PROJETS DEFIS SCIENTIFIQUES 2016

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Medi-SENS	UMR IRHS - SESaN	Béatrice Teulat	Exploring *Medicago truncatula* natural genetic diversity to study nitrogen use efficiency during seedling growth under low temperature and nitrogen supply
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PHY-ROSE

Project title: Phylogenomics of the genus Rosa - polyploidy and hybridization as diversification forces

Project duration: 36 months – Start date: 1/10/2016 End date: 31/12/2019

Key-words: Phylogeny, Complete Genome, Rosa

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Total cost of the project: 107 770 k€ (Do not include salaries others than those of the people hired for the project)
Financial support from « Objectif Végétal »: 107 770 k€

Summary:

The rose bush is the most economically important ornamental plant and the model plant for many studies about ornamental traits (flowering, fragrance, disease resistance). The genus *Rosa*, the taxonomic entity to which rose bush is assigned has a complex evolutionary history with interspecific hybridization events and polyploidizations. Little information about these events is presently available, while evolutionary history are mainly based on chloroplastic molecular markers. Recently, as part of an international consortium, co-coordinated by the GDO team, the first version of the genome sequence of rose cultivar (*Rosa chinensis* 'Old Blush') has been obtained. Moreover, as this cultivar is a vegetatively propagated hybrid between *Rosa chinensis* and *Rosa gigantea,* the sequence of 'Old Blush' provides an unrecorded access to gene variation between two poorly related species. **The objective of this project is to understand the evolutionary history of the genus *Rosa***, and especially to focus on the impact of hybridization and polyploidy in the history of the genus, and to benefit from the complete genome to identify the best molecular markers to resolve such phylogeny. From comparison of molecular sequences we will select the best suitable nuclear low copy genes to reconstruct the phylogeny of the genus. Test of these markers will exploit a set of eight sequences acquired through GenoRose project. Then the markers showing the higher sequence variations will by amplified on a sample representing 100 species of *Rosa*, using fluidigmic amplification of amplicons approach. The resulting sequences will be analysed using various phylogenetic tree-based and network-based methods, with a structured methodology regarding the sample (from diploid non hybrid taxa to polyploydes). This project will help to understand the relationships between the different species and to better predict the success of interspecific crosses (introgression of new sources of resistance from wild gene pools still underused), it will also allow looking of evolutionary changes in a set of ornamental characters.

Medi-SENS

Project title: Exploring Medicago truncatula natural genetic diversity to study nitrogen use efficiency during seedling growth under low temperature and nitrogen supply

Acronym: MeDi-SENS (Medicago Diversity, Seedling Establishment, Nitrogen use efficiency and Stress)

Project duration: 38 months – Start date: 1/11/2016 End date: 31/12/2019

Key-words: Medicago truncatula, natural genetic diversity, seedling establishment, heterotrophic growth, early autotrophic growth, low temperature, limitation of nitrogen supply, multistress, nitrate uptake, nitrogen remobilization, nitrogen use efficiency, GWAS

Coordinator: Béatrice Teulat, IRHS / SESaN, beatrice.teulat@agrocampus-ouest.fr

Total cost of the project: 114 604 € (Do not include salaries others than those of the people hired for the project : PhD or post-doc)
Financial support required from « Objectif Végétal »: 114 604 €

Summary:

Rapid and homogenous seedling growth is crucial for successful crop establishment. Pre-emergence growth at the expense of seed reserves and transition to autotrophy are two steps highly susceptible to environmental cues. Earlier (in the season) and northwards sowings are already practiced for some species and proved to be a good strategy of crop management modification in a context of global climate change. Furthermore the development of sustainable agriculture aims at decreasing nitrate runoff that pollutes waterways and can lead to eutrophication by decreasing nitrate fertilization. Taking into account these conditions and subsequent changes of crop management mean that seedlings will encounter more frequently low temperatures during the establishment phase in a rhizosphere environment characterized by low levels of nitrogen availability. Low temperatures as well as nitrogen limitation are two stresses either individually or in combination that are susceptible to compromise successful seedling establishment by affecting shoot and root elongation and biomass production. It is worth to note that nitrate not only affects seedling establishment as a nutrient but also acts as a signal molecule that shapes root system architecture.

