

JJC



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I-SITE CLERMONT
Clermont Auvergne Project



International Research Centre
Sustainable agroecosystems

INSTITUT SCIENCES
DE LA VIE, SANTÉ,
AGRONOMIE,
ENVIRONNEMENT
UNIVERSITÉ
Clermont Auvergne

Graduate Track:
*Changing
Environnements*



Place	Date	Session	Theme	Time	Speaker	Description		
Cézeaux	03/04/2023	8h45-9h15: Arrival						
		9h15-9h30: Opening remarks						
		Oral P.	Plant sequencing and data science	9h30-9h50	Javier BELINCHON MORENO	Genetic and functional diversity of the NLRome resistome in melon		
				9h50-10h10	Arnaud LIEHRMANN	DiffSegR: An RNA-Seq Data Driven Method for Differential Expression Analysis using Changepoint Detection		
				10h10-10h30	Johan HUNZIKER	Improvement of plant breeding by application of base editing tools in potato		
				10h30-10h50	Clémentine BORRELLI	Optimizing the breeding scheme of new grapevine disease resistant varieties by genomic and phenomic selections		
				10h50-11h00: Coffee break				
				11h00-11h20	Margot CORREA	Identification of transcription factors regulating the stress response in plants <i>via in silico</i> approaches		
				11h20-11h40	Emile MARDOC	Omics data integration using a new workflow and tool: case study on Poplar		
				11h40-12h00	Mamadou SENE	Building a sorghum grain: a transcriptome roadmap targeting protein digestibility		
				12h00-12h20	Mélanie LAVOIGNAT	Data integration to characterize gastric peptides resulting from bread digestion		
		12h30-14h00: Lunch break						
		Centre & Unit presentations		14h00-14h30	Emmanuel HUGO	Presentation of the INRAE centre		
				14h30-15h00	Jérôme SALSE	Presentation of the research unit GDEC		
		Posters		15h15-16h15	Poster Session			
		Oral P.	Plant-environment interactions	16h15-16h30: Coffee break				
				16h30-16h50	Benjamin HUBERT	Identification of defence mechanisms in dormant tomato seeds		
				16h50-17h10	Tiffanie SCANDOLERA	Impact of elevated CO2 levels on viral susceptibility/resistance: Phaseolus vulgaris as a model plant		
				17h10-17h30	Julien LEUENBERGER	Identification of <i>G. pallida</i> resistance haplotypes in potato		
		17h30-19h30: Moving to AppartCity /Free Time						
		19h30-20h30: Visit of Clermont-Ferrand city/Free Time						
		From 20h30 : Restaurant & Entertainment						

Lieu	Date	Session	Theme	Time	Speaker	Description	
Lemptégý	04/04/2023	8h00-9h00: Transportation to Lemptégý, Coffee, Welcoming					
		Oral P.	Plant diversity	9h00-9h20	Brice CHARLEUX	Involvement of TALEs in Xanthomonas campestris pv. campestris pathogenicity in cauliflower	
				9h20-9h40	Marie SERRIE	Designing resilient stone fruit trees via integrative phenotyping in low phytosanitary input orchards and association genetics	
				9h40-10h00	Maria Victoria GARCIA HERNANDEZ	Résistance au black rot (Guignardia bidwellii) chez la vigne et les espèces apparentées	
				10h00-10h20	Florent CORNET	Caractérisation fonctionnelle des protéines PsMAX1 dans la voie de biosynthèse des strigolactones chez le pois	
		10h20-10h40: Coffee break					
		10h40-13h00: Visit of the Volcano Lemptégý					
		13h00-15h00: Lunch break					
		Posters		15h00-16h00	Poster Session		
		Oral P.	Plant diversity	16h00-16h20	Baptiste IMBERT	Translational genomics for identifying biological functions linked to pulses stress resistance and adaptation to agroecological cropping systems	
				16h20-16h40	Christelle GINOT	Identification of traits of interest for Thinopyrum intermedium in different agronomic contexts: Combination of agronomic, ecophysiological and participatory approaches	
				16h40-17h00	Clovis PAWULA	Rosa gallica L. and other Gallica roses, origins and role in the genesis of cultivated roses	
				17h00-17h20	Mariana TISCARENO-ANDRADE	Searching for regulatory components of meiotic chromosomal movements during Prophase I	
		18h00: Return to Clermont-Ferrand					
		From 20h30: Dinner & Entertainment					

Lieu	Date	Theme	Time	Speaker	Description	
INRAE Croûel	05/04/2023	9h00: Arrival & Welcoming				
		Workshops	9h00-10h30	Ludovic BONHOMME	Pursuing with public research (intervention of researchers and teachers in the field of plant biology)	
				Hervé DUBORJAL	Pursuing with private research (intervention of researchers in the field of plant biology)	
				Annaïg BOUGUENNEC	Popularization: how to communicate research to the general public?	
				Valérie LEGUÉ	Data management: How to effectively manage thesis data?	
				Catherine RAVEL	Research and environmental issues: what place to give to environmental questions in your thesis?	
				Karen VERGNOL-REMONT	Living well with your thesis: what relationship with your supervisor?	
		BAP presentation	10h30-11h00	Norbert ROLLAND	Presentation of the INRAE department BAP	
		Visits of plateforms	11h00-12h30	Jacques LE GOUIS	Pheno3C facilities: high throughput phenotyping platform in the field under climatic constraints (water and CO2 control)	
				Charles PONCET	Gentyane platform: high-throughput genotyping and sequencing platform	
				Clément DEBITON	Biological Resource Center (CRB), which stores 27,000 straw cereal accessions	
				Guillaume CHARRIER	PIAF lab: tools and facilities for the physiological study of trees	
		12h40: Closing remarks & lunch				

Family name: BELINCHON-MORENO

First name: Javier

Year of PhD: 1st

Institute-city: INRAE GAFL, Avignon

Exploring the genetic and functional diversity of the NLRome/resistome in melon (*Cucumis melo* L.) using Nanopore Adaptive sampling (NAS).

Nucleotide-binding-site-leucine-rich-repeat (NLR) disease resistance genes encode the most important and one of the most variable family of plant resistance proteins. The characterization of their complete collection in a species (NLRome) would facilitate the creation of varieties with a very broad spectrum of resistance. However, although many efforts have been made, the specific role of each NLR gene remains largely unknown in melon. This lack of information is mainly due to their complex genomic structure and organization, frequently arranged in clusters that include a high level of presence-absence polymorphisms.

These structural singularities of NLR clusters make sequencing methods using short reads often ineffective to decipher them. To solve this problem, Nanopore Adaptive Sampling (NAS) is a cost- and labor-effective approach that provides long reads and selectively sequences pre-defined target regions.

Our project pursues the implementation of NAS for the sequencing and characterization of the NLRome of melon using ≈ 140 varieties. In a first step, we demonstrated the performance (= increase of enrichment) of the NAS approach against whole genome sequencing for the sequencing and assembly of the NLRome in two well-studied melon varieties, obtaining around 30X enrichment. We also assessed the transferability of our design to other varieties, in simplex- and multiplex-sequencing experiments. In the next steps of the project, we plan to continue exploring the possibilities of the NAS approach with a final goal of performing genetic association studies using the NLRome sequence variability of the ≈ 140 lines and their phenotypic variability against a group of selected pests and pathogens.

Family name: LIEHRMANN

First name: Arnaud

Year of PhD: 3rd

Institute-city: INRAE IPS2, Gif-sur-Yvette

DiffSegR: An RNA-Seq data driven method for differential expression analysis using changepoint detection.

To fully understand gene regulation, it is necessary to have a thorough comprehension of the transcriptome along with the enzymatic and RNA-binding activities that shape it. While many RNA-Seq-based tools have been developed to analyze the transcriptome, most only consider the abundance of sequencing reads along annotated patterns (such as genes). These annotations are typically incomplete and often lead to errors in the differential expression analysis.

To address this issue we present DiffSegR, an R package that enables the systematic discovery of transcriptomic differences between two biological conditions using RNA-Seq data. DiffSegR does not require prior annotations and uses a multiple changepoints detection algorithm to identify the boundaries of differentially expressed regions in the per-base log2 fold change.

In a few minutes of computation with DiffSegR we could rightfully predict the role of chloroplast ribonuclease MiniIII in rRNA maturation and chloroplast ribonuclease PNPase in (3'/5')-degradation of rRNA, mRNA, and tRNA precursor as well as the introns accumulation.

We believe DiffSegR will benefit the biologists working on transcriptomics as it allows access to information from a layer of the transcriptome that is not addressed by the classical differential expression analysis pipelines widely used today.

Family name: HUNZIKER

First name: Johan

Year of PhD: 5th

Institute-city: INRAE IGEPP, Ploudaniel

Improvement of plant breeding by application of Prime Editing in potato.

Plant scientists have rapidly adapted new genome editing tools for crop improvement, in complementation of traditional breeding approaches. The current technical challenge is to efficiently induce precise and predictable targeted point mutations valuable for crop breeding purposes. Several base editing tools such as Cytidine base editors (CBEs) and Adenosine base edition (ABEs) CRISPR/Cas9 derived tools have demonstrated their utility in plant breeding. However, such technologies proved their limits with non-predictable mutations introduced, resulting in difficulty to transfer known mutations between cultivars or species. Moreover, stable genomic integration of CRISPR/Cas9 components through *Agrobacterium*-mediated transformation faces rejection by regulators and customers for European field production. Trans-DNA might be difficult to eliminate, especially in vegetatively propagated plants such as potato. New technologies of precise base editing currently in development such as the Prime Editing, support the possibility to obtain expected DNA substitution in different species, without having to characterize large populations of mutants. In combination, new varieties can be created without requesting bacteria DNA insertion, with established protoplast transfection and regeneration protocols, associated with the use of Development Regulator (DR) genes, to stimulate the process of regeneration. In this project, the objective will be to transfer well-characterized mutations involved in PVY resistance, isolated from diploid Solanaceae such as pepper and tomato, to the tetraploid potato, by Prime Editing. For this, new tools for improving the efficiency of genome editing and plant regeneration are in development, with the aim to accelerate the transfer of interesting mutations based on translational biology.

Family name: BORRELLI

First name: Clémentine

Year of PhD: 1st

Institute-city: INRAE SVQV, Colmar

Optimizing the breeding scheme of new grapevine disease resistant varieties by genomic and phenomic selections.

French viticulture is facing the consequences of climate change and the challenge of reducing the use of phytosanitary products. One of the solutions to this scenario is the development of new grapevine disease resistant varieties with good agronomic and wine quality performances. The INRAE-ResDur breeding program was developed with this objective and has already released nine grape varieties carrying polygenic resistance against downy and powdery mildews. However, new approaches are needed to reduce the time of the breeding cycle, which is now about 15 years. The classical marker assisted selection (MAS) is difficult to be applied on traits with complex genetic architecture such as the ones related to wine quality and agronomic performance. The development and the use of breeding value prediction models such as those based on molecular markers (genomic selection) or infra-red spectra (phenomic selection) can help in targeting complex traits while reducing the length of breeding cycles. This nevertheless needs the establishment of a reference panel and statistical models in order to select the best individuals and improve the annual genetic gain in breeding programs. The aim of the thesis is to take advantage of the breeding plant material created during the INRAE-ResDur program with approximately 1100 genotypes which were phenotyped during multiple years and at multiple locations, in order to study the genetic architecture of agronomic and wine quality traits, to create breeding value prediction models based on molecular markers or NIRS and including prior information on genetic architecture of important traits and validate the model accuracy by making a comparison with the phenotypic data of the ResDur program.

Family name: CORREA

First name: Margot

Year of PhD: 3rd

Institute-city: INRAE IPS2, Gif-sur-Yvette

Identification des facteurs de transcription régulant la réponse aux stress chez les plantes via des approches in silico.

