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PhD / Postdocs Labex day 2022

- 11 October 2022 -

Final program

- 9.10 Bus arrival
- 9.15-9.30 Reception
- 9.30-9.45 Introduction by the LABEX ARBRE Director's unit Stephane Uroz and the organizing team
- 9.45-10.30 Speed dating
- 10.30-10.45 Coffee break
- 10.45-12.45 Round table: What to do after a PhD/postdoc? With the participation of Flora Todesco (President and co-founder of WETRUF), Nicolas Bilot (Chargé de mission valorisation des Services Ecosystémiques), Emilie Joetjer (Chargé de Recherche INRAE Grand-Est Nancy) and Thibaud Sauvageon (Médiateur scientifique).
- 12.45-14.00 Lunch
- 14.00-15.00 Regular + Flash talks
- 15.00-16.00 Poster session + Coffee break
- 16.00-17.15 Regular + Flash talks
- 17.15-17.30 Outro by the organizing team
- 17.45 Bus departs INRAE

First session of regular and flash talks 14.00-15.00

1. CiTIQUE: producing scientific quality-grade results with citizen on ticks in an open laboratory (Tous Chercheurs - Regular talk)

Jonas Durand

ABSTRACT: CiTIQUE is a participatory science program whose main goal is to study the ecology of ticks and tick-borne pathogens in order to improve prevention. Its principle is based on collaboration with citizens, who can report their tick bite and send the biting ticks to a laboratory. Most programs stop there, with citizens as data providers, but we decided to involve citizens more in the program by working with them in a research lab open to citizens, the Tous Chercheurs laboratory. There, citizens can participate in 2-days workshops during which they co construct a research question with a scientist and then analyze ticks to answer the question. Several participants told us that they were bitten by ticks brought home by their pets. Ticks take a while to bite their host and can move on to another. This raises the question: is there a risk to human health when humans come into contact with ticks from their cats or dogs? To date, more than 230 citizens have worked together to analyze ticks biting cats or dogs in France. More than 811 ticks have been identified morphologically using a citizen-friendly identification key. The quality of the results produced by the citizens was compared to that of scientific experts. 89% (624/811) of the ticks were adults and 12% (96/811) were nymphs. 83% (671/811) belonged to the genus Ixodes while Dermacentor were only found in dog biting ticks. Both genus are known vectors of human diseases such as Lyme disease. These results show that ticks brought by our pets can represent a risk for human health. Furthermore, these results show that it is possible to produce scientific-grade results by working with citizens in a research laboratory.

2. Metal-Chelating Peptides obtained from Enzymatic Hydrolysis of Protein Extracted from Fish Tilapia Scales (Oreochromis spp.) (LRGP - Flash talk)

Jairo Andrés CAMANO ECHAVARRIA

ABSTRACT: Bioactive peptides are biological compounds, naturally encrypted within protein sequence under an inactive form and released upon hydrolysis. Nowadays, these peptides have gained great interest due to their numerous bio functional activities reported in the literature, among them, metal-chelating activity. Due to the wide range of bio functional activities associated to metal ions, Metal-chelating peptides (MCPs) could offer several interesting applications. MCPs promote stability, absorption, and bioavailability of several dietary minerals (i.e., Ca2+, Zn2+, Fe2+ and Cu2+). Also. MCPs are antioxidant molecules, since they avoid pro-oxidant effect by complexing transition metal ions (Fe2+ and Cu2+). On the other hand, MPC can act as inhibitor of zinc-dependent enzymes related to the high blood pressure (i.e., angiotensin-converting enzyme, ACE), being suitable molecules for antihypertension treatments. Additionally, due to the zinc, copper and iron-chelation, and thus preventing their uptake by microorganisms, MCPs could present antimicrobial properties. Therefore, this research aimed to evaluate the use of Tilapia scale as protein source for metal-chelating peptide production. Tilapia scale protein was submitted to 3 enzymatic treatments. Results showed that Tilapia scale protein hydrolysates prepared using Alcalase®, and Alcalase® followed by Flavourzyme® showed the highest metal chelating properties, which was also associated with the high antioxidant and antihypertensive potentials. Both hydrolysates offer an effective strategy for obtaining metal-chelating peptides. Further research is needed to explore hydrolysates potential use as functional ingredients in food industry, and nutraceutical products.

3. TITLE: Evidence of the role of the siderophore Rhizobactin produced by *Caballeronia mineralivorans* PML1(12) in mineral weathering (IAM - Regular talk)

<u>Cintia Blanco Nouche</u>, Cédric Paris, Tiphaine Dhalleine, Marie-Pierre Turpault, Stephane Uroz

ABSTRACT: Forests are low human impact ecosystems that function on their own resources. Recycling and nutrient mobilization are so essential processes. Rocks and minerals are considered as an important reservoir of nutritive elements in such nutrient-poor ecosystems.

