dynamics under various human management practices. This communication aims at presenting results from different studies carried out by INRA and its partners in Corsica, raising questions on how to implement relevant disease strategies that take into account complex microbial communities and the presence of diverse pathogens under diverse management devices within an animal territory.

P29. Development of MetaXplor: a viral metagenomics database.

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Recent metagenomics-based studies have identified hundreds of unknown viruses living in environmental and ornamental hosts. While genomic, transcriptomic and metagenomic next generation sequencing (NGS) datasets are exponentially increasing, a large part of the virus-related sequences is probably still missed because (i) bioinformatics tools are still under-developed and (ii) our scientific community does not always share datasets. It is therefore crucial to better share, clean, store and analyze these datasets in order to better describe and characterize the virus diversity. In order to fulfill this objective, we have developed a novel Web-accessible NoSQL database – called MetaXplor – that archives reads and contigs obtained from viral metagenomics studies. This database also displays modules of (i) geolocalization of the samples, (ii) searches using similarity-based method (BLAST approaches), (iii) searches using Keywords:, and (iv) phylogenetic placements of the reads on reference phylogenetic trees.

Keywords: Viral metagenomics, bioinformatic tools, database

P30. Air microbiota in animal slaughterhouse: how metagenomic detection can help epidemiological studies?

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Introduction: Significant excess risks of lung cancer and haematologic neoplasms have been observed in slaughterhouse workers in eight New Zealand studies, and numerous studies conducted elsewhere. No specific causal agents have been identified, although a biological aetiology is suggested as the risk is highest in those areas where workers are exposed to live animals or to biological material containing animal urine, faeces or blood. This study aimed to assess the airborne bacterial microflora in the slaughterhouse environment in order to develop exposure categories for reanalysis of a meat workers' cohort. **Methods:** Bulk air samples (n=31) were collected for between 5 and 8 hours in five areas in both sheep and beef slaughterhouses using a SASS3100 sampler (fitted with a proprietary SASS filter) located between 0.5 and 2 meters from the worker. Nucleic acid was extracted from each filter and amplified using commercially available kits, then sequenced on an Illumina MiSeq instrument. Bioinformatics analyses conducted included comparative taxonomic analyses, gene function (including virulence factor) analyses, and principal component analyses to compare profiles in samples taken in different areas. **Result:** Of the bacteria identified over 95% were in the classes Actinobacteria, Firmicutes and Proteobacteria. Clear differences in all parameters were apparent in the different areas, however, and the full results of the comparative analyses and the development of exposure profiles will be presented. **Discussion/Conclusion:** This is the first study to show the air microbiota found in different workers' areas within an animal slaughterhouse. Different levels of exposure or different bacteria, with potential pathogens, could explain the different effects of biological exposures or possible route of transmission to meat workers. Metagenomic analysis of the composition of bioaerosol samples represents a promising method for the development of exposure categories for the epidemiological analysis of the effect of biological exposures in an occupational environment.

Keywords: Air microbiota, metagenomic detection, slaughterhouse

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