

The Second International Conference on Holobionts

Montréal, QC, Canada May 8-10, 2019

Final program

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Welcome

Welcome to the 2nd International Conference on Holobionts!

We have worked very hard to put together an exciting scientific program that will address all aspects of the holobiont. We will have keynote lectures from Seth Bordenstein, Monica Medina, Emma Allen-Vercoe, Richard Jefferson, David Relman and Paolina Garbeva, covering the evolution, ecology, communication, resilience and manipulation of holobionts. For the contributed presentations, we have tried to give priority to students and postdocs, and they will be competing for five presentation prizes.

We would like to thank our many sponsors for generously supporting this event. We were also helped by a very enthusiastic group of student volunteers. Without this help, we could not have put together such an exciting program.

Welcome to Montréal and to the 2nd International Conference on Holobionts!

Étienne Yergeau, chair François-Joseph Lapointe, co-chair

Local organizing committee

Hamed Azarbad, Institut national de la recherche scientifique Mohamed Hijri, Université de Montréal Simon Joly, Jardin botanique de Montréal Steven Kembel, Université du Québec à Montréal Corinne Maurice, McGill University Dominic Poulin-Laprade, Agriculture and Agri-Food Canada Marc St-Arnaud, Jardin botanique de Montréal Guylaine Talbot, Agriculture and Agri-Food Canada Alexandre Thibodeau, Université de Montréal Ruth Schmidt, Institut national de la recherche scientifique

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Faculté de médecine





Program At a glance

	Wednesday, May 8, 2019	Thursday, May 9, 2019	Friday, May 10, 2019
9:00		Resistance	Ecology
9:30			
10:00			
10:30			
11:00			
11:30			
12:00	Registration	Lunch	Lunch
12:30			
13:00	Welcome	Manipulation	Communication
13:30	Evolution		
14:00			
14:30			
15:00			
15:30			
16:00		Poster session	_
16:30			
17:00	Dinner on your own		
17:30			
18:00		Diner Cocktail	
18:30			
19:00			
19:30			
20:00	Holobiont Art-Sci event		
20:30			
21:00			
21:30			

Detailed program

Wednesday, May 8, 2019

12:00-13:00 Registration 13:00-13:15 Welcome

Holobiont and evolution

Chairs: François-Joseph Lapointe (U. Montréal) and Dominic Poulin-Laprade (Agriculture and Agri-Food Canada)

13:15-14:00 Keynote speaker

"On the Origin of Species by Means of Symbiosis"

Seth Bordenstein

Departments of Biological Sciences and Pathology, Microbiology, & Immunology, Vanderbilt University, USA

14:00-14:20 Schistocephalus solidus and threespine stickleback holobiont coevolution; the building blocks of local adaptation

Megan Hahn

Stonybrook University, USA

14:20-14:40 A single phage candidate gene for male killing in a bacterial endosymbiont

Jessamyn Perlmutter

Vanderbilt University, USA

14:40-15:00 Microbiota structure and dynamics in planorbid snails, vectors of the human

parasites Schistosoma spp.

Camille Huot

Université Perpignan, France

15:00-15:40 Coffee break

15:40-16:00 The rhizosphere metagenomes of ancestral and modern wheat cultivars grown

under low fertiliser inputs are not significantly different

Liliana Quiza

Institut national de la recherche scientifique, Canada

16:00-16:20 Identification of gut microbiota involved in fructose assimilation in Aedes

albopictus mosquito

Morgane Guegan

Université Lyon 1, France

16:20-16:40 Modeling the Role of the Microbiome in Evolution

Saúl Huitzil

Universidad Nacional Autónoma de México, Mexico

16:40-17:00 Prophage WO proteins in Wolbachia that hijack animal reproduction

Brittany Leigh

Vanderbilt University, USA

17:00-20:00 Dinner on your own

20:00-21:30 The Holobiont Art-Sci Event

Maison Notman, 51 Sherbrooke Ouest Montréal, QC, H2X 1X2

Thursday, May 9, 2019

Holobiont resistance and resilie	ence
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Chairs: Hamed Azarbad (INRS) and Marc St-Arnaud (U. Montréal)

9:00-9:45 *Keynote speaker*

"Microbial-host codevelopment in cnidarian holobionts from jellyfish to corals"

Mónica Medina

Department of Biology, The Pennsylvania State University, USA

9:45-10:05 Response of the wheat microbiome to three years of rainfall manipulations

Hamed Azarbad

Institut national de la recherche scientifique, Canada

10:05-10:25 Community ecology of the vaginal microbiome: the importance of host-microbe

interactions

Carmen Lia Murall

CNRS, France

10:25-11:00 Coffee break

11:00-11:20 Impact on plant microbiota of a recent shift in rice genetic diversity in a

sustainable century-old Chinese rice agrosystem

Pascal Alonso

CIRAD, France

11:20-11:40 Growth Phase-Dependent Physiology of Bacterial Isolates from the Human Gut

Microbiota

Will Jogia

McGill University, Canada

11:40-12:00 Complexity matters: soil food webs shape the willow holobiont response to

phenanthrene contamination

Sara Correa-Garcia

Institut national de la recherche scientifique, Canada

12:00-13:15 Lunch

Manipulation/Restoration of the holobiont

Chairs: Guylaine Talbot (Agriculture and Agri-Food Canada) and Alexandre Thibodeau (U. Montréal)

13:15-14:00 Keynote speaker

"Microbe managing by making drugs out of bugs"

Emma Allen-Vercoe

Department of Molecular and Cellular Biology, University of Guelph, Canada

14:00-14:20 Assessing bacterial activity and the role of bacteriophages in the human gut.

Corinne Maurice

McGill University, Canada

14:20-14:40 Creating synthetic wheat holobionts for increased drought resistance

Asmaâ Agoussar

Institut national de la recherche scientifique, Canada

14:40-15:00	Pushing the boundaries of shotgun metagenomics with genome reconstruction Julien Tremblay
	National research council, Canada
15:00-15:30	Coffee break
15:30-15:50	Bacteriophages regulate gut bacterial communities in child stunting in an age- specific manner
	Mohahammadali Khan Mirzaei
	McGill University, Canada
15:50-16:10	Does including a non-mycotrophic plant in agricultural rotation modifies the root and rhizospheric arbuscular mycorrhizal fungal microbiome of wheat, pea and canola?
	Jacynthe Masse
	Université de Montréal, Canada
16:10-16:30	Bacteriotherapy in healthy suckling piglets: success parameters to consider.
	Alexandre Thibodeau
	Université de Montréal, Canada
16:30-18:00	Poster session
18:00-20:00	Dinner cocktail

Keynote speaker

"Hormones and the Holobiont: Origins and some implications of hologenome theory"

Richard A. Jefferson

Queensland University of Technology (QUT)

Friday, May 10, 2019

Ecological principle	s in the holobiont
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Chairs: Corinne Maurice (McGill) and Steven Kembel (UQAM)

9:00-9:45 *Keynote speaker*

"Diversity, Stability, and Resilience in Mammalian Microbiomes"

David Relman

Department of Medicine - Med/Infectious Diseases, Stanford, USA

9:45-10:05 Into the wild: understanding the breadth of host-microbiome interactions

Christopher Greyson-Gaito

University of Guelph, Canada

10:05-10:25 Alternative stable states of a holobiont: the intestinal microbiota - host

relationship

Maarten van de Guchte

INRA, France

10:25-11:00 Coffee break

11:00-11:20 Gut bacterial physiology and activity along the progression to dysbiosis

Maria Taguer

McGill University, Canada

11:20-11:40 The symbiotic realm of marine cyanobacteria revealed by global ocean DNA

metabarcoding across plankton size fractions.

Ewen Corre

Station biologique de Roscoff, France

11:40-12:00 Fungi prevalence in human milk is influenced by the outdoor environment and

associated with bacterial composition

Shirin Moossavi

University of Manitoba, Canada

12:00-13:15 Lunch

Communication within the holobiont

Chairs: Ruth Schmidt (INRS) and Mohamed Hijri (U. Montréal)

13:15-14:00 Keynote speaker

"Communication from a distance"

Paolina Garbeva

Netherlands Insitute of Ecology, The Netherlands

14:00-14:20 The immunomodulatory treasure-trove of the gut microbiota

Naama Geva-Zatorsky

Technion – Israel Institute of Technology, Israel

14:20-14:40 Metatranscriptomic responses of the wheat holobiont to decreasing soil water

content

Pranav Pande

Institut national de la recherche scientifique, Canada

14:40-15:00 Microbially-driven trace gases dynamics in the rhizosphere

Anne de la Porte

Institut national de la recherche scientifique, Canada

15:00-15:30	Coffee break
15:30-15:50	Plant microbiome succession and assembly modulated by endogenous signal
	peptides in soybean (Glycine max)
	Itumeleng Moroenyane
	Institut national de la recherche scientifique, Canada
15:50-16:10	Development of a 3D printed platform for eavesdropping on intercellular
	signaling in the microbiome
	Caroline Birer
	University of Pittsburgh, USA
16:10-16:30	How can microbial volatiles help plants to withstand drought?
	Ruth Schmidt
	Institut national de la recherche scientifique, Canada
16:30	Closing remarks/End of the conference

Abstracts

Oral contributions

Creating synthetic wheat holobionts for increased drought resistance Asmaâ Agoussar

Impact on plant microbiota of a recent shift in rice genetic diversity in a sustainable century-old Chinese rice agrosystem

Pascal Alonso

Response of the wheat microbiome to three years of rainfall manipulations Hamed Azarbad

Development of a 3D printed platform for eavesdropping on intercellular signaling in the microbiome

Caroline Birer1

The symbiotic realm of marine cyanobacteria revealed by global ocean DNA metabarcoding across plankton size fractions.

Ewen Corre

Complexity matters: soil food webs shape the willow holobiont response to phenanthrene contamination

Sara Correa-Garcia

Microbially-driven trace gases dynamics in the rhizosphere Anne de la Porte

The immunomodulatory treasure-trove of the gut microbiota Naama Geva-Zatorsky

Into the wild: understanding the breadth of host-microbiome interactions Christopher Greyson-Gaito

Identification of gut microbiota involved in fructose assimilation in Aedes albopictus mosquito

Morgane Guégan

Schistocephalus solidus and threespine stickleback holobiont coevolution; the building blocks of local adaptation

Megan Hahn

Microbiota structure and dynamics in planorbid snails, vectors of the human parasites Schistosoma spp.

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Hormones and the Holobiont: Origins and some implications of hologenome theory Richard Jefferson

Growth Phase-Dependent Physiology of Bacterial Isolates from the Human Gut Microbiota

Will Jogia

Bacteriophages regulate gut bacterial communities in child stunting in an age-specific manner

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Does including a non-mycotrophic plant in agricultural rotation modifies the root and rhizospheric arbuscular mycorrhizal fungal microbiome of wheat, pea and canola? Jacynthe Masse

Assessing bacterial activity and the role of bacteriophages in the human gut.