However, the nutrients entrapped in these minerals are not directly available for the biosphere. In these circumstances, several bacteria have developed different strategies including the production of siderophores- to make trapped nutrients available. Despite all the new siderophores discovered each year, there is comparatively less empirical evidence for their role in mineral weathering (MWe). Strain PML1(12) from the genus Caballeronia has been demonstrated to weather minerals, to be effective at chelating iron and to regulate siderophore production according to the presence/absence of iron. Nevertheless, the genes involved, the type of siderophore and the role of this siderophore in MWe have not been demonstrated. In this study, we report the characterization of a Non-ribosomal peptide synthetase Independent Siderophore (NIS)-type biosynthesis gene cluster. Targeted mutagenesis associated to chelating assays and High-Performance Liquid Chromatography (HPLC) fractionation demonstrated the production of a single siderophore encoded by this NIS region. The use of HPLC and high-resolution and tandem mass spectrometry (HRMS and MS/MS) allowed to identify the siderophore as Rhizobactin. The evidence of the implication of the siderophore in

MWe was demonstrated during a hematite alteration assay. This study provides the first demonstration of the synthesis of a NIS siderophore in the genus Caballeronia and the involvement of this siderophore in mineral weathering. Our conclusions reinforce the idea that strain PML1(12) is particularly well adapted to nutrient-poor environment.

4. Metabolomic approaches to decipher the role of poplar glutathione transferases during infection by the rust fungus *Melampsora laricipopulina*. (IAM - Flash talk)

Laura Morette

ABSTRACT: Glutathione-transferases (GST) constitute a widespread superfamily of multifunctional enzymes involved notably in the detoxification of exogenous toxic compounds and in the synthesis or transport of specialized metabolites^{1,2}. Numerous studies have shown that their expression levels are strongly regulated under abiotic and biotic stress, especially in plants. The functions carried by GSTs rely either on the catalytic transformation of these molecules, mainly through glutathione conjugation reactions, or on their non-catalytic property known a "ligandin" dedicated to the transport or storage of various metabolites. However, the substrates or ligands and therefore the precise physiological functions of most GSTs are still unknown. The objectives of my thesis are i) to carry out an inventory of specialized metabolites present in preparation of secondary metabolite-enriched poplar extracts by metabolomic approaches, ii) to identify ligands and substrates of some poplar GSTs present in the previously analyzed extracts using in vivo and in vitro trapping approaches, iii) to characterize at the biochemical and structural levels the interactions of these molecules with some GSTs. During my first year PhD thesis, various extracts from poplar aerial organs (leaves, flowers, fruits, buds were analyzed by liquid chromatography coupled to tandem high resolution mass spectrometry. The molecular network generated by MetGem software allowed the identification of major families of metabolites such as flavonoids, known to be the ligands of GSTs³. This preliminary analysis will be applied in the near future to extracts obtained from poplar leaves infected by the pathogenic fungus Melampsora larici-populina, to help to decipher the role(s) and function(s) of poplar GSTs whose expression is upregulated during infection.

- 1 Lallement P.A., Brouwer B., Keech O., Hecker A., Rouhier, N., The still mysterious roles of cysteine-containing glutathione transferases in plants. Frontiers in Pharmacology, vol. 5 (2014)
- 2 Sylvestre-Gonon E., Law S.R., Schwartz M., Robe K., Keech O., Didierjean C., Dubos C., Rouhier N., Hecker A., Functional, Structural and Biochemical Features of Plant Serinyl-Glutathione Transferases, Frontiers in Plant Science, vol. 10 (2019)
- 3 Sylvestre-Gonon E., Morette L., Viloria M., Mathiot S., Boutilliat A., Favier F., Rouhier N., Didierjean C., Hecker, A., Biochemical and Structural Insights on the Poplar Tau Glutathione Transferase GSTU19 and 20 Paralogs Binding Flavonoids, Frontiers in Molecular Biosciences, vol. 9 (2022)

5. Assembly of the respiratory complex I in plants (IAM - Flash talk)

Alicia López-López, Jonathan Przybyla-Toscano, Olivier Keech, Nicolas Rouhier

ABSTRACT: Mitochondria is the organelle where the production of ATP takes place. ATP is the principal molecule for storing and transferring energy between the cells, being essential for life. For its production is necessary the mitochondrial oxidative phosphorylation (OXPHOS) system which is form by 5 protein complexes found in the inner membrane of the mitochondria. The first 4 complexes are part of the electron transport chain, that transfers electrons from complex I (that obtains 2 electrons and 2 H+ from oxidation of NADH) to complex IV, which forms H2O from the electrons that receives. This chain is important because induces the H+ pumping from the complexes to the intermembrane space, what creates a gradient that induces the activity of Complex V (ATPase synthase) that uses this gradient and ADP + phosphate (Pi) to generate ATP.

