SACLAY PLANT SCIENCES

SPS Conference 2013

Plant signalling in a changing environment

ABSTRACT BOOK

July 4-6, 2013
Evry, France

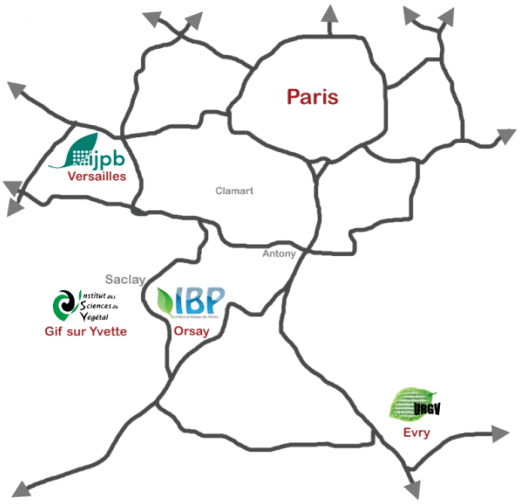
https://colloque.inra.fr/spsconference

The Saclay Plant Sciences LabEx

Network of research, teaching, training and innovation in plant sciences

SPS@versailles.inra.fr
www.saclayplantsciences.fr

4 research units located in the south and west of Paris:
- Institut de Biologie des Plantes (IBP, Orsay)
- Institut des Sciences du Végétal (ISV, Gif-sur-Yvette)
- Institut Jean-Pierre Bourgin (IJPB, Versailles)
- Unité de Recherche en Génomique Végétale (URGV, Evry).

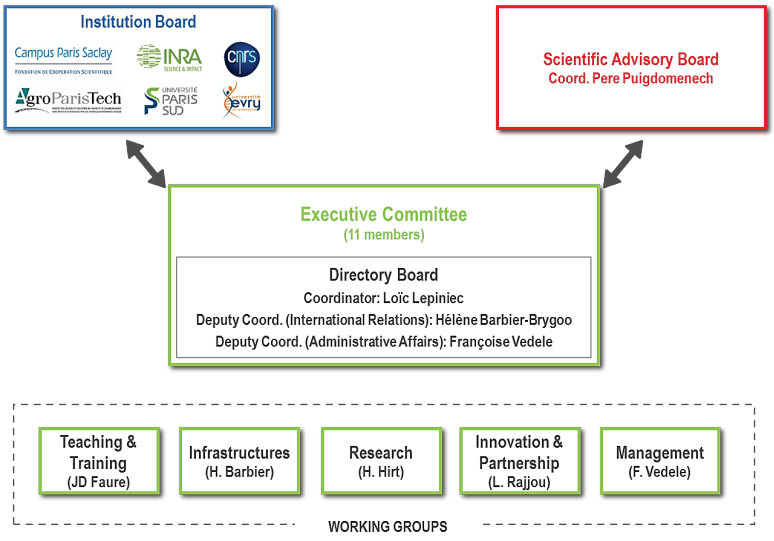


6 institutions:
- Coordinating Partner (FCS)
- AgroParisTech
- CNRS
- INRA
- University of Evry Val d'Essonne
- University Paris Sud

Scientific objectives
Understanding the genetic, molecular and cellular mechanisms that control plant physiology and development, as well as their interactions with fluctuating biotic or abiotic environments

Human and technical resources
About 700 people (among whom 400 permanent staff) with internationally renowned scientists
About 12 scientific platforms: cytology, imaging, chemistry, biochemistry, cell biology, metabolomics, transcriptomics, TILLING…
Plant growth facilities: ~ 5500 m² of greenhouses, ~ 100 growth chambers, security levels S1, S2, S3

Organization chart



3 scientific challenges:
1) Moving from a descriptive towards a more predictive biology
2) Understanding basic genetic, molecular and cellular mechanisms that control plant development and physiology
3) Developing tools and biotechnology for research, innovation and valorization

4 priority issues:
1) Improving the sustainability of crop plant production
2) Plants as factories: improving plant quality for food, feed, health, industry and environment
3) Plants to understand fundamental biological mechanisms
4) Developing new resources and tools for research and innovation.

Foreword

Welcome to the first international conference organized by the Saclay Plant Sciences LabEx !

As sessile organisms, all plants have the capacity to sense and respond to changing conditions in their environment to appropriately modify their development and physiology. During land colonization, different plant species have used diverse strategies to cope with largely variable environmental conditions. Although a great amount of knowledge has been accumulated in many areas of plant biology, the molecular mechanisms underlying signaling and adaptation to the environment are still largely unclear.

It is the aim of this conference to discuss the latest discoveries on how plants respond to changes in their abiotic and biotic environmental conditions. The conference will look at different strategies revealed by studying natural variation, highlighting novel insights into the genetic and epigenetic mechanisms, and showcase how this knowledge can be applied to improve crop production to meet the needs of the future.

We are happy to have you here in Evry and we hope that you will enjoy the quality of the talks and poster presentations of this conference.

We are looking forward to inviting you to other Saclay Plant Sciences international events !

The organizing committee:
Heribert Hirt
Martin Crespi
Michael Hodges
Herman Höfte
Annie Marion-Poll
Marie-Jeanne Sellier

Content

Program

Thursday July 4, 2013

2:00 PM	**Opening talk - Presentation of the Saclay Plant Sciences Laboratory of Excellence**	
Plenary lecture		
2:15 PM	**Paul Schulze-Lefert**	*Structure, functions, and evolution of the bacterial root microbiota*
Session 1: Biotic Interactions		
3:00 PM	**Thomas Boller**	*Peptide signals and their receptors in plant innate immunity*
3:35 PM	**Heribert Hirt**	*Role of MAP kinase signaling in plant-microbe interactions*
4:10 PM	**Coffee break**	
4:40 PM	**Sylvain Raffaele**	*Host and pathogen determinants in Arabidopsis Quantitative Disease Resistance against the necrotrophic fungus Sclerotinia*
5:00 PM	**Susana Rivas**	*Regulate and be regulated: tight control of plant defense responses by the Arabidopsis transcription factor MYB30*
5:20 PM	**Valérie Cotelle**	*CPK3, a Ca2+-dependent protein kinase regulated by 14-3-3 proteins, is required for sphingolipid-induced cell death in Arabidopsis*
5:40 PM	**Malik Mbengue**	*A conserved pathway mediates endocytosis of pattern recognition receptor*
6:00 PM	**Ana Victoria Garcia**	*The protein kinase OXI1 regulates salicylic acid-dependent plant immunity and cell death programs in Arabidopsis*
Poster session		
06:20 – 09:30 PM	**Poster session and Welcome cocktail**	

Friday July 5, 2013

Session 2: Abiotic Interactions

9:00 AM	**Phil Wigge**	*Ambient temperature sensing in plant development*
9:35 AM	**Anne Krapp**	*NIN-like proteins: key regulators of plant responses to nitrogen availability*
10:10 AM	**Johanna Molenaar**	*Unravelling drought responses in Arabidopsis thaliana using a world-wide natural population*
10:30 AM	**Bénédicte Wenden**	*Sweet cherry phenology in the context of climate change: a systems biology approach*
10:50 AM	**Benjamin Peret**	*Dissecting root architecture adaptation to phosphate starvation.*
11:10 AM	**Afif Hassairi**	*The extremophile grass Aeluropus littoralis: a source of candidate genes for improving salt and drought stresses in cereals*
11:30 AM	**Marieke Dubois**	*How drought affects leaf growth: ERF5/6 and DELLAs act together to regulate growth inhibition under stress*

11:50 AM	**Lunch**

Session 3: Biodiversity and natural variation

1:20 PM	**Johanna Schmitt**	*Natural variation in life history responses to climate in Arabidopsis thaliana*
1:55 PM	**Patrick Laufs**	*Genetic control of leaf shape*
2:25 PM	**Helen North**	*Exploiting natural variation in seed mucilage characteristics to identify novel genes involved in its production and function*
2:45 PM	**Manon Richard**	*The subtelomeric Khipu satellite repeat from Phaseolus vulgaris: lessons learned from the genome analysis of the Andean genotype G19833*
3:05 PM	**Sylvain Merlot**	*De novo transcriptome sequencing of the nickel hyperaccumulator Psychotria gabriellae and identification of PgIREG1 as a candidate nickel transporter involved in accumulation.*
3:25 PM	**Georgi Bonchev**	*Abundance of transposable elements affected by mating system in Arabidopsis lyrata*
3:45 PM	**Laurent Gentzbittel**	*Natural diversity in the model legume Medicago truncatula and the fungal pathogen Verticillium sp. allows identifying distinct genetic mechanisms for resistance to Verticillium wilt*

4:05 PM	**Coffee break**

Session 4: Translational biology

4:30 PM	**Rob Dirks**	*The making and use of chromosome substitution lines in plant breeding: comparison of isogenic hybrids and their respective methylation landscape*
5:05 PM	**Abdelhafid Bendahmane**	*Translational research, URGV experience*
5:40 PM	**Kent Bradford**	*Temperature-sensitive expression of LsNCED4 encoding an aba biosynthetic enzyme is required for thermoinhibition of lettuce seeds*
6:00 PM	**Andrew Lloyd**	*Meiotic gene evolution: can you teach a (duplicated) old dog new tricks?*
6:20 PM	**Raphaël Mercier**	*What limits meiotic crossovers?*
6:40 PM	**Hiro Nonogaki**	*Engineering abscisic acid metabolism and signaling in plants*

Poster session

07:00 – 8:30 PM	**Poster session (Food and drinks provided)**

Saturday July 6, 2013

Session 5: Organization and functioning of complex crop genomes and traits		
9:00 AM	**Jeff Bennetzen**	*Gene, centromere and transposon evolution in maize and its panicoid relatives*
9:35 AM	**Boulos Chalhoub**	*Polyploidy generates trait novelty and functional diversity in wheat*

Session 6: Epigenetics		
10:10 AM	**Ueli Grossniklaus**	*Epigenetic variation contributes to adaptation in selective environments*
10:45 PM	**Martin Crespi**	*Dynamic regulation of the epigenetic landscape by non-coding RNA*
11:05 PM	**Mathilde Orsel**	*The sense and anti-sense transcriptome of apple reveals the potential widespread regulatory control of gene expression through cis-acting si-RNA*
11:25 PM	**Christine Lelandais**	*MicroRNAs involved in root biomass and symbiotic interactions in the model legume Medicago truncatula*
11:45 PM	**Teddy Jegu**	*The SWI/SNF chromatin remodelling protein AtBAF60 directly controls the formation of a gene loop at the FLC locus in Arabidopsis*

12:05 AM	**Lunch**

Session 7: Hormones		
1:30 PM	**Dolf Weijers**	*Hormonal control of growth and patterning in the plant embryo*
2:05 PM	**Catherine Rameau**	*Strigolactones and other long range signals regulating shoot branching in pea*
2:40 PM	**Anouck Diet**	*Gibberellins control root growth and nodulation in Medicago truncatula*
3:00 PM	**Myckel Habets**	*Plant development requires dynamic microtubule localization of the PINOID kinase through a BT-KINESIN complex*
3:20 PM	**Annie Marion-Poll**	*Cell wall remodelling in hormonal control of seed dormancy and germination*

3:40 PM	**Coffee break**

Plenary lecture		
4:15 PM	**Ian Small**	*Controlling gene expression in energy organelles*
05:00 – 05:10 PM	**Closing Talk**	

Conference dinner

Departure: around 5:30 pm.

Return: around midnight.

Abstracts: Talks

PLENARY LECTURE

STRUCTURE, FUNCTIONS, AND EVOLUTION OF THE BACTERIAL ROOT MICROBIOTA

Davide Bulgarelli, Klaus Schläppi, Emiel ver Loren van Themaat, Nina Dombrowski, Matthias Rott, Yang Bai, and Paul Schulze-Lefert* (Max Planck Institute for Plant Breeding Research, D-50829 Cologne, Germany)

schlef@mpipz.mpg.de

Plants host distinct bacterial communities on and inside various plant organs. We show that roots of *Arabidopsis thaliana*, grown in different natural soils under controlled environmental conditions, are preferentially colonized by Proteobacteria, Bacteroidetes and Actinobacteria, and each bacterial phylum is represented by a dominating class or family. Soil type defines the composition of root-inhabiting bacterial communities and host genotype determines their ribotype profiles to a limited extent. Plant cell wall features provide a sufficient cue for the assembly of ~40% of the Arabidopsis bacterial root-inhabiting microbiota, with a bias for Betaproteobacteria. This root sub-community may not be Arabidopsis-specific but saprophytic bacteria that would naturally be found on any plant root or plant debris in the tested soils. A comparison of the bacterial root microbiota of *A. thaliana* with the microbiota from selected *A. thaliana* relatives grown under controlled environmental conditions or collected from natural habitats suggests the existence of an evolutionarily conserved core microbiota with species-specific footprints. These findings also imply that the bacterial root microbiota is surprisingly resilient to environmental changes. We have isolated > 40% of the root microbiota members from *A. thaliana* as pure bacterial cultures. This has allowed us to develop *in planta* test systems for single or combinations of microbiota members under laboratory conditions and to explore their functions in plant growth promotion and for plant health. We combine these *in planta* assays with whole-genome sequencing of microbiota members and assess their metabolic capacity using metabolic arrays to obtain first insights into the molecular basis of rhizobacteria interactions with Arabidopsis roots.

Further reading:

Bulgarelli et al., *Nature* 2012; 488(7409):91-5
Lundberg et al., *Nature* 2012; 488(7409):86-90
Bulgarelli et al., Annual Review of Plant Biology (2013), in press

SESSION 1:

BIOTIC INTERACTIONS

PEPTIDE SIGNALS AND THEIR RECEPTORS IN PLANT INNATE IMMUNITY

Thomas Boller (Botanical Institute, University of Basel, Switzerland)

thomas.boller@unbas.ch

Innate immunity is based, in a first instance, on the perception of characteristic microbial molecules, the so-called microbe-associated molecular patterns (MAMPs). We have identified and characterized two pattern recognition receptors involved in this process, namely the flagellin receptor FLS2 and the EF-Tu receptor EFR. These two receptors perceive highly conserved, characteristic peptide epitopes of bacteria. They are localized on the plasma membrane with an extracellular recognition domain and an intracellular protein kinase domain, and they contribute in important ways to the resistance of *Arabidopsis thaliana* against bacterial pathogens. In addition, the plants themselves produce peptides that function as danger signals. For example, *Arabidopsis thaliana* contains a group of seven peptides, called AtPEP1-7, which are recognized by the corresponding receptors PEPR1 and PEPR2. These receptors are similar in structure and function to FLS2 and EFR. Our current work addresses the question how AtPEPs and their receptors are integrated into the plant's innate immune system. Peptide signals are also important in plant development. In a recent study, it has been reported that the flagellin receptor FLS2 also perceives the peptide hormone CLV1, which is centrally involved in organization of the shoot meristem. In attempts to corroborate this finding,however, we found that FLS2 is completely blind to pure CLV1 peptide, but sensitive to minute amounts of contaminating flagellin-derived peptides. This highlights the selectivity and sensitivity of the plant's peptide receptors.

ROLE OF MAP KINASE SIGNALING IN PLANT-MICROBE INTERACTIONS

Heribert Hirt* (URGV Evry, France), Souha Berriri (URGV Evry, France), Jean Bigeard (URGV Evry, France), Marie Boudsocq (URGV Evry, France), Nicolas Frei dit Frey (URGV Evry, France), Ana Victoria Garcia (URGV, Evry, France), Nathalie Leonhardt (CEA Cadarache, France), Jean-Luc Montillet (CEA Cadarache, France), Stephanie Pateyron (URGV Evry, France), Jean Colcombet (URGV, Evry, France).

hirt@evry.inra.fr

Mitogen-activated protein kinase (MAPK) modules are highly conserved signaling modules in all eukaryotes. In Arabidopsis, MPK3, 4 and 6 are the best known MAPK pathways that are involved in stress responses, cell division control, cytoskeletal organization and developmental descisions. *mpk4* knockout (KO) plants are dwarfed and very sick, making it difficult to distinguish between cause and effect of its phenotype. To overcome this difficulty, we developed mutations triggering constitutive MPK4 activity and created transgenic lines allowing phenotypic studies on a WT-like plant (Berriri et al., 2012). By this approach, we confirmed that MPK4 functions as a negative regulator of pathogen defense, but our work also suggests that MPK4 interferes with stress signaling pathways at several distinct steps in pathogen-associated molecular pattern (PAMP)-triggered immunity (PTI) as well as in effector-triggered immunity (ETI). Our study shows that CA mutations are valuable complementary tools to identify novel MAPK functions (Hirt et al. 2012). MAPKs also play a role in mediating abiotic and biotic stress signals to close plant stomata and abscisic acid (ABA) has been suggested to regulate this process. Using genetic, biochemical, and pharmacological approaches in Arabdiopsis, we demonstrated that two distinct MAPK pathways exist to mediate PAMP and ABA signals, respectively (Montillet et al., 2013). The biotic pathway comprises the MAPKs MPK3 and 6 as well as the production of specific oxylipins by the 9-specific lipoxygenase LOX1 that functions upstream of the hormone salicylic acid. The pathway is negatively controlled by jasmonic acid and the pathogen effector analogue coronatine. The ABA pathway mediates abiotic signals via the protein kinases OST1 and the MAP kinases MPK9 and 12 and also sets the threshold for the biotic pathway. Finally, we show that the biotic and the abiotic pathways converge again at the level of the anion channel SLAC1 to execute stomatal closure (Montillet and Hirt, 2013). Overall, these data show the versatility of MAPK pathways in distinct physiological regulatory programs that respond to environmental signals.

Berriri S, Garcia AV, Frei Dit Frey N, Rozhon W, Pateyron S, Leonhardt N, Montillet JL, Leung J, Hirt H, Colcombet J. (2012) Constitutively Active Mitogen-Activated Protein Kinase Versions Reveal Functions of Arabidopsis MPK4 in Pathogen Defense Signaling. *Plant Cell* 24: 4281-93.

Colcombet J, Berriri S, Hirt H. (2012) Constitutively active MPK4 helps to clarify its role in plant immunity. *Plant Signal Behav*. 8:e22991

Montillet, J.-L., Leonhardt, N., Mondy, S., Tranchimand, S., Rumeau, D., Boudsocq, M., Garcia, A.V., Douki, T., Bigeard, J., Laurière, C., Chevalier, A., Castresana, C., Hirt, H. (2013) An ABA-independent oxylipin pathway controls stomatal closure and immune defense in Arabidopsis. *PLoS Biol.* 11(3):e1001513

Montillet, J-L., Hirt, H. (2013) New checkpoints in stomatal defense. *Trends Plant Sci.* 18(6):295-7

HOST AND PATHOGEN DETERMINANTS IN ARABIDOPSIS QUANTITATIVE DISEASE RESISTANCE AGAINST THE NECROTROPHIC FUNGUS SCLEROTINIA

Sylvain Raffaele* (LIPM, INRA Toulouse, France), Derry Voisin (LIPM, INRA Toulouse, France), Koanna Guyon (LIPM, INRA Toulouse, France), Claudine Balagué (LIPM, INRA Toulouse, France), Bruno Grèzes-Besset (Biogemma, Mondonville, France) Dominique Roby (LIPM, INRA Toulouse, France)

sylvain.raffaele@toulouse.inra.fr

Fungal plant pathogens are major and rising threats for global food security and environment sustainability. Necrotrophic fungi, that kill host cells for infection, are among the most devastating plant pathogens, however our knowledge on the molecular bases of their interaction with plants is one of the least advanced in the field of plant pathology. The White Mold pathogen *Sclerotinia sclerotiorum* is a cosmopolitan fungus featuring one of the broadest host range (>400 plant species). It infects notably wild and cultivated *Brassica* species, including the model plant *Arabidopsis thaliana*. We documented extensive variation in Quantitative Disease Resistance (QDR) to *S. sclerotiorum* in *A. thaliana* natural populations, opening the way to the molecular characterization of plant and pathogen determinants underlying this interaction. Using Genome Wide Association mapping we have pinpointed several candidate loci involved in plant QDR against *S. Sclerotiorum*. In parallel, we have conducted a refined *in silico* analysis of *S. Sclerotiorum* secreted proteins repertoire to identify candidate effectors altering the physiology of the host cell. Progresses towards the functional characterization of these molecular determinants will be presented. These results open new perspectives for the exploration of host and pathogen diversity to understand the nature and evolution of the molecular mechanisms underlying plant QDR and fungal pathogenicity in necrotrophic fungal infections.