Goals. This project aims to explore the natural genetic diversity of *Medicago truncatula*, a model legume species, to highlight adaptive physiological behaviors for nitrogen use efficiency (NUE) and to identify Quantitative Trait Loci (QTL) associated to successful seedling establishment under the above described new sowing conditions combining low temperature and low nitrate fertilizer supply. To our knowledge, the genetic variability of physiological behaviors in response to limitation of nitrogen for early growth stages has never been explored as well as the combined impact of nitrogen supply and low temperature stress.

Methodology. 1) Seedling growth in the dark will be characterized using a core collection of 192 *M. truncatula* natural accessions in four environmental conditions (with and without nitrate in the growth medium, at both optimal and low temperatures). Hypocotyl and radicle traits (lengths, ramification and biomass) as well as nitrate and total nitrogen (N) contents will be measured. All these characterizations will allow estimating the involvement of exogenous nitrogen and of N reserve mobilization in seedling growth as well as nitrate uptake for all accessions. Moreover, this strategy will allow highlighting potential groups with different behaviors in terms of NUE during heterotrophic growth under ample and limited nitrogen supply at both temperatures.
2) A genome-wide association study (GWAS) will be carried out to exploit linkage disequilibrium in the core collection, to study the genetic architecture of all the traits and to identify the strongest associated loci. This approach could also possibly allow highlighting some candidate genes that could be validated afterwards.

3) A small panel of genotypes selected to be representative of physiological behaviors will be then more finely analyzed. Organ elongation speed, relationships between genotype behaviors in the dark and under photoperiod will be investigated mimicking natural growing conditions. On this smaller panel special interest will be given also to identify, through ^{15}N labeling experiments, respective contributions of N reserves and exogenous source of N (nitrate) to seedling growth under low temperature, and whether there is a link between seedling fitness and on one hand, root system architecture and on the other hand, endogenous *vs* exogenous source of N. This fine analysis of genotype behaviors should also allow identifying physiological traits for ideotype(s) that can fit seedling establishment in crops under low temperature and/or nitrogen supply.

HAUDOR

Project title: Deciphering of Haustorium Development in Orobanche sp

Acronym: HauDOr

Project duration: 36 months – Start date: 01/10/2016 End date: 30/09/2019

Key-words: : Haustorium formation, Cytokinins, Orobanche, Evolution

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Total cost of the project: 128 k€ (Do not include salaries others than those of the people hired for the project : PhD or post- doc)
Financial support required from « Objectif Végétal »: 113 k€

Summary:

Understanding the biological functional networks in the rhizosphere is an important issue for the development of innovative strategies of plant production. Plants as sessile organisms offer a large contact surface between the soil and their roots and the release of organic materials from roots such as organic, amino and fatty acids, carbohydrates or flavonoids is getting more and more interest. It is now known to play a major role in the control of plant communications with others organisms in soils, notably with rhizosphere microorganisms. In return, the benefit of rhizosphere microorganisms producing plant regulators such as auxin and cytokinins for plant growth is also well documented. Such regulatory compounds correspond also to phytohormones that are synthesized and transported through the plants to allow a fine tailoring of its shape and functions. Interestingly, phytohormones can also be exuded from roots but evidences remain rare and the roles of such a process in the control of plant communication with rhizosphere microorganisms and other plants is poorly documented and understood. To date the only clues showing phytohormones act as plant-plant signaling cues within the rhizosphere are the involvement of strigolactones exuded from plant roots in the promotion of hyphal branching of the symbiotic arbuscular mycorrhizal fungi and the symbiotic interaction with Rhizobium, and in the germination of root-parasitic plants.

The plant - parasitic plant communication represents an outstanding opportunity in the present project for pioneer investigations on signaling of plant-exuded cytokinins within the rhizosphere. Indeed, recent data from our laboratory show that host-derived cytokinins promote the formation of the intrusive organ (*haustorium*) of the parasitic plant *Phelipanche ramosa* and are thus required for the establishment of the physical interaction. This will be achieved through the deciphering of the cytokinin signalization pathway in the parasitic plant following germination, and the crosstalk with other plant regulators present in the rhizosphere and known to affect the plant – parasitic plant communication, such as auxin, abscissic acid and strigolactones. This project should therefore shed a new light on the rhizosphere interaction subject which is actually of main interest by establishing hormonal plant-plant interaction as an ecologically important dialogue. In the context of this project, we therefore propose to use root parasitic plants as a tool to explore hormonal plant-plant signaling in the rhizosphere by focusing on cytokinin signaling and its effects on haustorium formation.