My project is mainly focused in the study of the assembly of Complex I (CI) in plants, the first entry point of electrons. CI is a protein complex that has a conserved L-shape structure that is characterized by the presence of a membrane hydrophobic arm in the inner mitochondrial membrane, where the reduction of the coenzyme Q to ubiquinol and H+ pumping takes place, and of a peripheral hydrophilic arm present in the mitochondrial matrix, where NADH oxidation and electron transfer occur. This electron transfer is possible due to the presence of 8 Fe-S clusters (two [2Fe-2S] and six [4Fe-4S] clusters) bound by 5 subunits (51 kDa, 75 kDa, 24 kDa, PSST and TYKY) of the 45 that are part of the CI. It is thought that the Fe-S clusters are first assembled in the subunits, and afterwards, the subunits are assembled stepwise in larger assembly intermediates which will form the whole holo-complex I. The synthesis of Fe-S clusters in mitochondria is carried out by the Iron-Sulfur Cluster (ISC) assembly machinery. A [2Fe-2S] cluster is first de novo assembled on the scaffold protein (ISU). It may be directly transferred to [2Fe-2S] cluster-containing subunits. Additional transfer and conversion steps and players are likely needed for [4Fe-4S] cluster-containing subunits. However, the exact mechanisms by which these five CI subunits obtain their Fe-S clusters from the transfer proteins of the ISC machinery still need to be elucidated.

To clarify these mechanisms, we have expressed in *Escherichia coli* and purified recombinant proteins corresponding to both CI subunits and possible transfer proteins. Whenever proteins are soluble and stable, we intend to perform *in vitro* Fe-S cluster reconstitution and transfer assays using either specific protein couples or multiprotein complexes that are physiologically relevant.

- 1. Przybyla-Toscano, J., Christ, L., Keech, O., and Rouhier, N. (2020). Iron-sulfur proteins in plant mitochondria: roles and maturation. J Exp Bot. doi:10.1093/jxb/eraa578
- 2. Ligas, J., Pineau, E., Bock, R., Huynen, M. A., and Meyer, E. H. (2019). The assembly pathway of complex I in Arabidopsis thaliana. Plant J 97, 447–459. doi:10.1111/tpj.14133.

6. Microbial community engineering tools for the valorization of new Biomolecules (LIBio - Regular talk)

Alexis Dijamentiuk

ABSTRACT: The microbial world harbors a great metabolic potential for the production of valuable biomolecules such as antimicrobials. Historically, most of the efforts at deciphering and engineering metabolic pathways for producing biomolecules of interest have been focused at the single-strain level. However, advances in microbial ecology have put forth that communities of microorganisms can display many useful functionalities, such as the production of valuable biomolecules through syntrophic interactions and cometabolism. In order to harvest this dormant repertoire, tools are needed for microbial community manipulation and engineering. Notably, culturing techniques are needed in order to recover the biological diversity that harbor potentially interesting activities. We proposed to evaluate the contribution of two cultivation systems for top-down microbial community engineering. Namely, a classical setup consisting of simple culture broth allowing bacterial interaction, and an invert emulsion system in which the inoculated culture broth is dispersed as droplets within a continuous oily phase that limits bacterial interactions. Communities have been propagated in these systems in a single propagation event or in semi-continuous propagation referred to as serial propagation. Cultivation of microbial communities altered their structure regardless of the cultivation system. Upon serial propagation, a classical setup leads to stabilization of community structures while invert emulsions lead to the rearrangement of community structures. Moreover, serial propagation in invert emulsions leads to the enrichment of subdominant taxa that are otherwise excluded from the communities. Finally, these two co-culture systems can lead to various communities from the same inoculum, with metabolic activities and molecular that remain to be explored. This highlights the interest of using both systems as complementary microbial community engineering tools for the valorization of biomolecules.

***********15.00-16.00 Poster session + Coffee break*********

Second session of regular and flash talks 16.00-17.15

1. Two-in-one plant defense peptides - characterization of both antifungal and elicitor activities of RISPs (IAM - Regular talk)

Julie LINTZ

ABSTRACT: Plants use defense peptides to resist pathogen attacks. RISPs (Rust Induced Secreted Peptides) are thermostable peptides that occur specifically in Salicaceae species (poplar and willow) and that exhibit a targeted antifungal activity against Pucciniales, the causal agents of rust diseases. Interestingly, RISPs also elicit physiological responses in poplar (Petre et al., 2016). In plant, only a handful of peptides combining both antimicrobial and elicitor activities have been described. Our work aims at characterizing the antimicrobial and the elicitor modes of action of RISPs. Regarding the antifungal activity, pull-down assays showed that purified RISPs bind to urediniospores of Pucciniales, and microscopy assays showed that RISP-GFP fusions accumulate at discrete areas onto the spore surface. Regarding the elicitor activity, RISPs genes cluster with Leucine Rich-Repeat Receptor-Like Protein (LRR-RLP) genes onto poplar and willow genomes, which we termed RALRs (RISP-Associated LRR-RLPs). We currently test the hypothesis that RALRs recognize RISPs to trigger immune signaling, by investigating the RISP-dependent activation of RALRs in Nicotiana benthamiana using protein biochemistry and molecular physiology approaches.