REGULATE AND BE REGULATED: TIGHT CONTROL OF PLANT DEFENCE RESPONSES BY THE ARABIDOPSIS TRANSCRIPTION FACTOR MYB30

Susana Rivas*, Daniel Marino, Joanne Canonne, Solène Froidure, Dominique Roby, Mickaël Pata, Amandine Léger (Laboratoire des Interactions Plantes-Microorganismes, Castanet-Tolosan, France), Cécile Pouzet, and Alain Jauneau (Plateforme Imagerie FR3450, Castanet-Tolosan, France)

Susana.Rivas@toulouse.inra.fr

Transcriptional regulation in host cells plays a crucial role in the establishment of plant defence and associated cell death (hypersensitive response; HR) in response to pathogen attack. The MYB transcription factor MYB30 was previously identified as a positive regulator of Arabidopsis defence and HR responses to bacterial pathogens (Vailleau *et al.*, 2002). MYB30 appears to modulate cell death-related lipid signaling by enhancing the synthesis of sphingolipid-containing very long chain fatty acids (VLCFAs) after bacterial inoculation (Raffaele *et al.*, 2008). We have shown that MYB30 is targeted by the *Xanthomonas* type III effector XopD resulting in suppression of MYB30-mediated plant defences and underlining the crucial role played by MYB30 in the regulation of plant disease resistance (Cannone *et al.*, 2011). In addition, the activity of MYB30 is tightly controlled by plant cells. First, in the presence of MYB30, the secreted phospholipase *At*sPLA2-□ is partially relocalized to the nucleus where the two proteins interact, leading to repression of MYB30-mediated HR and defence[Froidur] *et al.*, 2010). These data highlight the importance of cellular dynamics for defence-associated gene regulation in plants. Second, the RING-type E3-ubiquitin ligase MIEL1 (MYB30-Interacting E3 Ligase1) interacts with MYB30 in the plant cell nucleus, leading to MYB30 ubiquitination and proteasomal degradation, inhibition of MYB30 transcriptional activity and suppression of Arabidopsis defence and HR responses (Marino *et al.*, 2013). Third, MYB96, a MYB transcription factor within the MYB30 phylogenetic group, has been recently identified as an additional MYB30-interacting partner that positively regulates the establishment of defence (Pata *et al.*, submitted). Our work uncovers a MYB transcriptional rheostat that fine-tunes the initiation of defence-related cell death through the production of sphingolipid-containing VLCFAs. The complex transcriptional and interaction regulatory network involved in the temporal and spatial control of MYB30/MYB96-mediated plant defence responses will be discussed.

CPK3, A CA^{2+}-DEPENDENT PROTEIN KINASE REGULATED BY 14-3-3 PROTEINS, IS REQUIRED FOR SPHINGOLIPID-INDUCED CELL DEATH IN *ARABIDOPSIS*

Valérie Cotelle*, Christophe Lachaud, Elsa Prigent, Patrice Thuleau, Sabine Grat, Daniel Da Silva, Christian Brière, Christian Mazars (Laboratoire de Recherche en Sciences Végétales, Castanet-Tolosan, France)

cotelle@lrsv.ups-tlse.fr

In eukaryotic cells, sphingoid Long Chain Bases (LCBs) behave as second messengers involved in various processes including programmed cell death (PCD). In plants, induction of PCD by LCBs such as dihydrosphingosine (d18:0, DHS) or phytosphingosine (t18:0, PHS) has now been described. For example, Fumonisin B1 (FB1), a toxin produced by the necrotrophic fungus *Fusarium moniliforme*, induces plant PCD by accumulation of DHS and PHS, the two major LCBs in plants. However, the LCB pathway leading to PCD in plants is still enigmatic. Recently, we showed that DHS-induced PCD in tobacco BY-2 cells is controlled by nuclear calcium (Lachaud *et al.*, 2010, Cell Calcium 47, 92-100) and the mitogen-activated protein kinase (MPK6) was described as a transducer in the LCB-mediated PCD in *Arabidopsis thaliana* (Saucedo-García *et al.*, 2011, New Phytol. 191, 943-957). To get further insights into the still unknown signaling pathway of LCB-induced PCD in plants, we combined biochemistry and reverse genetics using the model plant *Arabidopsis thaliana*. We show that *Arabidopsis* CPK3, a member of the plant family of calcium-dependent Ser/Thr protein kinases (CDPKs or CPKs), plays a critical role in LCB-mediated cell death. We found that CPK3 dissociates from 14-3-3 proteins and is degraded during PHS-induced cell death. We also show that the equivalent of the Ser58 epitope in mammalian 14-3-3 *Arabidopsis* 14-3-3s is phosphorylated in a PHS and calcium-dependent manner by CPK3, thus identifying the plant kinase able to phosphorylate 14-3-3s at this site in the dimer interface. Moreover, gene knockout of CPK3 results in a FB1-resistant phenotype in *Arabidopsis*, revealing a new role for CPK3 as a positive regulator of plant PCD. On the basis of these findings, we provide a working model for how LCBs and calcium regulate CPK3 and 14-3-3 proteins in the context of plant cell death (Lachaud, Prigent *et al.*, 2013, Cell Death Differ. 20, 209-217).

A CONSERVED PATHWAY MEDIATES ENDOCYTOSIS OF PATTERN RECOGNITION RECEPTORS

Malick Mbengue[1]*, Fabio Gervasi[1], Alberto Macho[1], Sebastian Bartels[2], Thomas Boller[2], Cyril Zipfel[1], Takashi Ueda[3] and Silke Robatzek[1].

[1]The Sainsbury Laboratory, Norwich Research Park, Norwich, NR4 7UH, UK
[2]Institute of Botany, University of Basel, Hebelstrasse 1, 4056, Basel, CH
[3]Laboratory of Developmental Cell Biology, University of Tokyo, J.

malick.mbengue@tsl.ac.uk

Pattern recognition receptors (PRRs) are present at the plasma membrane and binding of their cognate ligands derived from microbes or plant endogenous peptides triggers signaling cascades and defense responses required for plant immunity. The FLAGELLIN SENSING 2 (FLS2) receptor is internalized via the endocytic pathway into *bona fide* endosomes when activated by its ligand flg22. This process requires the function of the co-regulatory receptor SOMATIC EMBRYO RECEPTOR KINASE 3 (SERK3), also required for activation of the plant hormone BRASSINOSTEROID INSENSITIVE 1 (BRI1) receptor (Robatzek et al., 2006; Beck et al., 2012). Perception of bacterial-derived elf18 and the plant peptide pep1 by the EF-Tu RECEPTOR (EFR) and the PEP RECEPTOR 1 (PEPR1), respectively, elicit highly similar responses compared to FLS2 signaling but their subcellular trafficking is unknown. Using live cell imaging in *N. benthamiana*, we show that FLS2, EFR and PEPR1 internalize into endosomal compartments co-localizing with RabF2B in a SERK3-dependent manner. Endocytosis of the receptors is highly ligand-specific as only their cognate ligands trigger the internalization of FLS2, EFR and PEPR1, respectively. Combined ligand treatments revealed that FLS2 co-localizes with PEPR1 at endosomes suggesting convergent endocytic trafficking of the PRRs. Moreover, we showed that the ligand-induced endosomal pool of FLS2 co-localize with the endosomal pool of BRI1 receptors, suggesting here a convergent endocytic trafficking between PRRs and non-PRRs. Taken together, our data suggests that ligand-dependent internalization RLKs occurs via a conserved endocytic pathway. In our current research we are focusing on endocytic regulators and motifs to further understand the mechanisms underlying this response and its potential role in signaling.

THE PROTEIN KINASE OXI1 REGULATES SALICYLIC ACID-DEPENDENT PLANT IMMUNITY AND CELL DEATH PROGRAMS IN ARABIDOPSIS

Ana Victoria Garcia* (URGV, Evry, France), Celine Forzani (Max Perutz Laboratories, Vienna, Austria), Kohki Yoshimoto (IJPB, Versailles, France), Claudine Balagué (LIPM, Toulouse, France), Eduardo Bueso (URGV, Evry, France), Jean Colcombet (URGV, Evry, France), Heribert Hirt (URGV, Evry, France).

garcia@evry.inra.fr

Whereas animal AGC kinases are key regulators of growth and cell death programs in response to environmental stresses, little is known about the function of AGC kinases in these processes in plants. Arabidopsis OXI1 is a Serine/Threonine kinase that belongs to the AGC family and is rapidly activated by oxidative stress and various signals mimicking pathogen attack (Garcia *et al*. 2012, Rentel *et al*. 2004). OXI1 is required for complete resistance against biotrophic pathogens and for the growth promotion conferred by the basidiomycete *Piriformospora indica* (Camehl *et al*. 2011, Garcia *et al*. 2012). Furthermore, *oxi1* mutant plants display reduced activation of the stress-induced MAP kinases MPK3 and MPK6 (Rentel *et al*. 2004). To further assess the function of Arabidopsis OXI1, we generated transgenic lines expressing tagged *OXI1* under its own promoter and selected lines with different levels of protein accumulation. Our results show that OXI1 over expression leads to age-dependent developmental defects, including reduced growth and severe leaf necrosis in adult plants. The appearance of necrotic lesions is associated with the misregulation of MAP kinase activities, over accumulation of reactive oxygen species and the defense hormone salicylic acid and the transcriptional reprogramming of defense and cell death regulatory genes. Furthermore, OXI1 over expressing lines display increased disease resistance to virulent *Pseudomonas syringae* pv *tomato* DC3000 (*Pst*) and avirulent *Pst* expressing AvrRps4 or AvrRpm1. Interestingly, we observe a misregulation of the cell death induced by avirulent pathogens and are currently investigating the underlying mechanism. In sum our results suggest that, similarly to the function of AGC kinases in animals, OXI1 plays an important role in controlling plant growth, disease resistance and cell death programs in Arabidopsis.

SESSION 2:

ABIOTIC INTERACTIONS

AMBIENT TEMPERATURE SENSING IN PLANT DEVELOPMENT

Philip A. Wigge*, Manoj Kumar, Matthew Box, Jaehoon Jung, Seong Jeon Yoo, Varodom Charoensawan, Anna Brestovitsky, David Schoepfer, Patrick Dickinson, Emma Sedivy, Sandra Cortijo, Yanniv Dorone (Sainsbury Laboratory Cambridge University, Cambridge, UK)

philip.wigge@slcu.cam.ac.uk

Plant development is highly responsive to ambient growth temperatures. Typical responses to warmer ambient temperatures in Arabidopsis include increased hypocotyl elongation and early flowering time. Research from multiple research groups has shown a key role for the bHLH transcription factor PIF4 in mediating warm temperature responses, but an outstanding question is how warm temperature information is integrated into PIF4 signalling activity [1-3]. We are currently pursuing two approaches to identify new components involved in temperature sensing. In a forward genetic approach, we are using a reporter line, *HSP70::LUC* in a large scale mutagenesis screen to identify mutants involved in temperature perception [4]. Secondly, we are using natural variation to identify differential responsiveness to temperature in growth. We will present initial results of these screens and our current working models for temperature perception in plants.

1. Gray, W.M., et al., *High temperature promotes auxin-mediated hypocotyl elongation in Arabidopsis.* Proc Natl Acad Sci U S A, 1998. **95**(12): p. 7197-202.
2. Koini, M.A., et al., *High temperature-mediated adaptations in plant architecture require the bHLH transcription factor PIF4.* Curr Biol, 2009. **19**(5): p. 408-13.
3. Kumar, S.V., et al., *Transcription factor PIF4 controls the thermosensory activation of flowering.* Nature, 2012. **484**(7393): p. 242-5.
4. Kumar, S.V. and P.A. Wigge, *H2A.Z-Containing Nucleosomes Mediate the Thermosensory Response in Arabidopsis.* Cell, 2010. **140**(1): p. 136-140.

NIN-LIKE PROTEINS: KEY REGULATORS OF PLANT RESPONSES TO NITROGEN AVAILABILITY

Chloé Marchive[1], François Roudier[2], Charlotte Renne[1], Yves Texier[1], Loren Castaings[1], Virginie Bréhaut[1], Camille Chardin[1], Eddy Blondet[3], Vincent Colot[2], Françoise Vedele[1], Christian Meyer[1], and Anne Krapp[1]*

[1] IJPB, INRA, France ; [2] ENS, CNRS, France ; [3] URGV, INRA, France

anne.krapp@versailles.inra.fr

Nitrogen is an essential macroelement for plant growth. Nitrate, beside its role as nutrient, acts as a signal molecule for triggering many adaptive responses to changes in N availability. How such nitrate specific mechanisms are regulated at the molecular level is poorly understood. We identified NIN-like protein 7 (NLP7), a member of the RWP-RK family of putative transcription factors, as an important element involved in the adaptation to N availability. *Nlp7* knockout mutants constitutively display several traits of nitrogen starved plants and *NLP7* expression pattern is consistent with a function in the sensing of N and translational fusions with the green fluorescent protein (GFP) show a nuclear localization for NLP7. Indeed, immediately after nitrate exposure, NLP7 accumulates in the nucleus and binds dozens of genes involved in nitrate signalling and assimilation leading to an altered response of many nitrate-regulated genes in the *nlp7* mutant background. Altogether, we propose NLP7 as a master regulator of early nitrate signalling.

UNRAVELLING DROUGHT RESPONSES IN ARABIDOPSIS THALIANA USING A WORLD-WIDE NATURAL POPULATION

Johanna Molenaar[1,2]*, Christine Granier[3], Dick Vreugdenhil[2], Joost Keurentjes[1]

[1]Laboratory of Genetics, Wageningen University, Wageningen, The Netherlands; [2]Laboratory of Plant Physiology, Wageningen University, The Netherlands; [3]Laboratoire d'Ecophysiologie des Plantes sous Stress Environnementaux, LEPSE-INRA, Montpellier

johanna.molenaar@wur.nl

Drought is one of the major causes of yield loss in agriculture. For centuries therefore breeders have tried to develop crops that are able to cope with water limited conditions without a yield penalty in both optimal and limiting conditions. To reach this goal it is useful to know the physiological mechanism behind the response of plants to drought and to have information on the genes involved in its regulation. We have grown a world-wide natural population of *Arabidopsis thaliana* in the Phenopsis Phenotyping Platform under control and moderate drought conditions. The drought response was registered by taking pictures twice a day and by determining the water content after 3,5 weeks. We found a large variation in drought response between the different accessions. For most accessions a reduction in rosette fresh weight was observed (up to 60%), but for some accessions we could not detect a measurable response to the drought. Genome wide association (GWA) mapping was performed (215k SNPs) on various drought related traits. More than 500 significantly associated unique SNPs were detected, distributed over the whole genome. Per chromosome between 9 and 18 regions of interest could be identified in which significant SNPs were located in close proximity. Candidate genes, underlying these regions, will be investigated to confirm their role in the drought response. Subsequent translation of findings towards crop species will enable marker-assisted breeding for varieties with a high performance even if a drought period is experienced.

SWEET CHERRY PHENOLOGY IN THE CONTEXT OF CLIMATE CHANGE: A SYSTEMS BIOLOGY APPROACH

Bénédicte Wenden*, José Antonio Campoy, José Quero-Garcia, Sophie Castède, Loïck Le Dantec, Teresa Barreneche and Elisabeth Dirlewanger

INRA, UMR 1332 Biologie du Fruit et Pathologie, Villenave d'Ornon, France
Univ. Bordeaux, UMR 1332 Biologie du Fruit et Pathologie, Villenave d'Ornon, France

bwenden@bordeaux.inra.fr

In temperate fruit trees, most key phenological stages are highly dependent on environmental conditions. In particular, correct timing for dormancy and flowering is essential to ensure good fruit production and quality. As a result, in a swiftly-changing environment, temperate fruit crop adaptation in many areas will be at risk in the coming decades. Global changes in environmental conditions include warmer winters and higher risks of frosts in the early spring, leading to a wide range of problems: flower and fruit set, sun-scald, cross-pollination or novel host-pest interaction.

With the final aim of developing ideotypes adapted to future environmental conditions, we present a research approach integrating phenomics and genomics into a predictive model for dormancy and flowering in a non-model fruit species that is sweet cherry. Phenology data from a wide range of sweet cherry genetic resources including flowering dates from all over Europe (supported by the COST action on sweet cherry that INRA-Bordeaux is leading), are being analysed to extract the main trends and select the best phenology models. In addition, a candidate gene research, related to bud dormancy and flowering, is on-going, associated with analyses from a sweet cherry RNAseq. Selected genes are further being studied by qRT-PCR performed on dormant buds and various tissues.

These results will allow formulating hypotheses related to the signalling pathways involved in the sweet cherry flowering response to environmental conditions. In a long term perspective, the predictive model will be built based on mathematical equations derived from these hypotheses.

DISSECTING ROOT ARCHITECTURE ADAPTATION TO PHOSPHATE STARVATION.

Benjamin Péret* (CEA Cadarache, France), Laurent Nussaume (CEA Cadarache, France), Thierry Desnos (CEA Cadarache, France)

benjamin.peret@cea.fr

The plasticity of plant developmental programs is incredibly complex. The root system is a fantastic model to study plant plasticity as the number and location of lateral roots is highly variable and strongly affected by the local environment. Amongst the numerous stimuli that can affect root architecture, our group focuses on the effect of phosphate (Pi) availability. Not only phosphate is an essential nutrient that is present in fundamental molecules such as ATP, DNA, RNA and phospholipids; it is also characterized by a low mobility in soil. As a result of its adsorption by cations, only a fraction of the total soil phosphate is readily available for uptake by root systems. In order to cope with these limitations, plants have developed strategies to perceive Pi, adapt their development and modify their physiology. At the root level, phosphate starvation triggers a rapid arrest of primary root growth, an induction of lateral root formation and elongation as well as production of numerous and longer root hairs. It has been suggested that modification of auxin fluxes due to primary root growth inhibition was responsible for lateral root induction. In this presentation, I will provide evidence that this is not the case. Using different approaches, I was able to uncouple these two responses. Manipulation of external versus internal Pi concentration and tissue specific complementation of a mutant affected in root responses to low Pi provided the evidence for two distinct sites of Pi perception in roots.

THE EXTREMOPHILE GRASS AELUROPUS LITTORALIS: A SOURCE OF CANDIDATE GENES FOR IMPROVING SALT AND DROUGHT STRESSES IN CEREALS

Walid Ben Romdhane[1,3], Rania Ben Saad[3], Adullah Al-Doss[1], Emmanuel Guiderdoni[2], Afif Hassairi[1,3]*

[1] King Saud University, college of Food and Agricultural sciences, PO box 2460, 11451 Riyadh, Saudi Arabia
[2] CIRAD, UMR AGAP, TAA108/03 34398 Montpellier cedex 5 France
[3] University of Sfax, Centre of Biotechnology of Sfax (CBS), LPAP, POBox 1117 3018 Sfax, Tunisia.

Drought and salt stresses are the major limiting factor of plant growth and crop yields. Tomorrow's plants will be required to yield more in spite of less favorable conditions and to limit the environmental impact of cultivation. To resolve the problem beside the use of traditional plant breeding, direct introduction of single or multiple genes into crops appears to be an attractive alternative. *The* C4 perennial halophyte monocotyledonous plant *Aeluropus littoralis* (Gouan) Parl exhibits high tolerance to salinity and drought. Thus, *A. littoralis* has the potential to become an important genetic resource for biotechnological strategies to improve abiotic stress tolerance in economically important crops such as wheat, barley, rice, maize... This will be facilitated by a small diploid (2n = 2x =20) genome of around 332 Mb.