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Fungi prevalence in human milk is influenced by the outdoor environment and associated with bacterial composition

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A single phage candidate gene for male killing in a bacterial endosymbiont J. I. Perlmutter

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Bacteriotherapy in healthy suckling piglets: success parameters to consider. Alexandre Thibodeau

Pushing the boundaries of shotgun metagenomics with genome reconstruction Julien Tremblay

Alternative stable states of a holobiont: the intestinal microbiota - host relationship. Maarten van de Guchte

Creating synthetic wheat holobionts for increased drought resistance

Asmaâ Agoussar¹, Hamed Azarbad¹, Étienne Yergeau¹

Drought is a limiting factor in agricultural production and will become a major problem with the predicted climate changes. The interactions between the plant and its microbiota (collectively known as a holobiont) could be the key to overcome this issue.

Our objective was to create a synthetic community from a collection of strains thoroughly tested for their beneficial effect on wheat drought tolerance. An extended culture collection of bacteria and fungi was created from leaves, roots, seeds and rhizosphere soils of field-grown wheat plants under reduced or normal precipitation levels (25 or 100% of the natural precipitation). The isolates were isolated on traditional growth media (TWYE, TSB, R2A and PDA) and specialized, environmentally relevant (soil extract, plant extract and artificial root exudates) growth media. We have isolated more than 500 bacteria and fungi. So far, we have characterized the strains for their growth under low water availability and for their capacity to promote root elongation. Synthetic communities will then be assembled and combined with various drought tolerant or sensitive wheat cultivars to create synthetic holobionts. These holobionts will then be compared in term of their capacity to sustain water-stress.

Future experiments will further zoom on the metatranscriptomic responses of these holobionts to water stress. Our results are expected to shed light on the microbial community characteristics necessary for improved water-stress tolerance of wheat holobionts.

¹INRS-Institut Armand-Frappier

Impact on plant microbiota of a recent shift in rice genetic diversity in a sustainable century-old Chinese rice agrosystem

<u>Pascal Alonso</u>¹, Laurence Blondin¹, Frédéric Mahé¹, François Massol², Denis Filloux¹, Elisabeth Fournier¹, Pierre Gladieux¹, Aurélie Ducasse¹, Jean-Benoit Morel¹, Pierre Mournet³, Julien Foruin³, Huichuan Huang⁴, Jin Bai hui⁴, Xiahong He⁴, Romain Ferdinand¹, Charlotte Julian¹, Emmanuel Fernandez¹, Christian Verniere¹, Philippe Roumagnac¹

¹BGPI, CIRAD, INRA, Montpellier SupAgro, Univ Montpellier, Montpellier, ²EEP, CNRS, Villeneuve d'Ascq, France, ³AGAP, CIRAD, Montpellier, France, ⁴The National Center for Agricultural Biodiversity, Yunnan Agricultural University, Kunming 650201, China

The century-old Chinese Yuanyang terraced rice paddy fields (YYT) developed by the Hani minority people has been studied for its long-term sustainability. These studies have suggested that the century-long maintenance of a high level of rice varieties diversity and a landscape mosaic structure of these varieties have contributed to the YYT sustainability. However, this sustainability is nowadays potentially challenged by the massive introduction of exogenous rice varieties in this region. We here focused on a YYT village where both traditional (T) and recently introduced (RI) rice varieties were equally cultivated and we asked two questions: How diverse were the rice varieties grown in this village? And how diverse were both microbiota and virome of these varieties? We inventoried leaf-, stem- and root-associated bacteria, fungi and viruses of 190 rice plants. We confirm that the rice varieties are split into two major lineages (T vs. RI varieties) and we reveal that the microbial diversity is primarily determined by the plant organ but not by the plant genotype. We finally show that the Southern rice black-streaked dwarf virus (SRBSDV) is prevalent in the studied area (15%) and that SRBSDV is significantly more prevalent in T varieties. This result suggests that the YYT agrosystem is not exempt of viral outbreaks, which is likely to drive a modification of the set of cultivated rice varieties. We hypothesize that the continuous introduction of rice varieties that do not modify the microbiota and the constant adjustment of cultivated rice varieties is reducing the burden of diseases.

Response of the wheat microbiome to three years of rainfall manipulations

Hamed Azarbad¹, Asmaâ Agoussar¹, Julien Tremblay², Etienne Yergeau¹

¹Centre INRS-Institut Armand-Frappier, Institut national de la recherche scientifique, Laval, Québec, Canada, ²Energy, Mining and Environment, National Research Council Canada, Montréal, Québec, Canada

Although water stress resistance is traditionally thought to be encoded in the plant genome, relationships with microbes can also enhance plant tolerance to stress. However, it is not known how this microbial facilitation develops through time and if it is transmitted from one generation to another. We hypothesized that plants exposed to water stress will associate with different microorganisms than plant not exposed to stress, and that some of these organisms will be transmitted to the next generation through the seeds. To test this hypothesis, we designed a multiyear field experiment where drought-tolerant and sensitive wheat genotypes were grown in a Quebec field that was never subjected to water stress. Soil water content (SWC) was passively manipulated using rain-out shelters removing 25%, 50%, and 75% of the precipitation. Seeds were harvested at the end of the growing season and seeded in the exact same plots the following year. DNA was extracted from soils before seeding (T0) and root, leaf, rhizosphere and seeds during each growing season and subjected to amplicon sequencing (16S rRNA gene and ITS). We present here the results from the first three years of the experiment (2016, 2017 and 2018). Wheat microbiome responses to the decreasing SWC were influenced by plant genotype and varied significantly through growing season and years. Our results demonstrate a clear differentiation in seed-associated microbiomes based on the genotype's sensitivity to water stress. These differences tended to increase over the years, suggesting some level of adaptation to low (SWC) through the seed microbiota.

Development of a 3D printed platform for eavesdropping on intercellular signaling in the microbiome

Caroline Birer1¹, Rosalie K. Chu2², Christopher R. Anderton2², Erik S. Wright1¹

¹Department of Biomedical Informatics, University of Pittsburgh School of Medicine, Pittsburgh, Pennsylvania 15219, United States, ²2Environmental Molecular Sciences Laboratory and Biological Sciences Division, Pacific Northwest National Laboratory, Richland, Washington 99354, United States

To understand complexities of the microbiome, we must first understand how microorganisms interact with each other. Cell-to-cell communication is a complex process that occurs through contact-dependent and contact-independent mechanisms using an immense repertoire of small molecules. For this reason, decrypting the "language of the microbiome" is a difficult task which remains underexplored.

Here we sought to analyze the collection of small molecules secreted by microorganisms, which presents a unique challenge due to the exometabolomes diversity and scale. To address this, we have pioneered a high-throughput method that allows us to capture the exometabolome and analyze it via mass spectrometry in a single device.

To separate cells from their exometabolome, we have developed a 3D printed device composed of two interlocking pieces separated by a 0.2 µm membrane. This system can be used to co-culture a variety of microorganisms in a controlled environment, with or without additional stimuli (e.g. pH, drug candidates, extracts). Our platform is compatible with high-throughput robotics used with direct-infusion mass spectrometry. We have validated our method with mixtures of 30 known compounds.

As a case study, we focused on the interactions among 5 microorganisms isolated from a small sample of soil. We have demonstrated our ability to detect excreted small molecules from each individual microbe, as well as unique features only biosynthesized during pairwise interaction.

The symbiotic realm of marine cyanobacteria revealed by global ocean DNA metabarcoding across plankton size fractions.

Ewen Corre^{1,2}, Nicolas Henry¹, Éric Pelletier², Tara Oceans Contributors^{1,2}, Colomban de Vargas¹

¹Sorbonne Université, CNRS, UMR 7144, Station Biologique, Place Georges Teissier, 29680, Roscoff, France, ²Genoscope, CEA, CNRS UMR Metabolic Genomics, Evry, 91057, France

Cyanobacteria form a large group of photosynthetic prokaryotes characterized by a high genetic diversity and broad range of habitats across latitudes. Some of them are also known as symbionts (cyanobionts) of larger organisms, such as plants or protists. However, the global diversity of cyanobacteria across organismal size-fractions is still unknown. In this study we identify and quantify cyanobioses in global marine plankton using data from the Tara-Oceans expeditions. We have generated a dataset of 480000 prokaryotic operational taxonomic units (OTUs based on 16S rRNA sequencing, V4-V5 region) based on 1354 samples that cover the tiny free-living cyanobacteria, the colonial taxa and the underexplored diversity associated with larger organisms or detritus. We then developed a semi-automatic method to identify the phylogenetic clusters that share a common "lifestyle": presence in the large size fractions (colonies, symbioses) versus small ones (free-living). The results were successfully corroborated by information extracted from the litterature. Overall, we found that 35% of all cyanobacterial OTUs (~25% of the reads) were preferentially distributed in the larger plankton size fractions (>20 µm). They consisted of colonial organisms (e.g. Trichodesmium spp., 35% of the OTUs) and known cyanobionts (e.g. Richelia intracellularis, 11%). The remaining and most abundant OTUs (~70% of the reads) belonged to phylogenetic clusters with no colony-forming representatives, suggesting that they live in aggregates or in symbioses with larger organisms. This analysis unveils a major diversity of putative cyanobionts that may provide fundamental metabolisms to larger plankton and thus play a critical role in ocean biogeochemistry.

Complexity matters: soil food webs shape the willow holobiont response to phenanthrene contamination

Sara Correa-Garcia¹, Vincenzo Corelli², Armand Séguin³, Étienne Yergeau¹

¹INRS-Institut Armand Frappier, 531 Boul. Des Prairies, Laval, QC, Canada, ²SP Building Loyola Campus, Concordia University, 7141, Sherbrooke St W, Montreal, QC, Canada, ³Laurentian Forestry Center, Natural Resources Canada, 1055 rue du P.E.P.S., Québec, QC, Canada.

The degradation of organic contaminants around plant roots, rhizodegradation, principally depends on the interactions within the plant holobiont at the level of the roots. There is mounting evidence suggesting that the diversity of the contaminated soils is linked to efficient phytoremediation. Indeed, a significant portion of the soil biomass is composed by soil invertebrates that dwell in the root environment and could be arguably part of the plant holobiont. We hypothesized that willows inoculated with a more complex invertebrate food web will thrive better in the presence of contaminants than willows that rely only on microbial communities or less complex food webs. A pot experiment was carried out, comparing eight food web complexity levels (microbial community (B), B+nematodes (N), B+springtails (C), B+earthworms (E), B+N+C, B+N+E, B+C+E, B+C+E+N) under contaminated (100mg·kg⁻¹ phenanthrene) and noncontaminated conditions. After 6 weeks of growth, plant shoots, roots and rhizosphere and bulk soil were sampled for amplicon sequencing analysis, plant morphological traits and phenanthrene levels. Our results showed a general decrease in willow biomass under phenanthrene contamination. Significant differences in biomass were also observed between the food web complexity treatments, with the non-contaminated*B+C+E+N treatment having the highest biomass and the contaminated*B treatments having the lowest biomass. Detailed analyses of the microbial communities will also be presented and linked to the plant phenotypic and soil contamination differences observed between the treatments. This experiment will inform us which food web complexity treatments are more efficient in promoting phenanthrene degradation, guiding future efforts to harness plant holobionts for phytoremediation.