Identifier des facteurs de transcription qui agissent ensemble pour réguler la transcription des gènes reste un challenge. L'équipe Réseaux Génomiques de l'IPS2 s'attache notamment à comprendre les interactions entre les facteurs de transcription (FT) et leurs gènes cibles lors de la réponse au stress. Suite à un stress, la plante va mettre en place une voie de signalisation pour s'adapter et survivre. Cette voie de signalisation entraîne l'activation de FT qui en se fixant sur des séquences d'ADN spécifiques vont activer ou non la transcription des gènes. Ces courtes séquences d'ADN (4 à 15 bases) ou motifs cis-régulateurs, sont situés majoritairement dans le promoteur proximal des gènes. La méthode PLMdetect (Preferentially Located Motif detection) a été développée pour identifier des motifs surreprésentés (PLM) dans les régions proximales des gènes (Bernard et al., 2010). Avec cette méthode j'ai identifié 93 motifs d'ADN spécifiques de cette réponse aux stress chez Arabidopsis (93 PLMs stress). J'ai caractérisé ces PLM stress à l'aide des données expérimentales (ChIP-seq, DAP-seq) et je montre ainsi qu'on peut lier un PLM stress à un FT. Actuellement, je finalise la méthode qui permettra d'identifier les FT candidats susceptibles de se lier à ces PLM stress. Dans un futur proche, à partir de ces PLMs stress, je souhaite identifier des couples de motifs qui sont simultanément présents dans les régions proximales des gènes afin de déterminer les facteurs de transcription co-actifs.

Bernard V. et al., 2010. TC-motifs at the TATA-box expected position in plant genes: a novel class of motifs involved in the transcription regulation. BMC Genomics
Une application de PLMdetect : Rozière J. et al., 2022. A comprehensive map of preferentially located motifs reveals distinct proximal cis-regulatory sequences in plants. Frontiers in Plant Science

Family name: MARDOC

First name: Emile

Year of PhD: 3rd

Institute-city: INRAE GDEC, Clermont-Ferrand

Omics data integration using a new workflow and tool: case study on poplar.

With the recent improvements in high throughput technologies, the number of omics data produced has soared. Hence, a new challenge is to exploit this large amount of data to answer biological questions unsolvable by uni/bi-variate methods, by analyzing the different omics (e.g. genomic, transcriptomic, methylomic, metabolomic...) data simultaneously: this multivariate approach is called (multi-) omics integration.

In this context, I present `cimDiablo_v2`, a new function based on `cimDiablo` from the R package `mixOmics`, which denoises the data while integrating them. After a brief introduction on omics integration, I will detail how `cimDiablo_v2` works and how to use it to answer different biological questions, with concrete applications on poplar datasets.

Family name: SENE

First name: Mamadou

Year of PhD: 1st

Institute-city: INRAE AGAP, Montpellier

Construction du grain de sorgho : une feuille de route transcriptomique ciblant la teneur et digestibilité des protéines.

Le sorgho est la 5^{ème} céréale mondiale pour la production de grains. Sa capacité à faire face aux contraintes biotiques et abiotiques pourrait en faire une source protéique végétale d'avenir dans un contexte de changement climatique. Cependant, la faible digestibilité des protéines de réserves du grain (appelées kafirines) par les protéases gastro-intestinales, potentiellement due aux structures de stockage de ces protéines (corps protéiques), représente un frein important à sa plus large utilisation pour l'alimentation animale et humaine. Les mécanismes moléculaires sous-jacents à la mise en place et aux modifications des corps protéiques contenant les kafirines sont encore peu connus. Dans ce contexte, notre étude vise à décrypter les mécanismes moléculaires impliqués dans la régulation de la teneur et de la digestibilité des protéines de réserve du grain de sorgho. Une approche transcriptomique a été réalisée sur les grains au cours de leur développement, suivie d'une analyse de réseaux de gènes (RG) avec le logiciel WGCNA. En parallèle, la teneur en protéine des grains et leur digestibilité ont été mesurées. Les résultats issus des RG ont été exploités pour détecter de nouveaux facteurs de transcription (FT) potentiellement régulateurs des mécanismes de mise en place de réserves protéiques. Nous avons identifié la présence en réseaux des gènes de kafirines avec des orthologues de FT déjà connus chez d'autres espèces. De nouveaux FT ont été également retrouvés dans ces réseaux. Pour la suite de ces travaux, nous envisageons de tester le rôle de ces FT par un système simplifié de surexpression dans des protoplastes.

Mots clés : Sorgho, grain, protéine, digestibilité, mécanismes moléculaires

Family name: LAVOIGNAT

First name: Mélanie

Year of PhD: 3rd

Institute-city: INRAE GDEC, Clermont-Ferrand

Data integration to characterize gastric peptides resulting from bread digestion.

Bread wheat (*Triticum aestivum*) is mainly consumed as bread, after milling grains into flour and breadmaking. Grain proteins are composed of albumins-globulins and storage proteins, the gliadins and glutenins. Thanks to their ability to form the gluten, storage proteins are indispensable for processing. Nevertheless, because wheat grain proteins are partially resistant to gastrointestinal enzymes, they are associated to several health issues as gluten related disorders. Therefore, protein digestibility is a key factor to consider. From our previous work, flour protein content and composition appear to influence bread protein digestibility.

In this context, our study aims at (i) clarifying whether peptides from bread digestates are linked to flour protein composition and (ii) identifying peptides discriminating accessions with a high or low bread protein digestibility.

Grains from 17 cultivars grown at two locations were milled into flour to measure protein content and composition. For each cultivar, breads were baked according to a standardized method, then digested in vitro with a dynamic gastrointestinal system TIM-1. After two hours of digestion (mid-digestion), the peptides from the gastric compartment were characterized and bread protein digestibility (%) was estimated based on a nitrogen balance.

Five and four cultivars were classified as having a high and low digestibility, respectively. Varieties with a digestibility close to the median were withdrawn. Applying multivariate analyses (multiblock Partial Least Squares-Discriminant Analysis - PLS-DA), we show that digested peptides would be linked to flour protein composition and that some peptides discriminate high and low digest cultivars.

Family name: HUBERT

First name: Benjamin

Year of PhD: 2nd

Institute-city: INRAE IRHS, Beaucouzé (Angers)

Identification of defence mechanisms in dormant tomato seeds.

Dormancy is an adaptive strategy that allows seeds to persist in the soil in the face of (a)biotic stresses to ensure germination and dispersal of the species. Our team's work has shown that imbibition of dormant *Medicago truncatula* seeds leads to the activation of a defence response (Bolingue et al., 2010). However, the defence pathways and their regulation during dormancy remain poorly understood. Here, we set out to identify the molecular pathways underlying defense activation in dormant tomato (*Solanum lycopersicum*) seeds. As a measure of seed defence, a method was developed that determines the antimicrobial activity in exudate from imbibing seeds against *Alternaria brassicicola* using nephelometry. Exudates from imbibing seeds that are primary dormant or in which dormancy has been reinduced by a heat treatment show antimicrobial activity. In contrast, during imbibition of seeds that are germinating, this activity is not detectable and only becomes evident at the seedling stage. Using the accessions of the tomato MAGIC population, we identified a large variation in the level of antimicrobial activity in the dormant seed exudates and this activity appears to be tissue specific. Current research focusses on dissecting the molecular pathways in dormant tomato seeds using transcriptomic and metabolomic analyses. These results will contribute to a better understanding how seeds defend themselves and will serve to develop new strategies of seed-borne pathogens management.

Family name: SCANDOLERA

First name: Tiffanie

Year of PhD: 2nd

Institute-city: INRAE IPS2, Gif-sur-Yvette

Impact of elevated CO₂ levels on viral susceptibility/resistance: *Phaseolus vulgaris* as a model plant.

One major concern of this century is the impact of climate change, notably on crop cultures. Experts of climate change have forecast an increase in atmospheric CO₂ level from 400 $\mu\text{L.L}^{-1}$ in 2014 to $\sim 1000 \mu\text{L.L}^{-1}$ in 2100 in the worst predictive scenario, as well as a raise of 3.3-5.7°C in temperature. Plants are directly impacted by these changes as well as pathogen populations including viruses.

In that context, an important question is to what extent the increase in CO₂ will affect plant-virus interactions, whether susceptibility or resistance?

The objective of our work is to study the impact of elevated CO₂ (eCO₂) level on viral susceptibility/resistance using common bean (*Phaseolus vulgaris* L.) as a model plant. We used the *P. vulgaris*/Bean pod mottle virus (BPMV, Comovirus) pathosystem to investigate the impact of eCO₂ on the level of susceptibility/resistance to BPMV using two natural genotypes BAT93 (resistant to BPMV) and Black Valentine (susceptible to BPMV).

In that aim, we analyzed viral titers in plants grown under eCO₂ using RT-qPCR. We also monitored the accumulation of salicylic acid (SA) using HPLC. Moreover, we studied the expression of genes encoding enzymes of the SA pathways, as well as genes encoding PR proteins and components of the RNA silencing pathway. All gene expressions were performed using RT-qPCR.

Our preliminary results show that both genotypes are more resistant/less susceptible under eCO₂ and this seems correlated with a higher accumulation of SA and with an increased expression of defense genes.

Family name: LEUENBERGER

First name: Julien

Year of PhD: 2nd

Institute-city: INRAE IGEPP, Plouarzel

Identification de d'haplotypes de résistance à *G.pallida* sur pomme de terre tetraploide.

Potato is a critical crop for global food security. However, it is vulnerable to various pests and diseases, including the quarantine cyst nematode *Globodera pallida*. Developing effective and durable resistant varieties is a crucial issue for the integrated management of nematodes, since the withdrawal of nematicides. To address this challenge, we conducted a Genome-Wide Association Study to identify key genomic regions associated with resistance to *G. pallida* in potato.

An INRAE potato panel, containing 276 pre-breeding clones carrying introgressions from different wild potato species, was studied. Using different types of molecular data (GBS, SolCAP SNP array, CAPS), a genotyping matrix of the panel was obtained with 27,147 filtered markers. *G. pallida* resistance was evaluated on all the clones, using Foot et al (1977) method. A genome-wide association study was conducted using a Multi-Locus Mixed Model and haplotypes were defined in intervals around markers showing a significant effect.

The GWAS analysis identified three main genomic regions, on chromosome 5 in a known hot-spot resistant region, on chromosome 9 in the region of the *GpaVI* locus previously unknown in this panel, and a newly detected region on chromosome 7 near a locus conferring resistance to the cyst nematode *G. rostochiensis*. Marker haplotypes significantly associated with resistance were identified in linkage disequilibrium blocks underlying all these regions.

These genomic regions can be tracked and combined using haplotype-assisted selection. This study provides valuable markers and sources of resistance for breeding of effective and durable resistant varieties against this quarantine pest.

Family name: CHARLEUX

First name: Brice

Year of PhD: 2nd

Institute-city: INRAE LIPME, Castanet Tolosan (Toulouse)

Involvement of TALEs in *Xanthomonas campestris* pv. *campestris* pathogenicity in cauliflower.

The *Xanthomonas* bacteria cause numerous diseases affecting commercially significant crop plants. Most *Xanthomonas* species translocate Transcription Activator-Like Effectors (TALEs) into plant cells using their type III secretion system. TALEs are a distinct class of bacterial effector proteins that serve as eukaryotic transcription factors to increase the expression of particular plant genes known as susceptibility genes (or S genes) for the bacteria benefit. According to recent studies, *Xanthomonas* species-specific disease management techniques can be developed by logically manipulating the host S genes.

Amongst the S genes targeted by *Xanthomonas* species are the SWEET genes, which encode for sugar transporters that are key to susceptibility in rice, cassava and citrus. It is speculated that the upregulation of these transporters in response to TALEs accelerates disease development by increasing the amount of nutrients supplied to pathogens and/or by contributing to sugar signaling for disease resistance.