We have constructed Suppression Subtractive Hybridization (SSH) cDNA libraries from root and leaf tissues of C4 perennial halophyte monocotyledonous plant *Aeluropus littoralis* grown in the presence of 300 mM NaCl. We have also constructed full-length cDNA library from salt stressed (300 mM NaCl) roots during 15 days. From these libraries we have isolated and sequenced 492 independent transcripts differentially expressed. Comparison using BlastX, 335 (68%) ESTs (Expressed Sequence Tag) were classified into putative known functions and unclassified proteins, 59 (12%) have homology only to unidentified homologous sequences. A total of 98 (20%) of the ESTs have no homologies to known sequences in the protein databases which can be considered as novel. We described here an example of a novel gene, *AlSAP*, and its promoter isolated from *A. littoralis*. The *AlSAP* gene has an intron at its 5'UTR. The AlSAP protein is characterized by the presence of two conserved zinc-finger domains A20 and AN1. *AlSAP* is induced not only by various abiotic stresses such as salt, osmotic, heat and cold but also by abscisic acid (ABA) and salicylic acid (SA). We generated tobacco plants expressing the *AlSAP* gene under the control of the duplicated CaMV35S promoter. These plants exhibited an enhanced tolerance to abiotic stresses. We found that the steady state levels of transcripts of eight stress-related genes were higher in *AlSAP* transgenic lines than in wild-type tobacco. To extend these findings to important crops, we generated marker-free transgenic durum wheat of the commercial cv. Karim and transgenic rice plants expressing the AlSAP gene. AlSAP wheat lines exhibited improved germination rates and biomass production under severe salinity and osmotic stress conditions. Following a long-term salt or drought stress greenhouse trial, AlSAP lines produced normally filled grains whereas wild-type (WT) plants either died at the vegetative stage under salt stress or showed markedly reduced grain filling under drought stress. Measurements of the RWC (relative water content) and endogenous Na^+ and K^+ levels in leaves of AlSAP plants, showed a lower water loss rate and a higher Na^+ accumulation in senescent-basal leaves, respectively, compared to those of WT plants. The expression of *AlSAP,* in rice cv. Nipponbare, enhances plant tolerance to cold, drought and salt stresses. Under a severe drought stress treatment (Fraction of Transpirable Soil Water down to 0.1), *AlSAP* lines exhibited enhanced *TE* (Transpiration Efficiency) and maintained a high *A* (Assimilation rate) value (22 µmol.m-2s-1) while these values dramatically decreased (*A*= 4 µmol.m-2s-1) in control plants which were subsequently unable to recover from the stress. Noteworthy, *AlSAP* rice plants yielded a similar and a 60% seed set under control and stress conditions respectively with regard to WT plants grown under control conditions. This indicates that *AlSAP* expression imposes no yield penalty and allows seed production even following a severe drought stress at the vegetative stage. Furthermore, *AlSAP* rice was shown to accumulate transcripts of a pilot set of eight stress-related genes at a significantly higher level than WT plants, both under control and stressed conditions. The results suggest that *AlSAP* expression generates stress tolerance in plants through maintenance of the photosynthetic apparatus integrity and by stimulating an endogenous adaptive potential which is not effectively accomplished in WT plants.

HOW DROUGHT AFFECTS LEAF GROWTH: ERF5/6 AND DELLAS ACT TOGETHER TO REGULATE GROWTH INHIBITION UNDER STRESS

Marieke Dubois* (VIB, Ghent, Belgium), Hannes Claeys (VIB, Ghent, Belgium), Aleksandra Skirycz (VIB, Ghent, Belgium), Katrien Maleux (VIB, Ghent, Belgium) and Dirk Inzé (VIB, Ghent, Belgium)

madub@psb.vib-ugent.be

Plant growth is a complex process that is continuously fine-tuned by the environment. Various abiotic stresses such as mild drought have been shown to alter leaf growth, but the underlying mechanisms remained largely unknown. Here, we describe the molecular mechanisms regulating osmotic stress-induced growth inhibition in Arabidopsis. We previously found that the ethylene precursor ACC is a very important early stress signal in developing leaves. Here, we show that ACC leads, through the MPK3/6 phosphorylation cascade, to the activation of the ethylene response factors ERF5 and ERF6, which act as master regulators of both growth inhibition by and tolerance to stress. Using inducible overexpression lines, it was found that they induce many stress tolerance genes, such as *STZ*, *MYB51* and *WRKY33*, but that also the gibberellin (GA)-degrading enzyme GA2OX6 is induced. The resulting decline in levels of the growth-stimulating GAs leads to stabilization of DELLA proteins, which are then responsible for mitotic exit and early onset of endoreduplication. Interestingly, the activation of the stress tolerance genes by ERF6 occurs independently from the ERF6-mediated growth inhibition. Together, these data fit into a leaf growth regulatory model in which ERF5 and ERF6 form the missing link between the previously observed stress-induced ethylene accumulation and DELLA-mediated cell cycle exit and execute a dual role by regulating both stress tolerance and growth-inhibition.

SESSION 3:

BIODIVERSITY AND NATURAL VARIATION

NATURAL VARIATION IN LIFE HISTORY RESPONSES TO CLIMATE IN *ARABIDOPSIS THALIANA*

Johanna Schmitt* (University of California, Davis, USA)

jschmitt@ucdavis.edu

The environmental signaling pathways controlling flowering time are well studied in the annual plant species *Arabidopsis thaliana*, and a number of quantitative trait loci (QTL) and candidate genes have been identified for flowering time variation. However, less is known about how natural variation at these loci is expressed in different natural seasonal environments or different climates across the species range. By growing selected recombinant inbred lines (RILs) in the field, we can measure the effects of known QTL on flowering time, life history expression (winter annual or rapid cycling), fitness, and population dynamics. The effects of known functional polymorphisms, notably loss-of-function alleles of *FRIGIDA* and *MAF2/3*, differ dramatically in their effects on flowering time and fitness across four European field sites. We also observe epistatic effects of these loci on flowering time and fitness in seasonal cohorts in New England. These effects depend strongly upon the timing of germination; different genotypes exhibit autumn-flowering vs. spring-flowering life histories in successive seasonal cohorts. Consequently, genotypes segregating different combinations of flowering time and seed dormancy QTL differ dramatically in life history phenology and population dynamics.

GENETIC CONTROL OF LEAF SHAPE

Patrick Laufs[*,1,2], Bernard Adroher[1,2], Nicolas Arnaud[1,2], Eric Biot[1,2], Thomas Blein[1,2], Arezki Boudaoud[3], Jasmine Burguet[1,2], Millan Cortizo[1,2], Alice Hasson[1,2], Annamaria Kiss[3], Marie-Capucine Lepeigneux[1,2]

[1] INRA, UMR1318, IJPB, Versailles, France,
[2] AgroParisTech, IJPB, Versailles, France,
[3] ENS, RDP Lyon, France

patrick.laufs@versailles.inra.fr

Leaves show a tremendous diversity in their sizes and shapes. Though, they all originate as small, finger-shaped primordia at the flanks of stem cells-containing groups of undifferentiated cells, the meristem. Leaf shape is established later during development, mainly as a result of differential growth of their margins. The formation of complexe shapes, such as those found in compound leaves that are formed by several leaflets, requires a coordinated delay of cell differentiation and the initiation of new growth axes in the young leaf primordium.
Here, we will present recent advances in the understanding of the regulatory networks controlling leaf development and how variations in these networks may have contributed to changes in leaf shape. We will exemplify this by looking at the roles of the *CUP-SHAPED COTYLEDON* genes during the development of simple and compound leaves.

EXPLOITING NATURAL VARIATION IN SEED MUCILAGE CHARACTERISTICS TO IDENTIFY NOVEL GENES INVOLVED IN ITS PRODUCTION AND FUNCTION

Helen North* (Institut Jean-Pierre Bourgin, Versailles, France), Marie-Christine Ralet (BIA, INRA, France), Susana Saez-Aguayo (Institut Jean-Pierre Bourgin, Versailles, France), Adeline Berger (Institut Jean-Pierre Bourgin, Versailles, France), Corinne Rondeau (IRSTEA, Rennes, France), Joseph Tran (Institut Jean-Pierre Bourgin, Versailles, France), Olivier Loudet (Institut Jean-Pierre Bourgin, Versailles, France), Juliette de Meaux (University of Münster, Germany), Christine Sallé (Institut Jean-Pierre Bourgin, Versailles, France), Annie Marion-Poll (Institut Jean-Pierre Bourgin, Versailles, France)

helen.north@versailles.inra.fr

Mucilage is a hydrogel formed from polysaccharides released from imbibed seeds. These polysaccharides are accumulated in the epidermal cells of the seed coat during seed development. The role of this mucilage remains to be determined, but its absence is not detrimental in laboratory conditions, making it an excellent model for the study of polysaccharide properties. We have previously shown that Arabidopsis mucilage is formed of a mixture of polysaccharides in two structurally distinct layers, both of which are mainly composed of the pectin rhamnogalacturonan I (Macquet *et al.,* 2007a). In order to gain insight into the function of seed mucilage, we have analysed over 300 Arabidopsis accessions for natural variation in mucilage release or soluble mucilage properties. Ten accessions were identified that do not release mucilage and whose seeds float. These accessions were affected in three different loci and eight independent mutations. A further four floating mucilage releasing (FMR) accessions were identified that are affected in another gene; all fourteen accessions originate from two specific geographic regions. Low-field NMR analysis of water mobility in and around seeds showed that imbibition is slower in seeds that produce mucilage, and that the function of released mucilage is not to improve the hydration rate of internal seed tissues. We propose that mutations causing seed mucilage to be produced and not released provide a selective advantage for seed dispersal by water. Two of the affected genes have been characterised and correspond to proteins that play a role in pectin maturation (Macquet *et al*., 2007b; Saez-Aguayo *et al*., 2013). Characteristics of soluble mucilage in accessions where it is released show little variability in mucilage composition; in contrast the amount and physicochemical properties of mucilage vary widely. Analyses have been carried out to link these variations with particular habitats.

THE SUBTELOMERIC *KHIPU* SATELLITE REPEAT FROM *PHASEOLUS VULGARIS*: LESSONS LEARNED FROM THE GENOME ANALYSIS OF THE ANDEAN GENOTYPE G19833.

Manon Richard* (Institut de Biologie des Plantes, Orsay, France), Nicolas Chen (Institut de Biologie des Plantes, Orsay, France), Vincent Thareau (Institut de Biologie des Plantes, Orsay, France), Stéphanie Pflieger (Institut de Biologie des Plantes, Orsay, France), Sophie Blanchet (Institut de Biologie des Plantes, Orsay, France), Andrea Pedrosa-Harand (Laboratory of Plant Cytogenetics and Evolution, Recife, Brazil), Aiko Iwata (Center for Applied Genetic Technologies and Institute for Plant Breeding Genetics, and Genomics, Athens, USA), Carolina Chavarro (Center for Applied Genetic Technologies and Institute for Plant Breeding Genetics, and Genomics, Athens, USA), Scott Jackson (Center for Applied Genetic Technologies and Institute for Plant Breeding Genetics, and Genomics, Athens, USA), Valérie Geffroy (Institut de Biologie des Plantes, Orsay, France)

manon.richard@u-psud.fr

Subtelomeric regions in eukaryotic organisms are known for harboring species-specific tandemly repeated satellite sequences. However, studies on the molecular organization and evolution of subtelomeric repeats are scarce, especially in plants. *Khipu* is a satellite DNA of 528bp repeat unit, specific of the *Phaseolus* genus, with a subtelomeric distribution in common bean, *P. vulgaris*. To investigate the genomic organization and the evolution of *khipu*, we performed genome-wide analysis on the complete genome sequence of the common bean genotype G19833. We identified 2,460 *khipu* units located at most distal ends of the sequenced regions. *Khipu* units are arranged in discrete blocks of 2-55 copies and are heterogeneously distributed among the different chromosome ends of G19833 (from 0 to 555 *khipus* units per chromosome arm). Phylogenetically related *khipu* units are spread between numerous chromosome ends, suggesting frequent exchanges between nonhomologous subtelomeres. However, most subclades contain numerous *khipu* units from only one or few chromosome ends indicating that local duplication is also driving *khipu* expansion. Unexpectedly, we also identified 81 *khipu* units located at centromeres. All the centromeric *khipu* units belong to a single divergent clade also comprised of a few units from several subtelomeres, suggesting that a few sequence exchanges between centromeres and subtelomeres took place in the common bean genome. The divergence and low copy number of these centromeric units from the subtelomeric units could explain why they were not detected by FISH (Fluorescence *in situ* Hybridization) although it cannot be excluded that these centromeric units may have resulted from errors in the pseudomolecule assembly. Altogether our data highlight extensive sequence exchanges in subtelomeres between non-homologous chromosomes in common bean and confirm that subtelomeres represent one of the most dynamic and rapidly evolving regions in eukaryotic genomes.

DE NOVO TRANSCRIPTOME SEQUENCING OF THE NICKEL HYPERACCUMULATOR *PSYCHOTRIA GABRIELLAE* AND IDENTIFICATION OF PgIREG1 AS A CANDIDATE NICKEL TRANSPORTER INVOLVED IN ACCUMULATION.

Sylvain Merlot[1,2]*, Laure Hannibal[2], Sara Martins[1], Hamid Amir[3] Michel Lebrun[4], Sébastien Thomine[1].

[1] Institut des Sciences du Végétal, Labex SPS, CNRS, Gif-sur-Yvette cedex, France.
[2] IRD, Laboratoire des Symbioses Tropicales et Méditerranéennes, Centre IRD Nouméa, New Caledonia.
[3] Laboratoire Insulaire du Vivant et de l'Environnement, Université de la Nouvelle-Calédonie, Nouméa, New Caledonia.
[4] Université Montpellier 2, Laboratoire des Symbioses Tropicales et Méditerranéennes, Montpellier, France.

sylvain.merlot@isv.cnrs-gif.fr

Nickel is an economically important metal and phytotechnologies such as phytoremediation and phytomining are developed to limit the impact of nickel mining on the environment. About 400 plant species scattered in 40 families are known to hyperaccumulate nickel. These hyperaccumulators and the underlying mechanisms of metal accumulation are receiving an increasing interest because of their potential use in sustainable phytotechnologies. However, despite the large diversity of nickel hyperaccumulators, our knowledge of the mechanisms of nickel accumulation is limited and restricted to temperate herbaceous Brassicaceae.
In order to broaden our knowledge of nickel accumulation, we used NGS technology to sequence the leaf transcriptome of *Psychotria gabriellae* (previously *P. douarrei*), a tropical nickel hyperaccumulator of the Rubiaceae family endemic from New Caledonia. More than 30.000 contigs were obtained after *de novo* assembly of the reads and about 18.500 contigs received a Gene Ontology annotation. Among candidate genes involved in nickel homeostasis, we characterized the activity of metal transporters from the NRAMP and IREG/FPN families. PgIREG1 expression in yeast increases nickel resistance by exporting nickel out off cells. In plants, PgIREG1 fused to GFP localizes in the tonoplast. PgIREG1 expression complements the nickel hypersensitive phenotype of the Arabidopsis *ireg2* mutant and increases nickel resistance compared to wild type. These results suggest that PgIREG1 might be involved in nickel accumulation in *P. gabriellae*. To support this hypothesis, we will analyze the expression of this gene in species related to *P. gabriellae* that do not accumulate nickel.

ABUNDANCE OF TRANSPOSBLE ELEMENTS AFFECTED BY MATING SYSTEM IN *ARABIDOPSIS LYRATA*

Georgi Bonchev* (University of Neuchâtel, Neuchâtel, Switzerland), Yvonne Willi (University of Neuchâtel, Neuchâtel, Switzerland)

georgi.bonchev@unine.ch

Transposable elements (TEs) comprise a substantial and dynamic part of plant genomes, yet the ultimate drivers of their abundance are still unknown. The abundance of TEs across the genome was proposed to be influenced by the host's mating system via its effect on ectopic recombination and effective population size. In this study, we compared TE abundance among North American populations of *Arabidopsis lyrata* varying in the mating system from predominantly outcrossing to predominantly selfing. We screened 10 individuals of 18 populations for fractions of TEs of 6 transposon families by Transposon Insertion Display (TID). TID is based on amplification of DNA fragments with one end being in a conserved TE region and the other end being a restriction cutting site usually lying outside of the TE. The three main results were: Among-population variation in TE bands was generally higher than within-population variation, presumably due to low gene flow among populations. Analysis of genetic distances revealed a split between eastern and western populations, clusters that were also found for microsatellites. Finally and most importantly, TE diversity was similar between outcrossing and selfing populations, completely in contrast with results based on microsatellites, suggesting that despite selfing populations having smaller effective population sizes, they seem to accumulate TEs. This supports the hypothesis that TEs are mostly slightly deleterious and accumulate under inbreeding.

NATURAL DIVERSITY IN THE MODEL LEGUME MEDICAGO TRUNCATULA AND THE FUNGAL PATHOGEN VERTICILLIUM SP. ALLOWS IDENTIFYING DISTINCT GENETIC MECHANISMS FOR RESISTANCE TO VERTICILLIUM WILT

Laurent Gentzbittel* (ECOLAB UMR 5245, INP-ENSAT, Toulouse, France), Cécile Ben (ECOLAB UMR 5245, INP-ENSAT, Toulouse, France), Samy Benameur (ECOLAB UMR 5245, INP-ENSAT, Toulouse, France), Maoulida Toueni (ECOLAB UMR 5245, INP-ENSAT, Toulouse, France), Azam Negahi (ECOLAB UMR 5245, INP-ENSAT, Toulouse, France), Ahmad Sarrafi (ECOLAB UMR 5245, INP-ENSAT, Toulouse, France), Marie-Claire Tardin (R2n, Druelle, France), Abed Sbeiti (ECOLAB UMR 5245, INP-ENSAT, Toulouse, France), Jean-Marie Prospéri (INRA, UMR AGAP 1334, Montpellier, France), Marie-Christine Gras (R2n, Druelle, France), Martina Rickauer (ECOLAB UMR 5245, INP-ENSAT, Toulouse, France)

gentz@ensat.fr

Verticillium wilt causes substantial yield losses in many crops including alfalfa (*Medicago sativa*). The model legume *Medicago truncatula*, a wild species which presents well-established genetic resources and high biodiversity was used as a host for studying resistance and susceptibility to *Verticillium albo-atrum* and *V. dahliae*.

The capacity of pathogenic fungi to adapt to new environments and hosts is a well-known threat to durability of resistant populations. In order to study the adaptive potential of *Verticillium* strains, the disease response of 25 *M. truncatula* genotypes was evaluated after root-inoculation with six *V. albo-atrum* and *V. dahliae* strains isolated from diverse plant species. Highly significant differences were observed among plant genotypes and fungal strains. This highlights that *M. truncatula* has a wide spectrum of susceptibility/resistance to *Verticillium* and suggests that the host range of this root pathogen may evolve.

To investigate the genetic variability of susceptibility to Verticillium within the *M. truncatula* species, the response to the most aggressive strain V31.2 was assessed in a collection of 237 lines from contrasted pedoclimatic regions. Disease functional parameters and plant colonization by the fungus varied largely among the lines. This biodiversity with regard to disease response encourages the development of association genetics and ecological approaches.

Further genetic analysis by a multi-cross, multi-strain, multi-site design revealed only few major QTLs suggesting that simple but distinct genetic mechanisms control Verticillium wilt resistance in different *M. truncatula* lines.

To further decipher the molecular mechanisms responsible for *M. truncatula* resistance to *V. albo-atrum*, transcriptomic studies comparing compatible and incompatible interactions were carried out. Data obtained with NGS technologies revealed candidate genes likely to play key roles in different crucial steps for resistance/susceptibility to Verticillium wilt.

SESSION 4:

TRANSLATIONAL BIOLOGY

THE MAKING AND USE OF CHROMOSOME SUBSTITUTION LINES IN PLANT BREEDING: COMPARISON OF ISOGENIC HYBRIDS AND THEIR RESPECTIVE METHYLATION LANDSCAPE.