Microbially-driven trace gases dynamics in the rhizosphere

Anne de la Porte², Etienne Yergeau¹, Philippe Constant²

¹Centre INRS-Institut Armand-Frappier, ²INRS-IAF

Legume plants are involved in a mutualistic relationship with soilborne bacteria, known as rhizobia, fixing atmospheric nitrogen (N2) in legumes' symbiotic organs, nodules. Legumes that host rhizobia lacking the hydrogenase uptake system release hydrogen (H₂) from their nodules as a by-product of N₂ fixation. H₂ is an energetic compound that is readily consumed by soil H₂oxidising microbes, thus potentially impacting rhizospheric community functioning. For instance, it has been shown that H₂ displays a soil fertilisation effect, but the underlying processes remain unknown. Our aim is to test the hypothesis that H₂ fertilisation effect is driven by changes in microbial-mediated processes related to nutrient turnover. Our objectives are (1) to record H₂ diffusion from nodules in the rhizosphere and its concentration gradients, as measured by gas chromatography, and (2) identify key microbial guilds whose biodiversity and activity is impaired by changes in H₂ concentrations. For the purpose of these experiments, we developed an innovative rhizotron system designed to collect gas samples in soil and examine spatial correlations between H₂ gradients and changes in microbial functions. Here, we present performance of rhizotron system to monitor trace gas diffusion and microbial turnover in soil. Case studies including soil alone, soil exposed to H₂ originating from artificial or legume plants are presented. Taken together, the results demonstrate that non-destructive and non-disturbing gas sampling in the rhizotron system permits temporal studies mimicking in situ conditions to study the impact of gas emission from plants on soil microbial community diversity and functioning.

The immunomodulatory treasure-trove of the gut microbiota

Naama Geva-Zatorsky¹, Noa Mandelbaum¹, Tal Avi-Gefen¹, Lillie Beck¹, Neerupma Bhardwaj¹

¹Technion, Faculty of Medicine

We are studying the gut microbiota as a forgotten organ that can complements its host, primarily host immune functions. We systematically characterized the role of over 50 human gut microbes from diverse genera and phyla, representing the gut microbiota diversity. We find most microbes to have immunomodulatory effects spanning from innate to adaptive responses, and with potential to be effective in a variety of diseases. Surprisingly, these effects were not encoded in microbial phylogenetic backgrounds (i.e. microbes from distant phyla could elicit similar effects and vice versa). We are mostly interested in dynamics of microbial consortia as an ecological interdependent microbial community. In order to study both microbe-host interactions and microbe-microbe interactions in a dynamic manner, we developed a tool to fluorescently label live anaerobic gut microbes in association with the mammalian host. We are continuously improving this labeling method and are studying spatial organization, microbial dynamics and mechanistic interactions with the host.

Into the wild: understanding the breadth of host-microbiome interactions

<u>Christopher Greyson-Gaito</u>¹, Timothy J. Bartley^{1,2}, Karl Cottenie¹, Will M.C. Jarvis³, Amy E.M. Newman¹, Mason R. Stothart⁴

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The microbiome has profound impacts on the ecology and evolution of the vast majority of organisms worldwide. The number of microbiome studies is exponentially increasing illustrating the massive interest and large unknowns in microbiome research. Understanding the ways that hosts and microbiomes shape each other's biology almost certainly relies on various ecological processes that may be limited or absent from highly controlled transplantation experiments. Yet, the potential biases due to highly controlled transplantation experiments has not been examined. A recent explosion in the number of studies conducting gut microbiome transplants now makes it possible to evaluate how the current literature is biased and how future studies might address fundamental knowledge gaps in host-microbiome interactions. Here, we assert that the hostmicrobiome research field should employ broader ranges of ecological conditions in transplant experiments. In doing so, conclusions will be avoided that are biased by experimental designs that limit the role of well-established ecological processes in shaping the biology of both hosts and microbiotic communities. We review the literature of microbiome transplant studies in nonhuman model systems to quantify the current literature bias towards rodent models and highly controlled conditions. Our work here considers the implications of these biases for understanding host-microbiome interactions and presents ways to eliminate these biases. Moving towards a wider breadth of studies that includes an EcoReal "space" is imperative for understanding the ecological and evolutionary interplay of hosts and microbiomes.

Identification of gut microbiota involved in fructose assimilation in *Aedes albopictus* mosquito

Morgane Guégan¹, Van Tran Van¹, Edwige Martin¹, Guillaume Minard¹, Benjamin Fel¹, Anne-Emmanuelle Hay¹, Laurent Simon², Mohamed Barakat³, Philippe Ortet³, Patrick Potier¹, Feth-el-Zahar Haichar¹, Claire Valiente Moro¹

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The Asian tiger mosquito Aedes albopictus is a current major public health concern because of its invasive success and its ability to transmit various pathogens. Although there is growing evidence on the role of microbiota in insects, little information is available in mosquitoes, notably on the interaction between the mosquito digestive process and its gut microbiota. Mosquitoes feed on plant nectar, which is mainly composed of fructose. We adapted for the first time the stable isotope probing approach to identify microbial communities involved in fructose assimilation. To that end, mosquitoes were fed with a ¹³C-labeled fructose solution for 24h. Comparative analysis between unlabeled and ¹³C-labeled DNA of microbes allowed to identify bacteria and fungi possibly involved in fructose metabolism. Common and/or specific genera were identified in males and females and some metabolic pathways associated with those microorganisms were highlighted. In parallel, quantification by droplet digital PCR in ¹³C-labeled DNA fractions showed that microbes assimilated fructose in higher proportion than the mosquito per se. This study is the first to investigate the trophic interactions between Ae. albopictus and its microbiota, thus highlighting the importance of the microbial component in the mosquito nectar feeding.

Schistocephalus solidus and threespine stickleback holobiont coevolution; the building blocks of local adaptation

Megan Hahn¹, Nolwenn M Dheilly¹

¹Stony Brook University

Next generation sequencing has given us the ability to study holobionts more intimately than ever before and it has become apparent that parasites interact with their host microbiome. Helminth parasites can alter the composition, diversity, and abundance of their host gut flora, and different parasite species impact the host microbiota differently. However, it remains unknown whether parasite-microbes interaction has evolved over time as a result of antagonistic coevolution with the host holobiont. Threespine sticklebacks are widely distributed and freshwater populations isolated in hundreds of distinct lakes evolve independently and host distinct microbiomes. Freshwater sticklebacks are infected by the cestode parasite Schistocephalus solidus that locally adapts to its fish host, thus providing the opportunity to study the adaptation of a parasite to its host holobiont. Using lab reared fish and parasites from three different populations from Alaska and Europe, we completed a fully reciprocal cross-infection experiment. The composition of the bacterial microbiome of control non-exposed fish, exposed but non-infected fish, and infected fish was characterized using 16S sequencing to determine whether the fish microbiome composition depends more on fish origin, exposure to S. solidus, or infection by S. solidus. This experimental design also allowed us to test whether parasites from different origin can differentially impact the fish host microbiome. We found that both fish and parasite origin play a role in determining the abundance and composition of stickleback gut microbiota and discuss our results in light of our knowledge of stickleback co-evolution with S. solidus.

Microbiota structure and dynamics in planorbid snails, vectors of the human parasites *Schistosoma* spp.

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Since the advent of molecular methods to study uncultivable microbes, more and more studies have revealed the importance of the microbiota in organism fitness and its influence on the relationship between parasites and their hosts through its effect on the immune response.

To decipher the interplay between microbiota, host and parasite, we have studied the interaction between the parasitic Platyhelminthes, Schistosoma spp., agents of the bilharzia, a neglected tropical disease affecting more than 200 million people worldwide, and their intermediate freshwater planorbid snail hosts.

Using 16S metabarcoding, we characterized the specificity of the bacterial microbiota for different mollusks' genera / species / strains, as well as its dynamics in the course of infection by the parasite S. mansoni in an allopatric and sympatric combination.

The microbiota's structure in naive snails differed according to the host identity, and following phylogeny, thus displaying a pattern of phylosymbiosis. Moreover, during a kinetic of infection by the parasite, a microbiota dysbiosis was observed. This change in microbiota structure and diversity was linked to modifications of the immune response and expression of antimicrobial peptides, especially biomphamacins, suggesting an interplay between host immune response to parasite infection and microbiota. These results are of particular importance to go further in the understanding of the relationship between microbiota and immune capabilities in invertebrates and support the importance of considering the holobiont in a host/parasite interaction.

Hormones and the Holobiont: Origins and some implications of hologenome theory

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Multicellularity requires coordination of spatially distal cells and temporally diverse actions. In vertebrates, much of this is mediated by hormones, frequently steroids, most of which are excreted through apocrine, exocrine and endocrine secretions as inactive conjugates of glucuronic acid or sulfate. Myriad, diverse microbiota populate the epithelia and through cleavage by diverse glucuronidases (GUS) and arylsulfatases (ARS), release the active form of the hormone which can then be resorbed or presented with altered kinetics, bioactivity and concentrations to diverse sites of action. The microbial populations thus modulate hormone action.

From 1980-1990, we developed and distributed GUS from *E. coli* as a reporter gene for plant, fungal and animal transgenesis and for microbial ecology studies. In our efforts to improve its efficacy, we explored glucuronide operon function in enteric microbes, and sought natural GUS variants in populations of soil, water, epithelial or fecal microbes that might have improved properties. We made field trips to Africa to isolate fecal and environmental microbes that were unlikely to be present due to human action and contamination. In parallel we revisited neglected literature on the function of the enzymes in vertebrate biology. In combination with stunning advances in microbial discovery and identification (e.g. David Ward's & Steve Giovannoni's discoveries of the ubiquity of uncultured microbes), and some neglected industrial R&D we were led to a major rethink on much of our framework around biology and evolution. science and society. This led to articulation of the hologenome theory of evolution at Cold Spring Harbor in 1994 and the extended proposal that hormone activity in metazoa and metaphyta was modulated by the dynamic population structures of associated microbes. The pleiotropic and powerful effect that hormone modulation could have to reorient and impact virtually all fitness-related traits, and indeed all reproductive activity of plants and animals, stimulated new insights into living systems. In this presentation I'll review the coincidences and congruences that led to the theory and some of the possible implications for science and society, in the form of some conjectures.