We identified the repertoire of *Xanthomonas* tal genes (TALome) from *Xanthomonas campestris* pv. *campestris* (Xcc), the causal agent of blackrot disease in Brassicaceae. To determine the transcriptome modifications induced by the Xcc TALEs, we did RNAseq experiments in cauliflower (*Brassica oleracea*). Interestingly, we demonstrated that Tal12a contributes to Xcc virulence on cauliflower, possibly by inducing the expression of the BoSWEET14b sugar transporter. We will present our latest results on the contribution of SWEET genes to susceptibility in cauliflower.

Family name: SERRIE

First name: Marie

Year of PhD: 2nd

Institute-city: INRAE GAFL, Avignon

Designing resilient stone fruit trees via integrative phenotyping in low phytosanitary input orchards and association genetics.

Fruit tree orchards are under attack from multiple pests and diseases incurring for significant damages and economic losses. The multiple sprays of pesticides throughout the entire life of the orchards is still a common practice to control these pathogens. It is therefore urgent to find alternative and durable solutions to reduce this massive use of phytosanitary products. We think that a promising but still misunderstood and under-exploited solution could rely on the breeding of resilient trees. My PhD project aims to define and characterize the phenotypic and genetic components of resilience in fruit trees.

To do so, two collections of two major stone fruit tree species (apricot and peach) have been deployed since 2017 in five contrasted environments in the south east of France and are maintained under low phytosanitary conditions. Through an integrative monitoring of the multiple symptoms of the diverse pests and diseases over the years, we will first decipher the different strategies and phenotypic manifestations of resilience. Thanks to the calculation of a “global biotic damage index” we already identified a few accessions which would be better adapted to low phytosanitary conditions. Different models of genome wide association studies (GWAS) will then be tested in order to identify genetic markers related to resistance or tolerance to these pathogens.

This will be a first step to try to understand the genetic architecture of the components of resilience in order to contribute in the long term to make resilience a concrete goal for breeders.

Family name: GARCIA HERNANDEZ

First name: Maria Victoria

Year of PhD: 1st

Institute-city: INRAE SVQV, Colmar

Resistance to black rot (*Guignardia bidwellii*) in grapevine and related species.

The deployment of new grapevine varieties resistant to downy and powdery mildew, intended to promote sustainable viticulture, allows the reduction in phytosanitary treatments. This new disease management leads, however, to the emergence of fungal diseases previously considered as secondary, the most detrimental of which is black rot, caused by the ascomycete *Guignardia bidwellii*. Black rot can infect all the green tissues of the vines during the growing season, and severe attacks can lead to significant losses of up to 80% of the harvest. Developing grapevine varieties that integrate also black rot resistance is therefore essential to progress in the agroecological transition in viticulture.

The main questions that will be addressed in this thesis are the availability of sources of resistance and the identification of new black rot resistance factors that can be used in grapevine breeding. In addition, the relationship between resistance to infection in leaves vs. bunches, the expression of black rot symptoms in the presence of downy or powdery mildew, and the effect of phenology on the severity of symptoms will be assessed.

In order to reach the different objectives, the thesis will be structured in three phases. A consolidation of data regarding the level of resistance to black rot from potential sources, followed by an analysis of biparental progenies to identify new resistance factors. The last phase will include an analysis of the expression of black rot symptoms under field conditions, considering the phenology stage, the organ affected and the presence of downy or powdery mildew.

Family name: CORNET

First name: Florent

Year of PhD: 4th

Institute-city: INRAE IJPB, Versailles

Caractérisation fonctionnelle des protéines PsMAX1 dans la voie de biosynthèse des strigolactones chez le pois.

Les strigolactones (SLs) sont des hormones végétales qui répriment la ramification. Elles sont également connues pour leurs activités au sein de la rhizosphère, comme la stimulation de la symbiose avec les champignons endomycorhiziens. Plus d'une trentaine de SLs naturelles aux structures diverses ont été caractérisées. Synthétisées en cocktail, elles sont souvent spécifiques de familles végétales. À partir de trans- β -carotènes, les premières étapes de la voie de biosynthèse sont communes à toutes les plantes et constituent le "CORE PATHWAY". Les voies de biosynthèse se diversifient ensuite et varient entre les espèces. La protéine MAX1 et ces homologues (CYP711A) ont un rôle central dans la diversification des SLs. Ces protéines CYP450 ont été bien décrites chez *Arabidopsis*, la tomate et le riz, mais leurs fonctions chez les autres espèces sont peu décrites. Mon projet se concentre sur les deux homologues PsMAX1 du pois chez lequel des simples et doubles mutants ont été obtenus. Nous effectuons des tests enzymatiques *in vitro* et des quantifications de SL *in planta* pour étudier les fonctions biochimiques et biologiques des PsMAX1. De manière inattendue, nos mutants simples et doubles ne présentent pas de phénotype d'hyperramification caractéristique des mutants SLs. Cependant, ces *Psmax1s* sont déficients en SLs canoniques présentent chez le pois, SLs qui seraient impliqués préférentiellement ou spécifiquement dans la signalisation rhizosphérique. Nos résultats suggèrent ainsi que le signal inhibiteur de ramification n'est pas l'une des SLs canonique décrites jusqu'à présent chez le pois mais que le véritable signal inhibiteur de ramification reste à découvrir.

Family name: IMBERT

First name: Baptiste

Year of PhD: 2nd

Institute-city: INRAE Agroécologie, Dijon

Translational genomics for identifying biological functions linked to pulses stress resistance and adaptation to agroecological cropping systems.

Legumes, and especially pulses, are an important source of protein for food and feed, and are appreciated for their positive impact on the “one health”. However, their unstable yields and their susceptibility to biotic and abiotic stresses highlight the need for varietal improvement in order to increase the cultivated areas and productivity. With the advent of sequencing technologies, a large pool of genetic and -omics resources, heterogeneous at the inter- and intra-species scale, is emerging. Thus, it is important to capitalize on these scattered heterogeneous data to develop translational research to boost breeding projects and crop diversification.

To meet this need, we undertook the development of OrthoLegKB, a NoSQL graph-based database using Neo4j, dedicated for translational research in pulses. Starting from genome sequences and annotation files, we inferred orthologous relationships between genes, and proposed associated syntenic blocks between the chromosomes of four cultivated crops, namely *Pisum sativum*, *Vicia faba*, *Lens culinaris* and *Vigna radiata*, and model legume *Medicago truncatula*. Available information on quantitative trait loci (QTL) for multiple traits are being included as well as expression data. The proposed modeling was evaluated in studying the conservation of a flowering-promoting gene. The main achievements as well as remaining challenges and perspectives will be discussed.

Family name: GINOT

First name: Christelle

Year of PhD: 1st

Institute-city: INRAE GDEC, Clermont-Ferrand

Defining ideotypes for *Thinopyrum intermedium* in different agronomic contexts: combination of participatory and experimental approaches.

Given the major role played by cereals in the world food security, improving the sustainability of grain production systems is a central challenge. The introduction of perennial grains such as intermediate wheatgrass (*Thinopyrum intermedium*) has been proposed as an alternative to annual cereals to diversify cropping systems and improve soil fertility. *T.intermedium* is grown for a number of production goals, agro-environmental and socio-economic services, in various pedo-climatic conditions and cropping systems. In this context, we question the ability of one crop variety to meet all these targets and conditions. Our aim is to design population ideotypes, considering the canopy performance of the crop, that would be suited to these different situations. Participatory approaches are used to consider stakeholders' objectives and constraints and define a set of ordered specifications for the crop. Moreover, agronomic and ecophysiological experiments are used to understand the effects of density of crop performance, at both plant level and per unit area, considering the trade-off between plant individual fitness and canopy performance. Stakeholders' specifications and knowledge about the crop functioning are then used to identify traits of interests in different agronomic contexts. The design of population ideotypes with a combination of approaches is proposed as a method of participatory breeding for developing suitable perennial grain varieties.

Family name: PAWULA

First name: Clovis

Year of PhD: 3rd

Institute-city: INRAE IRHS, Beaucouzé (Angers)

Rosa gallica L. and other Gallica roses, origins and role in the genesis of cultivated roses.

Rosa gallica L., a heterozygous tetraploid rose species, is protected in France and historically used in traditional medicine. This species was one of the first among the genus *Rosa* to be bred by humans in Europe. It is a principal ancestor of perfume and ornamental cultivars. However, the genetic diversity of this mainly European species has only been investigated at country or regional scales.

For the first time, we characterised *R. gallica* wild diversity over its natural range. In addition, we assessed the relationship between wild and cultivated *R. gallica*. This brought insights into the origins of French wild populations.

We gathered roses samples from France and abroad, collected in 244 wild populations, including 127 populations from 14 other countries. In parallel, microsatellite markers genotyped by sequencing and adapted to the genus *Rosa* were developed. These are able to reveal not only variations in SSR repeats but also SNPs and InDels. Subsequently, 1520 individuals regrouping wild and cultivated *R. gallica* and about 140 individuals of other *Rosa* species were genotyped. Using classical allele scoring and innovative k-mer based analyses, we showed that France is a crossroads for the diversity of *R. gallica*. The determination of clonal relationships confirmed the fertility of some individuals. This highlighted that few old varieties were traditionally grown in multiple locations in France and have survived successfully since their abandonment.

Our findings are already being used to assist the conservation of French populations, especially in the Centre-Val-de-Loire.

Family name: TISCARENO-ANDRADE

First name: Mariana

Year of PhD: 1st

Institute-city: INRAE IJPB, Versailles

Searching for regulatory components of meiotic chromosomal movements during Prophase I.

Meiosis is a cell division that produces the haploid cells at the origin of the gametes. This cell division is also essential, in most sexually reproducing eukaryotes, to introduce genetic variation from one generation to the other. This genetic variation is largely due to Crossing Over (CO) formation, which are genetic interchanges between homologous chromosomes. These COs are generated by the recombination process during the meiotic Prophase I. During this process, spectacular chromosome dynamics can be observed: the homologous chromosome recognition (pairing), their synapsis (polymerization of the synaptonemal complex), telomeres attachment and clustering to the nuclear envelop and finally drastic chromosome mobility. So far it is not well known in plants which are the molecular mechanisms that are driving these dynamics and how far they are regulating the recombination process. By live imaging we could achieve for the first time the characterization of the rapid chromosome movements at early stages of Prophase I in male meiocytes of *Arabidopsis thaliana* and we identified several main regulators of these movements. In addition, by 3D immunolocalization of nuclear envelop and chromosome markers, we achieved 3D reconstruction of the whole meiotic nuclei at different stage of development. In this way, it was possible to obtain a high-resolution localization pattern of some of these regulators during Prophase I and to more fully elucidate their role in chromosome dynamics.

Family name: BERNARDINO

First name: Jerome Monroe

Year of PhD: 1st

Institute-city: INRAE IRHS, Beaucouzé (Angers)

Demonstration of the role of AdPKS7 and AdPKS8 in aldaulactone production in the carrot necrotrophic pathogen, *Alternaria dauci*.

Quantitative disease resistance in necrotrophic pathosystems involves various mechanisms, including chemical warfare. *Alternaria dauci*, a necrotrophic fungus, synthesizes toxins conferring pathogenicity to its carrot host. Aldaulactone, a phytotoxic benzenediol lactone from *A. dauci*, has been shown to be central in both the pathogenicity of *A. dauci* and carrot partial resistance toward the fungus. Secondary metabolite biosynthesis is thus of prime importance in this interaction. Previous findings identified two candidate polyketide synthase genes – AdPKS7 and AdPKS8 in cluster 8 – as responsible for aldaulactone backbone biosynthesis. However, demonstrating the involvement of these genes in aldaulactone biosynthesis requires a more comprehensive approach, such as loss-of-function experiments. Here, we present the first successful attempt at transforming *A. dauci*. Using homologous recombination and a hygromycin resistance cassette, we produced *A. dauci* knockout mutants for two essential domains (acyltransferase and ketosynthase) of both PKS genes. We then analyzed the transformants' ability to produce aldaulactone through HPLC. Our results show an abolished aldaulactone production in all transformed strains when compared to the wild-type. Taken altogether, our findings strongly suggest that gene cluster 8 harboring AdPKS7 and AdPKS8 is responsible for aldaulactone biosynthesis. Supplementary approaches such as in planta pathogenicity tests and recombinant aldaulactone production in *Pichia pastoris* are underway.