Sara Movahedi[1], Laurens Deurhof[2], Erik Wijnker[3], Kees van Dun[1], Bastiaan de Snoo[1], Joost Keurentjes[2], Remco Ursem[1], Hans de Jong[2] and Rob Dirks[1*]

1 Rijk Zwaan Breeding, Fijnaart, The Netherlands.
2 IBMP, Strasbourg, France
3 Laboratory of Genetics, Wageningen University

r.dirks@rijkzwaan.nl

By suppressing recombination during meiosis and the subsequent regeneration of plants from spores that by chance obtained a full set of chromosomes, chromosome substitution lines are made.
Chromosome substitution lines, and especially libraries, offer a wealth of possibilities to study complex traits.
Epistatic interactions can easily be unraveled and genotypes can be reconstructed.
Isogenic hybrids can be made that cannot be reconstructed otherwise. By comparing the isogenic hybrids, the contribution of epi-genetics and genetics to a certain phenotype can be assessed, mapped and quantified. Methylation landscapes of isogenic hybrids and their unique parental lines can be studied which in turn addresses the relevance of methylation profiles.
Partial reconstruction of hybrids (sub-hybrid populations) allows the mapping of the contribution of every chromosome and chromosome combination towards the original hybrid.

TRANSLATIONAL RESEARCH, URGV EXPERIENCE

Abdel Bendahmane*, Marion Dalmais, Christelle Troadec, Fabien Marcel, Adnane Boualem (UMRGV, Evry, France)

bendahm@evry.inra.fr

National and international institutions have been engaged in large programs aimed at improving the nutritional or functional properties of the harvested plant for use in food, animal feed, or industrial products. This far, these efforts have been mainly carried out through classical breeding that involves crossing of chosen parental lines and subsequently selecting the offspring with the desired traits. At the molecular level, this strategy exploit genetic recombination, and thus it's limited to the same or closely related species at the best. Beside, crossing result in the introduction of unwanted traits together with the desired ones. Cleaning of the genetic material from the unwanted traits is usually hampered by the low frequency of recombination events and the large number of the needed backcrosses to purify the genetic materials. One of the major aims of plant biotechnology is the genetic engineering of improved crop plants. With this aim in mind, large-scale genome investigation projects have been undertaken in the model plant Arabidopsis and many important crop species, including rice, maize, soybean and tomato. Knowledge about plant growth, development and the molecular compositions of plant organs has increased tremendously, and the genes that control the function of the biological mechanisms involved also are in many cases identified. Consequently, the enormous amount of generated information has intensified the need for methods that permit to generate targeted genetic modification *in planta*. In model species such as *Physcomitrella*, yeast, mice and *Drosophila*, targeted gene modification is a routine tool for generating specific DNA sequence alterations that permit a greater understanding of gene function. Targeted gene modification also holds great promise for crop improvement. The most widely used gene targeted modification strategy use homologous recombination. In higher plants the efficiency of this method is extremely low and is thought to result from the double strand DNA breakage repair mechanisms that occur mainly through nonhomologous end joining. Based on this observation, targeted genetic modification strategies via nonhomologous end joining have been experimentally tested. While, these methods we shown to lead to induced frame shift mutations in the targeted genes, the efficiency is low and inapplicable in species recalcitrant to plant transformation. Besides, these methods do not allow with high efficiency site directed mutagenesis of a given nucleotide (11). Furthermore, when feasible, the time laps between the decision to mutate a given nucleotide and obtaining the mutate seed is at least three years. This duration is too long in the current molecular breeding schemas. On the other hand chemical mutagenesis combined with the TILLING approach is a straightforward and cost-effective way to identify induced alleles in a given sequence. To establish such tools in crop species, we have constructed reference mutant populations, developed HTP tools for rapid and systematic identification of mutations in target sequences and constructed a database, UTILLdb that contains phenotypic as well as sequence information on mutant genes. Our progresses in engineering leader alleles in different crop species will be presented.

TEMPERATURE-SENSITIVE EXPRESSION OF *LsNCED4* ENCODING AN ABA BIOSYNTHETIC ENZYME IS REQUIRED FOR THERMOINHIBITION OF LETTUCE SEEDS

Kent J. Bradford*, Heqiang Huo, Peetambar Dahal, Jason Argyris, Sebastian Reyes Chin-Wo, Fei-Yian Yoong, Richard Michelmore (University of California, Davis, CA, USA) and Claire McCallum (Arcadia Biosciences, Davis, CA, USA)

kjbradford@ucdavis.edu

Thermoinhibition, or failure of seeds to germinate when they are imbibed at temperatures above ~30ºC, is a common phenomenon in commercial lettuce (*Lactuca sativa*) cultivars such as Salinas. In contrast, seeds of an accession of *L. serriola* (UC96US23) do not exhibit thermoinhibition. Genetic analysis of recombinant inbred lines (RIL) derived from a Salinas by UC96US23 cross revealed that a QTL associated with high temperature germination (*Htg6.1*) contained *LsNCED4*, a key regulated gene in ABA biosynthesis. *LsNCED4* expression is elevated during late seed development in both Salinas and UC96US23, but increases (along with ABA content) during imbibition at high temperature only in Salinas seeds. Complementation of Arabidopsis *nced6/9* mutants indicated that *LsNCED4* genes from both genotypes encode functional proteins. Ectopic expression of Salinas *LsNCED4* under its native promoter resulted in thermoinhibition of UC96US23 seeds, whereas Salinas seeds in which *LsNCED4* had been silenced via RNAi germinated to as high as 40ºC. Similarly, a premature stop codon mutation or missense substitution in *LsNCED4* identified via TILLING also increased thermotolerance. Elevated *LsNCED4* expression was also induced in detached Salinas leaves by heat stress (40ºC for 1 h) but not by drought, while two other lettuce NCED genes (*LsNCED2* and *LsNCED3*) exhibited the opposite responses, indicating that NCED gene family members are responsive to distinct environmental signals. Transcriptomic analyses identified gene co-expression modules associated with temperature and ABA regulation of germination. A second thermoinhibition QTL from a different source and on a different chromosome (*Htg9.1*) has been identified, indicating that other genetic mechanisms are also involved. This work illustrates the ability to go from a QTL to a specific gene and develop native and mutant genes and markers to address a crop production limitation. Supported by USDA-NIFA Award 2008-35304-0472 and NSF Award 0820451.

MEIOTIC GENE EVOLUTION: CAN YOU TEACH A (DUPLICATED) OLD DOG NEW TRICK?

Lloyd AH[1,2]*, Ranoux M[3], Vautrin S[4], Glover N[3], Fourment J[4], Charif D[1,2], Choulet F[3], Lassalle G[5], Marande W[4], Tran J[1,2], Granier F[1,2], Pingault L[3], Remay A[3], Marquis C[1,2], Belcram H[6], Chalhoub B[6], Feuillet C[3], Bergès H[4], Sourdille P[3], Jenczewski E[1,2].

[1] INRA, UMR1318, Institut Jean-Pierre Bourgin, RD10, F-78000 Versailles, France
[2] AgroParisTech, Institut Jean-Pierre Bourgin, RD10, F-78000 Versailles, France
[3] INRA, UBP 1095, Genetics Diversity and Ecophysiology of Cereals, 234 Avenue du Brézet, F-63000, Clermont-Ferrand, France.
[4] INRA UPR 1258, Centre National des Ressources Génomiques Végétales, Castanet-Tolosan, France
[5] INRA, UMR1349-IGEPP, Le Rheu Cedex, France.
[6] INRA-CNRS-UEVE, Unité de Recherche en Génomique Végétale, Evry, France.

Andrew.Lloyd@versailles.inra.fr

Meiosis, the basis all breeding programs, evolved through specialization of duplicated DNA metabolism genes. In many economically important crops, whole genome duplication (WGD) has led to additional control of meiotic recombination and the opportunity achieve this altered regulation through diversification of duplicated genes. We show that meiotic genes return to a single copy more rapidly than genome-wide average following angiosperm WGDs and that the rate of loss decreases through time. Meiotic recombination genes in particular, show no evidence for further diversification, and we propose that they are passively lost, highlighting how WGDs may be resolved in the absence of selective duplicate retention. If “you can’t teach an old dog new tricks” it may be because most diploid species already have the tools required to correctly segregate chromosomes in a polyploid state. Meiotic adaption observed in established polyploids, and therefore future manipulation of polyploid recombination or generation of new polyploid crops, may require ‘fine-tuning’ the progression of meiosis or the expression of the underlying genetic architecture rather than mechanistic innovations.

WHAT LIMITS MEIOTIC CROSSOVERS?

Raphael Mercier (IJPB, INRA, Versailles, France)

Raphael.Mercier@versailles.inra.fr

Meiotic crossovers generate genetic diversity by creating new allelic combinations. Intriguingly the meiotic crossover rates are restricted within a very narrow range. A minimum of one crossover is formed per pair of homologous chromosomes per meiosis. This "obligate" crossover ensures correct segregation of the homologs at the first division leading to balanced gametes and ultimately favors fertility of an organism. Crossover frequency per meiosis does not typically vary above of the range of one to three crossovers per chromosome. A great deal has been discovered about how meiotic crossovers are formed. However, very little is known about what limits their formation despite an abundance of molecular precursors. Using a forward genetic screen specifically designed to identify mutants with increased meiotic crossover frequency (3 fold), we revealed that Fanconi Anemia Complementation Group M (FANCM) limits meiotic crossover formation in *Arabidopsis thaliana* by suppressing a normally minor pathway (Crismani et al, Science 2012). Thus, FANCM is a key factor imposing an upper limit on the number of meiotic COs, and its manipulation holds much promise for plant breeding.

ENGINEERING ABSCISIC ACID METABOLISM AND SIGNALING IN PLANTS

Hiro Nonogaki* (Oregon State University, Corvallis, USA), Mariko Nonogaki (Oregon State University, Corvallis, USA), Jessica G. Tran (Oregon State University, Corvallis, USA), Khadidiatou Sall (Oregon State University, Corvallis, USA)

hiro.nonogaki@oregonstate.edu

Abscisic acid (ABA) plays an essential role in plant signaling in a changing environment and is important for seed maturation programs, both of which are important for production of food and biofuel crops. The biosynthesis of ABA is regulated mainly by the rate-limiting enzyme NINE-CIS-EPOXYCAROTENOID DIOXYGENASE (NCED). Therefore, *NCED* genes are potential targets for modification to alter ABA levels in plants. The Plant Gene Switch System (PGSS), a chemically induced gene expression system, has been used to enhance *NCED6* expression in Arabidopsis seeds, which increased ABA levels in seeds more than 20 fold and suppressed seed germination. The same system can be used for conditional expression of *NCED6* in other plant organs, such as leaves and stems, to enhance drought tolerance and other responses of plants to environmental signals. Besides its role as a plant hormone, ABA has been suggested to have therapeutic potential for human health. The *NCED6* PGSS could provide a tool for molecular "pharming" to produce more ABA in plants. In addition, a system to cause spontaneous increase in ABA biosynthesis and signaling in seeds has been tested in our laboratory. This new approach can create a more convenient system to alter ABA metabolism and signaling pathways without an aid of chemical application.

SESSION 5:

ORGANIZATION AND FUNCTIONING OF COMPLEX CROP GENOMES AND TRAITS

GENE, CENTROMERE AND TRANSPOSON EVOLUTION IN MAIZE AND ITS PANICOID RELATIVES

Jeff Bennetzen*, Melanie Buser, Jeremy DeBarry, Taoran Dong, Matt Estep, Zack Farmer, Liang Feng, and Hao Wang (Department of Genetics, University of Georgia, Athens, USA)

maize@uga.edu

Most plant genomes are both highly complex and highly variable. Even within species, dramatic variation can be seen in genome size, mobile DNA content and relative genic organization. In the angiosperms, rates of change for some events, like genome expansion and contraction, are so high that a genome can be completely remodeled within a mere 2-3 million years. Most of this change seems to have little to no effect on gene or genome function, but the exceptions may be key factors in the evolution of novel organismal potential. Our lab has been investigating several components of genome change, primarily within the panicoid grasses (e.g., maize, Setaria, sorghum and teosinte), but using rice and other monocots as outgroups. I will present our results on several aspects of the grass genome evolution story, including centromere rearrangement and loss, gene pair rearrangement, intron gain/loss, the inheritance of "activatable" transposable element families, and the nature of small sequence changes across different domains within the rice genome.

POLYPLOIDY GENERATES TRAIT NOVELTY AND FUNCTIONAL DIVERSITY IN WHEAT

H. Belcram[1], S. Chalabi[1], H. Chelaifa[1], D. Arnaud[1], I. Mestiri[1], E. Floch[2], Xi Liu[2], J. Chiquet[2], C. Deveauchelle[2], C. Rizzon[2], O. Coriton[3], J. Jahier[3], Boulos Chalhoub[1]*

[1]Organisation and Evolution of Plant Genomes, Unité de Recherche en Génomique Végétale (INRA-CNRS-UEVE), Evry, France
[2]Laboratoire Statistique et Génome", UMR 8071, Evry France
[3]UMR1349 INRA-AgroCampus Ouest-Université de Rennes I, Institut de Génétique, Environnement et Protection des Plantes (IGEPP), 35653 Le Rheu, France

chalhoub@evry.inra.fr

In the important bread allohexaploid wheat (*T. aestivum*, AABBDD) crop, three divergent wheat genomes became 'coresident' in a single nucleus, through two successive allopolyploidization events.
In order to evaluate functional diversity and trait novelty with varying Ploidy in wheat, we have characterized several natural and synthetic wheat allopolyploids at the genetic, functional, and phenotypic levels. While most of these were shown to be relatively genetically stable, genome-wide analysis of gene expression showed that majority of genes exhibit same overall expression levels when comparing wheat allopolyploids and diploids suggesting important compensation, regulations and/or interactions between the constituting homoeologous gene copies.
We focused on understanding phenotypic and trait novelty in relation to the fate of duplicated homoeologous genes. Several polyploidy-related traits were studied. In particular, we investigated organization, evolution, and function of homoeologous gene copies of the important *Q/q* gene, known to have played a major role in domestication of polyploid wheat. Combined phenotypic and functional analysis indicate that the evolution of the *Q/q* loci in polyploid wheat resulted in the hyperfunctionalization of one homoeoallele (*5AQ*), a pseudogenization of a second homoeoallele (*5Bq*) that became non-protein coding RNA (npcRNA), but remains functional as affecting phenotype and expression of the other homoeoalleles, and a subfunctionalization of the third copy (*5Dq*), all contributing to the domestication traits. They interacts and crosstalk to each others, probably via epigenetic regulation.
The role of functional regulation and the fate of duplicated homoeologous genes in ingenerating diversity and trait novelty in allopolyploid crops will be discussed.

SESSION 6:

EPIGENETICS

EPIGENETIC VARIATION CONTRIBUTES TO ADAPTATION IN SELECTIVE ENVIRONMENTS

Ueli Grossniklaus* (University of Zürich, Zürich Switzerland), Chrisitian Heichinger (University of Zürich, Zürich Switzerland), Marc Schmid (University of Zürich, Zürich Switzerland), Lindsay Turnbull (University of Zürich, Zürich Switzerland), Bernhard Schmid (University of Zürich, Zürich Switzerland)

grossnik@botinst.uzh.ch

Climate change poses a significant threat to yield security and biodiversity and crop production as it is largely unknown to which degree plants can adapt to a rapidly changing environment. So far, most studies have been limited to study phenotypic responses to selection, while little is known about the underlying genetic or possibly epigenetic basis. In *Arabidopsis* a high rate of spontaneous variation in DNA methylation can occur in the absence of genetic variation and selection (Becker et al, *Nature* 480: 245; Schmitz et al, *Science* 334: 369). It is of great interest to determine whether natural epigenetic variation is subject to selection and contributes to adaptation in selective environments. We analyzed variation in selected phenotypic traits, genome-wide cytosine DNA methylation, and gene expression in two *Arabidopsis* genotypes that had undergone five generations of selection in experimental dynamic landscapes (Fakheran et al, *PNAS* 107: 19120) and compared them to their genetically identical ancestors. Selected populations showed significant differences in flowering time and the number of branches and fruits, differences that were maintained over two to three generations in the absence of selection. Resequencing on the genome of selected plants showed that indeed genetically identical plants, grown in a randomized design in the same environment can display distinct phenotypes. We identified differentially methylated cytosines, which were overrepresented in genes that regulate flowering time, epigenetic processes, development and morphogenesis. Moreover, differentially methylated genes were enriched in differentially expressed genes. Importantly, the same epigenetic changes were observed in independent experiments. Thus, epigenetic variation is subject to selection and may play an important role in the adaptive response of populations in rapidly changing natural environments.

DYNAMIC REGULATION OF THE EPIGENETIC LANDSCAPE BY NON-CODING RNA

M. Crespi*[1], F. Ariel[1], D. Latrasse[2], N. Romero-Barrios[1], T. Jegu[2], F. Bardou[1], A. Christ[1], C. Sorin[1], C. Lelandais[1], C. Hartmann[1] and M. Benhamed[2]

[1]CNRS Institut des Sciences du Végétal, Gif-sur-Yvette and Université Paris Diderot-Paris 7, Paris, France
[2]Institut de Biologie des Plantes, UMR8618 Université Paris-Sud XI, SPS Saclay Plant Sciences, 91405 Orsay France

martin.crespi@isv.cnrs-gif.fr

Eukaryotes exhibit a highly structured epigenome shaped in large part by different types of transcriptional activity. Epigenetic dynamics determine genome topology, conditioning transcriptional responses. Non-protein coding RNAs (npcRNA) represent an emerging class of riboregulators, which act either directly in a long form or are processed to shorter miRNA and siRNAs. They help to determine the epigenetic landscape, although the mechanisms by which they participate in shaping chromosome conformation remain largely unknown.
We have performed genome-wide bioinformatic analysis of transcriptomic databases and identified many *Arabidopsis* long npcRNAs. Eleven npcRNAs were antisense to protein-coding mRNAs whereas certain corresponded to miRNA or siRNA precursors. The expression pattern of a set of long npcRNAs in response to phytohormones and environmental stresses suggested a link with root growth and development. Interestingly certain 24nt siRNAs derived from these loci accumulate under the same conditions, suggesting an epigenetic link. The *Arabidopsis thaliana* long intergenic non-coding RNA *APOLO* is dynamically transcribed by the Pol II and the Pol V complexes in response to auxin. Chromosome conformation capture served to identify the formation of a chromatin loop between the *APOLO* locus and its neighbouring gene, a key regulator of auxin polar transport. The use of overexpressing, RNAi and mutant lines indicate that *APOLO* transcript levels affect the loop formation and the epigenetic profile of this region, notably modulating certain repressive histone marks. Long non-coding RNA may dynamically modulate chromatin loops to fine-tune promoter activity of neighbouring genes and modulate developmental plasticity.

THE SENSE AND ANTI-SENSE TRANSCRIPTOME OF APPLE REVEALS THE POTENTIAL WIDESPREAD REGULATORY CONTROL OF GENE EXPRESSION THROUGH CIS-ACTING SI-RNA.

Jean-Marc Celton (INRA IRHS, Angers, France), Mathilde Orsel* (INRA IRHS, Angers, France), Sylvain Gaillard (INRA IRHS, Angers, France), Maryline Bruneau (INRA IRHS, Angers, France), Sandra Pelletier (INRA IRHS, Angers, France), Marie Christine Le Paslier (INRA EPGV, Evry, France), Aurélie Bérard (INRA EPGV, Evry, France), Sébastien Aubourg (INRA URGV, Evry, France), Marie-Laure Martin-Magniette (INRA MIA, Paris, France), D Jasper G Rees (ARC, Biotechnology Platform, Onderstepoort, South Africa), Lionel Navarro (CNRS IBENS, Paris, France) François Laurens (INRA IRHS, Angers, France), Jean-Pierre Renou (INRA IRHS, Angers, France).

mathilde.orsel-baldwin@angers.inra.fr, jean-marc.celton@angers.inra.fr

Major challenges still reside in large-scale determination of gene annotation and expression. Indeed, despite constant improvements in the quality of gene prediction software, a genome annotation process remains complex and often leads to errors in gene prediction. Furthermore, characterizing the transcriptome of eukaryotic organisms is essential for studying transcriptional regulation and its impact on phenotype. In order to develop high-quality gene model and as an attempt to unravel the complexity of transcriptional and potential post-transcriptional regulation in apple (Malus x *domestica* Borkh) we have combined microarray analysis and high-throughput sequencing of mRNA and small RNA. Alignment of RNA-seq data from 19 apple tissues (representing over 700x) to the v.1 apple genome led to the detection of a substantial number of novel transcripts and exons, as well as alternative isoforms and chimeric transcripts. Based on v.1 apple genome, we designed a NimbleGen microarray chip allowing the detection of sense and natural anti-sense transcripts (NAT), and created a transcriptome atlas for 8 apple tissues. NATs were detected for up to 20% of predicted genes and varied greatly depending on GO category and tissue and/or developmental stage considered. Generating small RNA sequences from a fruit sample, we demonstrated that anti-sense transcript expression is correlated with the presence of short and long small RNA, and that the small RNA sequence coverage depends on the level of anti-sense transcript expression. This work revealed that the small RNA component of the apple transcriptome is more important than expected. The potential for cis-post transcriptional gene silencing *via* modifications of NAT expression and subsequent changes in the population of small RNAs is extensive and complex. It could play a major role in genome and transcriptome plasticity observed in plants in response to their changing environment.