Signaling of host-plant derived cytokinins in the parasitic plant will be studied both by a transcriptomic approach using NGS sequencing (RNA-seq) and by bioinformatic analysis of gene networks. In addition,

as a breakthrough approach, functional validation of candidate genes coding for major putative key actors in the parasitic plant will be performed using the targeted CRISPR (clustered regularly interspaced short palindromic repeat) - Cas9 genome editing strategy. Indeed, this strategy has been successfully used for efficient genome editing in human cell lines and animals (zebrafish, mouse) but applications in plants are still rare and need more evidences of successful. Involvement of the newly discovered regulators of such a plant-plant hormonal interaction will also be assessed in other plant – root parasitic plant interactions in order to conclude on the expected significance of this hormonal signaling in plant – parasitic plant communication.

ESCAPES

Project title: Effects of Secondary Contacts and Admixture on a Pathogen Epidemiological Success
Acronym: ESCAPES

Project duration: 36 months – Start date: 2/11/ 2016 - End date: 31/10/2019

Key-words: Epistasis, hybridization, fungal pathogen, secondary contact, quantitative genomics, QTL mapping, Dobzhansky-Muller Incompatibilities

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Total cost of the project: Total cost of the project: 120 770 € (Do not include salaries others than those of the people hired for the project: PhD or post-doc)
Financial support from « Objectif Végétal »: 112 770 €

Summary:

- Context

Secondary contacts with gene flow between divergent populations of pathogens dramatically increase risks of producing new combinations of genes leading to changes in epidemiological properties. Such an outcome however depends on the number and nature of genetics barriers to gene flow as well as the distribution of effects of interaction between loci (epistasis) in hybrids. Ecological barriers will condition the fitness of hybrids according to ecology (e.g. host). Conversely non-ecological barriers mainly depend on negative epistasis in new combinations of loci in hybrids. At last, positive epistasis would increase fitness of hybrids. Our project will focus on the secondary contact between populations of the apple scab, Venturia inaequalis, in wild and anthropized habitats of Kazakhstan that diverged 7,000 years ago. We previously identified phenotypic differences between these populations and genomic regions associated to genetic barriers to gene flow.

- Goals

Our PhD project will be a first attempt to understand the complex factors at work that determine the outcome of a contemporary hybridization in a fungal pathogen. This project is structured around two main objectives:

- First the PhD student will identify ecological and non-ecological genetic barriers to gene flow between strains of wild and anthropized habitats of Kazakhstan.
- Second, the project aims at evaluate the impact of hybridization on traits relevant for epidemiological efficiency. We particularly search for the distribution of epistasis effects in hybrids.

- Methodology

The biological material is composed of i) 90 strains already sampled in wild habitats of Tien-Shan mountains in Kazakhstan where hybrids have been found on the base of 181 quasi-diagnostic SNPs and ii) 200 offspring of an in vitro cross (to be available at the beginning of the PhD) between a strain from wild and a strain from anthropized habitats. Phenotypic measures will be performed on all natural and offspring strains on mycelial growth, sporulation rate, spore size, and pathogenicity on M. sieversii and M. x domestica. Detection of genetic barriers to gene flow will be performed thanks to full genome sequencing and association analyses using phenotypic classes previously identified. At last, analysis of quantitative genetics models parameter will be used to characterize the distribution of epistatic effects in hybrids.

NEMOFY

Project title: NEw MOlecular factors involved in Fungal pathogenicitY

Project duration: 36 months months – Start date: 2/11/2016 - End date: 31/10/2019

Key-words: fungal necrotroph, eisosome, epigenetic control, signaling pathway

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Total cost of the project: **112 770 k€** (Do not include salaries others than those of the people hired for the project: PhD or post-doc)
Financial support from « Objectif Végétal »: **112 770 k€**

Summary:

Context: Based on the Arabidopsis thaliana/Alternaria brassicicola pathosystem and using genome-wide transcriptomic analyses and functional approaches, we recently identified new fungal molecular determinants that may be involved in pathogenicity. We propose to study them in more detail in this project. The first factor is the associated-membrane structures called eisosomes. These structures would be in particular involved in the fungal adaptive response to severe hydric stress conditions consecutive to the gradual decrease in the water potential in maturing reproductive organs. We showed that the transcriptional control of eisosomal components was mainly mediated by the histidine kinase AbNik1, an upstream component of the high osmolarity glycerol (HOG) pathway, which is also the molecular target of dicarboximide and phenylpyrrole fungicides in A. brassicicola. The second factor is the induction of a chromatin remodeling process during the infection process, as strongly suggested by the over-expression during this step of a pool of genes involved in histone modification, DNA methylation or nucleosome structuration.