Family name: BACHELET

First name: Fanélie

Year of PhD: 2nd

Institute-city: INRAE Agroécologie, Dijon

Caractérisation du transporteur de sulfate vacuolaire SULTR4 chez le pois : rôle dans l'établissement de la composition protéique des graines

La contribution du sulfate vacuolaire à l'élaboration du rendement et de la qualité des graines de pois est étudiée en ciblant l'unique gène SULTR4 de pois qui code un transporteur permettant l'efflux de sulfate de la vacuole vers le cytosol. Ce transporteur a été caractérisé par une analyse phylogénétique de sa séquence, une simulation de sa structure 3D, et une étude de sa localisation intra-cellulaire. Un criblage d'une population TILLING (Targeting Induced Local Lesions IN Genomes) a permis d'identifier cinq mutants *sultr4*, dont le phénotype en condition de carence en soufre a été étudié. Deux mutants pénalisés dans leur rendement en graines ont été retenus pour une étude fonctionnelle. Cette étude a révélé que la composition protéique de leurs graines est modifiée même en condition non limitante en soufre : la quantité relative d'albumines PA1, protéines très riches en acides aminés soufrés, est plus faible. Des études d'expression ont montré que le gène SULTR4 s'exprime lors du développement des graines de pois : il pourrait donc fournir du sulfate à l'intérieur même des graines. Les graines des mutants *sultr4* accumulent la même quantité de soufre que le type sauvage, mais leur quantité de sulfate est plus élevée ; ce qui suggère une utilisation limitée du sulfate dans les graines de ces mutants. Pour analyser plus finement l'impact de ces deux mutations, la structure 3D du transporteur SULTR4 porteur des mutations a été simulée et la stabilité des ARNm et de la protéine sont en cours d'étude.

Family name: BANOUH

First name: Meriem

Year of PhD: 2nd

Institute-city: INRAE GDEC, Clermont-Ferrand

Low impact of polyploidization on transcriptome in synthetic allohexaploid wheat.

Bread wheat is a recent allohexaploid (genomic constitution AABBDD) that emerged through a hybridization between tetraploid *Triticum turgidum* (AABB) and diploid *Aegilops tauschii* (DD) less than 10,000 years ago. The hexaploidization can be re-created artificially, producing synthetic wheat that has been used to study immediate genomic responses to polyploidization. The scale of the consequences of polyploidization, and their mechanism of establishment, remain uncertain.

Here we sampled several synthetic wheats from alternative parental genotypes and reciprocal crosses, and examined transcriptomes from two different tissues and successive generations. We did not detect any massive reprogramming in gene expression, with only around 1% of expressed genes showing significant differences compared to their lower-ploidy parents. Most of this differential expression is located on the D subgenome, without consistency in the direction of the expression change. Homoeolog expression bias in synthetic wheat is similar to the pattern observed in the parents. Both differential expression and homoeolog bias are tissue-specific. While up to three families of transposable elements became upregulated in wheat synthetics, their position and distance are not significantly associated with expression changes in proximal genes

While only a few genes change their expression pattern after polyploidization, they can be involved in agronomically important pathways. Alternative AB parents can lead to opposite changes on D located genes, which is relevant for harnessing new diversity in wheat breeding. Tissue specificity of the polyploidization-triggered expression changes indicates the remodelling of transcriptomes in synthetic wheat is plastic and likely caused by regulome interactions rather than permanent changes. We discuss the pitfalls of transcriptomic comparisons across ploidy levels that can inflate the deregulation signal.

Transcriptomic response to polyploidization in synthetic AABBDD wheat is modest and much lower than some previous estimates. Homoeolog expression bias in wheat allohexaploids is mostly attributed to parental legacy, with polyploidy having a mild balancing effect.

Family name: CHIR

First name: Laurine

Year of PhD: 1st

Institute-city: INRAE LEPSE, Montpellier

Physiological determinants of genetic variability in response to high temperatures in grapevines.

Mediterranean viticulture is particularly threatened by climate change. The average rise in temperature leads to disturbances in the functioning of the grapevine, its development and the composition of the grapes. The impact of extreme heat episodes, however, remains poorly understood. In particular, temperature peaks are accompanied by irreversible burns on leaves and bunches with differences in sensitivity between grape varieties. We have already detected regions of the genome involved in these variations of sensitivity in grapevines. Different processes may be involved, including disruption of water transport in the leaves or necrosis induced by oxidative stress. The objective of my work will be to investigate the main mechanisms that determine these burns in order to better identify their genetic determinants. On the one hand, we will study candidate genes underlying the causal polymorphisms previously identified through genetic and bioinformatics approaches in grapevine, and the homologous candidate test in *Arabidopsis*. On the other hand, we will deploy a mechanistic approach to decipher the respective contributions of hydraulic disruption and oxidative stress in the development of symptoms, using an ecophysiological characterization on grapevine leaves. This project will strengthen our understanding of plant responses to high heat events and their link to water relations within plants, a major issue in improving plant response to multiple abiotic stresses.

Family name: COLOMBO

First name: Michel

Year of PhD: 3rd

Institute-city: INRAE AGAP, Montpellier

Evolution of cultivar mixture's associated genetic effects in modern durum wheat germplasm.

Mixed cropping has been suggested an efficient approach to realize an agro-ecological transition. Yet current breeding programs neglect biological interactions between cultivar mixtures especially competitive interaction. One way to account for biological interactions is to measure the general mixing ability: the additive contribution of a cultivar to mixture performance. The general mixing ability can be further decomposed into a direct genetic effect (DGE, effect of a genotype on himself) and an indirect genetic effect (IGE, effect of a genotype on the neighboring plants). DGE and IGE enables to characterize genotype behavior within mixture and when related to functional traits to describe biological interaction functions. Characterizing the relative contribution of IGE, DGE and GMA as well as the evolution of these genetic terms since the green revolution in different level of plant-plant interactions might be interesting to establish future breeding strategy adapted to mixture performance. In this talk we characterize the relative contribution of IGE and DGE to GMA in a field experiment for several yield components. Then to infer biological interactive function at play in our mixtures we explore the relation between functional traits (specific leaf area, plant height and heading date) and IGE, DGE and GMA. Last, we proposed to study the evolution of IGE, DGE, and GMA since the green revolution of the 60's. These results are important to promote breeding programs adapted to varietal mixture.

Family name: DROUAULT

First name: Justine

Year of PhD: 2nd

Institute-city: INRAE LEPSE, Montpellier

Reconsidering Photoperiod Sensitivity for Maize Adaptation to Climate Change.

Flowering time, fundamental to the fitness of crop species, is controlled by integrated networks of external (environment) and internal (plant) signals. Originating from the tropics, historical maize was initially flowering under short photoperiod ($\sim <13$), while long-day conditions caused photoperiod sensitivity (PS) resulting in delayed flowering time and poor fitness. Maize adaptation to temperate environments (long-days), by selection on PS variations, followed by breeding on narrow pools of diversity, has reshaped the phenotypic space for flowering time regulation. This has reduced temperate maize's plasticity which putatively constrains its adaption to future environments. We hypothesize that reintroducing partial PS in temperate breeding pools could expand the phenotypic space and benefit maize adaptedness. However, to exploit the potential of PS, its genetics regulation and ecophysiological response has to be better understood. The PS response will be explored at the genotypic and haplotypic scales across a photoperiod gradient using existing datasets. Separately, the results of interaction between tropical PS alleles and temperate background will be evaluated on flowering time and yield related traits, using NILs that consists in temperate lines containing PS haplotypes. Thus, the allelic variation underlying PS and the potential pleiotropic effect of those alleles will be highlighted. Ultimately, this will be used to parametrize and validate a crop growth model and simulate PS response in temperate maize under current and future climatic scenarios. The model's outcomes will serve to assess whether PS is expected to improve maize productivity and/or yield stability today and in the future.

Family name: FONTAINE

First name: Florent

Year of PhD: 2nd

Institute-city: INRAE IJPB, Versailles

Functional and Structural study of lipid droplet addressing domains for production and extractability of recombinant/heterologous proteins

Lipid droplets (LDs), organelles with a neutral core and a phospholipid monolayer, are found from yeast to plant (Chapman et al., 2019; Ischebek et al., 2020). Originating from the endoplasmic reticulum (ER), they contain numerous proteins on their surface, coming either from the ER or the cytosol (Renne et al., 2020; Pyc et al., 2021). LDs are also back on the screen with new plant biotech companies which are using them to easily purify their protein of interests (POI). To do so, they use recombinant oleosins, the major proteins found on seed LDs, to anchor POI on LD surface (Nykiforuk et al., 2006). Yet, multiple questions are still on hold. Are there some common structural determinants that target proteins to LD? Is it possible to improve POI targeting by using other protein than oleosin?

To answer these questions, different approaches are used. First of all, we perform microscopy screening in planta of a collection of proteins and peptides described as located on LDs in various organisms. Second, these proteins and peptides are analysed in silico to determine their folding. The objective is to determine if a link between LD localisation efficiency and protein folding could be done. This work will hopefully help to identify a robust targeting domain to plant LDs and also for POI purification.

Family name: FREY

First name: Caroline

Year of PhD: 1st

Institute-city: INRAE GDEC, Clermont-Ferrand

Control of molecular and physiological mechanisms by the Stb16q resistance gene in wheat.

Septoria Tritici Blotch (STB), a wheat disease caused by the fungus *Zymoseptoria tritici*, is one of the most treated diseases in the world. To decrease its impact on crops, there are other means known as genetic resistance. The genes involved in this resistance are called Stb genes. Today, the resistance mechanisms of these genes are not well known. There are 22 known major resistance genes, and recently Stb16q has been cloned. This gene codes for a protein of the CRK (cysteine-rich receptor like kinase) family and confers a strong resistance to *Z. tritici*. STB16 is a receptor protein with two extracellular copies of DUF26 domain, a transmembrane domain and an intracellular kinase domain. Stb16q confers a full broad-spectrum resistance because of its involvement in the closure of stomata. To better understand the mechanisms mediated by Stb16q, we will try to identify the role of stomata in STB16-mediated resistance to STB by looking at the expression of the Stb16q gene in wheat leaves, and analyze the molecular modifications induced by STB16 at the stomatal level. In addition, we will study the signals activating the resistance mechanism mediated by Stb16q by trying to find the lineage of the extracellular domain of the protein.

Family name: ILYAS

First name: Amina

Year of PhD: 1st

Institute-city: INRAE IPS2, Gif-sur-Yvette

Beneficial effect of *Enterobacter* sp. SA187 under elevated CO₂ conditions in *Arabidopsis thaliana*.