MICRO-RNAs INVOLVED IN ROOT BIOMASS AND SYMBIOTIC INTERACTIONS IN THE MODEL LEGUME *MEDICAGO TRUNCATULA*

Pilar Bustos[1], Jérémie Bazin[1,2], Damien Formey[3], Erika Sallet[3], Jean-Christophe Combier[3], Christophe Roux[3], Andreas Niebel[3], Caroline Hartmann[1,2], Jérome Gouzy[3], Martin Crespi[1] and Christine Lelandais-Briere[1,2] *

[1] ISV (Institut des Sciences du Végétal), CNRS, F-91198 Gif sur Yvette
[2] Université Paris Diderot, Sorbonne Paris Cité, F-75205 Paris.
[3] LIPM, CNRS-INRA, Castanet-Tolosan, France.

christine.lelandais@isv.cnrs-gif.fr

Root architecture and biomass are crucial for crop productivity and acquisition of soil resources. Plants can adapt their root architecture to the soil environment by controling lateral root formation, growth and/or root hair density. Most plants are also able to establish symbiotic interactions with arbuscular mycorrhizal fungi, which help them to cope with nutrient deficiency. In addition, in conditions of nitrogen starvation, legumes (Fabaceae) develop particular root lateral organs through interactions with rhizobia, the nitrogen fixing nodules. A major family of riboregulators is composed of small RNAs (21-24 nucleotides) including microRNAs. These regulatory RNAs play a variety of roles in plant development through their integration into ribonucleoprotein complexes (e.g. the RISC complex) and determine the stability and/or translation of their target mRNAs.
To analyse miRNA diversity in roots and nodules of the model legume *Medicago truncatula*, we prepared small RNA libraries from nitrogen-fixing nodules and roots under a variety of biotic interactions including rhizobial and mycorrhizal symbionts (Lelandais-Brière *et al.,* 2009; MIRMED ANR project, France). This allowed us to identify more than 200 conserved and new miRNAs, including a large part specific to legumes. Using statistical analyses, *in situ* hybridization and promoter-GUS fusions, we characterized the spatio-temporal expression of specific miRNA variants during root and nodule development. Among the most abundant miRNA in root tips, we studied in detail two isoforms of miR396. In *A. thaliana*, this miRNA regulates leaf growth and polarity by repressing transcription factors of the Growth Regulating Factors family (GRF). In *M. truncatula*, mtr- miR396 regulates 6 *MtGRF* genes but also 2 bHLH transcription factors. Moreover, the two miR396 genes are differentially expressed in roots and nodules, suggesting a possible diversification of these miRNA variants and a complex regulatory network. Over-expression of miR396 in roots led to reduced growth and cell division activity in the RAM, while its inactivation using a mimicry construct gave an opposite phenotype and particularly strongly enhanced root biomass. Furthermore, we showed that this miRNA positively controls mycorrhization (Bazin et al., 2013). Our results thus highlight a new regulatory network controlling root architecture and biomass in legumes.

THE SWI/SNF CHROMATIN REMODELLING PROTEIN ATBAF60 DIRECTLY CONTROLS THE FORMATION OF A GENE LOOP AT THE *FLC* LOCUS IN ARABIDOPSIS

Teddy Jégu* (Institut de Biologie des Plantes, Orsay France), David Latrasse (Institut de Biologie des Plantes, Orsay France), Marianne Delarue (Institut de Biologie des Plantes, Orsay France), Heribert Hirt (URGV Plant Genomics Research, Evry France), Séverine Domenichini (Institut de Biologie des Plantes, Orsay France), Federico Ariel (Institut des Sciences du Végétal, Gif-sur-Yvette France), Martin Crespi (Institut des Sciences du Végétal, Gif-sur-Yvette France), Catherine Bergounioux (Institut de Biologie des Plantes, Orsay France), Cécile Raynaud (Institut de Biologie des Plantes, Orsay France), and Moussa Benhamed (Institut de Biologie des Plantes, Orsay France).

teddy.jegu@u-psud.fr

SWI/SNF complexes mediate ATP-dependent chromatin-remodelling to control gene expression. Many components of these complexes are evolutionary conserved and several subunits of Arabidopsis SWI/SNF complexes are involved in the control of flowering, a process depending on the floral repressor *FLOWERING LOCUS C* (*FLC)*. AtBAF60 is a SWI/SNF subunit and, in this work, we showed that AtBAF60, via a direct targeting of the floral repressor *FLOWERING LOCUS C* (*FLC),* induces a change at high order chromatin level and represses both the photoperiod and the autonomous flowering pathways. AtBAF60 accumulates in the nucleus and regulates the formation of the *FLC* gene loop by modulation of histone density, composition and post-translational modification. Physiological analysis of *AtBAF60* RNAi mutant lines allowed us to propose that this chromatin remodelling protein creates a repressive chromatin configuration at the *FLC* locus. Our results revealed a new plant protein controlling chromatin structure to regulate flowering.

SESSION 7:

HORMONES

HORMONAL CONTROL OF GROWTH AND PATTERNING IN THE PLANT EMBRYO

Dolf Weijers (Laboratory of Biochemistry, Wageningen University, Dreijenlaan 3, 67-3HA Wageningen, the Netherlands)

dolf.weijers@wur.nl

Both growth and tissue patterning are processes that occur continuously during plant life. A key question is how these are coordinated in space and time to generate plant shape and function. Hormones, notably auxin and cytokinin, play a central role in both growth and patterning, but the mechanisms through which they act are largely unknown. We use the early Arabidopsis embryo as a simple and highly predictable model in which growth and patterning are intricately coordinated. I will discuss our recent work aimed at understanding the cellular basis for the establishment of multicellular patterns in 3D, as well as its genetic control. I will describe a genetic network that integrates auxin and cytokinin activity to drive the initiation of the first vascular tissue precursors, and transforms a small uniform procambial cell population into a growing, patterned bi-symmetric vascular bundle.

STRIGOLACTONES AND OTHER LONG RANGE SIGNALS REGULATING SHOOT BRANCHING IN PEA

Catherine Rameau(1)*, Alexandre de Saint Germain(1), Yasmine Ligerot(1), Guillaume Clavé(2), Jean-Paul Pillot(1), Christelle Troadec(3), Abdelhafid Bendahmane(3), Sandrine Bonhomme(1), François-Didier Boyer(1,2)

(1) Institut Jean-Pierre Bourgin, UMR1318 Institut National de la Recherche Agronomique (INRA)-AgroParisTech, Route de Saint-Cyr (RD 10), F-78026 Versailles Cedex, France
(2) Centre de Recherche de Gif, Institut de Chimie des Substances Naturelles, UPR2301 Centre National de la Recherche Scientifique (CNRS), 1 avenue de la Terrasse, F-91198 Gif-sur-Yvette Cedex, France
(3) Unité de Recherche en Génomique Végétale, INRA/CNRS, CP5708, 91057 Evry Cedex, France

Catherine.Rameau@versailles.inra.fr

Shoot branching results from the tight regulation of axillary bud outgrowth located at most leaf axils. For each axillary bud along the stem, endogenous and environmental signals (e.g. light, nutrition status) are integrated for maintaining the bud in a dormant state or inducing its outgrowth. The classical theory of apical dominance, based on decapitation experiments and exogenous hormone applications, supported the importance of auxin produced by the shoot apical meristem for inhibiting the buds below. Cytokinins from the roots antagonize auxin in the regulation of shoot branching. The use of pea branching mutants and grafting experiments demonstrated the existence of a novel signal acting as a branching inhibitor, now identified as strigolactones (SL). Our metabolism studies using tritiated-labelled SL suggest that SL-derived compounds rather than SLs are moving from root to shoot to inhibit branching. Evidences for further long distance signals, with a strong influence on shoot branching will be shown here, such as the shoot-to-root *RMS2*-dependent feedback signal, and the *RMS3*-dependent graft-transmissible signal which can partially suppress branching independently of SLs. The cloning of these genes demonstrates that *RMS2* encodes the pea homolog of the auxin-receptors AFB4/5 and RMS3 encodes an α/β-hydrolase shown to be the SL-receptor. Our results suggest that other long distance signals involved in the tight regulation of branching are auxin and the RMS3 protein moving in the phloem sap. Current experiments for a better understanding of their action in the regulation of shoot branching will be presented.

GIBBERELLINS CONTROL ROOT GROWTH AND NODULATION IN *MEDICAGO TRUNCATULA*

Anouck Diet[1,2] *, Camille Fonouni-Farde[1], Florian Frugier[1].

[1]Institut de des Sciences du Végétal – CNRS Gif sur Yvette, France.
[2]Université Paris Diderot- Paris 7, France.

anouck.diet@isv.cnrs-gif.fr

The adaptation of root system architecture to environmental constraints is a major agricultural trait. Legumes, in addition to root branching through lateral roots, can develop symbiotic interactions with soil bacteria of the Rhizobiacae family to form another secondary root organ, the nitrogen-fixing nodule. Nodule formation is regulated by several plants hormones including cytokinins which are necessary and sufficient to activate nodule organogenesis. Gibberellins (GAs) are plant growth regulators associated with several plant growth and development processes, such as seed germination, stem elongation, flowering and fruit development. GAs are also involved in pea and *Lotus japonicus* nodulation with opposite behaviours, since GAs have respectively a positive and a negative role.
We therefore aim to understand how gibberellins and their signaling pathway interfere with root and symbiotic nodule development in the model legume *M. truncatula*.
Exogenous application of GAs negatively control lateral root formation and root growth. Confocal microscopy experiments reveal that meristems of root treated with GAs are shorter and display a reduced number of cells. Concerning the nodulation process, *M. truncatula* plants treated with GAs exhibit a reduced number of nodules. Expression of *NIN* and *ERN1* symbiotic genes was reduced upon GA treatment. These results suggest that GAs play a negative role in the regulation of nodule organogenesis. Activation of the GA signaling pathway using an RNAi construct targeting three *DELLA* genes, encoding for negative transcriptional regulators of the GA pathway, led to a decrease in *M. truncatula* nodule formation. We are also currently analyzing the cross-talk between GAs and cytokinins in the regulation of the nodulation process using a mutant impaired in the cytokinin receptor CRE1.

PLANT DEVELOPMENT REQUIRES DYNAMIC MICROTUBULE LOCALIZATION OF THE PINOID KINASE THROUGH A BT-KINESIN COMPLEX.

Myckel Habets* (Leiden University, Leiden, The Netherlands), Carlos Galván-Ampudia (University of Amsterdam, Amsterdam, The Netherlands), Hélène Robert-Boisivon (CEITEC, Brno, Czech Republic), Rene Benjamins (Utrecht University, Utrecht, The Netherlands), Marcelo Kemel Zago (Universidade Federal de Ciências da Saúde de Porto Alegre, Porto Alegre, Brazil), Christa Testerink (University of Amsterdam, Amsterdam, The Netherlands), Remko Offringa (Leiden University, Leiden, The Netherlands)

m.e.j.habets@biology.leidenuniv.nl

Polar cell-to-cell transport of the plant hormone auxin (PAT) by the asymmetrically localized PINFORMED (PIN) transporters generates local auxin minima and maxima that are essential for plant development. The direction of PAT correlates with the polar distribution of PINs at the PM, which is determined by phosphorylation of the central PIN hydrophilic loop by the AGC3 protein kinases PINOID (PID), WAG1 and WAG2. We identified a PID interacting protein complex consisting of the BTB AND TAZ DOMAIN (BT) scaffold protein PINOID BINDING PROTEIN 2 (PBP2)/BT1, and the PBP2/BT1 BINDING KINESINs PBK1 and PBK2. Furthermore, we obtained evidence that PID relocalizes to the microtubule cytoskeleton upon activation through phosphorylation by the 3-phosphoinositide-dependent protein kinase 1 (PDK1). In contrast to wildtype PID, both loss- (S to A) and gain- (S to E) of-phosphorylation mutant PID versions only partially complemented the *pid/wag1/wag2* triple mutant embryo and inflorescence phenotypes, indicating the importance of dynamic PDK1-mediated control of PID throughout plant development. Overexpression of *PID* in the *pbk2* mutant background resulted in defects in pollen formation. This result is reminiscent of the male and female gametogenesis defects observed previously in the *bt2 bt3* double mutant, and suggests a possible role for the BT-PBK complex in sequestering PID during gametogenesis.

CELL WALL REMODELLING IN HORMONAL CONTROL OF SEED DORMANCY AND GERMINATION

Julien Sechet, Grégory Mouille, Adeline Berger, Helen North and Annie Marion-Poll* (Institut Jean-Pierre Bourgin, UMR 1318 INRA–AgroParisTech, Versailles, France)

annie.marion-poll@versailles.inra.fr

Dormancy and germination are two mechanisms under the control of the hormonal balance between abscisic acid (ABA) and gibberellins (GA), ABA induces and maintains dormancy while GA activates germination. After dormancy release, germination results from the protrusion of the radicle through the surrounding layers (endosperm and testa). It is thus essential to understand how cell wall remodelling regulates embryo growth and envelop properties and interferes with the hormonal control of germination and dormancy.

Mutant screens for germination resistance to paclobutrazol, an inhibitor of GA biosynthesis, led to the isolation of a mutant called *452*, besides the previously described mutant *aba4* (North et al., 2007, Plant J 50, 810). This mutant is not only tolerant to paclobutrazol but also displays other characteristics such as reduced dormancy and tolerance to sucrose, which are typical phenotypes of ABA deficient mutants. However ABA levels in *452* dry seeds are not reduced, but rather slightly higher than in wild type seeds. Fine mapping of the mutation and 'next generation sequencing' allowed the identification of the candidate gene, which encodes an α-xylosidase involved in xyloglucan maturation. These hemicelluloses are described as important regulators of cell wall properties (elasticity and rigidity).

The precise role of xyloglucan maturation in seed properties has not yet been elucidated. Therefore the composition in xyloglucans and their tissue-specific distribution have been analysed during seed development and germination. Seed phenotypes of several mutants impaired in xyloglucan synthesis or maturation have also been characterized in order to bring out the impact of xyloglucan branching on germination and dormancy. The hormonal regulation of these xyloglucan modifications is currently studied.

PLENARY LECTURE

CONTROLLING GENE EXPRESSION IN ENERGY ORGANELLES

Ian Small (Australian Research Council Centre of Excellence in Plant Energy Biology, The University of Western Australia, Perth, Australia)

ian.small@uwa.edu.au

Energy from photosynthesis drives plant growth and development, but this energy supply varies considerably depending on external conditions such as light levels, temperature and water availability. As a consequence of this variability, plants have evolved numerous mechanisms to control energy metabolism so that they can adapt to changes in growth conditions. The primary sites of energy metabolism are within mitochondria and chloroplasts and depend on massive protein complexes composed of a complex mix of nucleus-encoded and organelle-encoded subunits. The biogenesis of the plant's energy system thus depends on successfully synchronizing gene expression in three separate genetic compartments. Such synchronization depends on signals transmitted between the compartments whose nature is still largely unknown, and on mechanisms for controlling gene expression within the organelles that are only just becoming understood. We have shown that the expression of many (probably all) genes within mitochondria and plastids is reliant on nucleus-encoded RNA binding proteins of the pentatricopeptide repeat (PPR) family. The sequence-specific RNA binding ability of these proteins is remarkable; each protein binds only a single target sequence, or close variants thereof. PPR proteins display a number of obvious parallels with cytosolic microRNAs: they recognize short, single-stranded stretches of RNA by primary sequence; binding can stabilize, destabilize or even promote cleavage of the target RNA; binding can promote or prevent expression of the target RNA. The varied roles of these proteins in plants will be explained, as well as the prospects for designing custom proteins for manipulating desired RNA targets.

Abstracts: Posters

SESSION 1:

BIOTIC INTERACTIONS

1-01

A TANDEM AFFINITY PURIFICATION (TAP)-BASED STRATEGY TO STUDY THE MEKK1-MKK2-MPK4 MODULE IN ARABIDOPSIS THALIANA

Jean Bigeard* (URGV, Evry, France), Jean Colcombet (URGV, Evry, France), Delphine Pflieger (LAMBE – Université d'Evry Val d'Essonne, Evry, France), Stéphanie Pateyron (URGV, Evry, France), Heribert Hirt (URGV, Evry, France)

bigeard@evry.inra.fr

Mitogen-activated protein kinase (MAPK) proteins are present in all eukaryotes. They function in signal transduction modules where a MAPK kinase kinase (MAPKKK) activates by phosphorylation a MAPK kinase (MAPKK) which in turn activates by phosphorylation a MAPK. In plants, MAPK cascades are involved in various processes such as stress responses, hormone signaling and development. In *Arabidopsis thaliana*, the signal transduction pathway composed of MEKK1-MKK1/MKK2-MPK4 is a key regulator of defense, repressing cell death and immune responses. This pathway has been well studied at the genetic level and some substrates of MPK4 have been identified, but relatively little is known about the protein regulation of this cascade. Are there MAPK scaffold proteins like in yeast and mammals? Are there proteins implicated in the spatial distribution of MAPK cascades? To try to identify proteins involved in the regulation of the MEKK1-MKK1/MKK2-MPK4 signal transduction pathway, a tandem affinity purification (TAP)-based strategy was employed. Stably transformed plants were first obtained where a TAP-tagged MAPK was expressed in the corresponding *mapk* knock-out genetic background. TAP experiments with tagged MEKK1, MKK2 and MPK4 were then realized in several replicates and purified proteins/complexes were identified using liquid chromatography coupled to tandem mass spectrometry (LC-MS/MS) and Mascot software. A total of 11 core and 69 differentially associated proteins were identified as putative regulators or substrates of the MEKK1-MKK2-MPK4 module. Several candidates were cloned and are currently tested by yeast-two hybrid and BiFC assays to clarify direct and indirect interactions between the different components and the MAPK module. In parallel, knock-out and tagged lines of selected components were isolated and are undergoing molecular phenotypic analysis in the context of the MPK4 pathway.

1-02

BSA-NGS COMBINED STRATEGY FOR FASTER IDENTIFICATION OF MICRO-TOM TOMATO GENES AFFECTED IN PVX RESISTANCE

Aurélie Andrieu (Unité de Recherche en Génomique Végétale, Université d'Evry Val d'Essonne, Evry, France) Guillaume Beaumont (Unité de Recherche en Génomique Végétale, Evry, France), Chantal Faure (UMR 1332 Biologie du fruit et Pathologie, Villenave d'Ornon, France), Rémy Bounon (Etude du Polymorphisme des Génomes Végétaux, Evry, France), Aurélie Bérard (Etude du Polymorphisme des Génomes Végétaux, Evry, France), Aurélie Chauveau (Etude du Polymorphisme des Génomes Végétaux, Evry, France), Marie-Christine Le Paslier (Etude du Polymorphisme des Génomes Végétaux, Evry, France), Dominique Brunel (Etude du Polymorphisme des Génomes Végétaux, Evry, France), Thierry Candresse (UMR 1332 Biologie du fruit et Pathologie), Abdel Bendahmane (Unité de Recherche en Génomique Végétale, Evry, France), Bénédicte Sturbois*(Unité de Recherche en Génomique Végétale, Université d'Evry Val d'Essonne, Evry, France)

sturbois@evry.inra.fr

The Potato Virus X (PVX) resistance gene, Rx, was cloned in *Solanum tuberosum*. Rx encodes a CC-NBS-LRR like protein and its presence leads to extreme resistance (ER) upon PVX infection (Bendahmane *et al*., 1999). The Rx gene was introduced in the Micro-Tom cultivar genome, leading to PVX resistance. An EMS-mutagenized Rx Micro-Tom population was produced and challenged with a partially breaking strain of PVX. Five mutants with a clear surviving phenotype were selected. Genetic and molecular characterizations led to the identification of five independent mutations: 3 are recessive and 2 are dominant. None of the mutations are localized on the Rx gene (Sturbois *et al*., 2012).