Did the advent of agriculture and the concomitant rise in sedentary and concentrated populations cause massive inbreeding depression of the microbiome and become the origin of disease - plant, animal and human - as dysbiosis? Is the germ theory of disease opportunistically right but structurally wrong? Did commensalism and community tactility/touching - the hallmarks of social behavior - cause harmonization and reinforce convergent microbial populations and thus congruent hormone action and hence behavior? Has all of evolutionary thought been compromised by scale bias? Is it reasonable that the logic of evolution is embedded in macro-organisms (e.g. anything we can see) rather than the vast majority of all living systems that we can't? Is the holobiont - in the case of all systems that have experienced the post agricultural microbial collapse - really a merobiont, with at best metastable populations that do not reflect an empirical steady state? Is Darwin's natural selection an 'edge-case' in evolution, working well at medium physical and temporal scales, but not truly reflecting the primacy of information-state persistence that could describe microbial

life? Could the hologenome theory and the microbiome provide the missing mechanism to the late 19th century's other great evolutionist - now largely forgotten - Pyotr Kropotkin, who proposed 'Mutual Aid: a factor in evolution" in 1902? Could the real driver of evolution not be replication and reproduction, but persistence and pooing? Can the role of macro-apobionts be largely as dynamic scaffolds to recruit, select, nurture, amplify and disseminate microbial populations?

Growth Phase-Dependent Physiology of Bacterial Isolates from the Human Gut Microbiota

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The human gut microbiota is a complex community of microorganisms heavily implicated in human health. Much of the current understanding of this community relies on metagenomic techniques, which have identified many of the bacteria commonly found in the gut, but do not provide any information about the physiology and metabolism of these bacteria. Here, we use bacterial isolates from the human gut microbiota and link bacterial physiology to growth by employing single-cell techniques used in other microbial ecosystems. We use the relative bacterial nucleic acid content, determined by staining with SybrGreen, as an indicator of bacterial activity, and we assess cell membrane damage as another component of bacterial physiology. We hypothesized that bacterial physiology, as characterized by nucleic acid content and cell damage, was dependent on growth phase for these bacterial isolates. Our results indicate that some bacteria remain invariably low nucleic acid bacteria (LNA) throughout their growth, while others are more dynamic and switch between high nucleic aid bacteria (HNA) and LNA. Bacterial growth was not limited to only HNA bacteria or only LNA bacteria, suggesting that replication is physiologically distinct from relative activity. Cell damage, when present, was highest in stationary phase, when growth is absent or at a balance with cell death. Overall, these findings show that cell damage, when present, is growth phase-dependent, and that nucleic acid content is growth phase-dependent for most bacterial isolates. The more static isolates, however, may be an invariable part of the less active subset of the human gut.

Bacteriophages regulate gut bacterial communities in child stunting in an age-specific manner

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Child stunting is a significant health concern in low and middle-income countries, affecting 22% of children under the age of five worldwide. Delayed and reduced growth, or stunting, results from nutrition deficiencies, genetics, and is linked to an altered gut microbiota. Recent studies have shown that gut bacteria of stunted children are different from their normal counterparts, generally with lower Firmicutes/Proteobacteria ratio. While the role of phages in regulating bacterial populations in the environment is well studied, their role in gut dysbiosis and stunting is unknown. We sampled thirty stunted and thirty healthy Bangladeshi children under 48 months old from two age groups with distinct diets. We separated phage and bacterial communities from fresh fecal samples and proceeded with in vitro anaerobic cross-infections, where phages and bacterial communities from children with different health status were incubated overnight in the nutrientrich media. Phage and bacterial abundances were determined via epifluorescence microscopy, and changes in their diversity assessed using 16S and shotgun sequencing. We identified that phage and bacterial community diversity and abundances change with age and health status. In the youngest cohort, Bacillus phages were most abundant in healthy children, while Lactococcus phages dominated in stunted children: whereas, in the older cohort, healthy children had more Microviridae phages and their stunted counterparts were colonized mostly with Salmonella phages. Data from our cross-infections show that phages can also regulate the bacterial diversity and abundance in an age-specific manner, suggesting an intervention time window for microbiome manipulation under the age of 24 months.

Prophage WO proteins in Wolbachia that hijack animal reproduction

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Mosquito-borne viruses impact hundreds of millions of people worldwide, and many diseases they cause lack effective therapeutics. Consequently, many governments and institutions are using novel biocontrol strategies that curb virus transmission based on genetic or symbiont modification. The World Health Organization endorsed pilot releases of mosquitoes harboring the endosymbiotic bacteria Wolbachiabecause it confers insect resistance to many RNA viruses and encodes a natural drive system that allows it to spread through mosquito populations, a process known as cytoplasmic incompatibility (CI). CI is characterized by an unknown sperm modification set in the testes that results in early embryonic death. The identification of the prophage WO basis of CI has opened the door for avenues to functionally interrogate the two genes, cytoplasmic incompatibility factors A and B(cifA, cifB). Recently, we developed antibodies to these two phage WO protein products and determined their localization patterns within testes and ovaries of infected and transgenic Drosophila. Through immunogold labeling and immunohistochemistry, we demonstrate the extracellular localization of the Cifs throughout spermatogenesis and oogenesis. Moreover, identification of active phage WO particles and vesicles within Wolbachiainfected testes raise new hypotheses about possible routes of Cif protein transport out of Wolbachia. Identification of key ligands through immunoprecipitation and mass spectrometry provides further clues into interacting proteins. Localization of the Cif proteins within Drosophilareproductive tissues and identification of interacting proteins uncovers the timing, location and potential mechanism of the prophage-induced sperm modification behind this critical symbiosis strategy at the forefront of worldwide vector control efforts.

Does including a non-mycotrophic plant in agricultural rotation modifies the root and rhizospheric arbuscular mycorrhizal fungal microbiome of wheat, pea and canola?

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With \$26.7 billion injected into the economy each year, canola is the most profitable crop in Canada. In western Canada, Canola is grown in rotation with other crops, being most often cereals and pulses. Canola is a non-mycotrophic plant that does not actively associate with arbuscular mycorrhizal fungi (AMF). It is recognized that AMF allow mycotrophic crops to access a larger pool of nutrients and increase plant health and productivity. However, the widespread use of canola in rotations raises concerns on the loss of soil microbial diversity and increased susceptibility of crops to pathogens. We hypothesized that crop rotations with higher frequency of mycotrophic plants could modify the root and rhizospheric microbiome of the crops present in the rotation and increase the overall productivity of the production system. However, the effect of the frequency of each rotation-species on the microbiome and productivity of each crop are still poorly understood. The main objective of this research is to characterize the AMF community of wheat, pea and canola under different rotation systems in order to understand the effects of crop frequencies on the root and rhizospheric microbiome of each crop and on plant productivity. To do this, three rotation systems (intensifying canola, cereals or pulses over four years) were tested in a complete random blocks design. The rotation system and the growth stages had an impact on the biodiversity and structure of AMF communities in all crops. However, only wheat has seen its productivity increased in the pulse-intensified rotation.

Assessing bacterial activity and the role of bacteriophages in the human gut.

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The human gut is home to trillions of microbial cells, bacteriophages (viruses specific to bacteria), fungi, and eukaryotes; collectively referred to as the gut microbiota. The gut microbiota is key to human health: it is central to our digestion, synthesizes essential vitamins, metabolizes therapeutic drugs, and shapes host immunity. Yet we have no clear understanding of the metabolic activities performed by individual members of this complex community, or how they interact with each other. Despite the progress made in determining the gut microbiome's diversity and genetic potential, most studies rely on sequencing-based approaches that are challenging and limited to determine activity. In addition, few studies include other members of the gut microbiota, such as bacteriophages. These are critical gaps in our functional understanding of this complex community that hinder our attempts to manipulate this community. Using original combinations of single-cell and sequencing approaches in an ecological theoretical framework, we aim to better characterize the active bacterial members of the gut microbiota and determine how bacteriophages alter bacterial communities. Collectively, our projects aim to explore human health from a microbial standpoint. Ultimately, our goal is to increase our understanding of the ecological processes and interactions between the different members of the gut microbiota, focusing on bacteria and phages, in order to modulate them for health purposes.

Fungi prevalence in human milk is influenced by the outdoor environment and associated with bacterial composition

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Background: Fungi constitute an important yet frequently neglected component of the human milk microbiota.

Method: We studied the milk mycobiota of 271 mothers in the multicentre CHILD cohort. Milk was collected at 3-4 months postpartum. Mycobiota was analysed by Illumina ITS2 sequencing. Maternal, infant, and home environment characteristics were obtained by questionnaire. Population density was computed using the 2006 census data for each residential address. Associations were examined using $\chi 2$ test, logistic regression, and linear discriminant analysis. Bacteria-fungi co-occurrence analysis was performed using CoNet.

Results: Overall, 61 (22.5%) samples tested positive for fungi. Candida parapsilosis (60% of positive samples), Alternaria eichhorniae (50%), and Rhodotorula mucilaginosa (43%) were the most prevalent fungi. Mothers residing in Vancouver had the highest prevalence of milk fungi (32% vs. 18-21% in Edmonton, Toronto, and Winnipeg). Higher population densities (OR=1.87 95%CI 1.02-3.48 for above vs. below median) and summer season (OR=3.94, 95%CI 1.53-11.56 for summer vs. spring) were associated with higher prevalence of fungi. There was no difference based on mode of breastfeeding, maternal BMI, delivery mode, or indoor mould. Bacterial taxonomic clusters were significantly associated with presence of fungi independently of city and season. Members of Actinobacteria, Bacilli, and γ -Proteobacteria were enriched in samples with detectable fungi. Within positive samples, Candida was negatively associated with multiple bacterial genera.

Conclusion: Fungi were detected in a minority of human milk samples and their presence was associated with various bacterial and environmental parameters. Ongoing research will assess the impact of milk mycobiota on infant gut microbiota and health.

Plant microbiome succession and assembly modulated by endogenous signal peptides in soybean (*Glycine max*)

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Managed agricultural ecosystems are unique systems where crops and microbes are intrinsically linked and selected to promote crop health and increase production. This study focuses on successional development of the plant microbiome and assembly processes that delimit them, and aims to 1)tests for evidence of niche differentiation and to what extend plant signal peptides influence community assembly and 2)create a successional model that can predict these niche shifts as they occur during plant development. To this end. Glycine max var. Pioneer plants were grown in an environmental chamber till seed maturation. Microbiome and xylem sap samples were collected at the various developmental stages: emergence, growth, flowering, and seed development. Community structure and abundance were assessed with amplicon sequencing. The sap was purified and signal peptides were identified using nano LC-MS/MS. Abundance was highest in the epiphytic and rhizosphere communities, and community structure varied according to-plant organ, plant developmental stage, and life history (epiphytic or endophytic). Machine learning models correctly predicted the abundance and distribution of key taxa. Phylogenetic niche shifts were detectable across plant organs and developmental stages. Lastly, niche-based processes delimited distribution and assembly of endosphere and rhizosphere communities. We demonstrate that by understanding the mechanistic processes that assemble microbiome communities, it becomes not only possible to identify key microbes that are essential for promoting plant growth and health, but also key intervention points where the microbiome community can be engineered to be composed of microbes that can mitigate plant stresses and influence longterm management practices.