Goals: A first part aims to investigate the role of eisosomes in virulence of fungal necrotrophs. We also plan to explore the link between eisosomes and signaling pathways when the fungus is exposed to various environments. A second part of the project aims to evaluate the importance of heterochromatic landscapes for fungal virulence. We plan to identify the key players and the environmental conditions (exposure to defence metabolites, hydric stress…) controlling this process. Then, the impact of this potential epigenetic regulation on gene expression will be evaluated.

Methodology : We plan to use functional genomic approaches by generating mutants that are deficient for eisosomal components or for key players in heterochromatin assembly and maintenance, and compare their behaviour to those of the wild-type strain. Moreover, production and distribution of eisosomes along the membranes and chromatin condensation states will be examined under various stress conditions using confocal and epifluorescence observations of mutants producing fluorescently labelled proteins.

BRAM

Project title: Functional-structural modelling of bud burst patterns in rosebush: integration of the role of sugars and hormones.

Acronym: BRAM (BRAnching Modelling)

Project duration: 36 months – Start date: 01/11/2016 End date: 31/10/2019.

Key-words: Bud, Branching, Regulation networks, Dynamical systems, Functional-structural plant modelling, Sugars, Hormones.

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Total cost of the project: 118 k€ (Do not include salaries others than those of the people hired for the project: PhD or post-doc)
Financial support from « Objectif Végétal »: 22 k€. The project benefits from an INRA scholarship.

Summary:

To minimize the impact of agriculture on environment, resource supply should be tightly adjusted to production goals. Computer models predicting plant growth response to environment are powerful tools to optimize resource supply according to plant needs, especially within controlled environments (greenhouses…). Developing such tools implies a better understanding of branching regulation at the plant scale because it is an important variable of plant plasticity that determines productivity and the visual quality of ornamental plants.

The project aims, for the first time, at building a comprehensive model accounting for the role of sugars, together with hormones, in the regulation of bud burst patterns along rosebush primary axis under contrasted light environments. On the contrary to hormones, sugars are a very recently identified player in apical dominance and its mode of action is not known yet. As photosynthetically-derived nutrients, sugars are internal players that have the potential to mediate a part of environmental effects. We propose to develop a new model to understand environmental regulation of branching, calibrated for simple rosebush plants under different light environments. Such model will be defined in a generic way to be adapted easily to other economically important species. It will also open new prospects to understand branching behavior of adult plants, and later, to apprehend the complexity of urban environment and its effect on plant architecture.

The methodology will consist in implementing physiological processes common to different species in a computer functional-structural model. We have recently developed a model at the bud scale that simulates bud burst response to both sucrose and hormones. This bud model will be integrated in a plant architecture to simulate bud burst patterns at the scale of the plant. For that, we will couple the bud model with simulation of spatial and temporal dynamics of hormones and sugars within plant architecture. This involves the use of existing models of sugar and hormone dynamics, and requires physiological and molecular experiments to gain knowledge about how sugars interact with hormonal network. Such a multidisciplinary approach will be achieved through a consortium between biologist experts in the physiological regulation of bud outgrowth (ARCH-E) and computer scientists with strong expertize in modelling of plant morphogenesis (Inria Virtual Plants).

SCALD

Project title: *Déterminisme précoce d'une maladie majeure de conservation du fruit: l'échaudure superficielle de la pomme*

Project duration: 36 mois

Coordinator: Mathilde.orsel-baldwin@inra.fr

Total cost of the project: 96 000 €.
Financial support from « Objectif Végétal »: 0 €.
The project benefits from an INRA scholarship.

Summary:

L'échaudure superficielle ou « superficial scald » est un désordre physiologique qui se manifeste par le brunissement de la surface des pommes pendant ou après la conservation des fruits au froid, les rendant ainsi non commercialisables. L'apparition des symptômes dépend de nombreux facteurs tels que la variété, les conditions climatiques en production, les traitements post-récolte ou encore les conditions de conservation. Le déterminisme du déclenchement de l'échaudure reste largement inconnu et l'interdiction progressive des traitements chimiques de protection et/ou leurs coûts prohibitifs font que cette maladie de conservation est devenue un problème majeur pour la filière, en particulier pour certaines variétés phares du marché telle que Granny Smith. Le brunissement de la peau des fruits résulte de la nécrose des cellules de l'hypoderme des fruits, et de l'épiderme dans les cas les plus sévères. Il résulte d'un processus oxydatif induit par la mise en conservation au froid et est corrélé avec l'accumulation de produits de l'auto-oxydation de l'α farnésène tels que des triènes conjugés (Ctols) et le 6 methyl 5 hepten 2 one (MHO). Toutefois les quelques approches ciblées sur l'expression des gènes impliqués dans les voies de biosynthèse de l'α farnésène, des polyphénols et de quelques enzymes antioxydants ne permettent pas de conclure sur la cascade d'évènements aboutissant au développement des symptômes d'échaudure plusieurs mois après la récolte. En effet, à ce jour il n'existe pas d'approche intégrative permettant de comprendre le développement de l'échaudure dès les premiers stades suivant la récolte, prenant en compte la variation annuelle de la susceptibilité à l'échaudure ainsi que le stade de maturité des fruits lors de la récolte.

Le sujet proposé vise à identifier la, ou les voies de signalisation conduisant au développement des symptômes de l'échaudure des fruits. En corollaire cela permettra d'identifier des marqueurs précoces de la mise en place du phénomène comme une aide au pilotage de la récolte et de la conservation des variétés sensibles en fonction des conditions climatiques de l'année, voire d'identifier des pistes pour la recherche d'itinéraires techniques d'évitement de l'échaudure.

DEEP DIVING

Project title: DoEs host domEstication Promote effectors DIVersification IN pathoGens?

Project duration: 24 months – Start date: 15/01/2017 End date: 14/01/2018

Key-words: effectors diversification, wild populations, emergence disease, Transcriptomic, domestication, hybridization

Coordinator: Le Cam Bruno, IRHS, bruno.lecam@angers.inra.fr

Partner: Lemaire Christophe, IRHS, christophe.lemaire@univ-angers.fr

Total cost of the project: 43 218 € (post-doc)
Financial support from « Objectif Végétal »: 40 100 €

Summary:

Since the beginning of agriculture, wild plant species have been hybridized to generate new crops with higher agronomic and economic added values. However, little is known about **how host domestication may influence the evolution of pathogens and how it may even favour the emergence of new diseases.** Dessert apple Malus x domestica is a hybrid between several wild Malus species. The principal contributor is M. sieversii, which is endemic in Central Asia. M. sylvestris and M. orientalis, which are endemic in Europe and Caucasus, respectively, are secondary contributors. The main pathogen of M. x domestica, the fungus Venturia inaequalis, also originates from Central Asia. Although V. inaequalis can infect all these Malus species, isolates obtained from the different hosts differ in their pathogenic behaviour. Indeed, previous studies showed that virulence and aggressiveness have probably been gained during host domestication.

Here, we want to understand how successive hybridization events between Malus species, i.e. host domestication, have modified the pathogenicity determinants of V. inaequalis. We will focus on the polymorphism of protein effectors, which are known to be key determinants of the virulence of fungal pathogens.

830 putative protein effectors are predicted in the genome of V. inaequalis. However, in order to work on genes that are actually expressed during infection of the host and therefore might play a role in the interaction, we will take advantage of transcriptomic data obtained during the infection already available in the team. Polymorphism of the effector candidates in V. inaequalis populations will be measured thanks to the genome sequence available for 42 isolates that were sampled on the different Malus species (M. x domestica, M. sylvestris, M.orientalis and M. sieversii). Using coalescence analyses, we will determine the origin of effector candidate alleles that are segregating in strains that infect domestic apple trees. Using several tests like ABBA-BABA, **we will evaluate if new alleles were either introgressed from a local population of V. inaequalis preexisting on wild Malus trees or originated from de novo adaptation.**

OBAUC

Project title: Ornamental bush Architecture under urban conditions

Project duration: 15 months – Start date: 1/10/2016 End date: 31/12/2017

Key-words: Urban context, plant architecture response, water, light, compacted soil, transpiration model

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Partner 2: Fabrice Rodriguez, IRSTV – LEE, Fabrice.rodriguez@ifsttar.fr

Total cost of the project: 31.3 k€
Financial support from « Objectif Végétal »: 25k€

Summary:

Context: Plants in cities are living in a stress-inducing environment characterized by unfavorable aerial conditions (high temperature, low air humidity, blast of wind, light reverberation, infrastructure shade…) and soil conditions (compaction, limited volume, contamination by heavy metals and polycyclic aromatic hydrocarbons…). Then, city managers are looking for plant species able to adapt in such an environment, but also able to guarantee aesthetic features, with low inputs and simple management. However, proposing plant species models fulfilling such conditions requires the understanding of plant development, depending on these abiotic environmental factors.