To identify one of the dominant mutants, named #1179, a BSA-NGS (Bulk Segregant Analysis-Next generation Sequecning) strategy was developed using a segregating population from the #1179 X Rx cross. The F2 progeny was phenotyped after inoculation with the partially breaking PVX strain. Two pools were created according to their resistance or their susceptibility. The goal of this approach is to detect differential SNP between the resistant and the susceptible pools. After DNA extraction, these pools, the parental lines and the WT Micro-Tom were sequenced by Hiseq Illumina®. All the reads were mapped to the reference genome of tomato (The Tomato Genome Consortium, 2012). CLC Bio software was used to detect SNP(s) corresponding to #1179 mutation. After subtraction of common SNP from the #1179-R pool, potential candidate genes which may be linked to PVX resistance will be identified.

The same approach will then be pursued to find out candidate genes for the #2135 recessive mutation.

1-03

CHARACTERIZATION OF ARABIDOPSIS THALIANA MUTANTS RESISTANT TO THE TYPE THREE-SECRETED EFFECTOR DspA/E.

Alban Launay[1]*, Manon Moreau[2], Oriane Patrit[2], Régine Vedel[2], Alexandre Degrave[3], Mathilde Fagard[1]

[1]Institut Jean-Pierre Bourgin, UMR1318 INRA-Agroparistech, 78026 Versailles, France
[2]Laboratoire des interactions plantes-pathogènes, UMR217, 75005 Paris, France
[3]IRHS, UMR1345 INRA-Université d'Angers, Agrocampus-Ouest, 49070 Beaucouzé, France

alban.launay@gmail.com

The phytopathogenic bacterium *Erwinia amylovora (Ea)* is the causal agent of the fire blight disease on the *pyreae* tribe of *Rosaceae* (apple tree, pear tree...). This disease is characterized by necrotic symptoms on leaves and fruits which gives a "burned" aspect to the plant. Pathogenicity of *Ea* depends on a type-three secretion system (Barny *et al.,* 1990), a syringe-like protein complex which allows the bacteria to translocate effectors into the cell, for instance the effector DspA/E essential for the pathogenicity. On the host plant, this effector is required for the induction of necrosis, an oxidative burst (Venisse *et al.*, 2003) and suppresses callose-deposition (DebRoy *et al.,* 2004). However, the molecular function of DspA/E in the plant cell is not yet known. Our team chose to use the model plant *Arabidopsis thaliana* to study the role and the function of DspA/E in the plant cell. Indeed, *Ea* induces necrotic symptoms and multiplies transiently *in planta* (Degrave *et al.,* 2008) and it is DspA/E-dependent (Degrave *et al.,* 2013). Moreover, construction of transgenic plants which express DspA/E under the control of an estradiol-inducible promotor, allowed us to show that DspA/E is able to restore the growth of an *Ea dspA/E* mutant (Degrave *et al.,* 2013). In parallel we showed that DspA/E represses plant protein synthesis when it expressed in the transgenic lines and injected by *Ea.* These data are coherent with an increase of bacterial multiplication when plant protein synthesis is inhibited by cycloheximide (Moreau *et al.,* 2012). To understand DspA/E's toxicity mechanisms in the plant cell, an EMS mutagenesis was performed on one of ours transgenic line carrying DspA/E. The progeny from this mutagenesis line was sown on estradiol to identify individuals resistant to DspA/E toxicity. We identify 4 resistant mutants, which were characterized. Three of these mutants belong to the same complementation group. We sent 2 mutants for a whole genome sequencing.

1-04

IDENTIFICATION AND CHARACTERIZATION OF HOST GENES IMPLICATED IN POTEXVIRUS RESISTANCE IN SOLANACAE.

Germain Gourdon[1], Aymeric Leveau[1], Zoé Terret[1], Gregori Bonnet[2], Julien Bonnet[3], Céline Labourey[2], Abdelhafid Bendahmane[1], Bénédicte Sturbois[1].

[1] URGV (UMR1165-INRA- UEVE ; ERL CNRS, 2, rue Gaston Crémieux, 91000 EVRY, France
[2] Syngenta Seeds SAS, 346, Route des Pasquiers 84260 SARRIANS France
[3] Syngenta Seeds SAS , 12, Chemin De L'Hobit 31790 SAINT SAUVEUR, France

gourdon@evry.inra.fr

Plants are continually challenged by various pathogens. The Rx-mediated resistance against Potato virus X (PVX) illustrates a defense mechanism known as the gene-for-gene model (H. H. Flor, 1971), in which the recognition of the PVX coat protein (CP) the elicitor, by the product of the Rx1 gene, a NBS-LRR protein determines the outcome of the interaction (A. Bendahmane *et al.,* 1995). Moreover, previous studies have shown that the CP of other Potexviruses (NMV, CymMV, WClMV and PepMV) can induce the Rx-mediated resistance (I. Baurès *et al.,* 2008).The minimal fragment elicitor of PVX CP has been characterized and used as bait in a Yeast Two Hybrids (Y2H) screening. Two potential interactors have been identified: a transcription factor and a chloroplastic protein.
Therefore, the resistance mechanism and the two PVX-CP identified interactors, will be analyzed regarding an endemic pathogen of tomato crop: the Pepino Mosaic Virus (PepMV).
In order to go one step further, five tomato (Solanum lycopersicum) cDNA libraries have been constructed after PepMV inoculation with various time conditions. Using the CP, the Triple Gene Block protein 1, 2 and 3 (TGBp1, TGBp2 and TGBp3) of PepMV as baits in an Y2H screening, a list of host proteins interacting with viral proteins have been established. Following deeper analyses, seven interactors have been selected for upcoming functional validation experiments such as transient gene silencing (VIGS- viral induced gene silencing) or over/under-expressing lines. The Tomato TILLING platform held at URGV is also currently exploited for validation purpose.

1-05

IDENTIFICATION AND FUNCTIONAL CHARACTERIZATION OF A *BRACHYPODIUM DISTACHYON* UGT INVOLVED IN DETOXIFICATION OF THE *FUSARIUM GRAMINEARUM* MYCOTOXIN, DEOXYNIVALENOL

Pasquet J-C[1]*, Schweiger W[2], Macadré C[1], Morlet C[1], Tharreau V[1], Saindrenan P[1], Adam G[2] and Dufresne M[1]

[1]Institut de Biologie des Plantes, Université Paris Sud, Orsay France
[2]BOKU, University of Natural Resources and Life Sciences, Tulln, Austria

jean-claude.pasquet@u-psud.fr

Fusarium head blight is a major cereal disease mainly due to the pathogenic toxinogenic fungus *Fusarium graminearum*. During its development *in planta*, the fungus produces mycotoxins harmful to humans and animals, among which deoxynivalenol (DON). No specific resistance to *F. graminearum* has been underlined but a number of quantitative trait loci (QTL) have been identified in bread wheat and barley. Genetic, transcriptional or metabolic analyses have correlated some of these QTLs to functions involved in DON detoxification, among which some UDP-glycosyltransferases (UGTs). The novel model plant *Brachypodium distachyon* is an ideal system to better understand detoxification mechanisms by cereals in response to DON. We have established that the interaction between these two partners is compatible. Mycotoxin production quantitatively affects the colonization of *B. distachyon* spikes, an intermediate situation to that known in wheat or barley.

Recently, a barley UGT has been identified as being potentially involved in DON detoxification. The search for orthologous genes in *B. distachyon* by sequence homology and phylogenetic analysis has identified six candidate genes. Expression monitoring of these genes by qRT-PCR has shown that the *Bradi5g03300* gene is the preferential candidate: its expression is strongly induced 3 hours after the mycotoxin application and 72 hours after an inoculation by *F. graminearum.* Moreover, this gene is much less induced in response to a mutant strain unable to produce the mycotoxin (Δ*tri5*). Functional analyses *in planta* have been undertaken through the construction of *B. distachyon* lines exhibiting either a mutated allele or an altered expression of this gene. We have screened the *B. distachyon* TILLING mutants' collection and identified three mutant lines. These mutants exhibit an increased sensitivity to the mycotoxin and to *Fusarium*, at the root and spike levels, respectively. Analysis of lines overexpressing the UGT-encoding gene is underway and will be presented.

1-06

IDENTIFICATION OF A RECEPTOR LIKE KINASE INVOLVED IN *MEDICAGO TRUNCATULA* NODULATION

Carole Laffont* (Institut des Sciences du Végétal, Gif-sur-Yvette, France), Virginie Mortier (Department of Plant Systems Biology, Ghent University, Belgium), Sofie Goormachtig (Department of Plant Systems Biology, Ghent University, Belgium), Florian Frugier (Institut des Sciences du Végétal, Gif-sur-Yvette, France).

carole.laffont@isv.cnrs-gif.fr

Root system architecture is crucial to adapt plant growth to changing soil environmental conditions and consequently to maintain crop yield. In addition to branching through lateral roots, legumes roots can develop under nitrogen starvation another lateral organ, the nitrogen-fixing nodule, through a specific interaction with symbiotic bacteria. Nodule formation comprises two tightly interconnected processes: bacterial infection and organ development which are both under complex phytohormonal controls. In addition, it was shown that legumes can control nodule number through an autoregulation of nodulation (AON) pathway, involving systemic signal exchanges between roots and shoots (1). In *Medicago truncatula*, CLE (CLAVATA/ Embryo Surrounding Region) peptides are involved in nodule establishment but also control nodule numbers by interacting with the AON pathway. These peptides interact with Leucin Rich Repeat- Receptor Like Kinases (LRR-RLK) and we have identified one of these receptor as potentially regulating nodulation. Progress in the characterization of this pathway will be presented.

1-07

IDENTIFICATION, IN CROP SPECIES, OF HOST PARTNERS IMPLICATED IN POTATO VIRUS X (PVX) RESISTANCE

Aymeric Leveau (URGV, Evry, France), Aurélie Andrieu (URGV, Evry, France), Grégori Bonnet (Syngenta, Sarrians, France), Julien Bonnet (Syngenta, Saint Sauveur, France), Abdelhafid Bendahmane (URGV, Evry, France), Bénédicte Sturbois* (URGV, Evry, France)

sturbois@evry.inra.fr

The Rx gene confers an extreme resistance against common strains of PVX (potato Virus X) (Ritter, E. *et al.* 1991). Two loci of Rx, Rx1 and Rx2, have been identified in different potato varieties. In this biological system, the recognition step involves an indirect interaction between the RX protein and the PVX capsid protein, the elicitor (Bendahmane, A. *et al.,* 1995). In the RX/PVX pathosystem, the molecular cascade and the mechanisms conferring the resistance remain unclear. However it has been shown through different strategies, that RX1 protein could interact directly with RanGAP2 and that RX accumulation involves other proteins like HSP90, SGT1 or RAR1 (Hubert, D.A. *et al.,* 2003 ; Azevedo, C. *et al.,* 2002 ; Sacco, M.A *et al.,* 2007 : Tameling, W.I. and Baulcombe, D.C., 2007).
In order to identify host proteins implicated in Rx-mediated resistance, a yeast two hybrid strategy has been performed. The bait was a minimal fragment of the PVX CP that has been identified through serial deletions as done before for other Potexvirus coat protein (Baures, I. *et al.,* 2008). This minimal fragment has been shown to be sufficient to elicit the defense reaction when transiently expressed in wild type or Rx1-expressing *Nicotiana*. The prey has consisted of a cDNA library from wild type or Rx1-expressing *Nicotiana* infiltrated or not with the minimal PVX elicitor.
The screening of several millions of clones has lead to the identification of 12 potential interactors. A transcription factor and a chloroplastic protease have been studied in more details. Their interactions have been confirmed in yeast, in vivo by BiFC and their implication in Rx resistance has been pursued through transient over-expression in wild type or Rx1-expressing *Nicotiana*. The TILLING platform is also exploited to identify mutants for the two main interactors. The results are in agreement with their implication in a general resistance mechanism against viruses.

1-08

MECHANOSENSITIVE CHANNELS IN LEGUME ROOT HAIR: ROLE IN SYMBIOSIS WITH RHIZOBIUM

Daniel Tran* (Institut des Sciences du Végétal, Gif-sur-Yvette, France), Marjorie Guichard (Institut des Sciences du Végétal, Gif-sur-Yvette, France), Lydia Alloul (Institut des Sciences du Végétal, Gif-sur-Yvette, France), Tiffanie Girault (Institut des Sciences du Végétal, Gif-sur-Yvette, France), Jean-Marie Frachisse (Institut des Sciences du Végétal, Gif-sur-Yvette, France)

Daniel.tran@isv.cnrs-gif.fr

The root hair plays crucial roles in water and nutrient uptake, and thus in plant development. In Legumes, it also plays a central role in the establishment of the nitrogen-fixing symbiotic interaction with rhizobia, which has a crucial importance for sustainable agriculture as it largely contributes to limitation of nitrogen fertilizers. During the early symbiotic interaction between the model legume *Medicago truncatula* root hair and its microsymbiont *Sinorhizobium meliloti*, crucial events in term of signalling and mechanics happened. The bacterial infection initiated by bacterial adhesion to root hairs will lead to hair curling that entraps the bacteria. The hypothesis of the involvement of physical sensors at the surface of the root hair during these early events is worth considering. In plant, the molecular nature of protein mediating mechanoperception is still elusive. Mechanosensitive ion channels belonging to MSL (Mechanosensitive Small-conductance Like) family represent suitable candidates to be involved in this process leading to cell signalling. MSL proteins have been recently characterized in Arabidopsis in our laboratory (Haswell et al., 2008). These proteins are channels activated by membrane stretching. They behave as microprobes able to transduce a mechanical force into an electrochemical signal allowing the plant to respond to mechanical stimulations. In Medicago, we have identified 13 members related to MS channel with *in silico* analysis, amongst which 8 strong putative candidates were divided into two clades, as observed in Arabidopsis. The expression analysis of one clade by semi-quantitative PCR, revealed that these candidates are present in root hair and other root tissues. Preliminary results showed that transient expression in a silenced-system provides a channel activity dependant on membrane tension for one MSL. Further investigations are in progress, in order to get a functional characterization of these root hair channels and to obtain an integrated view of their role in early symbiosis. This study is supported by a grant (CAROLS) of the French ANR.

1-09

MEDICAGO TRUNCATULA AS A MODEL TO STUDY VERTICILLIUM WILT IN LEGUME PLANTS: REGULATORY MECHANISMS AND INTERACTION WITH ROOT SYMBIOSIS

Martina Rickauer (Université de Toulouse, INPT, Laboratoire d'Ecologie fonctionnelle et Environnement, Castanet-Tolosan, France)

martina.rickauer@ensat.fr

Verticillium wilt, caused by the soilborne fungus *Verticillium albo-atrum* (*Va*) affects more than 200 plant species including alfalfa. We study the interaction between the model legume *Medicago truncatula* and *Va* V31-2, a strain isolated from alfalfa to understand the mechanisms involved in the response to this root pathogen. Due to their N-fixing symbiosis with rhizobia, legume plants have an important role in agro- and ecosystems. In addition to presenting well-established genetic resources and high biodiversity, *M. truncatula* enables to investigate putative cross-talk between disease and symbiosis. Adaptation to root pathogens is likely to have effects on symbiosis and vice versa.

Root inoculation of *M. truncatula* line F83005.5 with a spore suspension of V31-2 induces typical wilt symptoms whereas line Jemalong A17 is resistant to the fungus (Ben et al., 2013). Studies with a GFP-expressing *V. albo-atrum* strain showed that symptoms and colonisation pattern in infected susceptible *M. truncatula* plants were typical of Verticillium wilt. Resistant A17 plants eliminated the fungus from their vessels between 4 and 7 days after inoculation, whereas other lines showed tolerance (weak symptoms, but presence of fungus). Pretreatment of the susceptible line F83005.5 with various plant hormones showed that some protected against subsequent inoculation. Differential expression of hormone-regulated genes suggests that plant hormones play a role in resistance. To further study regulatory mechanisms of the response to *Va* V31-2 we produced libraries of mRNA and small RNA from roots of inoculated lines A17 and F83005.5. Sequence analysis of these libraries is under way.

Several mutants of the resistant line A17, impaired in distinct steps of rhizobial symbiosis, were affected in their response to *V. albo-atrum*, which suggests that mechanisms involved in the establishment of symbiosis or disease might have some common regulatory control points.

1-10

MICROBIAL SIDEROPHORE: A TOOL TO STUDY THE CROSSTALK BETWEEN IRON REGULATION AND IMMUNITY RESPONSES OF ARABIDOPSIS THALIANA

Aude Aznar[1-5]*, Nam Phong Kieu[2], Nicolas Chen[3], Martine Rigault[1-5], Sébastien Thomine[4], Dominique Expert[5], Alia Dellagi[1-5]

[1] UMR1318 INRA AgroParisTech, Institut Jean Pierre Bourgin, 78026 Versailles cedex, France.
[2] Laboratory of Phytopathology, Faculty of Bioscience Engineering, Ghent University, 9000 Ghent, Belgium.
[3] AGROCAMPUS OUEST Centre d'Angers, UMR1345 IRHS, SVAH, 49071 Angers, France.
[4] Institut des Sciences du Végétal, UPR2355 CNRS, 91198 Gif-sur-Yvette, France.
[5] UMR217 INRA-AgroParisTech-UPMC, Laboratoire des Interactions Plantes-Pathogènes, 75231 Paris cedex, France

aude.aznar@versailles.inra.fr

Iron is essential for most forms of life. It is required in metabolic processes as respiration or photosynthesis. However, iron is weakly bio-available in environment and toxic in its free form because it can form reactive oxygen species by catalyzing the Fenton reaction.
Under iron deficiency, microbes secrete low molecular weight molecules with high iron affinity, called siderophores. The ferri-siderophore complexes are specifically recognized at the cell envelope and generally transported inside the microbial cell where iron is made available. Siderophores are required for the full virulence of several micro-organisms including bacteria and fungi infecting animals or plants. In our laboratory, we have shown that chrysobactin is a siderophore secreted by the phytopathogenic enterobacterium *Dickeya dadantii* and is required for systemic progression of soft rot symptoms on Arabidopsis and *Saintpaulia ionantha*. We have also shown that deferrioxamines are siderophores secreted by the phytopathogenic enterobacterium *Erwinia amylovora* and are required for full virulence on apple seedlings and flowers.
Previous results showed that purified siderophores infiltrated into Arabidopsis leaves trigger: i) a transcriptional activation of root genes involved in iron uptake and ii) a defense signal which leads to the accumulation of salicylic acid as well as the transcriptional activation of PR genes in leaves.
To further decipher this cross-talk between iron metabolism and immunity regulation, we studied a set of defense markers in siderophore-treated plants. We also have been interested in the establishment of immunity mechanisms in the *irt1-1* mutant, impaired in its ability to transport iron from soil to root cells. Results of this work will be presented.