2. Characterization of *Phanerochaete chrysosporium* mutants resistant to *Bagassa guianensis* wood extractives (IAM - Flash talk)

<u>Delphine Noël</u>, Duy Vuong Nguyen, Antonio Fernández-González, Emilie Rezer, Nadine Amusant, Eric Gelhaye, Mélanie Morel-Rouhier, Rodnay Sormani

ABSTRACT: Wood decaying fungi are ever more attractive as they possess an array of enzymes able to degrade lignocellulosic material. However, during wood degradation processes, fungi have to cope with toxic molecules released from wood and defined as "extractives". Thousands of wood extractives molecules have been discovered but very little is known concerning their putative antifungal activity. In that context, woods from tropical species appear as models of choice to study wood extractives toxicity due to their strong durability and high amounts of extractives. To highlight the molecular targets of such extractives in fungi, a collection of Phanerochaete chrysosporium mutants was generated using UV mutagenesis and screened for resistance to Bagassa guianensis wood extractives. Among the 34 isolated mutants, allelic series of AGR57 7124 mutations have been identified for which 13 mutants display the same allele. Ectopic expression of that allele in wild type genetic background confers resistance to B. quianensis wood extractives. Moreover, those mutants have been used as genetic tools to prove the protective role of extractives in wood: mutant resistant to B. quianensis wood extractives are able to mineralize B. guianensis wood while the WT cannot. Based on a phylogenetic analysis, the AGR57_7124 gene codes for an orthologous to the human DENND6 protein. To decipher the molecular mechanism leading to this resistance phenotype, the functional characterization of PcDENND6 is undergoing.

3. Understanding Poplar-Microbe Interfaces: From model systems to model synthetic communities (IAM - Flash talk)

Aline Sauvage

ABSTRACT: Thousands of species of fungi occur in the soil of temperate and boreal forests, but only a small fraction is able to form symbioses and bring in benefit to tree growth. Ectomycorrhizal fungi (ECM), but also endophytes with positive, negative or neutral effects on their hosts, are important members of the tree microbiome. Thanks to few in vitro model systems, significant progress has been made in the elucidation of signals that are critical for ECM development but still, very few is known about endophytic interactions and the interplay of these two interactions. The objective of this project is to develop an experimental setup allowing to track the colonization of the root system of Poplars in controlled conditions by single to multiple microorganisms in order to extend our molecular studies from simple in vitro systems to more complex systems. Here, we show preliminary microscopy results of Poulus tremula x alba roots colonized with a community of ECM and endophyte fungi in in vitro conditions. This system could be further explored to test more complex communities in wild type and transgenic Poplar plants.

4. Dynamic sampling and inference for a smart forest monitoring with application to the french national forest inventory (LIF - Regular talk)

DUONG Ho Kim Trinh **Supervisors:** BOURIAUD Olivier, CHAUVET Guillaume

ABSTRACT: Since the early twentieth century, National Forest Inventories (NFIs) have been implemented in many countries to quantify and describe the forest resources. In the past decades, forests have experienced a rapid increase in disturbances – storms, fires, insect damages, and so on – that have strongly impacted on their state. Thus, new objectives for NFIs consist in monitoring the forest where disturbances occurred, to see how the forest reacts and if management action is required to repair. While the methods used in NFIs were conceived at a time when the forests' dynamic was low, they now have to face new challenges: e.g., increased reporting frequency and responsiveness, necessary to monitor forest response to these increased disturbances. These new objectives therefore challenge their sampling and estimation methods.

The French NFI methods are the closest to mentioned above objectives, because the sampling intensity can be tuned yearly and spatially. Though they have been implemented for 15 years, these methods have never been examined and formalized according to the sampling theory, and they have never been evaluated. Therefore, the main purpose of my PhD thesis is to describe, develop and optimize the sampling and estimation strategies of the French NFI with the accuracy, flexibility, and responsiveness in order to adapt to the current critical context.

A first part of my PhD work consists in describing the French NFI sampling and estimation methods under the design-based paradigm. The French NFI makes use of a spatial systematic grid to create annual samples, and a set of random points are selected each year at the first phase. Based on photo-interpretation, a stratified sample is selected at the second phase. Then, post-stratification is applied to estimate the parameters of interest. Artificial data as well as inventory data arising from 15 years of survey are available. They will be used to evaluate sampling and estimation strategies, by using the R software. In particular, one of the objectives of my thesis is to propose other possible estimators, with improved accuracy.

Key words: sampling; estimation; forest inventory; post-stratification; variance

5. Influence of slow stomatal responses on leaf and ecosystem fluxes and water-use efficiency (SILVA - Flash talk)

Yasin GUNDESLI

ABSTRACT: Stomatal opening allows the plant to absorb the optimal amount of carbon for photosynthesis while limiting water loss. Unlike photosynthesis, stomata respond slowly to changes in environmental factors. This has been studied only at the leaf level. One of the goals of my PHD is to characterize the importance of the decoupling between photosynthesis and stomatal conductance on the dynamics of water and carbon fluxes at the ecosystem scale. We will base our work using kinetic data measured at the leaf level under controlled conditions and in the forest at the Fr_Hes ICOS ecosystem station.