Goals: The aim of the project is to characterize plant-environment inter-relations, to understand and predict plant development. The specific goals are to analyze the combined effects of soil compaction, water supply restriction and shading on plant transpiration, plant biomass as well as aerial and root architecture. Modelling of plant transpiration and biomass production will be assessed.

Methodology: For this starter project, we will mimic urban environmental conditions in a greenhouse, with opened roof. Urban climate will be replicated from the Nantes Urban Environment Observatory (ONEVU) database (hydrological and meteorological data). Three environmental factors will be tested using rose bush as plant model. Modalities will include restrictive irrigation, vertical shading screen to represent urban infrastructures, and soil type. The control modality will be an uncompacted soil under water comfort condition (soil water content at field capacity) and without shade. Rose growth will be followed during one year. Several parameters around the climate, plant ecophysiology and architecture, and the soil will be measured. An existing soil-plant-atmosphere model predicting plant transpiration under water restriction will be used and adapted in the present project.

PREDIRE

Project title: Constitutive level of basal defences in healthy apple cultivars as predictors of multi-resistance and performance of plant defence stimulators

Acronym: PREDIRE (PREDIction of REsistance in apple)

Project duration: 36 months – Start date: 1 October 2016 End date: 30 September 2019

Key-words: basal defences, constitutive expression, structural annotation, functional annotation, sequence diversity, RT-qPCR, primers, expression QTL, genome-wide association mapping, SNP, apple, scab, fire blight

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Total cost of the project: 24 978 k€
Financial support from « Objectif Végétal »: 24 978 k€

Summary:

Apple is susceptible to numerous diseases and pests that request intensive chemical controls with much concern for human health and environment. Breeding new resistant varieties and/or stimulating plant defences in commercial orchards are thus major stakes. Especially, increasing the level of basal defences against a large range of bioagressors instead of focusing on complete resistance against particular ones is a major goal to efficiently decrease the number of treatments. We hypothesize that the constitutive level of basal defences (i.e., before any induction) is driving the plant capacity to reach a sufficient resistance level to control a pathogen or pest attack, possibly with the help of Plant Defence Stimulators (PDS). In several plant models, constitutive defences have actually been associated to quantitative resistance and are highly suggested to predict the protective efficiency of PDS. Accurate assessment of the constitutive level of defence-related genes at the transcriptional level is thus a major stake, which is challenged by the genetic variability of the corresponding gene families especially when a large germplasm has to be screened. The genomic resources recently developed in apple at IRHS Angers (new genome sequence, accurate annotation, resequenced panel, SNP genotyping array), the germplasm resources present on-site, and the skill of the involved teams in bioanalysis, quantitative genetics and functional analysis of defences with or without PDS allow us to explore the genetic variability of defence gene sequences and to decipher the genetic control of constitutive defences thanks to expression QTL (eQTL) analysis.

Goals: The genetic variation of apple constitutive defences at the transcriptional level will be assessed and characterized in a large range of apple cultivars (several hundreds of old apple cultivars from various European origins). We want to perform a proof-of-concept that the constitutive expression level of some particular defence genes (i) can be accurately assessed with RT-qPCR in a very large apple germplasm, (ii) is subject to heritable genetic variation, (iii) can be subject to eQTL analysis thanks to genome-wide association studies (GWAS), and (iv) is potentially correlated to apple scab and fire blight partial resistance and to PDS protective efficiency. The major technical challenge is to define robust conserved primer pairs allowing the accurate assessment of gene expression of multigenic families in a large germplasm by RT-qPCR.

Methodology: The experimental approach is subdivided in 4 tasks, with the 5th task devoted to the dissemination of results:

1- Characterizing in silico a set of max. 50 apple defence-related gene families (bioanalysis, functional annotation of subfamilies)
2- Designing and validating robust RT-qPCR primer sets for 15-20 families in 30 apple cultivars
3- Assessing the genetic variation of the constitutive expression level of 3 candidate defence-related genes and performing GWAS on 400 cultivars with the help of available high-density genotyping data (487k Axiom array)
4- Correlating the constitutive expression levels of defence genes to scab and fire blight resistance or to the PDS protective efficiency against these diseases on either 30 cultivars (x 15-20 genes) or 400 cultivars (x 3 genes)