Community ecology of the vaginal microbiome: the importance of host-microbe interactions

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Clinical studies have shown that, unlike the gut microbiome, highly diverse vaginal communities are more susceptible to pathogen invasions. It is conjectured that Lactobacillus-dominated communities drive down diversity through lactic acid production and are, thus, effective at preventing infections and benefiting the host. However, simply adding Lactobacillus species as probiotics is not yet a reliable treatment of bacterial vaginosis (a highly diverse, dysbiotic, anaerobe community state). We study why using a model of the vaginal microbiome adapted from community ecology theory. We run perturbation analyses to test community state stability and compare the results to change point inference of published longitudinal data of vaginal microbiome abundances. In particular, we ask what processes drive switches in dominance between Lactobacillus-dominated vs. anaerobe-dominated community states. We find that across 38 women, 97% of statistically significant switches in dominance (SSD), from Lactobacillus- to anaerobe-dominated (or vice versa), happened either during menses or peak estrogen, suggesting host resources play a role. Model analyses show that resource-based interactions are stabilizing, making community states quite resistant to perturbations. However, increasing the number of direct microbe-microbe interactions in the system increases susceptibility to perturbations (such as surges in lactic acid production or antibiotics). Finally, we confirm the expectation that lactic acid is a powerful perturbation but sustained changes in resource availability (i.e. press, not pulse, perturbations) are more effective at driving changes in dominance. Consequences for potential treatments for bacterial vaginosis are discussed.

Metatranscriptomic responses of the wheat holobiont to decreasing soil water content

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Prolonged and recurring droughts are affecting the wheat productivity in western Canada. Time taken by conventional approaches of wheat breeding to generate new and successful varieties cannot match the rate of declining wheat yields. Alternatively, a promising approach is to manipulate the plant-associated microbiota resulting in the rapid improvement in wheat resistance to water stress. However, the combined and coordinated responses of the wheat and its microbiota (the wheat holobiont) to water stress are poorly understood. The principal aim of this experiment was to determine the metatranscriptomic response of the wheat holobiont to water stress in the field. For this, wheat was grown in the field and received 100%, 75%, 50% or 25% of the natural precipitation using rain exclusion shelters. RNA was extracted from the rhizosphere soil and root samples collected in July, at the peak of the growing season, Root eukaryotic and rhizosphere prokaryotic mRNA libraries were constructed independently and sequenced using Illumina HiSeq. The preliminary results show that the absolute abundances of the transcripts of both the prokaryotic as well as the eukaryotic components of the holobiont are affected by the level of precipitation in the wheat rhizosphere soil. Most significantly different transcripts are related to transcription, post translational modification, protein turnover and chaperons. Further analysis will provide an in-depth characterization of the genes and pathways involved in drought tolerance of the wheat holobiont. Our long-term objective is to engineer the wheat holobiont for increased drought resistance and resilience.

A single phage candidate gene for male killing in a bacterial endosymbiont

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Wolbachia are maternally-transmitted bacteria that infect almost half of all arthropods and many nematode species worldwide. In arthropods, these bacteria selfishly manipulate host reproduction to enhance the fitness of infected, transmitting females, thereby facilitating their own spread through the host population. One such phenotype is male killing, where the sons of infected females are selectively killed. This reduces competition among their surviving, Wolbachiatransmitting sisters. Despite significant impact of Wolbachia on animal reproduction, evolution, and vector control, the microbial genes underlying most reproductive manipulations remain elusive. Here, we demonstrate the discovery of a single gene in the eukaryotic association module of Wolbachia's prophage, WO, which kills male Drosophila embryos. The gene, hereafter denoted WO-mediated killing (wmk), causes male lethality when transgenically expressed in uninfected Drosophila melanogaster. Specifically, transgenic expression of wmk results in a female-biased sex ratio, reduced hatching of male embryos, and several male-biased cytological defects during early embryonic development that are typical of Wolbachia-induced male killing. The discovery of wmk as a Wolbachia male-killing gene candidate establishes new hypotheses for the role of phage genes in eukaryotic biology and potentially advances efforts to use male killing for the control of arthropod pests and vectors.

The rhizosphere metagenomes of ancestral and modern wheat cultivars grown under low fertiliser inputs are not significantly different

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The use of chemical fertilizers in intensive agriculture and recent breeding efforts mostly carried out under high levels of fertilizers has led to the hypothesis that modern wheat cultivars might have lost their capacity to associate with soil microbes that would help them acquire nutrients from the soil under low fertilization conditions. To test this hypothesis, ten ancestral and modern wheat cultivars belonging to two wheat species (Triticum aestivum, T. turgidum-) were seeded in a field experiment under low fertilization conditions. At the post-anthesis stage, the rhizosphere soil was collected, its DNA extracted and sent for shotgun metagenomic sequencing. In contrast to our hypothesis, there was no significant difference in the rhizosphere metagenomes of the different cultivars, and this held true when focusing the analyses on specific taxonomic or functional categories of genes. The main factor influencing the rhizosphere metagenome was space, with samples collected from different parts of the field showing significant differences in their genetic and taxonomic content. This variability was also visible for the yields and other agronomic parameters measured at the end of the growing season, which results in significant correlations between these parameters and N and P cycling genes, which would need to be further scrutinized to ascertain their roles in wheat nutrition and growth. Our study showed for the first time that the rhizosphere metagenome of wheat is stable across a wide variety of genotypes, which suggests that modern genotypes did not lose the ability to associate with beneficial nutrientcycling soil microbes.

Modeling the Role of the Microbiome in Evolution

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Multicellular organisms host a great variety of microbes. Most microbes are not pathogenic and some even perform essential functions for the survival of their host (such as absorption of nutrients, defense against pathogens, or training of the immune system, among others). It is known that many microbes cannot survive outside of their host, so their fate is linked to the success of their host to adapt and reproduce. This has motivated theories in which the holobiont (the system consisting of the host and its microbes) is considered as a unit of selection in evolution. However, the role that microorganisms play in the adaptation process of their host is still under debate. This has generated the need for models that explain the role of microorganisms in evolution. In this talk I will present a mathematical model based on genetic regulatory networks that explores the consequences of the "hologenome hypothesis," namely, of considering the holobiont as a unit of selection. Our model shows how symbiotic relationships between multicellular organisms and microbes arises. It also shows the need for specialization of the microbiota and the emergence of complexity in both the dynamics and the structure of the holobiont.

How can microbial volatiles help plants to withstand drought?

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Climate change resulting in higher temperatures leads to enormous losses in yields for major crops, including wheat. A promising and sustainable approach to improve wheat yields under suboptimal conditions stems from volatiles organic compounds (VOCs) producing soil microbes. Terpenes are likely to play important roles in driving the plants resistance to drought. To test this hypothesis, we first seeded wheat plants in pots containing soil with contrasting water stress history and grew them under different water availability for four weeks. The rhizosphere soil was then collected, its DNA extracted and sent for shotgun metagenomic sequencing. The results showed the overarching effects of soil water stress history and the comparatively little effect of short-term water stress. Many genes involved in important water-stress-related processes, including VOC production, showed significant differences in their abundance between the treatments. Second, we have developed a spatially-resolved plant cultivation apparatus named "rhizotron" that allows the simultaneous measurement of microbial and plant genes and VOC emissions with high spatial and temporal resolutions. This system will show which taxa, genes and VOCs are significantly linked to each other and which ones are the main responders to water stress in the rhizosphere of wheat. The causality of the links uncovered in these two experiments will be confirmed by applying VOCs to field-grown wheat plants, setting the basis for VOCmediated microbial engineering strategies.

Gut bacterial physiology and activity along the progression to dysbiosis

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During homeostasis, the gut microbiota provides many critical functions to the human host. Conversely, when the gut microbiota is in dysbiosis, it is correlated to a wide range of diseases. Dysbiosis is currently defined as a disease-specific shift in bacterial community diversity. A specific, functional-based description of dysbiosis is lacking, as well as the understanding of how the gut microbiota reaches this dysbiotic state. We hypothesize that there is a bacterial succession to dysbiosis where changes in bacterial physiology and activity occur before the onset of dysbiosis, specifically in the active bacterial population. These changes in bacterial physiology and activity are what lead to the change in community composition that is currently the endpoint of dysbiosis characterization. Using single-cell techniques such as fluorescently activated cell sorting and 16S sequencing (FACS-Seg), and bioorthogonal non-canonical amino acid tagging (BONCAT), we follow the dynamics of bacterial physiology and activity during the progression to dysbiosis. Specifically, we look at relative nucleic acid content, membrane damage, and translation. Our preliminary data show that upon dextran sodium sulphate (DSS) induced colitis in mice, changes in bacterial physiology occur on the same day as the onset of symptoms, which both precede the onset of dysbiosis. The active population of the gut microbiota is specifically enriched for Akkermansia muciniphila, highlighting the importance of low-abundant bacteria in promoting the disease state. Characterizing changes in bacterial physiology and activity during the progression of intestinal dysbiosis will allow for the identification of windows for successful therapeutic intervention.

Bacteriotherapy in healthy suckling piglets: success parameters to consider.

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Fecal microbial transplantation (FMT) is considered as a strategy to modulate gut microbiota to increase the robustness of piglets. The objective of this research was to characterize the effectiveness of FMT to modulate the establishment of intestinal microbiota in healthy new-born suckling piglets. An equal mix of fecal material harvested from healthy suckling and fully weaned piglets was used to prepare FMT. For the experience, 2 low birth weight (1.13±0.13 kg) and 2 high birth weight (1.70±0.09 kg) animals were selected from 10 litters. FMT material was suspended in bovine colostrum and orally administered to half the selected piglets at 3, 4, 8, 9 and 10 days of life. Treated piglets remained under their sow with the rest of the litter throughout the experiment. At 21 days of age, piglets were euthanized and ileal, caecum as well as colon contents were recovered. Microbiota was surveyed by 16S rRNA amplicon (V4 region) sequencing followed by Dada2, Phyloseg and MaAsLin for analysis. Results revealed that administration of FMT affected the piglet's colon microbiota. However, as the sow had a strong imprint on piglets' microbiota, a large variation was observed between samples originating from different litters. Therefore, when using sow and FMT as dependent variables, MaAsLin analysis indicated that most differences were associated to sows. In conclusion, FMT to piglets had the potential to modulate mainly hindgut microbiota and considering the sow is crucial for a successful FMT procedure. Further studies are necessary to determine FMT effect on lifelong pig's health.