The atmospheric concentration of CO₂ is constantly and rapidly increasing and this is expected to provide a fertilization effect on plant productivity. However, this effect is only observed initially because plants acclimate to these conditions upon longterm exposure. This is believed to be due to lower photosynthetic activity, sink strength and nitrogen (N) content. In this context, the interaction of *Arabidopsis thaliana* and the beneficial bacteria *Enterobacter* sp. SA187, known to increase plant abiotic stress through activation of the Ethylene signaling pathway, was studied to observe its potential of improving plant development by avoiding the photosynthetic acclimation to elevated CO₂. A significant increase in plant biomass (up to 51%) has been observed through fresh weight phenotyping, along with a restored C/N ratio similar to ambient conditions. Transcriptomic analyses highlighted that *Enterobacter* sp. SA187 inoculation seems to alleviate primary metabolism inhibition in response to eCO₂ and activates the SA and Ethylene phytohormonal pathways, confirmed by RT-qPCR analyses. The confirmation of these hormonal pathways is still to be confirmed through functional characterization. More aspects are still to be explored to validate the transcriptomic interpretation. Concluding from initial results so far, establishing the SA187 beneficial microbe-plant interaction appears to be a good strategy for improving plant growth and development under elevated CO₂ conditions.

Family name: LAPORTE

First name: Antoine

Year of PhD: 2nd

Institute-city: INRAE BFP, Bordeaux

Study of the metabolism of the Pinot Noir on several Climats of Burgundy.

In this thesis, I am trying to determine the metabolic differences in the grapes between several plots of vines in burgundy, from few days after flowering to the harvest date. I'm using whole genome metabolic networks and multi-omics approaches such as targeted and untargeted metabolomics, transcriptomics and possibly genomics.

To do so, I'm sampling whole berries on the plots throughout all the growing and maturation process until harvest.

I am then generating data by targeting and quantifying metabolites of interest on the HiTMe platform (Bordeaux Metabolome). I'm also doing untargeted metabolomics (LC-MS) to help build a precise metabolic network for each of the plots, by acquiring a better idea of the biomass generated by this species.

I will also extract RNA for each of the plots at each of the sampling dates to generate constraint-based models.

Then, by comparing fluxes generated in those networks with the help of bioinformatics tools, I'll try to isolate and characterize unique metabolic behaviors inside the plots.

Family name: MAIN

First name: Oscar

Year of PhD: 2nd

Institute-city: INRAE IJPB, Versailles

Drier and smaller but not necessarily less productive: can agronomic losses caused by water stress in French forage maize be compensated by an increase in quality?

With dwindling global freshwater supplies and increasing water stress, agriculture is coming under increasing pressure to reduce water use while maximising use efficiency. Plant breeding requires high analytical capabilities. For this reason, near-infrared spectroscopy (NIRS) has been used to develop prediction equations for whole-plant samples, particularly for predicting dry matter digestibility, which has a major impact on the energy value of forage maize hybrids and is required for inclusion in the official French catalogue. Although the historical NIRS equations have long been used routinely in seed company breeding programmes, they do not predict all variables with the same accuracy. In addition, little is known about how accurate their predictions are under different water stress environments. Here, we examined the effects of water stress and stress intensity on agronomic, biochemical, and NIRS predictions in a set of 13 modern forage maize hybrids under four different environmental conditions, including two water stress levels. We compared the reliability of NIRS predictions for basic forage quality traits obtained using the historical NIRS equations and the new equations we recently developed. We found that NIRS predictions were affected to varying degrees by environmental conditions. We also showed that forage yield gradually decreased as a function of water stress, while both dry matter and cell wall digestibilities gradually increased. By combining forage yield and dry matter digestibility, we were able to quantify digestible yield and identify varieties with different strategies, raising the exciting possibility that there are still important potential selection targets to identify.

Family name: MARSAN

First name: Laurie

Year of PhD: 2nd

Institute-city: INRAE SVQV, Colmar

Characterization of the Rpv2 locus, conferring total resistance against downy mildew in grapevine, and of its avirulence factor.

Since the 19th century, European viticulture is confronted to several diseases imported from North America through contaminated grapevine materials. Downy mildew is a major grapevine disease that, in absence of chemical protection, causes serious yield loss and reduced berry quality. Thus, viticulture is a major consumer of phytochemicals which have negative effects on environment and health. An alternative to fungicides is using varieties carrying resistance factors to pathogens. *Vitis rotundifolia*, an American grapevine species, is totally resistant to downy mildew due to the presence of several resistance loci. Genetic mapping of resistance loci in *V. rotundifolia* cv. Trayshed highlighted two major loci named Rpv1 and Rpv2, for resistance to *Plasmopara viticola*, the oomycete responsible of downy mildew. Rpv2, which induces total resistance to downy mildew by stopping infection at the primary hypha state, is localized on chromosome 18 in an interval containing a cluster of TIR-NB-LRR (TNL) genes. Functional annotation of the genes in the interval and their expression analysis during the infectious cycle allowed the identification of two TNLs as strong candidate genes for Rpv2 resistance. Functional validation to determine which one of the two candidate genes confers the resistance associated to Rpv2 is in progress. At the same time experiments to determine the avirulence gene associated to Rpv2 are ongoing. Knowledge of the Rpv2 resistance mechanism and of its potential to stand pathogen evolution will permit us to evaluate whether this gene would be suitable for integrating future breeding programs for new resistant grapevine varieties.

Family name: MHAMDI

First name: Oumayma

Year of PhD: 1st

Institute-city: INRAE IJPB, Versailles

The identification of the genes and metabolites controlling plant-plant interaction (Allelopathy).

Allelopathy is defined as the effect of a donor plant on another receiver plant through the liberation of metabolites from the roots, which affect, positively or negatively, the growth of the nearby plants (germination, growth of seedlings...). The SAS team has developed a protocol called Plant-Soil feedback (PSF) to study the plant-plant interactions, especially the allelopathic effect, and distinguish it from the competition for nutrients, light, and water. Using PSF and the model plant *Arabidopsis thaliana*, Genome-Wide Association Studies (GWAS) were performed to explore the genetic basis of allelopathy. The GWAS experiment has identified multiple loci/genes potentially involved in allelopathy. After functional annotation, the BGLU30 gene, a member of the glycoside hydrolyze family involved in glucosinolate catabolism, was selected for further studies. The glucosinolates and derived molecules are specialized metabolites identified as defense compounds and allelochemicals. To validate this candidate gene, we will use genetics and molecular biology approaches. Mutants of glucosinolates biosynthesis pathway (such as the quadruple mutant *myb28 myb29cyp79b2 cyp79b3 =gkO*) and mutants of transport (such as the double mutant *gtr1 gtr2*) are used to validate our results. Furthermore, we will use analytical chemistry approaches to analyze the root exudates composition and isolate the allelopathic compounds and determine their identity.

Family name: MOBARAK

First name: Térance

Year of PhD: 2nd

Institute-city: INRAE IRHS, Beaucouzé (Angers)

Nitrogen status modifies apple response to PRI.

Plant resistance inducers, or PRIs, are an alternative to pesticides but their efficiency highly depends on plants' physiological status. We focus our research on the possible interactions between apple (*Malus domestica*) nitrogen status and ASM, a functional salicylate analog known to partially protect apple against various pests. Our hypothesis is that nitrogen limitation could improve, diversify and/or perpetuate ASM efficiency against these pests.

In order to test this hypothesis, apple seedlings were grown in greenhouses under controlled sub-irrigation. 4 weeks old apple seedlings were subjected to short (9 days), or long (12 days) nitrogen limitation before N resupply, or kept under sufficient nitrogen as control. Plants were then treated or not with ASM just before N resupply or 24h after. Two days after treatment, young leaves were inoculated with *Erwinia amylovora* the causal agent of fire blight, a bacterial disease. Samples were collected before and four days after ASM treatment on not inoculated plants and two days after treatment on inoculated plants for molecular analysis. This experiment has been repeated three times.

First results show a strong interaction between plants' nitrogen status and ASM efficiency with N-limited plants twice as much protected against fire blight than control plants. The expression level of 29 well known defence genes usually responding to ASM treatment (qPCR analysis) could not explain the different phenotypes. Considering that, analysis of transcriptome and metabolome (primary and secondary metabolites) are currently underway in order to explain the strong protection phenotype observed with ASM x N-limited plants. In parallel, experiments with *Venturia inaequalis* (the causal agent of apple scab, a fungal disease) are being conducted to verify whether a similar interaction between ASM treatment and nitrogen limitation also exists and results in a disease-reducing phenotype with this pathogen.

Family name: MOREL

First name: Marine

Year of PhD: 1st

Institute-city: INRAE EGFV, Bordeaux

Rootstock regulation of scion mineral status: the relationship between rootstock parentage and petiole mineral content.

Grapevine is grown grafted in most of the world largely because of Phylloxera. Rootstocks not only provide tolerance to Phylloxera, but also ensure the supply of water and mineral nutrients to the scion. Rootstocks are an important means of adaptation to environmental conditions. To aid this adaptation, we can exploit the large diversity of rootstocks used worldwide.

Vitis vinifera cvs. Cabernet-Sauvignon, Pinot noir, Syrah and Ugni blanc were grafted onto 55 different rootstock genotypes and planted in a vineyard as three replicates of five plants. In 2020 and 2021, petioles were collected in the cluster zone with six replicates per combination. Petiolar concentrations of 13 mineral elements were determined at veraison. In 2020, Mg deficiency severity was visually scored on each plant; a visual score between 0 and 3 was assigned. Rootstocks were also grouped according to their parentage when at least 50 % of a genetic background is present.

Rootstock genotype showed a significant influence on the petiole mineral element composition. Rootstock effect explained from 8 % for Al to 42 % for S of the variance. The genetic background Vitis riparia seems to increase the probability of P or Mg deficiency. The petiole Mg measurements we related to Mg deficiency symptoms, but severity of Mg deficiency symptoms varied depending on the scion cultivar. The differences in mineral status conferred by rootstocks did not show significant correlations with vigor or fertility.

The study of such an experimental design has shown that the rootstock effect is higher than the scion effect for the large majority of mineral elements. The joint evaluation of magnesium levels by petiole analyzes and observations of the intensity of deficiency symptoms underlines for the first time the variability of the thresholds of satisfactory mineral nutrition. Fertilization choices must therefore take the rootstock into account.

Keywords : Vitis, climate change, plant material, mineral status, genetic background, rootstock x cultivar

Family name: PENG

First name: Shuang

Year of PhD: 2nd

Institute-city: INRAE IJPB, Versailles

Integrated Proteomics and Metabolomics Emphasized Nutritional Benefits Driven by Lentil (*Lens culinaris* Medik) Seed Germination.

The lentil plant is a small, upright plant that grows to a height of about 2-3 feet. It produce seeds that are high in protein and dietary fiber. They are a good source of several important nutrients, including iron, folate, and vitamin B1. Lentil seeds are used in a variety of dishes, including soups, stews, and salads, and they can also be ground into flour and used in baked goods. Lentil plants are an important food crop in many parts of the world and are grown on a large scale for both human and livestock consumption.

The genome of *Lens culinaris* has been sequenced and analyzed by researchers. The lentil genome is relatively small, with an estimated size of around 502 million base pairs. It contains around 25,000 protein-coding genes, which are responsible for the production of proteins in the plant.

Analysis of the lentil genome has provided insights into the genetic basis of important agronomic traits in this crop, such as seed size and shape, seed coat color, and disease resistance. It has also helped researchers to identify genes that are involved in the synthesis of important nutrients and compounds in lentil seeds, such as protein, fiber, and antioxidants.

Overall, the analysis of the lentil genome has helped to improve our understanding of the biology of this important food crop and has the potential to contribute to the development of improved varieties of lentil in the future.

Family name: SACCARAM

First name: Chandrodhay

Year of PhD: 2nd

Institute-city: INRAE IJPB, Versailles

Seed exudation - driving force for microbial growth and interaction in the spermoosphere?