6. Study of nutrients translocation in Oak tree rings depending on soil fertility and forest management (BEF - Flash talk)

Caroline Jaozandry

ABSTRACT: This thesis project aims to identify and understand the patterns of translocations in tree rings by relying on observation and experimentation networks. Translocations of mineral elements from mature or senescent organs to storage or growing organs are one of the natural mechanisms that allow trees to produce biomass on soils that are generally infertile and unsuitable for agriculture. In a context where pressures (anthropogenic activities, air pollution and climate change) are increasing on forests, together with degrading mineral nutrition, knowledge of this mechanism is of major importance for the sustainable management of forests, whether public or private. This project is based on three large national observation networks (RENECOFOR) and experimentation (by manipulating soil fertility - positive: AMMENDMENT and negative: MOS of the IN-SYLVA infrastructure) for which a certain number samples are already available (wood cores) and will be supplemented by specific campaigns (for example to cover all the size classes of trees). In addition to the cognitive aspect, the results of this thesis project will provide the forest manager with an additional diagnostic tool regarding possible biomass exports depending on the type of soil. It will also allow the proof of the concept to be demonstrated for subsequent and wider deployment on the scale of the territory.

7. Climate-sensitive Forest models and forest management: a literature review (SILVA - Regular talk)

Modeste Meliho

ABSTRACT: Forest ecosystems are facing the negative impacts of climate change, which will increase in the future according to different climate projections scenarios. In this context, climate-sensitive forest modelling is gaining momentum to support forest decisions. The objective of this review is to assess how the most recent climate-sensitive forest models (CSFMs) address the issue of forest management and adaptation to climate change. A grid has been developed to analyze model attributes in order to diagnose which model can be used depending on the issue addressed. CSFMs are described according to several description criteria, including the modeling approach, the representation of the forest cover, the processes represented, etc. Different forest decisions have been described at different scales. The description of CSFMs and their potential use as decision support tools have been illustrated by twenty CSFMs developed in France. Great attention must be paid to the choice of a model, which is a determining factor in the production of simulations to support forest decisions. There are some gaps to fill in terms of forest model's development and in their use as decision support tools. Regeneration and mortality processes are considered in a limited way in process-based models. There is a lack of forest fire models that represent the forest cover. Communication and collaboration between forest modelers and decision-makers need to be strengthened to facilitate the development of models that are more oriented towards issues of forest management and adaptation to climate change. To address models' comparison and structural uncertainties issues, multi-model comparative analysis approaches can be an interesting complement.

8. Structure and recent dynamics of the industrial roundwood trade from a network analysis perspective (BETA - Regular talk)

Valentin MATHIEU, David W. SHANAFELT

ABSTRACT: While the use of wood products is increasingly linked to sustainability and climate change, forest resources are characterized by an uneven distribution between countries and different regional forest dynamics. Redistributing surpluses resulting from overproduction of wood products to countries with demand deficits to meet sustainability issues can be achieved through trade. The literature offers a comprehensive study of the international timber trade, but few studies to date consider the physical structure of the trade network in their analyses. We study the international industrial roundwood trade from 1997 to 2018 by adopting a network theoretic approach which allows a deeper understanding of the inherent structure of the trade of wood products and complements the existing literature. We expect the global industrial roundwood trade network to follow global trends in economic growth, but to also be sensitive to major economic and political events, and natural disasters. Our results showed that the industrial roundwood trade network increased in value while varying slightly in size, and became more tightly connected over the study period. In particular, its composition in terms of exporters and importers has been varying over the studied period. We also observed variation in network structure over time that can be explained by three broad categories of events: economic disruptions, political events, and natural disasters. Lastly, our results yield implications for forest policy, as the structure of the network can dictate potential cascades of local or regional shocks to the global trade market. For a country, reasonable strategies such as diversifying trading partners or improving national industrial capacity to process roundwood would limit the impact of shocks locally and over the whole network. JEL Codes: C60 General Mathematical Methods; Q23 Forestry; Q27 Issues in International Trade.

KEYWORDS: Globalization; Trade; Wood products; Network theory; Forest policy; Unweighted networks

Poster session

1. Forest trees species shape the biochemistry of soil organic matter under the control of their associated microbial communities (IAM)

KHALFALLAH Fadwa

ABSTRACT: Soil organic matter (SOM) is the largest carbon reservoir on the continents. It contains three to four times more carbon than vegetation. SOM content is particularly high in forest ecosystems, which cover 30% of Europe. The tree species is one of the main factors influencing the storage and biochemistry of soil organic matter, through its litter quality and the species-specific microbial communities. The objectives of our study were (i) to compare the effect of tree species on the biochemical quality of SOM (ii) to investigate the impact of the

biochemical quality of SOM on microbial communities and vice versa. We hypothesized, that the tree effect on SOM biochemical quality was regulated by the pedoclimatic conditions. Also, we thought that the microbial community enriches the SOM with N compounds and some specifics polysaccharides. In this aim, we sampled forest floor and topsoil (0-10 cm) in seven monospecific plantations of different tree species in five European sites (total of 24 conditions). We characterized the organic matter using a technique combining pyrolysis and gas chromatography (Pyro-GC/MS). We also described the microbial communities present in these same samples using metabarcoding (16S and ITS Miseq) and we measured microbial functions involved in SOM decomposition and nutrient mobilization through enzymatic activities. Our Pyro-GC/MS results revealed that some organic compounds were tree species-specific, and others were site-specific. In particular, we showed that N-fixing tree species and

arbuscular mycorrhizal (AM) tree species increased the proportion of N-compounds in SOM. These biochemical results will be discussed in relation to microbial community and functional diversity data.