Pushing the boundaries of shotgun metagenomics with genome reconstruction

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Our group's focus is to use nucleic acid sequencing-based approaches to investigate microbial community structure in a variety of different environments and under different conditions. One of our research interests is to investigate the potential of indigenous bacteria to naturally metabolize hydrocarbons in the context of an oil spill event. Over the past several years, the throughput of modern sequencing technology platforms has dramatically increased with lower costs for sequencing and data generation but higher costs and complexity for data computation and downstream processing. This high throughput of data production has significantly increased the complexity and scale of shotgun metagenomic sequence data analysis, which can make overall biological interpretation of sequencing data overwhelming. Here we will address methodology and key concepts in the analysis of shotgun metagenomic sequencing data obtained from laboratory experiments simulating conditions relevant to oil spills in natural ecosystems. A particular emphasis will be put on bioinformatics data mining methodology enabling genome reconstruction/binning from raw sequencing data leading to high resolution biological insights.

Alternative stable states of a holobiont: the intestinal microbiota - host relationship.

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Chronic immune-mediated diseases with a connection to intestinal microbiota status are rapidly expanding and often extremely difficult to cure. We recently proposed the existence of a predisease state, more susceptible to disease development, of the intestinal microbiota-host ecosystem to account for this rapid expansion. We further postulated that health, pre-disease and disease states represent alternative stable states, to explain the difficulty of curing (pre-) disease. Here we show in a rat model system that alternative stable states indeed exist in the intestinal ecosystem, and that inflammation in a context of diet-induced low microbiota diversity can prompt stable state-transition. Based on these results, we propose a theoretical framework providing new insights in the interplay between host inflammatory status and microbiota status, to predict behavior of the ecosystem and guide (pre-) disease prevention and therapeutic strategies.

Poster contributions

37 - The pharyngeal microbiota among healthy adult prisoners in Ghana

Emmanuel Kobla Atsu Amewu

44 - Links between the microbial communities composition, functions and processes and wheat yields and grain quality

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37 - The pharyngeal microbiota among healthy adult prisoners in Ghana

<u>Emmanuel Kobla Atsu Amewu</u>¹, Cynthia Kyerewaa Adu-Asiamah^{1,2}, Samuel Terkper Ahuno^{1,2}, Alexander Kwarteng^{1,2}

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Asymptomatic colonisation of the pharynx and nasopharynx has been identified as the most important risk factor for subsequent invasive disease by bacterial pathogens. Prisons in Ghana are overpopulated, in addition to other significant challenges with health implications, and thus poses a high risk for transmission of infectious diseases, such as meningitis. Colonisation of meningitis-causing bacterial remains to be fully characterised in the meningitis belt. A crosssectional study was conducted with 205 volunteers at the Kumasi Central Prisons (KCP) to assess the pharyngeal colonisation of meningitis causing-bacteria and association with years of incarceration. Pharyngeal swabs were plated on CNA agar for isolation of Group A and B streptococci, Staphylococcus aureus and Streptococcus pneumoniae, and GC agar for the isolation Neisseria species. Streptococcal grouping was conducted for Group A and B streptococci, catalase and dry spot test for S. aureus and optochin test for S. pneumoniae. Oxidase test and gram staining was conducted to identify Neisseria spp. We observed a total colonisation of 52.20%, where Neisseria spp. were highest 49.76% (102/205), followed by S. aureus, 3.90% (8/205) and S. pneumoniae, 0.49% (1/205). There was an inverse association (Chi Square) between colonisation and years of incarceration for only Neisseria spp. (p = 0.0143). The study shows high colonisation of meningitis-causing bacteria among inmates in the KCP and a higher carriage among inmates who have been in prison for fewer years. There is the need for further studies in this area and development of health policies, which include vaccination of at risk inmates nationwide.

44 - Links between the microbial communities composition, functions and processes and wheat yields and grain quality

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Nitrogen nutrition is the most important factor for wheat yields and grain quality. Despite this central role, nitrogen fertilization decisions are often taken based on crude soil physicochemical measurements that ignore the biotic component of the soil. Indeed, soil microorganisms are the main responsible for the nitrogen cycle and their activities make the nitrogen more or less available for plant uptake. The main objective of this project is to develop a predictive model for wheat yields and grain quality based on microbial parameters. In order to parametrize this model, we sampled 1) over 70 commercial wheat fields of the province of Quebec at the beginning of the growing season and 2) the same experimental field every two weeks throughout one growing season. We have measured 1) potential processes related to hydrogen and CO oxidation and CO₂ and N₂O production, 2) community-level physiological profiles (Biolog), 3) soil nutrient, 4) microbial community structure and diversity (16S rRNA gene and ITS) and 5) wheat yields and grain baking quality. Current analyses are aiming at uncovering the parameters that are best linked to the yields and quality variables, and test their robustness across time and space. Overall, our project will result in a model explicitly taken soil microbes into account, which could be used to better inform agronomist and farmers of their N fertilization needs.

54 - Characterization of the replicating gut bacteria in a mouse model

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The naturally occurring microorganisms in the intestines, called the gut microbiota, are essential for host health. Mainly consisting of bacteria, the composition of the gut microbiota is altered in disease, though little is known about how these changes arise. One factor leading to changes in microbial composition during disease is a change in bacterial replication. However, there are no established techniques for identifying individual replicating bacterial members in a complex community such as the gut.

In this study, we aimed to address this gap by optimizing an experimental pipeline to identify replicating gut bacteria using 5-ethynyl-2'-deoxyuridine (EdU) click chemistry coupled with flow cytometry. As a proof of concept, we first conducted EdU labeling of three known gut bacterial isolates: Escherichia coli, Enterococcus faecalis, and Bacteroides fragilis.

We then extracted fecal bacteria from healthy C57BL/6 mice and show that with a 3-hour incubation in bovine heart infusion (BHI) broth with 50 uM EdU, up to 47% of bacteria are replicating cells. Furthermore, restriction fragment length polymorphism on the V4 region of the bacterial 16S rDNA shows that our incubation time with EdU does not significantly alter the original bacterial population. Next, we separated EdU⁺ and EdU⁻ cells using fluorescence-activated cell sorting and identified the number of cells required to accurately reflect gut bacterial diversity. This will enable the valid characterization of these populations using 16S rDNA sequencing. These findings confirm the validity of EdU labeling for identifying replicating gut bacteria.

48 - Comparing methods to assess the biodiversity of soil microbes

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Plant productivity and community composition varies according to the microbial biodiversity in the soil. This diversity can be monitored using Next-Generation Sequencing: DNA is extracted from the soil of a plant's root system, and genetic barcodes for the microbes of interest, are amplified by PCR. These amplicons can then be sequenced, and the reads analyzed via a bioinformatics pipeline. To mitigate any errors that may have been introduced to the sequence data by PCR, or the sequencing reaction, retained sequences have typically been grouped together into operational taxonomic units (OTUs) based on 97% to 100% similarity. OTUs are usually then matched, or clustered, together into taxonomic groups, capturing the microbial biodiversity represented in the sequence data. However, OTUs simultaneously have high false positive rates—they overestimate diversity—and high false negative rates, by being unable to accurately discriminate real biological diversity from errors near the OTU-defining cut-off. Moreover, OTUs identified in different studies are not directly comparable. The recent DADA2 pipeline, however, identifies taxa sequences with as little biological variation as one or two nucleotides, and correctly discriminates real diversity from errors. Since sequences can always be compared between analyses, unlike OTUs, DADA2's output may be more valuable for future biodiversity studies. Thus, an objective of my project is to evaluate how previously generated OTU data from plant roots relates to the sequences identified in the same data by DADA2. Here, I will present our preliminary results comparing both pipelines.

38 - The microbiome of the rhizosphere of canola variates between years.

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The microbes that live in the close environment of plants form the plant microbiome. The core members of this microbiome, i.e., those that are always present, could be of great importance for plant health. However, most microbiome studies analysed a snapshot of a microbial community, while microbial communities are subject to variation across time. Thus, it is necessary add a temporal dimension to the structure of the microbiome to assess the existence of a 'core'. As canola is of economic interest in Canada and since Brassicaceae have simpler rhizosphere microbial communities, it was used as a model plant to study the core-microbiome of the rhizosphere. We tested the hypothesis that canola has a core microbiome that persists through time. To do so, we took advantage of a long-term experiment with rotation systems ranging from canola monocultures to different levels of rotation diversifications. The field experiments were organized in randomized complete block designs with four blocks and were repeated at three locations across western Canada at Lacombe, Lethbridge (AB), and Scott (SK). The canola phase of the rotation systems was sampled in 2013 and 2016. The results show that the rhizosphere microbial community structures changed markedly between 2013 and 2016, with species that fit the core-microbiome concept ranging from 6 to 21.

50 - Composting matters at the Montreal Botanical Garden: a unique species-level microbial characterization

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Organic wastes such as biosolids, green and food residues can make up over 5% of municipal greenhouse emissions during decomposition. Composting has the potential to reduce greenhouse gas production as well as provide sustainable fertilizer. However, our understanding of how microbial communities change to drive the complex chemical and biological processes within this diverse environment is insufficient to generate a consistent final product.

Composting windrows of four different age (1.5, 3, 12 and 24 months), as well as stable feedstock litter, were sampled (4 replicates) at the Montreal Botanical Garden for physicochemical, composition and metagenomic analysis. The V4 region of the 16S ribosomal RNA gene was amplified and sequenced using Illumina Mi-Seq technology. ANCHOR, a newly developed pipeline offering species-level resolution with integrated differential abundance analysis was used to analyse the sequenced data. A total of 2,757,474 sequences were obtained across the 20 extracted samples.

Substantial variation in temperature, oxygenation, C:N ratio and fermentation product liberation was observed throughout the compositing process. Characterization of significantly differentially abundant OTUs allowed us to identify specific species of bacteria involved in the different phases of organic matter transformation.

These changes were linked directly to modification of nitrogen dynamics and carbon mineralisation niches as well as the temperature profile. This informed understanding of microbial colonisation could be used for targeting optimisation of composting practices.

Understanding these specific composting community dynamics can lead to improved waste management strategies which have the potential to significantly reduce the negative impact of humans upon the environment.

6 - RNA viruses associated with Threespine stickleback and the cestode parasite Schistocephalus solidus

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It has recently become apparent that microbes can play key roles in metabolism, immune responses and even reproduction of their host organisms. However, we still know very little about the interactions between the parasite holobiont and the host holobiont. The threespine stickleback and its tapeworm parasite Schistocephalus solidus constitute an important model for ecological and evolutionary studies of host-parasite interactions thanks to the large number of isolated and divergent populations that are experimentally tractable. We have purified viruses from fish and tapeworm tissues and used a metagenomic approach to sequence their genomes. We report the discovery of a new species of positive strand RNA virus, of the genus picornaviridae in sticklebacks and of two new species of negative strand RNA viruses, a chu-like virus and a rhabdo-like virus, associated with S. solidus. We used diagnostic PCRs to investigate the prevalence and mode of transmission of the Threespine Stickleback picornavirus (TSPV), the Schistocephalus solidus Chuvirus (SsCV) and the Schistocephalus solidus Rhabdovirus (SsRV). We used experimental infections to test the effect of SsRV and SsCV on S. solidus infection success and on host and parasite phenotypes. Our results suggest that host and parasite associated viruses must be considered in the future as the viruses could interfere with results of behavioral, physiological, or immunological studies that employ this fish and tapeworm as model system. The discovery of viruses associated with both species provide an opportunity to investigate the role of viruses on the co-evolution of the system.