Seeds play a fundamental role in the reproduction and dispersal of higher plants. Seed quality is a key element for agricultural performance. The seed vigour is defined by rapid and homogeneous germination, right seedling establishment, and the capability to adapt to biotic and abiotic constraints imposed by the environment. The seed coats and endosperm are favourable structures for the protection of the embryo throughout its development, its conservation till germination and seedling emergence. During germination *sensu stricto*, from water uptake to radicle protrusion, the seed releases exudates on its tegument surface consisting of complex mixtures of organic and inorganic molecules. While many studies previously described exudates from plant tissues (e.g. root, stem, leaves), the scientific literature is quite limited concerning exudates from germinating seeds. Plant exudates have been described to have functional properties such as antimicrobial or antioxidant activities. Our preliminary works indicated that the germinating seed releases many metabolites, peptides, proteins and small RNAs during the early period of imbibition. The objective of the thesis will address the characterization of the germinating seeds exudates and the identification of molecules displaying bioactivities on the biotic environment and / or on the seed embryo to improve its vigour. The results of this work will be included in prospective studies on research of new products derived from plants for seed treatment applications (e.g. coating, pelleting or priming).

Family name: SAIDI

First name: Somia

Year of PhD: 2nd

Institute-city: INRAE URGI, Versailles

Characterization of transposable elements in Pangenomes.

The impact of Transposable Elements (TEs) in a genome is explored by searching for insertions events. Individuals (or accessions) of the same species independently undergo TE insertions causing inter-individual genetic variability.

This variability between individuals is the basis of the natural selection that leads to an increased adaptation of individuals to their environment. A way to search for the potential role of TEs in host adaptation is through a pangenomic approach. The TE pangenome is described by (i) TE insertions present in all individuals of the species (core-genome), (ii) insertions present only among a subset of individuals (dispensable-genome) or (iii) ecogenome when the individuals share the same environment, and finally (iv) insertions specific to an individual.

Current pangenome analysis methods are based on the alignment of reads from different accessions of the species to an assembled reference genome. But, the advent of the third-generation sequencing makes now possible to approach this question on several assembled genomes of the same species.

I will present a new pipeline which identify the TEs in pangenome compartments from several assembled genomes. There is therefore no dependency on a reference genome. This new pipeline identifies TE copies shared by a group of individuals.

This pipeline has been tested on *Brachypodium distachyon* and compared with results from Minigraph, a pan-genome graph method to identify sequence variations.

Family name: VIOLET

First name: Martha

Year of PhD: 1st

Institute-city: INRAE LEPSE, Montpellier

Multi-criteria optimization of cover crop and management practices of grapevine resistant varieties in the context of climate change.

This study is dedicated to the analysis of the nutritional (C, N) and water functioning of grapevine subjected to contrasted genotype x environment x management interactions. The main output is to define new multi-factorial 'ideotypes' in the context of climate change and biodiversity loss.

The genotype effect will be analysed investigating the behaviour of four varieties resistant to fungal diseases. The environment will be modified with different inter row practices: spontaneous or sown cover crop, bare soils. 'Soil cover' treatments will be coupled with contrasted crop load conditions, through modifications in foliage height and pruning practices. Measurements will involve medium to high throughput phenotyping methods (near infra-red spectrometry, leaf fluorescence, imaging, T-Lidar). Together with these measurement, more precise ones at the organ scale on water (transpiration), nitrogen, carbon (photosynthesis) will be performed. A special focus will be given on the impact of management practices on carbon and nitrogen allocation between vegetative and reproductive organs in order to determine the best equilibrium between water consumption, yield and sugar accumulation in berries. A functional structural modelling approach will be used to simulate the performances of each system. This modelling approach will allow computing light interception, water use and carbon production at the plant and vineyard scales. This will help defining the relative importance of the different types of traits (architectural/functional) on the multi-performances of the system. Finally, it is expected to define some 'proxy' that could be quickly estimated and used to assess the 'performances' of the whole agro-ecosystem.

Family name: ZHAN

First name: Xi

Year of PhD: 2nd

Institute-city: INRAE EGFV, Bordeaux

Role of reactive oxygen species (ROS) and the redox system in grapevine berries exposed to heat stress.

In the context of climate change, it is becoming increasingly urgent to better understand the mechanisms mobilized by grapevine to cope with high temperatures and recurrent heat waves. Our team has produced a large set of molecular data to better define the impact of high temperatures on the development and metabolism of grapevine berries. It seems that the redox status could strongly modulate the consequences of heat stress (HS) on fruit development and its potential quality at harvest. Therefore, my PhD study aims to determine the impact of reactive oxygen species (ROS) and the redox system on the metabolic behavior of grapevine berries exposed to HS.

Specifically, the aims of my study are 1) to generate metabolomic profiling of berries exposed to HS in both microvine and grapevine fruit cuttings; 2) to conduct a functional characterization of candidate genes associated with the HS/redox balance by exploiting the CRISPR/Cas9 genome editing technology on microvine; and 3) to evaluate the thermotolerance of 51 grapevine cultivars by assessing heat injury with OJIP method.

Family name: ADAMIK

First name: Larissa

Year of PhD: 2nd

Institute-city: INRAE GDEC, Clermont-Ferrand

Deciphering the specific responses of wheat to Fusarium head blight under fluctuating irrigation.

Fusarium head blight (FHB), primarily caused by *Fusarium graminearum*, is a prominent disease of small grain cereals impacting both yield and grain quality. In wheat, substantial efforts are still produced to find efficient and sustainable genetic resistances, especially focusing on susceptibility factors as new and original sources. However, despite the important role of the abiotic environment in the establishment of the disease, only few FHB studies have already included the role of future climate scenarios on the expression of such molecular determinants driving the infection process. Focusing on fluctuating water regimes, this work initiates the phenotypic and molecular characterization of the FHB-induced responses in bread wheat facing soil water deficit just before the time of infection.

The setup of mild drought conditions of different durations during spike development demonstrated a specific and non-additive response to the combination of the constraints. A strong decrease of FHB symptoms in plants suffering from previous reduced watering during at least 6 days have been observed compared to well-watered plants. No impact from strain aggressiveness has been reported but the longer drought conditions were in place, the less symptoms could be observed. Spike samples were taken and will further make possible the examination of the underlying molecular mechanisms specifically set up upon drought as compared to well-watered plants, based on metabolite profiling and large-scale dual-transcriptomic approaches.

Family name: AL BOLBOL

First name: Mohamad

Year of PhD: 1st

Institute-city: INRAE AGAP, Montpellier

Deciphering the molecular mechanisms of gibberellin-mediated flowering control in apple (*Malus domestica* Borkh).

Alternate bearing habit is an annual cyclical change in floral intensity and crop load that leads to important yield losses, affecting the agronomical and economical suitability of fruit production in France, Europe and worldwide. However, only a little is known about its genetic control. In many fruit trees such as apple, alternate bearing is related to fluctuations in flowering induction, and it is believed to be controlled by the phytohormone gibberellin (GA). In the annual model plant *Arabidopsis*, GA acts as a potent promoter of floral induction by negatively regulating DELLA proteins, which in turn inhibit the activity of floral promoter regulatory proteins, such as SQUAMOSA PROMOTER BINDING-LIKE (SPL) transcription factors. Nevertheless, in perennial woody plant species, GA represses floral induction, and DELLA proteins might act as co-activators of unknown floral promoter proteins. By using apple as a tree model, I am going to study the molecular mechanisms by which GA inhibits flowering in apple, a biological process that has a tremendous impact in alternate bearing and crop production.

Family name: BOMSEL

First name: Zoé

Year of PhD: 2nd

Institute-city: INRAE IJPB, Versailles

The interphasic regulation of cortical microtubules for the establishment of plant cell division plane.

The plane of cell division is a crucial element of tissue organization and morphogenesis in plants. Division is regulated by multiscale interactions, among which the cell cycle, the plasma membrane and the cytoskeleton play major roles. During the cell cycle, microtubules (MT) organize into two cortical arrays in interphase (ICMT in G1, to G2, and the PPB in late G2), then in two mitotic arrays.

The preprophase band (PPB) is a pre-mitotic ring-shaped MT array that accurately predicts the site and plane of cell division. The PPB-deficient *trm678* mutant was mostly able to maintain cell division orientation[1], indicating that the plane of division might rather depend on interphasic MT arrays on which the PPB would later align with. To further elucidate the integrative role of interphasic cortical MT arrays in the regulatory pathway of cell division, we propose to produce a spatiotemporal reference framework of plant cell division.

Lines expressing fluorescent markers for several subcellular structures were produced in *Arabidopsis thaliana*. High-resolution confocal imaging of cycling cells will allow for the analysis of MT arrays relative to many parameters, such as the cell cycle, the position of the nucleus, the morphology of the cell and the position of the cells surrounding it. Mutant lines of proteins of the TTP complex[2], an important regulator of MT arrays, were obtained, and will be implemented in this analysis pipeline. This will lead to the determination of stages and transitions, defining the sequential events in an average cycling cell and identifying the critical steps for the establishment of correct plane of division.

[1] Schaefer, Estelle et al. "The preprophase band of microtubules controls the robustness of division orientation in plants." *Science* (New York, N.Y.) vol. 356,6334 (2017): 186-189. doi:10.1126/science.aal3016

[2] Spinner, Lara et al. "A protein phosphatase 2A complex spatially controls plant cell division." *Nature communications* vol. 4 (2013): 1863. doi:10.1038/ncomms2831

Family name: CHERON

First name: Floriane

Year of PhD: 2nd

Institute-city: iGReD, Clermont-Ferrand

The meiotic challenges of having multiple sets of chromosomes.

Meiosis is the cell division that promotes genetic diversity among the offspring. It consists of halving the number of chromosomes sets by two rounds of division. The first round is very specific to meiosis where there is an exchange of DNA between homologous chromosomes via a process called the homologous recombination. In allopolyploids, which result from the hybridization between two closely related species, this process is more complex as allopolyploids possess two or more sets of non-identical homeologous chromosomes. Recombination between homeologous chromosomes can create association of several chromosomes, disrupt the faithful segregation of homologous chromosomes, and leads to the production of aneuploid, genome disorganization in the offspring and reduced fertility. First generation allopolyploids are thought to have been strongly selected to overcome the challenge of controlling meiotic recombination by limiting homeologous recombination to favor recombination between homologous chromosomes, but the molecular mechanisms remain to be elucidated. In the laboratory, we can recreate the allopolyploid species *Arabidopsis suecica* which closely imitate unevolved allopolyploids. By comparing meiosis of both unevolved and evolved allopolyploids of *Arabidopsis suecica*, we show using FISH that, centromere dynamics at early meiosis is altered in unevolved plants. Using genome-wide meiotic crossover frequency and MLH1 foci quantification, we demonstrate a reduction in homologous recombination in unevolved plants. In parallel, through FISH analysis, we observe frequent chromosomes associations belonging to different subgenomes showing an increase of homeologous recombination in unevolved plants. Together, these data reflect a problem in the choice of the recombination partner.

Family name: COQ--ETCHEGARAY

First name: Domitille

Year of PhD: 3rd

Institute-city: INRAE BIOGECO, Bordeaux

Differentiated gene expression response of specialized metabolites-related genes among five modern bread wheat (*Triticum aestivum*) cultivars to the infection of *Fusarium graminearum*.

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2. Univ. Clermont-Auvergne, INRAE, GDEC, F-63000 Clermont-Ferrand, France

Fusarium head blight (FHB) is one of the major diseases of modern bread wheat cultivars causing major quality losses and sanitary risks by the production of mycotoxins in grains. This disease is caused by the infection of wheat grains by pathogens such as *Fusarium graminearum*. The response and resistance of wheat cultivars to the infection by *F. graminearum* have been widely investigated and susceptible and resistant cultivars have been identified. One response of wheat to the infection is an important shift of gene expressions related to defence mechanisms and master regulators of plant response to biotic and abiotic stresses. One of these defence mechanisms is the production of chemical compounds also called specialized metabolites. Our hypothesis is that genetic variation among cultivars results in different gene expression responses to the infection and may contribute to contrasting responses of specialized metabolism pathways. Our goal here was to identify contrasting gene expression responses among cultivars upon infection by *F. graminearum*. Specifically, we studied the response to the infection of *F. graminearum* of five modern wheat cultivars with different levels of susceptibility. We performed RNA-seq analysis and obtained 990 genes (p -value < 0.001) with contrasting expressions responses among cultivars at 72 hours post inoculation. Among those genes, 111 specialized metabolites-related genes were less upregulated upon infection in the two cultivars compared to the three others. Genes with contrasting patterns of expression in response to infection by *F. graminearum* were enriched in genes from specialized metabolite pathways, such as the glutathione metabolic pathway known to contribute to the detoxification of xenobiotics.