1-11

PERCEPTION OF SYMBIOTIC LIPO-CHITOOLIGOSACCHARIDES BY PLANTS

Marie Cumener[1], Luis Buendia[1], Judith Fliegmann[1], Virginie Gasciolli[1], Sylvie Camut[1], Christophe Lachaud[1], Sandra Uhlenbroich[2], Carole Pichereaux[3] Michel Rossignol[3], Sophie Canova[4], Jean-Marie Beau[4], Boris Vauzeilles[4], Eduardo Martinez[5], Hugues Driguez[5], Sylvain Cottaz[5], Sébastien Fort[5], Julie Cullimore[1], Jean-Jacques Bono[1] and Benoit Lefebvre[1]*

[1]INRA-CNRS, LIPM, Castanet-Tolosan; [2]UPS-CNRS, LRSV, Castanet-Tolosan; [3]FR3450 Plateforme de Protéomique, Toulouse; [4]ICMMO, UMR8182, Orsay; [5]CNRS, CERMAV, Grenoble

benoit.lefebvre@toulouse.inra.fr

Lipo-chitooligosaccharides (LCOs) were first identified as secreted by rhizobial bacteria and essential for establishment of the root nodule symbiosis in many legumes. LCOs have been more recently shown to be secreted as well by a symbiotic arbuscular mycorrhizal (AM) fungus, colonizing roots of many plants. Receptor-like kinases (RLKs) from the lysin motif (LysM) subfamily are involved in LCO perception in legumes and have been shown to play a role in the establishment of the Rhizobium and the AM symbioses in the non-legume *Parasponia*.
We have characterized in detail NFP and LYK3, two LysM-RLKs from *Medicago truncatula* essential for the root nodule symbiosis, including their kinase activities, post-translational modifications, interacting partners and LCO binding properties. No high affinity binding of radiolabelled LCOs to LYK3 and/or NFP expressed in tobacco BY2 cells was detected, although binding to endogenous tobacco proteins was observed. Similarly, *NFP* and *LYK3*-independent LCO-binding sites have been detected in *M. truncatula*. We are seeking to identify the corresponding LCO-binding proteins in order to study their functions in the legume *M. truncatula* and in the non-legumes, *Brachypodium distachyon* and to *Solanum lycopersicum*.

1-12

RESISTANCE SIGNALLING IN STEMS OF TWO *CAPSICUM ANNUUM* CULTIVARS WITH DIFFERENT DEGREES OF SENSITIVITY TO *PHYTOPHTHORA CAPSICI.*

M. Emilia Requena* (Department of Plant Biology, University of Murcia, Murcia, Spain), Catalina Egea-Gilabert (Department of Agrarian Science and Technology, E.T.S.I.A., Polytechnic University of Cartagena, Murcia, Spain), Ana Requena (Department of Agrarian Science and Technology, E.T.S.I.A., Polytechnic University of Cartagena, Spain), M. Emilia Candela (Department of Plant Biology, University of Murcia, Murcia, Spain)

mariemi@um.es

The interaction of two varieties of pepper (*Capsicum annuum* L.), one resistant (cv. Serrano Criollo de Morelos-SCM) and the other sensitive (cv. California Wonder-CW), with the pathogenic oomycete *Phytophthora capsici* L. was studied. The plants were infected with the oomycete by decapitating the stems when they had 5-6 true leaves. The depth of penetration of the oomycete hypha and production of reactive oxygen species (O_2^- and H_2O_2) along the stem six days post-infection were studied. Both varieties developed a hypersensitive reaction and generated ROS as a defence mechanism. Both, the ROS production intensity and oomycete penetration depth are significantly higher in SCM. Three zones could be identified in the inoculated stems: "necrotic" at the inoculation front (5.3 mm in SCM and 89.3 mm in CW), "intermediate", just below the necrotic zone (measuring about 20 mm), followed by a "healthy" zone. In the intermediate and healthy sections, a series of 200 micron thick cross sections were made to measure ROS. 500 micron thick sections were made to measure the depth of oomycete penetration, placing them in Petri dishes with PDA medium and measuring mycelial growth after 48 h. The pathogen penetrated 2 sections (1 mm) in SCM and 21 sections (10.5 mm) in CW. To measure ROS production, O_2^- was measured with NBT and H_2O_2 with DAB. The O_2^- was only detected in the first two sections and always with higher intensity in SCM, while H_2O_2 was very intense in the vessels of the first six sections of SCM, diminishing in intensity until the tenth section. In CW, H_2O_2 intensity was lower in the xylem but more intense in the phloem from section 2 until section 7. Because of SCM was able to overcome the infection by inhibiting the pathogen's growth, while CW succumbed to the disease, it seems that H_2O_2 behaves as a messenger that intervenes in the signalling to activate the defence response.

1-13

RHIZOBIUM/LEGUME SYMBIOSIS: DECIPHERING THE SYMBIOTIC CONTROLS OF PLANT DEFENSES

Benjamin Gourion* (Institut des Sciences du Végétal, Gif-sur-Yvette, France), Marie Bourcy (Institut des Sciences du Végétal, Gif-sur-Yvette, France), Anne Cayrel (Institut des Sciences du Végétal, Gif-sur-Yvette, France), Fathi Berrabah (Institut des Sciences du Végétal, Gif-sur-Yvette, France), Alexis Eschstruth (Institut des Sciences du Végétal, Gif-sur-Yvette, France), Pascal Ratet (Institut des Sciences du Végétal, Gif-sur-Yvette, France)

Benjamin.Gourion@isv.cnrs-gif.fr

Most legumes species are able to establish symbiosis with soil bacteria referred to as rhizobia. During these interactions, the legumes house several thousands of rhizobia into the cells of a newly formed organ: the nodule. In nodules, molecular nitrogen is reduced by rhizobia for the benefit of the plants. This allows the legume to develop onto nitrogen poor soil without fertilizer supply. In contrast to what normally occurs when bacteria massively invade plant tissues, no visible defense reactions are triggered in legume infected nodules. Now that the perspective of transferring symbiotic nitrogen fixation capacity to non-legume crop plants is returning in the discussions, unraveling the molecular mechanisms underlying the symbiotic control of plant defenses is of particular interest. Using forward and reverse genetics, two genes inhibiting defense-like reactions in *Medicago truncatula* nodules were identified. Phenotypical analysis of the corresponding mutants suggested the existence of two signaling pathways controlling plant defenses in nodules. One pathway is required for nitrogen fixation in all tested conditions. The other is dispensable for the symbiotic process in some environments. The existence of permissive and restrictive conditions for the corresponding KO mutants indicate that environment shape the genetic requirement for nitrogen fixation. The predicted proteins corresponding to the two genes will be presented as well as elements suggesting a link between them and the MAMP triggered immunity signalization.

1-14

SPLICE VARIANTS OF THE SIP1 TRANSCRIPTS PLAY A ROLE IN NODULE ORGANOGENESIS IN LOTUS JAPONICUS

Chao Wang, Hui Zhu, Liping Jin, Tao Chen, Longxiang Wang, Heng Kang, Zonglie Hong, Zhongming Zhang* (State Key Laboratory of Agricultural Microbiology, Huazhong Agricultural University, Wuhan, China)

zmzhang@mail.hzau.edu.cn

SymRK-interacting protein 1 (SIP1) has previously been shown to interact with the symbiosis receptor kinase, SymRK, in *Lotus japonicus*. A longer variant of the SIP1 transcript, SIP1L, was isolated and characterized. SIP1L contains an additional 17 amino acids that make its C-terminus a complete heat shock protein 20 (Hsp20)-like domain. In contrast to SIP1S, the longer splicing variant SIP1L could not interact with SymRK. Both SIP1L and SIP1S transcripts could be detected in developing nodules and other plant tissues, although the former was always more abundant than the latter. SIP1L and SIP1S formed heteromeric protein complexes, which were co-localized in the plasma membrane, cytoplasm and nuclei. Expression of SIP1-RNAi in transgenic hairy roots resulted in impairment in the nodule and arbuscular mycorrhizal development, suggesting an important role of SIP1 in the common symbiosis pathway. Overexpression of either SIP1L or SIP1S increased the number of nodules formed on transgenic hairy roots, indicating a positive role of SIP1 in nodulation. The SIP1S-like transcript was not detected in other higher plants tested, and the SIP1L-like proteins of these plants were capable of interacting with the SymRK orthologs. It is proposed that the loss of the ability of SIP1L to interact with SymRK in Lotus is compensated by the expression of a shorter splicing variant, SIP1S, which binds SymRK and may play a role in relaying the symbiosis signals to downstream cellular events.

1-15

THE INVOLVEMENT OF JASMONIC ACID IN THE REGULATORY ROLE OF AM FUNGI ON ROOT HYDRAULIC CONDUCTIVITY.

Beatriz Sánchez-Romera* (Estación Experimental del Zaidín (CSIC), Granada, Spain), Juan Manuel Ruiz-Lozano (Estación Experimental del Zaidín (CSIC), Granada, Spain), Jose María García-Mina (CIPAV-R&D Roullier Group, Pamplona, Spain), Ángel María Zamarreño (CIPAV-R&D Roullier Group, Pamplona, Spain), and Ricardo Aroca (Estación Experimental del Zaidín (CSIC), Granada, Spain).

beatriz.sanchez@eez.csic.es

The arbuscular mycorrhizal (AM) fungi establish symbiotic association which provides benefits to the host plants. It is known that it can improve water status of the plant, modifying stomatal conductance, root hydraulic conductivity (L) and the expression and abundance of aquaporins to protect the plants against abiotic stresses. On the other hand, jasmonic acid (JA) is a very important hormone for its effect in the regulation of stomatal opening and in their defensive role against various stresses. Recent studies showed that JA affects the development and establishment of colonization by AM fungi.
The aim of this research was to investigate the involvement of jasmonic acid in the regulatory role of AM fungi on root hydraulic conductivity (L). For this, tomato plants deficient in JA (*def-1*) and WT plants were grown, without AM inocolum or with *Rhizophagus irregularis* as AM fungi inoculum. Besides, we had two water treatments, well watering conditions and drought. It has been seen: 1) that neither drought nor JA-deficient plants had an effect on the percentage of AM colonization. 2) The AM roots showed smaller size than no AM roots, except *def-1* plants under well-watering conditions. 3) L was higher in AM plants than in no AM plants, except again, in *def-1* plants under well-watering conditions. These no AM *def-1* plants had higher L than no AM WT plants. The L results correlated with phosphorylation state of PIP2 aquaporins and AIA root concentration.
Conclusion. Regardless of soil water status, the fungal colonization capacity was not affected by JA deficiency in the roots of *def-1* plants. However, the effectiveness of the fungus to improve L and regulate phosphorylation of aquaporins under well irrigation conditions was limited. Instead, *def-1* plants behaved similar to WT in drought conditions, indicating that under stress conditions, the AM fungi are able to find an alternative way to regulate plant water status independent of JA.

1-16

THE PGPR STRAIN *PHYLLOBACTERIUM BRASSICACEARUM* STM196 INDUCES REPRODUCTIVE DELAY AND PHYSIOLOGICAL CHANGES THAT RESULT IN IMPROVED DROUGHT TOLERANCE IN ARABIDOPSIS

Justine Bresson* (INRA-SUPAGRO, Montpellier, France), Fabrice Varoquaux (Université de Montpellier II, Montpellier, France), Thibaut Bontpart (INRA-SUPAGRO, Montpellier, France), Bruno Touraine (Université de Montpellier II, Montpellier, France), Denis Vile (INRA-SUPAGRO, Montpellier, France)

bresson@supagro.inra.fr

Understanding how biotic interactions can improve plant tolerance to drought is a challenging prospect for agronomy and ecology. Plant growth promoting rhizobacteria (PGPR) are promising candidates but the phenotypic changes induced by PGPR under water limitation remain to be elucidated. We investigated the effects of *Phyllobacterium brassicacearum* STM196 strain, a PGPR isolated from the rhizosphere of oilseed rape, on two accessions of *Arabidopsis thaliana* with contrasting flowering phenology. We measured multiple morpho-physiological traits related to plant growth and development in order to quantify the added value of the bacteria to drought response strategies of Arabidopsis. We confirmed the PGPR-effect of STM196 on Arabidopsis in soil conditions. A delay in reproductive development induced by the bacteria resulted in a gain of biomass independently of the accession and of the watering regime. Coordinated changes in transpiration, ABA content, photosynthesis and development resulted in higher water use efficiency and a better tolerance to drought of PGPR-inoculated plants. Our findings give new insights into the ecophysiological bases by which PGPR can confer stress tolerance to plants. Rhizobacteria-induced delay in flowering time could represent a valuable strategy for increasing biomass yield, whereas rhizobacteria-induced improvement of water use is of particular interest in multiple scenarios of water availability.

1-17

TOWARDS UNDERSTANDING HOW SYMBIOTIC MOLECULES STIMULATE LATERAL ROOT DEVELOPMENT IN *MEDICAGO TRUNCATULA*

Violaine Herrbach* (LIPM, Toulouse, France), Clare Gough (LIPM, Toulouse, France), Sandra Bensmihen* (LIPM, Toulouse, France)

Violaine.Herrbach@toulouse.inra.fr

The model legume *Medicago truncatula* is able to establish two types of root endosymbiosis: the arbuscular mycorrhizal symbiosis (AMS) with mycorrhizal fungi and the rhizobium-legume symbiosis (RLS) with soil bacteria. Our team demonstrated that both rhizobial and mycorrhizal partners of *M. truncatula* produce lipo-chitooligosaccharides (LCOs), called Nod Factors and Myc-LCOs, respectively. In addition to their symbiotic roles, these LCOs were shown to stimulate lateral root formation (LRF) in *M. truncatula*. Little is known about the cellular, physiological and genetic mechanisms involved in this LRF stimulation by LCOs. Furthermore, in contrast to *Arabidopsis thaliana*, LRF in *M. truncatula* is not well documented, despite the specificies of this plant, in particular the ability to form original root organs called nodules during RLS. In this study, we aim at understanding the cellular mechanisms of LRF stimulation by LCOs. To highlight *M. truncatula* specificities, we first present a precise description of the successive stages of LRF in this plant using reporter genes and histological sections, and show that LRF in *M. truncatula* differs from the description in *A. thaliana*. Based on this description, we look at any differences in the cellular events leading to LRF stimulation by LCOs, especially to understand whether LCOs are active on a particular stage of LRF. Given that auxin is the major phytohormone involved in LRF and that rhizobial LCOs have been shown to interfere with auxin transport, we are also interested in understanding any interaction between symbiotic LCOs and the auxin homeostasis or signaling pathways in the context of LRF. This aspect is being assessed using pharmacological and transcriptomic approaches. Taken together, these analyses should help us to understand how symbiotic LCOs interfere with the *M. truncatula* root developmental program to stimulate LRF.

1-18

EVALUATION OF ANTIFUNGAL ACTIVITY OF CRUDE EXTRACTS FROM *ZIZYPHUS SPINA CHRISTI L. DESF*. AND *PEGANUM HARMALA L*. ON FUNGAL SPECIES DEVELOPMENT

Khaldi Achraf* (Laboratory of valorization of vegetal Resource and Food Security in Semi Arid Areas, University of Bechar, Algeria), Meddah Boumedienne (Institute of Natural Science and Life, University of Mascara, Algeria), Moussaoui Abdallah (Laboratory of valorization of vegetal Resource and Food Security in Semi Arid Areas, University of Bechar, Algeria), Gouri Saif (Laboratory of valorization of vegetal Resource and Food Security in Semi Arid Areas, University of Bechar, Algeria)

achrafsystemdz@yahoo.fr

This work studies the antifungal activity of medicinal plants extracts from the algerian Sahara (South-West of Algeria): *Zizyphus spina Christi L. Desf.* and *Peganum harmala L*. Aerial part from each plant is used for extraction by three solvents: water, methanol and hexane. Among all solvents, methanol gave the best extraction yield for *Peganum harmala L*. (24, 02%) and water for *Zizyphus spina Christi L. Desf*. (25, 27%). The phytochemical screening indicates that our plants are relatively rich in active consisting: flavonoids, saponins, sterols, terpenes, steroids and tannins. Antifungal activity of the different extracts was studied witch respect to seven fungal strains. The results of method direct contact on mycelial growth, shows that the three extracts of *Peganum harmala L* were more active against fungi with extracts of *Zizyphus spina christi L. Desf*. With a concentration of 1500 µg/1ml the most important effect for the 1st plant was observed by the methanol extract, whose, *F. oxysporium*, *Alternaria*, *A. ochraceus* and *Cladosporium* are most sensitive, with percentages of inhibitions respectively of 68,67%,71,15%, 80,39% and 95%. Whereas for the 2nd plant the extract most active was the hexanic extract, the most sensitive stocks are *F. oxysporum* and *A. Niger* with an inhibition of 80,95% and 50%. In addition to the growth of the mycelium, the various extracts of two plants showed, in vitro, a antifungal activity at least important on the two other developmental stages, germination and the sporulation, of all fungi, is showed an inhibiting action of the spores which exceeds 50% of all fungi. The evaluation of germination showed that the extracts methanolic and hexanic of *Peganum harmala L* are more active on the germination of the spores. On the other hand for *Zizyphus spina christi L. Desf* the efficiency was observed with the aqueous extract.

SESSION 2:

ABIOTIC INTERACTIONS

2-01

A FORWARD GENETIC APPROACH TO DISSECT SIGNALLING MECHANISM IN RESPONSE TO SALINITY STRESS IN RICE

Apekshita Singh* (Amity University, Noida, UP, India), Ashwani Pareek (Jawaharlal Nehru University, New Delhi, India)

apek28@gmail.com

Being sessile, plants are constantly exposed to a variety of environmental cues and to survive, they must need to perceive any change in external or internal environment and respond effectively. To achieve that, there must be a very intricate signalling network that needs to be explored, to develop crop varieties capable of growing in changing environmental conditions. In the present study, we have utilised forward genetics approach to identify various components of signalling pathway in response to salinity stress in rice. For this, a large randomly mutated population has been created using physical mutagens (gamma rays) and screened for response to salinity stress. As a first step, the gamma irradiation (at different doses) was done in seeds as well as seed derived calli in parallel. The irradiated seeds were propagated in field to raise M1 plants and M2 seeds were collected. The notable phenotypic differences were obtained in M1 like plant height, early and late growth, panicle sterility, altered grain shape and size, varying size of the panicle and flowering time etc. Similarly, calli were also propagated till M1 generation and seeds were collected. The range of phenotypic variability confirmed the mutagenic effect. The salinity screening was performed with M3 seeds grown in hydroponic system by analysing various parameters such as root length, shoot length, Na^+/K^+ ions ratio and fresh weight. The potential lines are being selected and will be used for generating mapping population by crossing with parental line.

2-02

ALTERATION OF ROOT HYDRAULIC PROPERTIES BY RHIZOBIAL SYMBIOSIS

Ricardo Aroca* (Estación Experimental del Zaidín, CSIC, Granada, Spain), Vinicius Ide Franzini (Embrapa Amazônia Oriental, Belém, Brazil), Juan Manuel Ruiz Lozano (Estación Experimental del Zaidín, CSIC, Granada, Spain), Rosario Azcón (Estación Experimental del Zaidín, CSIC, Granada, Spain)

raroca@eez.csic.es

It is known that symbiosis with nitrogen fixing bacteria and legumes may enhance plant host tolerance to salinity. Most of the studies dealing with the interaction between legumes and nodule forming bacteria were focused to the effect of salt on the nitrogen fixation process. However how rhizobial symbiosis improves water status under salt stress conditions has been less studied. Here it was studied how inoculation of *Phaseolus vulgaris* plants with the rhizobial strain CIAT 899 modifies plant water relations under unstressed and salt stressed conditions. It was found that inoculated plants had higher leaf relative water content than non inoculated plants under NaCl conditions. Also, stomatal conductance was higher in inoculated than in non inoculated plants under both unstressed and salt stressed conditions. Thereafter, the capacity of absorbing water was estimated by calculating root hydraulic conductivity (L). While L was lower in inoculated plants than in non inoculated ones under unstressed conditions, salt treatment only inhibited L in non inoculated plants. These differences could not be explained by different root gene expression of plasma membrane aquaporins (PIP). However, the PIP protein amount under saline conditions was higher in inoculated roots than in non inoculated ones. Moreover, roots of inoculated plants accumulate less Na^+ ions than non inoculated ones under salt stress conditions. Therefore, rhizobial symbiosis ameliorate the water status of *P. vulagris* plants by diminishing the changes caused by salt stress on water relation parameters like stomatal conductance and L and by inducing the accumulation of PIP proteins in their roots and decreasing the accumulation of Na^+ ions in root tissues. Here is reported by the first time how rhizobial symbiosis may alter root hydraulic properties under any environmental conditions.

2-03

ANALYSIS OF GENE-EXPRESSION IN CHLOROPLASTS OF ARABIDOPSIS SEEDS DURING STRATIFICATION.