2. Understanding drought stress resilience in the Fagus sylvatica – *Cenococcum geophilum* ectomycorrhizal symbiosis (IAM)

Huayong Wang

ABSTRACT: With increasing temperature, climate change will likely lead to decreased soil water content in forest ecosystems. After several unusually dry years in France and Switzerland, many beech diebacks have been reported. Therefore, developing forest management approaches to improve the drought resistance of forest trees becomes increasingly important. Ectomycorrhizal (ECM) fungi form mutualistic interactions with the roots of forest trees. They increase the root uptake area with the soil and facilitate water and nutrient uptake from the soil. The ectomycorrhizal fungus Cenococcum geophilum (Cq) is distributed worldwide and is prominent in ecosystems under abiotic environmental stresses (Obase, Douhan et al. 2016). It appears to be more resilient to drought stress than other ECM fungi (Jany et al. 2002). It can form ectomycorrhiza with a high number of alpine herbs and woody plants (both Gymnosperms and Angiosperms) and is often a major component of ectomycorrhizal fungal communities (LoBuglio, 1999; Trappe, 1962). Therefore, this beneficial symbiont is a good candidate to assess the ability of ECM fungi to enhance drought adaptation of trees. In my Ph.D. research project, I aim to assess whether promoting establishment of Cg ectomycorrhizal roots can protect beech plantlets from drought stress. I will (1) perform a drought resistance screening using 100+ Cg isolates isolated from Europe, China, Japan and USA to identify drought resistance isolates; (2) conduct an inoculation experiment in growth chambers with selected Cg isolates inoculated to both Scots pine and European beech seedlings and then, (3) assess the drought resistance of ectomycorrhizal beech trees under different water potential levels. Furthermore, I will use (4) comparative transcriptomics and genome wide association studies (GWAS) of selected Cq isolates to identify the gene networks involved in drought resistance. The overarching aim of this research is (1) to identify Cg isolates having a high mycorrhization ability with beech, but also (2) able to increase the drought resistance of the host.

3. Electrostatic spray drying: a new technology to stabilize probiotics (LIBio)

Preethi Jayaprakash

ABSTRACT: Probiotics are described as 'live-organisms which, when administered in adequate amounts, confer a health benefit on the host'. Interest in the consumption of foods that contain probiotics is increasing and demanding in the market. Their viability must be maintained during manufacture, storage, and delivery to exert their health benefit. Dried form of probiotics is usually prepared by using freeze drying at low temperatures, which is expected to have high viability and to cause less injuries to probiotics. Meanwhile, this technique is considered to be expensive and time consuming. Alternatively, spray drying can be utilized as drying method to encapsulate probiotics but the use of high temperatures causes them to denature and decrease their viability. There is recently introduced spray drying technique with electrostatic force, promises to encapsulate the probiotics at lower temperature (between 35 – 80°C) and increase their viability. The aim objective of this study, to encapsulate Lactobacillus rhamnosus (LGG) with 25% Maltodextrin DE19 by using these three drying techniques and their viability after drying is compared at laboratory and semi-industrial scales. Microparticles are stored at 20°C and their viability was studied for 2 months. The results were significantly different between the technologies rather than process scale. Approximately 100% of viability is obtained with freeze drying and electrostatic spray drying whereas their viability in conventional spray drying had dropped down twice the rate. Microparticles from laboratory scale after two months of storage, showed their viability drop at 0% for spray drying, 56% for

freeze drying and 81% for electrostatic spray dryer. The semi-industrial scale seems to follow the same trend. Thus, the drying conditions influences the viability of probiotics and the choice of drying technique is essential especially in larger scale industries. From this study, electrostatic spray drying presented similar results of freeze-drying technique and thus, can be a good alternative for probiotic applications.