Family name: EL GHAZZAL

First name: Zineb

Year of PhD: 1st

Institute-city: INRAE URP3F, Poitiers

The conception of a lucerne variety ideotype for living mulch use.

Living mulch that is defined by the co-culture of a perennial plant species with an annual cash crop, is an option for the diversification of cropping systems. The living mulch provides a soil cover for several years, and has several beneficial services by improving physical, chemical and biological soil properties and reducing weed occurrence. When the perennial species is a legume, such as lucerne, its symbiotic nitrogen fixation capacity may increase the availability of soil nitrogen. Current lucerne varieties have been selected for forage production, they are vigorous and, as a result, there is strong competition with wheat, producing negative effects on wheat yield. The genetic diversity within the *Medicago sativa* species complex is extremely wide, ranging from winter-dormant and creeping accessions to erect, winter-active ones. Our hypothesis is that the large diversity is sufficient to design an alfalfa ideotype adapted to living mulch use. Thirty populations of lucerne, cultivated or wild, belonging to the subspecies *sativa* or *falcata*, diploid or tetraploid, were collected and sown in a split-plot design with the wheat variety "Geny" in four treatments: "pure lucerne", "alfalfa with wheat", "pure wheat" and finally "pure wheat well fertilised". The morphological and phenological traits related to the plant architecture (number of stems, height, leaf surface, growth habit) and the growth dynamics, which have an impact on the competition with the wheat and consequently its yield, are measured. The characteristics that describe the ideotype of a lucerne suitable for living mulch use will be identified.

Family name: IQBAL

First name: Shehyar

Year of PhD: 1st

Institute-city: INRAE UMR1158 UMR Transfrontalière
BioEcoAgro, Estrées-Mons/Laon

**Miscanthus even more beneficial for the environment:
validation of key variables for nitrogen recycling in
Miscanthus sinensis and QTL detection.**

Miscanthus is a perennial grass that belongs to the family Poaceae. Miscanthus x giganteus biomass is an interesting ecological alternative as a source of renewable energy, green chemistry, or bio-sourced products. Its nutrient recycling allows it to have low nitrogen fertilization needs, which would limit environmental impacts. It is an ideotype for biomass production due to its high yield coupled with efficient nitrogen recycling, including strong spring remobilization (J Leroy thesis, 2021). But today, only one genetic background of Miscanthus x giganteus is mostly cultivated in France, which can present a risk at the slightest hazard. Miscanthus sinensis is an interesting alternative to diversify the varietal offer because of a huge genetic variability, good tolerance to abiotic stresses, and high intraspecific and interspecific variability (Zub H and Brancourt-Hulmel). My thesis has a double objective, associated with two complementary aspects related to ecophysiology and genetics: (i) to characterize spring and/or fall remobilization in Miscanthus sinensis and to identify and validate indirect measurement variables, (ii) to evaluate the extent of genetic variability and the genetic parameters of these variables and to identify the chromosomal regions involved. A protocol has been established to analyze the variability of spring and fall remobilizations in Miscanthus sinensis progeny. Characterization begins at the end of winter. We have highlighted potential indicators (indirect measures of variables) and will further validate them this year. We have already observed high genetic variability and heritability for height and we anticipate discovering a similar pattern for nitrogen-related traits.

Family name: LACROIX

First name: Martin

Year of PhD: 1st

Institute-city: INRAE IJPB, Versailles

Relationship between RNA silencing and DNA damage/repair.

Post-transcriptional gene silencing (PTGS) is triggered when double-stranded (ds)RNAs are processed into 21- and 22-nt short interfering (si)RNAs by DICER-LIKE (DCL)2 and 4 enzymes. After loading into ARGONAUTE (AGO) proteins, mainly AGO1, siRNAs guide the cleavage of homologous single-stranded (ss)RNAs. What causes the production of dsRNAs and the activation of PTGS remains a key question. Exogenous threats such as viruses produce dsRNAs during their replication, which could activate PTGS. They also produce large amounts of RNAs that are considered as aberrant by the cell. Aberrant (ab)RNAs usually are destroyed by the RNA quality control (RQC) pathway. However, when RQC is saturated by an excess of abRNAs (for example during virus infection), abRNAs are transformed into dsRNAs by endogenous RNA-dependent RNA polymerases. Transgenes that are strongly expressed under the control of viral promoters also activate PTGS, likely because they produce an excess of abRNAs. In contrast, endogenous protein-coding genes (PCGs) generally do not activate PTGS. However, when RQC is saturated during virus infection or impaired by mutations, PCGs produce siRNAs referred to as *rqc*-siRNAs. In Arabidopsis, 5000 out of the 25000 PCGs are capable of producing *rqc*-siRNAs. What provokes the production of abRNAs that are converted into dsRNAs is not fully understood. Different elements brought us to hypothesize that DNA damage and/or DNA repair mechanisms could have a role in abRNAs production and thus, PTGS triggering. By combining genetic and biomolecular tools, I aimed to analyze the potential role of DNA integrity on siRNA production in Arabidopsis.

Family name: LAMBELIN

First name: Laurine

Year of PhD: 2nd

Institute-city: INRAE IRHS, Beaucouzé (Angers)

Towards the identification of quantitative resistance mechanisms of rose to black spot disease.

Black spot disease, caused by the hemibiotrophic fungus *Diplocarpon rosae*, is one of the main foliar diseases of garden roses. Resistant varieties have proven to be an efficient alternative to the use of fungicides, but the genetic basis of resistance is not well known. Yet, understanding the underlying mechanisms of resistance is essential for an effective and sustainable deployment of resistant varieties. The *Rosa x wichurana* genotype exhibits resistance to black spot disease that is stable across multiple environments and genetic backgrounds. Analysis of an F1 progeny from a cross between the susceptible genotype *Rosa chinensis* 'Old Blush' and *Rosa x wichurana* showed that its resistance results mainly from the combined action of two QTLs, located on chromosomes 3 and 5. Characterizing the interaction of each of these QTLs and their combination with *D. rosae*, at the histological and molecular levels, is crucial to progress towards cloning the underlying genes and identifying markers for their use in breeding. To better understand the action of each QTL and their combination, individuals from the F1 progeny have been selected to present only one of the QTLs or the combination of the two QTLs. Based on their level of resistance/susceptibility and their genotype, some of these individuals have been retained for RNA sequencing and microscopic observations of the first stages of infection. Examining and comparing their reactions at both transcriptomic and histological levels will be useful to identify the mechanisms underlying each QTLs.

Family name: LAPOUS

First name: Romane

Year of PhD: 1st

Institute-city: INRAE IRHS, Beaucouzé (Angers)

Investigation of molecular mechanisms implicated in apple scab quantitative resistance using untargeted metabolic QTL mapping.

Apple scab, caused by the fungus *Venturia inaequalis*, requires more than 20 phytochemical treatments per year in apple orchards. Diversifying the molecular mechanisms involved in genetic resistance in new varieties should contribute to more sustainable pest management. Thus, studying the potential role of secondary metabolites in quantitative disease resistance is a key element in trying to better understand how to combine these different molecular pathways and enhance genetic resistance. To study the genetic control of secondary metabolism in apple leaves, metabolic signals obtained by mass spectrometry coupled to liquid chromatography, have been studied without or after inoculation in a biparental population, which exhibits well-described resistance QTL (rQTL) to apple scab. A new automatic pipeline aiming at detecting QTL has been proposed and led to the identification of 1,026 constitutive metabolic QTL (mQTL) from the analysis of 2,261 metabolic signals. A mQTL hot-spot was found on linkage group (LG) 16, and was shown to be associated with phenolic compounds. In addition, co-localisations between a rQTL and four constitutive mQTL on LG 11 were identified. A new dataset describing 2,158 metabolic signals obtained after fungal infection was recently acquired. Qualitative comparison of the two datasets will allow us to detect metabolic signals specifically induced by the pathogen and then to study their variation among 256 genotypes by mQTL mapping. Further analysis will then open up new avenues on the role of constitutive and induced genetic resistance in domestic apple to address sustainable scab management.

Family name: LOUGMANI

First name: Célia

Year of PhD: 1st

Institute-city: INRAE GQE, Gif-sur-Yvette

Genomic responses to climate change of two emblematic crop stone fruit trees, the apricot tree and the almond tree, and of their wild relatives.

Understanding the processes of adaptation is a major aim in evolutionary ecology. It could help in better understanding how species emerged but also in predicting species responses to global change, and whether or not they will be able to cope with it. Domestication left strong footprints in the genomes and thus provides a good model to study adaptation. Transposable elements are repeated elements that can generate mutations and structural variants, and may be critical for population adaptation and crop domestication. The role of transposable elements in genome modelling during adaptation and domestication have been seldomly studied in long-lived perennial crops. In this PhD project, we will unravel the role of transposable elements during the domestication of two emblematic crop stone fruit trees from the genus *Prunus*, growing in temperate climates: the almond tree and the apricot tree. First, we will reconstruct the domestication histories of the almond tree and the apricot tree. Second, using population genomics, we will investigate whether transposable elements were recruited during the domestication of apricot tree and almond tree. Then, we will predict genomic offset under climate change of cultivated and wild populations of apricot tree. The results of this study will provide a starting point for adapting food production systems to climate change, based on climate scenarios, and will help in better understanding evolutionary processes involved in the domestication and environmental adaptation of perennial crops.

Family name: MONFORT

First name: Manon

Year of PhD: 2nd

Institute-city: INRAE IJPB, Versailles

FLOREA: FLOWering Regulators gene Editing to improve soybean regional Adaptability..

Europe has a deficit in sources of protein and rely on imports. One objective of the European commission is to increase domestic protein production and European soybeans can play a critical role to reach this goal.

Soybean is a short-day plant with photoperiod sensitivity which limits the natural geographical distribution to a narrow range of latitude. Breeding efforts are underway to expand soybean cultivation. However, there is limited availability of soybean varieties adapted to European conditions and adaptation remains a central issue.

Adapting soybean to long-day conditions at higher latitudes requires early flowering and a reduction of sensitivity to photoperiod. Flowering control involves many genes which makes a classic breeding approach hazardous and time consuming. Here, we propose to use new genome editing techniques to generate genetic diversity on the gene network involved in soybean flowering time to produce new, earlier soybean varieties that would allow it to be grown in a larger part of Europe.

The approach consists in the simultaneous mutagenesis of twelve soybean flowering repressors by CRISPR-Cas9, followed by phenotypic screening without a priori. The mutant lines obtained will exhibit a diversity of allelic combinations on the targeted genes. After a screening based on the criterion of flowering time, the causal mutations in the lines of interest will be identified by sequencing. Plants showing an earlier flowering time phenotype while keeping important agronomic traits will be selected and tested in field to identify varieties that can be used in more varied European environments than current varieties.

Family name: PATIN

First name: Etienne

Year of PhD: 1st

Institute-city: INRAE EGFV, Bordeaux

Genetic diversity of root traits in *Vitis* sp. and potential for grapevine rootstock varietal innovation.