47 - METHYLOBACTERIUM TEMPERATURE ADAPTATION EXPLAINS ITS SEASONAL DIVERSITY CHANGES

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Methylobacterium is one of the most abundant bacterial genera of the phyllosphere. Despite its potential importance for host plant function, little is known about its finer-scale niche adaptation. Here, we investigated factors that shape the diversity of Methylobacterium at a finer evolutionary scale. We isolated and identified 186 Methylobacterium strains from 36 trees from seven species sampled four times from summer to fall across two sites in the province of Québec (Canada). Using a highly polymorphic, Methylobacterium-specific marker that we developed from gene rpoB. we revealed that a considerable and previously underestimated diversity of Methylobacterium colonizes the surface of leaves in temperate forests. This diversity was structured according to sites, tree species, and time, suggesting that beside its ubiquity, Methylobacterium diversity might reflect tight and long-term adaptation to the distinct environments it occupies and to their seasonal changes. We cultured very different subsets of Methylobacterium diversity from the same leaf, depending upon the temperature of isolation (20°C or 30°C). For example, one the most abundant Methylobacterium lineages (likely representing a new species) was almost exclusively isolated at 20°C. We measured growth of 75 representative strains for different temperature treatments and found strong association between lineages and overall growth profile. Most strains performed better at 20° than 30° but different taxa showed subtle and significant differences in growth performance when subject to temperature increase or decrease. These observations suggest that not only seasonal changes affecting habitat, but also direct adaptation to temperature variations shape Methylobacterium adaptive response to local climatic variations.

39 - Response of the Bat Skin Microbiota to Different Treatments Against the Fungal Disease White Nose Syndrome

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The white-nose syndrome, caused by the fungus Pseudogymnoascus destructans, has killed millions of hibernating North American bats since 2006. The skin microbiota almost certainly interacts with the causative agent of the disease as it infects the skin, inducing in turn ulceration and premature death. Understanding the effects of the white nose syndrome on the skin microbiota of vulnerable species such as the little brown bat (Myotis lucifugus) is crucial for management. On the other hand, investigating potential treatments against the disease is important as it could alter the skin microbial community. Our first objective was to study skin microbiota in a controlled environment in order to explain how the fungus may affect the holobiont. Our second objective was to characterize the microbiota in the context of two promising new treatments for white nose syndrome management: 1) polyethylene glycol, and 2) chemical inhibitors of the proteases. We surveyed hibernating little brown bats, which were experimentally inoculated with the fungus during winter 2016-2017. We explored bacterial, archaeal and fungal community of the skin using high-throughput 16S rRNA and ITS gene sequencing. As far as we know, this study is the first to investigate the complete skin microbiota under controlled conditions and potential treatments against the white nose syndrome. Our preliminary results support the hypothesis of fungal infection and treatment disruptive effects on the skin microbiota, which could potentially be harmful to bats. Skin microbiota should be carefully assessed in future treatment assays to minimize disruption and potential negative consequences on holobiont health.

16 - Effect of colistin on healthy piglet intestinal microbiota

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Escherichia coli post-weaning diarrhea (PWD) is a problematic disease in pig production, as it leads to economic losses through pig's growth retardation and death. One ethological agent of this disease, enterotoxigenic E. coli (ETEC) expressing the F4 fimbriae (ETEC: F4) has developed multiple resistances to antibiotics. Therefore, new treatments options are needed. One such option is the use of colistin. The aim of this study was to evaluate the impact of colistin on the healthy piglet intestinal microbiota. The experimental study was conducted during a 35 days trial, using two groups of 12 healthy weaned piglets. The first group was treated by individual oral administration of colistin for 5 consecutive days and the second group served as control. DNA was extracted from fecal samples collected at day 0 (before treatment), day 3 (during treatment), day 7 (after treatment) and day 35 (after colistin mandatory withdrawal period). A metagenomics approach (PCR V4 rRNA gene Illumina MiSeg sequencing analyzed by Mothur) was used to compare the beta-diversity between the two groups. According to the Jaccard and Yue-Clayton indexes, colistin did not affect the microbiota of healthy pigs. Only few differences on rare OTU were observed between the two groups, at day 35. These results are in accordance with the narrow spectrum of action of colistin, being active only against Gram-negative bacteria. Because of the mass treatment of all piglets in the farm following a PWD episode, it was interesting to evaluate the impact of colistin on the intestinal microbiota of healthy animals.

53 - Evaluation and use of EcoFABs as a platform to screen and analyze commercial product

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Plant-microbe interactions are fundamental for plant health and productivity. The interactions may underly plant-holobiont-based strategies for sustainable agriculture. The dynamic interactions between plants and microbes are challenging to study in a manner that is agriculturally-relevant and reproducible. Fabricated ecosystems, EcoFABs (www.eco-fab.org) are convenient devices that facilitate detailed investigations of plant-microbe interactions including plant developmental exo/endo-metabolomics and microbial kinetics within researcher-defined environments. The multi-laboratory ring-trial using Brachypodium distachyon in EcoFABs showed the statistical consistency of the system. In the current study, we aimed to assess the feasibility of using EcoFABs to rapidly screen the efficacy of a commercial microbial consortia and gain insight into the mechanisms using targeted and untargeted metabolomics. SYNERGRO (Concentric Aq) is a proprietary microbial formulation that improves crop yields and replenishes agricultural soil. Here we present the first successful use of EcoFABs to grow the high-value crops; collard greens (dicot, Brassica oleracea) and white spear onion (monocot, Allium fistulosum). Both crops showed increased biomass and root architecture with SYNERGRO. Additionally, metabolomics analysis will be used to profile the metabolomes associated with SYNERGRO performance within the EcoFABs under various conditions including abiotic stresses. Overall, our study highlights the feasibility of using EcoFABs to rapidly determine key features (chemical, microbial or plant physiological) to establish a product performance assay and to potentially identify the mechanisms of commercial products.

51 - The role of plant miRNAs in shaping the rhizosphere microbiota

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Plant associated microbiota have been thought to be impacted mainly throughout organic signaling molecules such as sugars or organic acids released in the rhizosphere. Recent data demonstrated the involvement of small RNAs (miRNA) in animals - gut microbiota interaction (Liu et al. 2016). These miRNAs have a key role in regulating gene expression and are thus considered as essential regulators of biological processes. It is well known that plants produced a great diversity of them to control their own growth and development as well as their responses to stress or pathogens. We thus hypothesized that miRNAs could be involved in the interaction processes between the plant and its rhizospheric microbiota. Using next-generation sequencing technologies and advanced bioinformatic analyses, we aim for the first time to demonstrate that higher plants use this genetically mobile information to shape the diversity and activity of their rhizosphere associated communities of bacteria and fungi. An experiment was conducted using Arabidopsis thaliana and Brachypodium distachyon, where we extracted and sequenced the small RNAs from the rhizopshere of these two plants and from the bulk soil in unplanted control pots. We observed that both rhizospheres were enriched in small RNAs and we identified miRNAs specific to each plant and others that were common to both of them. These latter could constitute universal regulators of rhizospheric microbiota. These preliminary results are encouraging and we are currently running complementary experiments to validate our hypothesis of this plantrhizospheric microbiota communication throughout miRNAs.

27 - Life in the Gutter: Exploring Wastewater Microbial Communities Involved in a Wastewater Treatment Plant

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Three trillion litres of wastewater are generated annually in Canada, making effective treatment one of the most nationally important biology-based industrial processes. Advances in sequencing and bioinformatics technologies provide new opportunities to illuminate the complex microbial communities underpinning wastewater treatment. To investigate microbial community change throughout wastewater treatment, three stages were sampled: primary wastewater (PW), activated sludge (AS) and wastewater treatment plant effluent (EF). Wastewater constituents and characteristics were measured and 16S ribosomal RNA gene sequencing with a non-clustering bioinformatics pipeline, Anchor, was used to profile the microbial community. Of 1,109 OTUs identified, 470 were confidently annotated as species, 361 at genus level, 82 at family level, 41 at order level, 13 at class level and 141 were poorly characterised. Across all samples, the most prominent phyla included Proteobacteria, Firmicutes and Bacteriodetes, with Firmicutes shifting from PW (28%) to EF (7%). Differentially abundant analysis revealed strains of prominent human gastrointestinal tract species, such Facalibacterium prausnitzii, were lost from PW and replaced in AS by species such as the denitrifying Sterolibacterium denitrificans and sulfur oxidising Sulfuritalea hydrogenivorans. Although EF chemical analysis suggested significantly reduced bacterial load, diverse species including common faecal inhabitants Prevotella copri and Bacteroides uniformis as well as common water inhabiting bacteria were detected. In-depth understanding of the microbial communities which enable wastewater treatment opens up potential strategies for improving process efficiency as well as generates novel opportunities to clean persisting contaminants. Such impact could help reduce the economic and environmental burden of undertreated wastewater release into Canadian waterways.

24 - Surface metabolome together with sea water temperature and trace metals contamination shape the surface microbiote of the Mediterranean brown seaweed holobiont Taonia atomaria

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In marine ecosystems, macroalgal surfaces are prone to be colonized by complex communities of microorganisms. Seaweeds and epiphytic microbes have developed a wide range of interactions defining an integrated entity called holobiont. Especially, algal surfaces constitute a privileged area of chemical interactions with the associated microbiome. Advances in technologies for microbiome sequencing and metabolome analysis have enabled these research fields to raise, allowing a better understanding of the impact of surface metabolites released by algae on their epiphytic microbial communities.

The cosmopolitan brown alga Taonia atomaria (Dictyotaceae) was shown to produce surface antifoulants. The impact of environmental factors on its surface metabolome and epiphytic microbial communities of was investigated during its occurrence period (February to July) on five contrasted sites (North-Western Mediterranean Sea). Densities of heteroprokaryotic cells and bacterial alpha-diversity indexes showed a continuous increase during survey period whatever the sites. A distinct beta- and a higher alpha-diversity were observed in comparison to communities from surrounding sea water and biofilms formed on abiotic surfaces. The increase of sea water temperature was found to drastically impact the community composition for all sites (e.g increase of Bacteroidetes). Moreover, in two sites with distinct trace metal contamination profiles, specific bacterial taxa (e.g. Rubritaleaceae) were observed at the surface. In line with previous works, strong correlations between epibacterial communities and surface metabolome were supported using a multi-omic approach. Thus, several metabolites (e.g. DMSP and proline) already reported for their ecological significance, seemed to play a key role in the colonization process during summer.