4. Dynamics of the subalpine forests in the French Pyrenees over the last two centuries (SILVA)

Noémie Delpouve

ABSTRACT: The upper subalpine forest line is characteristic of mountain landscapes. This limit is influenced by global change and the subalpine forest dynamics has consequences on carbon sequestration. We studied subalpine forest in its extent with its upper line shift, and in its structure with the transition zone from closed to open forest. The studied period goes from the forest minimum to its current maximum. We focused on the French Pyrenees because it is the first digitized area in the map which dates back to 1850 ("Etat-Major" map). We used two other maps from 1994 and 2010, made by the National Forest Inventory according to aerial photographs interpretation. We estimated the elevation of subalpine forest lines for each date, in 125 municipal districts covering the study area. We then calculated the difference in elevation between two consecutive dates. For a subset of 60 districts, we estimate the ecotone width as the distance between open and closed forests lines, and assess its variation between 1994 and 2010. We observed an upward shift of forest line in most of the municipal districts between 1850 and 1994 (82) %), and after 1994 (76%). On average, the forest line raised of 125 m between 1850 and 1994, and of 50 m after 1994. Hence, the forest line raised on average of 10 m per decade between 1850 and 1994 and of 35 m per decade after 1994. For the 60 districts where we study the ecotone width, the shift velocity was on average of 40 m per decade for open forest line, and of 70 m per decade for closed forest line. An ecotone contraction was observed in 2/3 of the cases, with a contraction velocity of 30 m per decade, between 1994 and 2010. Our study shows an acceleration of subalpine forest line upward shift. Moreover, the faster upward shift of closed forest lines leads to ecotone contraction. This may be due to climate change acceleration, if we suppose a constant land abandonment during the studied period. While forest line upward shift acceleration and ecotone contraction may be a threat for biodiversity, they also represent strong potential sink of carbon for long term sequestration.

5. Original multifunctional derivatives from biosourced polyphenols for cosmetic or pharmaceutical applications (LERMAB)

Georges Eid

ABSTRACT: The decrease in easily accessible petrochemical resources has aroused, over the past ten years, a growing interest in the use of raw materials of renewable origin. First of all, bio-sourced chemistry developed strongly around annual agro-resources; it is also turning more and more towards the use of lignocellulosic biomass from wood because it constitutes a particularly abundant source of renewable carbon on earth and whose use does not compete with food resources. The primary wood processing industry generates large volumes of waste each year which is currently either recycled to other sectors such as paper mills or the panel industry, or used as a source of energy, and therefore towards markets with low added value. The project is situated in this context of sustainable development and enhancement of wood industry co-products through the exploitation of secondary metabolites present in wood, such as phenolic compounds, and more specifically flavonoids which are indeed of interest for their biological activities. The objective of this work is to functionalize accessible and abundant compounds in order to obtain polyfunctional compounds with 2-in-1 properties and thus be able to simplify cosmetic formulations. The project was first initiated on the basis of model flavonoids such as catechin, rutin and naringenin, on the one hand because of their accessibility and on the other hand because of their aromatic structure suggesting that these molecules have a strong propensity for self-assembly. The structural modifications envisaged also present other interests. This is in fact, on the one hand, to exacerbate their biological activities by modulating their hydrophilic / hydrophobic balance, to facilitate their passage through biological barriers and on the other hand, to facilitate their formulation by providing them self-assembly properties. Functionalization has been considered by two routes: by chemical hemisynthesis and/or by enzymatic catalysis (Saik, 2017). The objective is to obtain two types of compounds, in this case bi- and tri-modular compounds. We have thus been able to access bi-modular compounds, by conventional esterification between a fatty acid and the aliphatic hydroxyl of a flavonoid such as catechin. We have also prepared trimodular compounds by chemical hemisynthesis by combining different amino acids, as well as fatty acids of variable length with catechin. Three different trimodular structures have thus been synthesized, in order to obtain different types of amphiphilic compounds, such as conventional amphiphilic compounds, or geminis, or double-stranded ones. The amino acids used were chosen either as models or because of their accessibility, such as lysine, a by-product of the sugar industry, or glutamic acid, present in many plant and animal proteins. The tristrand homologs derived from rutin and naringin, associating a carbohydrate instead of the amino acid could also be prepared by enzymatic catalysis. The antioxidant activity of the synthesized compounds was evaluated by two methods: by UV-visible spectroscopy by evaluating the disappearance of the DPPH radical, as well as by the method of characterization of the inhibition of the oxidation of methyl linoleate. The antiproliferative activity of these compounds was also tested. Finally, their self-assembly properties were also evaluated, in particular by determining their surface activity. All synthesized compounds exhibit antioxidant activity; on the other hand, they have differences in water solubility and different surfactant properties between derivatives based on carbohydrates or based on amino acids.

6. Populus MYC2 orchestrates root transcriptional reprogramming of defences to impair *Laccaria bicolor* ectomycorrhiza development (IAM)

Jose Eduardo Marqués Gálvez

ABSTRACT: The jasmonic acid (JA) signaling pathway plays an important role in the establishment of the ectomycorrhizal symbiosis (ECM) between Laccaria bicolor and poplar. We previously showed that the L. bicolor effector MiSSP7 induces the stabilization of the poplar JAZ6, a JA corepressor protein that binds to Populus MYC2.1 and MYC2.2, orthologs of the Arabidopsis MYC2 transcription factor (TF), blocking their activity. Here we showed that both TFs play a central role in root colonization by L. bicolor mycelium, since their overexpression decreased the formation of the Hartig net, the hyphal network involved in symbiotic nutrient exchanges. By combining RNA sequencing and DNA Affinity Purification sequencing (DAP-seq) analysis, we identified a core set of JA-responsive genes directly activated by poplar MYC2.1 and MYC2.2, that need to be bypassed by the fungi to colonize root apoplastic spaces. These genes encode for other TFs, receptor-like kinases and many defense-related proteins, including terpene synthases (TPS). Monoterpenes produced by some of these TPS impact L. bicolor growth and ECM formation, suggesting a role for poplar root monoterpenes as negative regulators of in planta fungal growth and ECM symbiosis.