The European vine *Vitis vinifera* is mostly grafted in most of the world, largely because of its sensitivity to *Phylloxera*. Rootstocks currently used today are accessions or hybrids of American *Vitis* species naturally tolerant to *Phylloxera*. They were selected more than 100 years ago and have a narrow genetic background. In the current global change context, new threats may limit the sustainability and production in viticulture, in particular, drought is one of the most important risks in this context. The *Vitis* genus has many wild species around the world but few of them are mainly used as rootstocks. Consequently, there is a need to explore the diversity of grapevines to identify genetic resources with resistance to drought. For this purpose, the aim of my thesis is to characterize the response to drought of wild *Vitis* sp. used as rootstocks for their use in breeding programs. The root system will be phenotyped using image and metabolomic analyses. With this information we aim to establish a link between some metabolomic profiles and complex root traits related to drought resistance of wild *Vitis* sp. with the obtained results we aim to improve the high-throughput phenotyping of roots and pre-breeding methods.

Family name: PETIOT

First name: Valentine

Year of PhD: 1st

Institute-city: iGReD, Clermont-Ferrand

Role of SRS2 helicase in mitotic and meiotic recombination in *Arabidopsis thaliana*.

Homologous recombination (HR) is a set of universally conserved DNA break repair mechanisms, essential for maintaining genomic integrity and ensuring genetic diversity. In somatic cells, HR is used to repair DNA breaks caused by environmental stresses and endogenous factors, such as stalled and collapsed replication forks. In meiotic cells, HR plays a key mechanical role by ensuring faithful chromosome segregation and shapes genetic diversity through the formation of crossover (CO) and non-crossover recombination events. Recombination has thus important consequences for fertility, genome evolution, adaptability to a changing environment, and is key to plant and animal breeding and agronomical applications.

A crucial step of HR is the search for and invasion of, an intact homologous DNA molecule that will be used as a template to repair the break. This step is catalyzed by the RAD51 and DMC1 recombinases along with several cofactors (such as DNA helicases). One such factor is SRS2, an helicase identified in *Saccharomyces cerevisiae* that plays a key role in recombination by controlling the assembly of RAD51 at DNA breaks. Although SRS2 is conserved in plants, its function has not been fully characterized. This study aims to characterize the role of SRS2 in the repair of DNA breaks during mitosis and meiosis in the model plant *Arabidopsis thaliana*. We will present our data showing that AtSRS2 has no major role during mitotic recombination but that its absence increases the strength of crossover interference (the mechanism that prevents a crossover recombination events to occur next to another) during meiosis.

Family name: PREVOT

First name: Cécile

Year of PhD: 2nd

Institute-city: INRAE EGFV, Bordeaux

Unraveling the complexity of high temperature tolerance by characterizing key players of heat stress response in grapevine.

In the context of climate change, it is becoming urgent to clarify the mechanisms used by the vine to cope with extreme heatwaves. Our team has produced an important set of molecular data to better define the impact of high temperatures on grape development. The interest is to select and characterize genes that may play a key role in the adaptation processes involved in grapevine facing high temperature. This will contribute to a better understanding of the molecular mechanisms involved in the heat stress response in a perennial plant species of agronomic interest and should ultimately lead to the selection of resilience biomarkers that could be used to improve and/or select grapevine cultivars better adapted to the ongoing climate change.

Family name: PULLARA

First name: Sara

Year of PhD: 1st

Institute-city: INRAE LPCV, Grenoble

Chloroplast biogenesis: towards the role of localized translation in Arabidopsis.

Chloroplasts are a major component of plant cells. Until recently, all nuclear-encoded proteins destined to chloroplast were believed to possess an N-terminal and cleavable chloroplast targeting peptide, and to engage the TOC/TIC machinery. However, recent studies have revealed that alternative routes also exists and identified a series of nuclear-encoded proteins imported via such pathways. Recent proteomic studies, conducted by our team (Bouchnak et al., Mol Cell Proteomics 2019), identified a list of cyto-ribosomal subunits associated to chloroplasts, thus suggesting that localized translation might occur at the chloroplast surface. We were recently able to isolate plastid-associated cyto-ribosomes and to decipher their composition when compared to purified whole cell cyto-ribosomes. Interestingly, these plastid-associated cyto-ribosomes contain a few non cyto-ribosomal proteins which might participate to the control of localized translation at the chloroplast surface. These non cyto-ribosomal proteins were first fused to GFP to analyze their subcellular location. Then, we isolated Arabidopsis knock-out mutants affected in the expression of two of these specific proteins and initiated their phenotypical characterization. Finally, with the aim to identify the nature of the nuclear-encoded mRNAs that are translated by these chloroplast-associated cyto-ribosomes, the identification of mRNAs trapped within these plastid-associated cyto-ribosomes was performed. Surprisingly, very few of these mRNAs code for chloroplast proteins.

Family name: SALOMON

First name: Sarah

Year of PhD: 2nd

Institute-city: INRAE LPCV, Grenoble

Betaine Lipids overexpression in higher plants.

During evolution, photosynthetic organisms have developed specific strategies to respond to the fluctuations of their environment. Among these various external stresses, nutrient scarcity like phosphate starvation is frequently observed and leads to drastic changes in the lipid metabolism. Phosphorus is a fundamental nutrient for cell physiology and is highly remobilized during such situation to maintain growth and development. During phosphate deprivation, phospholipids (PL) are degraded to provide phosphate resource for the cell. These lipids are then replaced by non-phosphorus lipids to maintain membrane integrity. In plants, digalactosyldiacylglycerol (DGDG) synthesized in chloroplast envelope is known to replace the phospholipid phosphatidylcholine (PC) in extra-plastidial membranes (Jouhet et al., 2004). Another remodelling mechanism can occur in algae, a diverse group of photosynthetic organisms living in ocean and freshwaters. In these organisms, the PC degraded during phosphate starvation is replaced by betaine lipids (BL), a class of glycerolipid present only in algae, some fungi and lower plants. It has been shown that BL can entirely replace PC in extra-plastidial membranes in marine algae (Abida et al., 2015) whereas DGDG can only replace 20% of the PC in terrestrial plants. Thus, BL seem to be a better substitute for PC during phosphate starvation and their disappearance in higher plant raises many questions. The goal of this project is to have a better understanding of BL roles and evolution in plant kingdom by overexpressing BL in plant.

Family name: SIGURET

First name: Clea

Year of PhD: 3rd

Institute-city: INRAE GDEC, Clermont-Ferrand

Consequences of polyploidy events on angiosperm genome evolution.

In plants, polyploidy is a widespread phenomenon with major consequences on genome evolution. It is considered as a major process of speciation, diversification and ecological adaptation of plants. Polyploidy is the inherited condition of having more than two complete sets of chromosomes resulting from whole genome duplication. Polyploidization is quite common in the plant kingdom. About half of current flowering plants are polyploids.

The aim of this study is to understand the polyploidy impact on evolution and conservation of flowering plants by comparing the genomes of 84 angiosperm species. Does polyploidization drive sequence diversity? Can we determine divergence time and the age of polyploidization events from sequence diversity?

Here, using comparative genomics, we identified 81,700 conserved gene families between the 84 species (including 61% with at least two species). Since these species have gone through at least one round of duplication, it is assumed that they should have few or no conserved single-copy genes. However, 3,249 gene families containing at least 50 species with singleton genes were identified. The number of single copy genes varies widely between species. Indeed, after a polyploidization event, there is a gradual loss of duplicated genes, as the genome tends to return to a diploid state. Thus, single copy genes were used as an index to categorize recent or ancient polyploids. Similarly, the analysis of GC content and amino acid composition showed that the sequence profiles in monocots and eudicots are different while polyploidy does not impact sequence diversity. Finally, using synonymous substitution (Ks) rates, we were able to estimate divergence time (speciation) and infer the age of polyploidy events over nearly 250 million years of angiosperm evolution.

Family name: TAVERNIER

First name: Flora

Year of PhD: 2nd

Institute-city: INRAE AGAP, Montpellier

Real-time monitoring of sugar accumulation in the grapevine berry with near-infrared spectroscopy.

Context and purpose of the study -It becomes particularly urgent to decipher the physiological mechanisms underlying the impact of climate change on berry ripening and to select new genotypes keeping an adequate balance between sugars and acids despite the increase in summer temperature. Classical genetic studies consist in studying the diversity of traits of interest, possibly in response to constraints, in order to identify alleles of interest. This type of study is particularly complex to implement regarding grape composition, which displays marked developmental changes during berry ripening, this phenological variation being itself genetically controlled. It is therefore essential to phenotype berries from various varieties at the same developmental stage, so that the differences observed are not due only to differences in maturity level. In this context, it is crucial to develop non-destructive tools for monitoring ripening and berry development at high throughput. This is further complicated by the noticeably asynchronous development of berries within clusters.

Material and methods - Here, we report the use of near-infrared spectroscopy (NIRS), using a portable device in the field, to study the accumulation of sugar and acids in berries of ten grapevine varieties, over two years. We sequentially acquired spectra on single berries from 50 clusters all along ripening, from the green stage to over-maturity, collecting a subset of these berries weekly, for quantifying sugar and acid concentrations with HPLC.

Results - These data were used to train calibration models between spectra and sugar or acid concentrations using a partial least square regression method (PLSR). We cut the data into a training set (% of the data) and a validation set (% remaining), by using the Kennard-Stone algorithm. These models proved to be quite accurate within cross-validation settings. They were further applied to predict sugar and malic acid concentration, which were the best predicted (with a determination coefficient (R^2) of 0,74 and 0,66 respectively), on the berries that were followed with NIRS but not collected for HPLC measurements. This enabled the reconstruction of developmental trajectories of individual berries during the whole ripening period. These results pave avenues for genetic and physiological studies of berry ripening which are critical for selecting and developing new varieties in the context of climate change.

Keywords: Grapevine, Near Infrared Spectroscopy, Single berry, Sugar, Organic acids

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Role of peroxisomes and peroxisomal β -oxidation in lipid remodeling and plant adaptation to phosphate starvation.

Phosphate (Pi) starvation is a frequent nutrient stress significantly impacting plant growth and crop yield. To adapt to Pi starvation, plants activate different mechanisms to increase Pi uptake from soil and to remobilize intracellular reserve. For example, during Pi starvation, phospholipids (PL) located in extra-plastidial membranes are degraded to release Pi and recycled to synthesize digalactosyldiacylglycerol (DGDG) in plastids. Then, DGDG is transferred to other organelles like mitochondria to substitute PL in order to maintain the integrity and functions of organelle membranes. However, the mechanisms involved in membrane lipid remodeling adaptation to Pi starvation remain poorly understood. In our lab, we have previously identified a mitochondrial lipoprotein complex called MTL (mitochondrial transmembrane lipoprotein) involved in phospholipids-DGDG replacement between plastid-mitochondria membrane contact sites (MCS) during Pi starvation. The composition of the MTL complex is dynamic and varies during Pi starvation. In previous studies, we demonstrated that Tom40, a protein of the mitochondrial OM, is present in MTL complex. To further identify candidates located at plastids-mitochondria MCSs, we based on Tom40 protein and performed co-immunoprecipitation (co-IP) with Tom40-HA. Unexpectedly we found peroxisomal proteins that are involved in β -oxidation, a lipid degradation pathway, as partners of Tom40. These results suggested that peroxisomes and peroxisomal β -oxidation pathway might play a role in lipid remodeling. Thus, the aim of my PhD project is to investigate the role of peroxisomes and peroxisomal β -oxidation in lipid metabolism and plant adaptation to Pi starvation.

In my present work, I optimized peroxisomes purification protocol in order to analyze how the lipid composition of peroxisomes is modified in response to Pi starvation. Then, to understand the role of β -oxidation in lipid remodeling, I will analyze the phenotype and the efficiency of lipid remodeling in β -oxidation mutants during Pi starvation. In addition, I will measure the level of β -oxidation in those lines grown in presence and absence of Pi to see if variation occurs in response to Pi starvation.

Key words: phosphate starvation, lipid remodeling, peroxisomes, β -oxidation