49 - Complex impacts of neonicotinoid pesticides on the phyllosphere bacterial communities of soybean and corn

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The phyllosphere (aerial surfaces of plants including leaves) is a dynamic habitat for microbiota. It is exposed to various disturbances and environmental changes. Non-pathogenic microbiota, such as leaf beneficial bacteria, play a critical role in plant growth and health, which can lead to an increase in food crop performance. There is a crucial need to understand the dynamics of plant-microbial composition in agroecosystems and the community resilience to external parameters such as agricultural practices (e.g. pesticide application). During the last decades, a family of systemic insecticides, called neonicotinoids, has been widely used to control foliar and soil early-season pests, such as soybean aphids. Few studies have focused on the non-target and temporal effects of these chemicals on agroecosystem microbiota. In this study, we aimed (i) to quantify soybean and corn leaf bacterial community composition and (ii) to explore bacterial variation and temporal dynamics in a three-year soybean/corn crop rotation and in response to neonicotinoid seed treatment. Our initial approach using 16S rRNA gene amplicon sequencing suggests that neonicotinoids have complex effects on phyllosphere bacterial composition. These impacts vary among host species and growth stages, as well as over time (during the growing season and among years). We discuss the implications of our findings for predicting agroecosystem responses to pesticide use and crop rotations.

57 - Resistome diversity in intestinal microbiome of pigs raised without antibiotics or with penicillin as a prophylaxis

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Antimicrobial resistance is a major public health menace recognized worldwide and livestock animals are suggested to be a significant source of antimicrobial resistant bacteria and antimicrobial resistance genes (ARG) that may reach humans either by direct contact with the animals, consumption of meat or through the environment. The aim of the study was to assess the diversity of ARGs found in feces of commercial pigs raised in four distinct husbandry settings. One of the settings followed conventional practices, involving prophylactic administration of penicillin, while pigs in the other three settings were reared without the use of antibiotics. For each husbandry setting, feces of two groups of 84 days old growers originating from seven litters were sampled (26 to 35 animal per group). Fecal metagenomic DNA was extracted and pooled by group. The pooled DNA was then subjected in parallel to shotgun-metagenomic sequencing and targeted-metagenomic sequencing using the myBait capture system with a custom pool of probes targeting 4,009 ARGs. With both methods, ARGs conferring resistance to tetracycline were found to be most prevalent which was expected given the prominent use of tetracycline in swine production. The two methods were found to be complementary as although the baiting approach allowed for the qualitative identification of more than twice the number of individual ARG clusters compared with shotgun sequencing, the latter provided the quantitative proportions of ARGs within and between samples in addition to determining the phylogenetic composition of the microbiome which provides important information on potential agents of infectious disease.

55 - Screening for antimicrobial potential of bacterial isolates associated with Porites astreoides, the dominant Caribbean coral-reef species.

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Marine invertebrates, as holobionts contain symbiotic bacteria that coevolve and develop antimicrobial substances. These bacteria are an underexplored source of new bioactive molecules. Within the coral microbiome, bacteria with antagonistic activity against pathogens could be considered as putative BMC (Beneficial Microorganisms for Corals), promising tool to protect or improve coral health. We culture and characterize bacteria associated from dominant Porites astreoides coral species of Guadeloupe, in an effort to understand what secondary metabolites might be released. A total of 26 independent bacterial colonies were isolated based on colony morphology and 16S rDNA sequence analyses. The collection of coral bacterial isolates was screened for antimicrobial activity against two marine pathogens Vibrio aestuarianus and Vibrio splendidus. Our results showed that 39% of the isolates exhibited antimicrobial activity on both marine pathogens. We taxonomically identified these strains and found that members of the Vibrio genera were the most representative producers of antimicrobial substances. The variability observed among the different coral bacterial isolates, revealed that the production of antibiotic substances not only is strain-specific, but in many cases may also depends on other parameters. Our results revealed that within the microbial flora of the Caribbean dominant coral species Porites astreoides, the existence of beneficial bacteria species that might play a role in their fitness and their resilience among Caribbean coral reefs. Further investigation, with the addition of Porites astreoides microbiome member in the BMCs would represent a novel approach for minimizing coral mortality in the context of increasing environmental pressure.

45 - Bacteriophages modulate ulcerative colitis-derived bacterial communities

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Inflammatory bowel diseases (IBD), which include Crohn's disease (CD) and ulcerative colitis (UC), are thought to arise from an inappropriate interaction between the immune system and commensal bacteria. Bacteriophages (phages), which occupy the gut at similar abundances to bacteria, are overlooked in most studies. Phages have been shown in several ecosystems to be powerful modulators of bacterial communities. In the context of IBD, the phage-bacteria interactions in the gut remain poorly understood. We propose that phages alter the abundance and diversity of UC-derived bacterial communities, similar to other systems. To address this hypothesis, we isolated bacteria from UC patient fecal samples and administered these to germfree mice. Following colonization of bacteria, we administered phages derived from healthy or UC patient fecal samples and induced colitis in these mice using dextran sodium-sulfate (DSS). In comparison to the no phage control, the virus-to-bacteria ratio was elevated in mice given phage treatments, suggesting that healthy and UC-derived phages are infectious. In support of these findings, 16S rRNA gene sequencing revealed differences in compositional dissimilarity between bacterial communities given healthy or UC-derived phages relative to the no phage control. Furthermore, in mice given UC-phages, there was a 15% decrease in the phyla Bacteroidetes, which was maintained during the colitis period. Together, our data suggest that phages in the gut can modulate whole communities of UC-derived bacteria. By understanding how phages modulate their host communities in the context of IBD, our data may guide therapeutic interventions focused on using phages to manipulate the gut microbiota.

28 - Holobiosis in an urbanizing world

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Urban landscapes are among the most ubiquitous and fastest growing ecosystems in the world, and organisms colonizing these environments are subject to a suite of novel stressors and selective pressures. While this makes urbanization a major conservation concern, these same properties mean cities can be opportunistically leveraged as an experimental system to study holobiont evolution in a free-living context. Using a 16S amplicon sequencing approach, we tested for microbiome convergence among replicated urban and forest populations of wild eastern grey squirrels. We observed that urban populations hosted bacterial OTUs which were absent in adjacent forest populations and converged upon similar inferred metagenomic functional profiles. Namely, urban squirrels exhibited a reduction in the relative abundance of fibrolytic taxa but increases in taxa specializing on host-derived nutrients. These inter-city patterns were mirrored at within city spatial scales, such that squirrels in forest fragments embedded in an urban landscape hosted bacterial communities more akin to squirrels in forests 300km distant than to those occupying bordering urban habitat. We speculate that access to anthropogenic food sources contributed to these patterns. However, we also demonstrate that measures of stress physiology more parsimoniously explained major variation in the bacterial microbiome than the environment from which a squirrel originated. We conclude that although the holobiont's external environment can shape constituent microbial communities, in many cases it does so indirectly, environmental affects filtered through host physiology. Thus, uncovering the impacts of urbanization on eco-evo dynamics that shape the holobiont requires an understanding of microbial ecology and host physiology.

46 - Flavonoid function in phytobiome of land and sea plants

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The concept of holobiont is changing our understanding of ecology and evolution of plants. The symbiotic and pathogenic microbial associations profoundly affect the host plant growth, development, fitness and survival under various climatic conditions. Major shifts in the microorganismal population and their interactions within the holobiont contribute to the host health and are affected by temperature (soil, water and air), light quality (from sunrise to sunset), water depth (micro and macroalgae). One other mechanism of holobiont functioning is the presence of flavonoids and the signal exchange between the host and the microorganisms for colonization. Apart from signal exchanges, the flavonoids presence governs evolutionary significance in retaining specific microbiome of the host and also as a function of hereditary endocellular symbiogenesis. UV-protectant flavonoids were of significance in the establishment of land plants, most of which are hypothesized to have been acquired from microalgae. They also function as chemical markers in cells that lack polarity and for high light adaptation. In this talk, the flavonoids of land and sea plants are compared to understand the multiple functions that they impart in the holobiont.

30 - Induction of Prophages of Human Gut Bacteria by Xenobiotics

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Bacteriophages are major contributors to the human gut microbiota: they are found in similar abundance to their bacterial hosts and contribute up to 17% of the human fecal metagenome. In the gut, bacteriophages are commonly found in bacterial genomes as prophages. Prophages are ecologically important as they can modulate their bacterial host's phenotype. Prophages are not an evolutionary dead-end for bacteriophages, as they can switch back to lytic replication through induction. Induction is typically caused by a stress response of the bacterial host to either environmental or cellular factors.

It is currently unknown what gut-specific environmental conditions might trigger prophage induction. We are investigating the inducing potential of xenobiotics in the human gut, as our previous work found that they could up-regulate prophage induction genes in this system. Hypothesis: Xenobiotics induce prophages leading to detectable changes in bacterial and phage communities in the gut.

We are tested several xenobiotics distributed over different drug classes: antibiotics, anticancer, and non-steroidal anti-inflammatory drugs. We screened these xenobiotics against multiple human gut bacterial isolates belonging to the major phyla of the human gut (Actinobacteria, Bacteroidetes, Firmicutes, and Proteobacteria) and show that induction is xenobiotic and strain-dependent. We identify many xenobiotics capable of inducing prophages, including non-steroidal anti-inflammatory drugs (Tolmetin and Diclofenac). As regulators of bacterial communities, bacteriophages are likely influenced by everyday xenobiotics in the human gut

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21 - Effect of crop rotation on N2-fixation and nitrification genes expression in the microbiome of brassicaceous crops

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All plant organs are colonized by bacteria and fungi forming the plant microbiome, and the association between the plant and its microbiome functions as a holobiont. N is a limiting element in canola production while the prokaryotic genes nif H and amoA are keys in N2-fixation and nitrification processes in the soil. Understanding these processes in the root microbiome is a key to intensify crop production in a sustainable way. This study aims to identify the rotation system with best efficiency of N2-fixation and nitrification in the crop microbiome by quantifying the expression of genes involved in these processes. Field experiments in the Canadian Prairies were established using five brassicaceous crops grown on plots previously planted with wheat, lentil or left in fallow. Roots and rhizosphere soils were sampled at blooming for RNA extraction. Nif H and amoA genes expressions were determined by qRT-PCR. Results showed higher expression of archaeal amoA in rhizosphere, while bacterial amoA and nif H had higher expression in roots. A previous lentil year allowed the highest overall expression of nif H and amoA genes in brassicaceous crops. Higher expression of nif H was found in all crops except LL canola, with the highest lentil effect on N₂-fixation found in ethiopian mustard. Both archaeal and bacterial amoA genes expression was highest in false flax and lowest in polish canola microbiomes. This experiment showed that previous-year treatment affect N₂-fixation and nitrification in the microbiome whose expression varied with crop species.

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