7. Etude et conception d'ingrédients xylo-sourcés à haute valeur ajoutée pour le marché de la cosmétique (LERMAB)

Pauline Gérardin and Floriane Mangin

ABSTRACT: Les travaux présentés font partie intégrante du projet « ResiNoeud ». Ce projet intègre le principe de l'économie circulaire et a pour objectif de créer de nouvelles applications à forte valeur ajoutée basée sur la valorisation de co-produits de l'industrie du bois, et plus

particulièrement des noeuds, pour la production d'ingrédients et de cosmétiques naturels anti-âge intégrant une activité anti-pollution. En effet, l'industrie de première transformation du bois génère chaque année un grand nombre de déchets, aujourd'hui peu valorisés ou valorisés vers des marchés de faible valeur ajoutée. Les noeuds sont connus pour renfermer des quantités importantes de composés extractibles pouvant être potentiellement valorisés, directement ou après modification, dans le domaine de la cosmétique notamment grâce à leurs propriétés antioxydantes ou antibactériennes. Le vieillissement de la peau (derme et épiderme) serait dû au stress oxydatif, qui implique des molécules réactives liées à l'oxygène

(Reactive Oxygen Species - ROS) et qui endommagent l'ADN ou encore les composants des membranes cellulaires. Ce ne sont cependant pas des composés nocifs, puisque nous en avons besoin lors de la communication cellulaire. Ils le deviennent lorsqu'ils sont produits en excès et que l'équilibre dans l'organisme est rompu. Les antioxydants exogènes sont alors précieux pour aider notre organisme à lutter contre les radicaux libres. L'autre rôle des antioxydants en cosmétiques est de protéger la formule du produit et ainsi éviter son oxydation. Il y a une forte demande pour les conservateurs naturels, en raison des effets secondaires des parabènes utilisés récemment rapportés. De plus, dans un contexte d'une volonté de simplification des formulations cosmétiques, il est également nécessaire de mettre au point des composés polyfonctionnels. Cette étude porte sur la valorisation des noeuds d'essences résineuses fournis par des industries de première transformation du bois. Elle est divisée en deux grands axes : d'une part, l'analyse phytochimique des extraits, l'optimisation de l'extraction et de la purification des molécules d'intérêt et, d'autre part, leur modification chimique permettant d'apporter des propriétés supplémentaires. Enfin, pour déterminer leur intérêt en cosmétique les propriétés biologiques et physico-chimiques des extractibles, bruts ou modifiés, sont étudiés.

8. What processes lie behind the thermophilization and β -diversity change of understory communities? Uncovering hidden patterns of thermophilization and β -diversity change (SILVA)

Jeremy Borderieux

Climate change is already reshuffling forest communities toward an increase in warm-adapted species, at the expense of cold-adapted species, the so-called thermophilization. Thermophilization has been widely studied as an increment of the mean climatic optimum (in $^{\circ}$ C) of species present in a given community. However, such measures overlook the demographic dynamics at play, extinction of present species and colonization of new species, and the impact of these dynamics on landscape-scale diversity, β -diversity.

We used the French National Forest Inventory to pair geographically close floristic plots done ten years apart. This balanced pairing across the territory is then used to infer thermophilization and β - diversity changes at the Sylvoecoregion scale, a unit that divides French forests based on homogenous soil and climate condition. We then partitioned both of these changes into four components; extinction and colorization, of cold and warm-adapted species. We also assessed the relationship between those components and climate.

We found a significant thermophilization of 0.10° C/decades, driven almost exclusively by the extinction of cold-adapted species. Thermophilization was faster in Mediterranean climates (up to 0.25° C/decades), due to a greater contribution of extinction. While we didn't observe a significant β -diversity change at the Sylvoecoregion scale, we found a previously hidden pattern with opposite directions: Extinction of cold-adapted species and colonization of warm-adapted species are detrimental to β -diversity.

Those results confirm previous thermophilization studies but complement them by integrating the community dynamic underlying them. An extinction-driven thermophilization challenges the idea that thermophilization is a signal of community adaptation, as it reflects the dieback of already present species. We did not observe significant β -diversity change, but the detrimental effect on β -diversity of cold-adapted species loss reveals that rare species are as much impacted as common species, as a result, warm-adapted species may become more common.

Climate change have visible impact on ecosystems, such as thermophilisation but also subtle or slow changes, such as β -diversity change. By studying hidden pattern, we can deepen our understanding of the processes at play, as well as detects early indicators of visible changes.