Livia Merendino* (LPCV, Grenoble, France), Sumaira Kousar (LPCV, Grenoble, France), Mustafa Malik Ghulam (LPCV, Grenoble, France), Rosa Pipitone (Università degli studi di Palermo, Italy), Florence Courtois (LPCV, Grenoble, France), Fabien Chevalier (LPCV, Grenoble, France), Thomas Pfannschmidt (LPCV, Grenoble, France) and Silva Lerbs-Mache(LPCV, Grenoble, France).

livia.merendino@cea.fr

Seed dormancy is a specific trait of many plant species to avoid germination in adverse environmental conditions. In dormant seeds, the temporary arrest in growth is often released only upon most-chilling (stratification). In the laboratory, seeds of *Arabidopsis thaliana* are usually stratified, by imbibitions on plates in darkness at 4°C, to synchronize germination in a population. The molecular bases of this process are not well understood.
Plastids are cell organelles in photosynthetic organisms, where essential functions occur. In dicotyledonous plants the plastid genome is transcribed by three RNA polymerases: RPOTp and RPOTmp, which are encoded by the nucleus, and PEP, which is encoded by the plastid itself.
Aim of our work was to study the role of the plastid gene-expression in the synchronization of seed germination during stratification in Arabidopsis. For that, we have analyzed the transcription of a selected group of plastid genes in dried and stratified seeds. Our data indicate that during stratification there is a strong synthesis of the ribosomal 16S RNAs by RPOTmp from a specific promoter, called PC, and also of the mRNAs coding for the PEP subunits by RPOTp. Germination tests using the *rpoT* mutants indicate that RPOT-mediated transcription of the plastid genome is required for efficient synchronization of seed germination. We can speculate that in stratified seeds, activation of the two RPOT polymerases allows producing the necessary plastid RNA stocks for a rapid and efficient development of the plantlet as soon as the imbibed seeds are exposed to the optimal conditions for germination (light at room temperature).
Interestingly, the activation of the PC promoter for synthesis of the 16S rRNAs was also observed in chloroplasts of 7 day old plants which have been exposed to a cold stress for few days. Our data suggest that the usage of the PC promoter both in seeds and in plantlets is activated by the cold treatment more than by a developmental signal.

2-04

CONSTITUTIVELY ACTIVE MAPKs AS A NEW TOOL TO UNDERSTAND MAPK SIGNAL TRANSDUCTION IN PLANTS

Baptiste Genot*, Heribert Hirt and Jean Colcombet (URGV, Evry, France)

genot@evry.inra.fr

Mitogen-activated protein kinases (MAPKs) are key regulators in plant signal transduction (Colcombet and Hirt, 2008). In *Arabidopsis thaliana* the most studied MAPK pathways are the MPK3, MPK4 and MPK6 signaling cascades which are activated by the PAMP (Pathogen-associated molecular pattern) flagellin. In our lab we recently identified mutations which render MAPKs constitutively active (CA). We first created MPK4-CA lines and confirmed the role of MPK4 as a negative regulator of defense responses and the production of ROS (reactive oxygen species) and SA (salicylic acid) (Berriri *et al.,* 2012). We are now studying MPK3, MPK6 and MPK11 using both knock out and constitutively active MPK lines under their own or estradiol-inducible promoters. The function of these MAPKs is studied with respect to obvious phenotypic alterations as well as their role in innate immunity. The comparison of MAPK gain-of-function with MAPK loss-of-function (T-DNA insertion) mutants provides a powerful tool to understand the roles of the different MAPKs in the absence of their upstream signals or signaling components. Constitutively active MAPKs will also be important tools to identify the respective MAPK substrates and thereby understand how and which target genes are regulated by MAPKs. This enlarged MAPK toolbox can help to provide answers that go beyond classical genetic analysis with loss-of-function or knockout mutants.

2-05

DIFFERENTIAL PHOSPHORYLATION OF CHLOROPLAST MOVEMENT PROTEINS BETWEEN STRONG LIGHT AND DARK IN ARABIDOPSIS THALIANA

Edouard Boex-Fontvieille* (Institut de Biologie des Plantes, Orsay, France), Mathieu Jossier (Institut de Biologie des Plantes, Orsay, France), Marlène Davanture (UMR de Génétique Végétale du Moulon, Gif-sur-Yvette, France), Michel Zivy (UMR de Génétique Végétale du Moulon, Gif-sur-Yvette, France), Michael Hodges (Institut de Biologie des Plantes, Orsay, France) and Guillaume Tcherkez (Institut de Biologie des Plantes, Orsay, France)

edouard.boex-fontvieille@u-psud.fr

Chloroplasts can quickly move in any direction in response to the quality, intensity and position of the incident light. Under low-intensity light, they move to the periclinal walls of palisade cells to maximize light-harvesting (accumulation response). In contrast, chloroplasts move away from strong incident light to the anticlinal walls to avoid photodamage (avoidance response). Recently, a meshwork of short chloroplast-actin filaments (cp-actin) situated at the interface between chloroplast and cytosolic membranes were visualized in *Arabidopsis thaliana* plants expressing an actin-binding protein fused to fluorescent protein. These cp-actin filaments play a major role in chloroplast relocation and their regulation depends on the blue light photoreceptor phototropin. To date, a dozen proteins have been described to be involved in cp-actin filament regulation by reverse genetic approaches. However, the downstream signaling cascade from light perception by phototropin to chloroplast movement remains unclear. In this study, we have investigated the phosphorylation status of proteins involved in chloroplast movement between a strong light condition (avoidance response) and darkness. We took advantage of a gas-exchange system with liquid N_2 spraying for instant sampling and nanoLC-MS/MS based phosphoproteomics to characterize phosphorylated peptides of *Arabidopsis thaliana* rosette leaves. We report 42 new phosphorylation sites and show concerted changes in 23 phosphorylation sites between the two conditions. Our results suggest a key role of phosphorylation in chloroplast photorelocation.

2-06

DISSECTING DEVELOPMENTAL IRON DEFICIENCY RESPONSES IN ARABIDOPSIS ROOTS USING NATURAL VARIATION

Santosh Satbhai*, Bonnie Wohlrab, Wolfgang Busch (Gregor Mendel Institute of Molecular Plant Biology, Vienna, Austria)

santosh.satbhai@gmi.oeaw.ac.at

Plant development is highly plastic and is profoundly influenced by the environment. The sessile life style of plants has led to the development of diverse mechanisms required for the perception and response to fluctuating environments. It has yet much to be learnt on how environmental conditions tunes plant growth and development. We set out to use the growth response upon iron (Fe) deficiency to identify genes that integrate environmental and internal information into the regulation of plant development. To this end, we have phenotyped more than 400 *A. thaliana* accessions for root growth traits under Fe deficient conditions. We observed remarkable variation of growth responses in Fe Deficiency conditions in those accessions. We then used those data to conduct genome wide association (GWA) mapping to identify genome regions that are involved in controlling growth responses to Fe deficiency. One of the novel candidate gene identified is a member of the Protein Phosphatase 2C (PP2C) family. A T-DNA mutant line showed significantly reduced root growth rate. Moreover, expression analysis revealed that known iron homeostasis genes (such as AtIRT1, AtFRO2, AtFIT1) were mis-regulated in the mutant line. This study demonstrates the use of natural variation to identify Fe-deficiency-regulated genes in Arabidopsis, and identified novel genes with potential new roles in signaling during Fe deficiency.

2-07

FUNCTIONAL ANALYSIS OR TWO EUCALYPTUS CBFS (C-REPEAT BINDING FACTORS) INVOLVED IN COLD TOLERANCE

Phi Bang Cao* (Laboratoire de Recherche en Sciences Végétales, UMR 5546 UPS/CNRS, Auzeville/Castanet-Tolosan, France), Hong Chien Nguyen (Laboratoire de Recherche en Sciences Végétales, UMR 5546 UPS/CNRS, Auzeville/Castanet-Tolosan, France), Hélène San Clemente (Laboratoire de Recherche en Sciences Végétales, UMR 5546 UPS/CNRS, Auzeville/Castanet-Tolosan, France), Christiane Marque (Laboratoire de Recherche en Sciences Végétales, UMR 5546 UPS/CNRS, Auzeville/Castanet-Tolosan, France) and Chantal Teulières (Laboratoire de Recherche en Sciences Végétales, UMR 5546 UPS/CNRS, Auzeville/Castanet-Tolosan, France)

cao@lrsv.ups-tlse.fr

The Eucalyptus, which is the most planted hardwood species worldwide, is a persistent and non-dormant plant. Permanent exposure of leaves to temperature changes implies specific adaptive mechanisms to withstand frost events. Eucalyptus grandis genome sequence was released in 2011, allowing genome-wide analyses of genes families. CBF transcription factors play a prominent role in the cold response pathway in binding CRT/DRE (C-RepeaT/Dehydration Responsive Element) located on the promoters of target genes. Generated in our lab, CBF-overexpressing Eucalyptus lines were found to exhibit, at in vitro stage, an increased stress tolerance and altered phenotype including developmental modifications (Navarro et al, 2011), as was described for different plant species. That suggests that the cold response and CBF pathway involved various gene families supposed to be involved in frost tolerance (cell protection) or developmental changes under stress. This poster presents in silico prediction at the genome scale of the CBFs putative target genes through identifying elements CRT/DRE in their promoter. From the whole families manually annotated in Eucalyptus genome, the putative cold and CBF-responsive genes were selected both according to CRT/DRE presence in their promoter and their representation in the EST collection of cold-acclimated E. gunnii (Keller et al, 2009). In addition to in silico analyses, the expression of the selected genes were quantified in CBF-overexpressing Eucalyptus lines. Finally, the poster presents the first data about CBF-regulon in Eucalyptus.

2-08

IDENTIFICATION AND CHARACTERIZATION OF THE ChLoride CHANNEL, AtCLCg, INVOLVED IN SALT STRESS RESPONSE IN *ARABIDOPSIS THALIANA*

Chi Tam NGUYEN[1], Astrid AGORIO[1], Sébastien THOMINE[1] and Sophie FILLEUR[1,2] *

[1] Institut des Sciences du Végétal, CNRS, 1 avenue de la Terrasse 91190 Gif sur Yvette - France.
[2] Université Paris 7 Denis Diderot, U.F.R. Sciences du Vivant, 35 rue Hélène Brion, 75205 Paris Cedex 13 France.

sophie.filleur@isv.cnrs-gif.fr

In plant cells, anion channels and transporters are essential for key functions such as nutrition, ion homeostasis and, resistance to biotic or abiotic stresses. In *Arabidopsis thaliana*, members of the ChLoride Channel (CLC) family localized on the tonoplast are required for nitrate homeostasis (AtCLCa, AtCLCb) or involved in salt tolerance (AtCLCc).
We characterized the chloride channel AtCLCg in *A. thaliana*. We found that the protein is localized in the tonoplast. When grown on 75 mM NaCl, *atclcg* KO mutants showed a decrease by 20% of total plant fresh weight and an over-accumulation of chloride in shoots by 21% compared to wild-type. A similar phenotype was found in the presence of 75 mM KCl for *atclcg* mutants, but no difference was detected in response to 140 mM mannitol. These results suggest that the NaCl hypersensitivity phenotype of *atclcg* mutants depends on the ionic component and not the osmotic effect of salt stress.
Since AtCLCg and AtCLCc share a high degree of homology, approximately 75% of identity at protein level, and both are necessary for NaCl response. We constructed an *atclcc atclcg* double KO mutant. Phenotypic analysis showed that these two mutations do not have additive effect on 75 mM NaCl. Gene expression analysis revealed that: (i) *AtCLCg* and *AtCLCc* are strongly expressed in mesophyll cells and stomata respectively and (ii) *AtCLCg* is repressed in the *clcc* mutant background, and conversely. Altogether these results demonstrate that both AtCLCc and AtCLCg are not redundant and form part of a complex NaCl auto-regulatory network.

2-09

IDENTIFICATION AND FUNCTIONAL STUDY OF A NOVEL MAPK MODULE INVOLVED IN ABA SIGNALING

Axel de Zelicourt* (URGV - Plant Genomics Research, Evry, France), Agyemang Danquah (URGV - Plant Genomics Research, Evry, France), Marie Boudsocq (URGV - Plant Genomics Research, Evry, France), Nicolas Frei dit Frey (URGV - Plant Genomics Research, Evry, France), Stephanie Pateyron (URGV - Plant Genomics Research, Evry, France), Jean Colcombet (URGV - Plant Genomics Research, Evry, France), and Heribert Hirt (URGV - Plant Genomics Research, Evry, France)

axel@evry.inra.fr

The phytohormone abscisic acid (ABA) regulates diverse cellular processes and transduces signals to protect plants from abiotic stresses. Among the molecular elements working in ABA signaling, the mitogen activated protein kinase (MAPK) cascades play important roles in regulating the signaling network. To date, however, only a handful of MAPKs have been identified and characterized in ABA signaling. To identify these key factors in stress tolerance, we have performed bioinformatics analysis on existing transcriptomic data on abiotic stresses and have identified several candidate genes in *Arabidopsis thaliana*. Among them, two closely related *MAPKKKs* were selected due to their induction by ABA and abiotic stresses. In two ABA insensitive mutants, *pyr1*/*pyl1*/*pyl2*/*pyl4* and $hab1^{G246D}$, the ABA- and NaCl-dependent expression of the two *MAPKKKs* was strongly reduced, indicating that these two kinases act downstream of the core ABA signaling complex. Using yeast-2-hybrid and BiFC assays, only one MAPKK and several MAPKs were identified as the downstream pathway components of the two MAPKKKs. Moreover, in mesophyll protoplasts, we could reconstruct the whole MAPK module and show that the module is activated after ABA treatment. Genetic evidence showed that, the MAPKK *knock out* mutant is hypersensitive whereas the constitutively active MAPKK lines are resistant to ABA, NaCl and mannitol stress. While the double mutant of the two MAPKKKs was not affected in germination, seedlings were more susceptible to salt stress, a phenotype that is shared with the MAPKK mutant. Taken together, these results suggest that we have identified an entire MAPK cascade that is involved in the regulation of plant responses to abiotic stress in an ABA-dependent manner.

2-10

IDENTIFICATION OF NOVEL CANDIDATES INVOLVED IN TEMPERATURE PERCEPTION IN ARABIDOPSIS

Manoj Kumar* and Philip A. Wigge (Sainsbury Laboratory, Cambridge University, Cambridge, United Kingdom)

Manoj.Kumar@slcu.cam.ac.uk

How plants and other species perceive the temperature is still poorly understood at the molecular level. However, plants are unique considering the fact that being non-motile they quickly need to acclimatise to change in temperature to survive, suggesting prevalence of very precise temperature perception mechanisms operative in the plant system. But the temperature sensing research in plants is largely focused on the temperature stress tolerance, overlooking the fact that the plants are also affected within a non-stress range of temperature. The gap in this knowledge was enlightened with two recent reports revealing the role of H2A.Z nucleosome in temperature perception and the contribution of PIF4 transcription factor in regulating flowering time in response to change in ambient temperature. With the increasing availability of advanced molecular biology and genomics tools in plant research, many new reports emerged demonstrating the effect of temperature in hormone signalling, flowering time, the circadian clock, light signal transduction, cold and heat acclimatisation, metabolism and growth and cell defence mechanisms. This suggests that there must be an intricate network functioning between the site of temperature perception (the thermosensor) and the downstream gene networks that might be distinct for different tissues within a plant. In order to understand the thermosensory stimuli and the pathways and to be able to manipulate them in future, it would be valuable to identify novel genes that are responsible for temperature perception under non-stress levels. We have established the use of *HSP70::LUCIFERASE* reporter lines as an indicator of the thermosensory status of the plant. A preliminary mutagenesis screen with *HSP70::LUC* has been very fruitful, and we have identified many new potential candidates, which might act as thermosensors or be involved in maintaining different physiological pathways in response to change in ambient temperature.

2-11

IDENTIFICATION OF REGULATORY FACTORS WITH A DUAL IMPLICATION IN LEGUME SYMBIOTIC NODULE SENESCENCE AND ROOT ABIOTIC STRESS RESPONSE

Théophile Kazmierczak* (Institut des Sciences du Végétal, Gif-sur-Yvette, France), Audrey Macquet (Institut des Sciences du Végétal, Gif-sur-Yvette, France), Roberta Loh (Universidade Federal do Rio de Janeiro, Brésil), Carole Laffont (Institut des Sciences du Végétal, Gif-sur-Yvette, France), Marcio Alves Ferreira (Universidade Federal do Rio de Janeiro, Brésil), Martin Crespi (Institut des Sciences du Végétal, Gif-sur-Yvette, France), Florian Frugier (Institut des Sciences du Végétal, Gif-sur-Yvette, France), Véronique Gruber (Institut des Sciences du Végétal, Gif-sur-Yvette, France ; Université Paris Diderot, Paris, France)

Theophile.Kazmierczak@isv.cnrs-gif.fr

To overcome environmental stresses, the legume root system is able to adapt its architecture through root branching and the formation of nitrogen-fixing nodules. These nodules result from a symbiotic association between roots and soil rhizobia, allowing plant growth in nitrogen poor soils. A critical step in stress responses involves transcriptional regulation, and previous transcriptomic analyses identified several transcription factors rapidly regulated by salt stress in *Medicago* roots, including some belonging to the NAC (NAM /ATAF/ CUC) family (Gruber at al., 2009). Interestingly, a specific *NAC* gene, Mt*NAC969*, revealed a differential regulation of Mt*NAC969* in roots and nodules in response to salt stress. The expression of Mt*NAC969* was additionally induced in symbiotic nodule after a nitrate treatment provoking their senescence. Accordingly, Mt*NAC969* RNAi nodules showed a premature senescence, based on dedicated markers and detailed histology (de Zélicourt et al., 2012). Altogether, these results demonstrate that the Mt*NAC969* transcription factor is differentially regulated by environmental cues depending on the organ considered (roots and nodules) and has a dual function in root adaptive response to salt stress and in symbiotic nodule senescence.
To understand mechanisms controlling both root salt stress response and nodule senescence in more details, we initiated a bioinformatic screen on available public transcriptomes to search for genes with a similar expression pattern than *MtNAC969*. These studies will also contribute to establish the regulatory network controlled by Mt*NAC969*. Progress in the characterization of selected regulatory genes will be reported.

2-12

IMPORTANCE OF NAD HOMEOSTASIS FOR PLANT GROWTH AND RESISTANCE TO ADVERSE ENVIRONMENTAL CONDITIONS

Linda de Bont* (Institut de Biologie des Plantes, IBP, Orsay, France), Pierre Pétriacq (IBP), Marie Garmier (IBP), Marina Pfalz (Laboratoire Ecologie Systématique Evolution, ESE, Orsay, France), Kevin Phan (IBP), Thomas Poitevin (IBP), Juergen Kroymann (ESE), Guillaume Tcherkez (IBP), Michael Hodges (IBP), Bertrand Gakière (IBP)

Linda.de-bont@u-psud.fr

As sessile organisms, plants must adapt to changing environmental conditions that affect their optimal growth and development. Adaptation relies on metabolites such as NAD (nicotinamide adenine dinucleotide), a redox compound involved in many cellular functions such as respiration and nitrogen assimilation. Recently, NAD has also been implicated in signaling that involves net consumption of the molecule. These processes require NAD synthesis and could be influenced by cellular NAD concentration. In plants, L-aspartate oxidase (AO) is the first committed enzyme engaging NAD biosynthesis. Literature highlights that AO gene expression is up-regulated during developmental and stress conditions. While these events imply a large NAD consumption, the single gene encoding AO is an interesting target to deregulate NAD production in Arabidopsis. T-DNA homozygous mutant lines for AO (*mAO*) are being used to study the impact of an NAD decrease on plant physiology and stress resistance. *mAO* plants have decreased levels of AO transcripts and reduced NAD/P(H) contents that disrupt the global redox state of the plant cell, and impact overall metabolism. Indeed, these mutants exhibit a C/N metabolite imbalance and perturbations in C/N assimilatory activities. These modifications result in dwarf phenotype and pale green leaves, pointing out the critical role of NAD for harmonious growth and development. Additionally, these mutants show altered responses to various abiotic (nitrogen limitation, drought and heat) and biotic (aphids, bacteria) stresses, emphasizing NAD effects on plant adaptation to various environmental perturbations.

2-13

INCREASED TOLERANCE TO HIDRIC STRESS OF LAVANDULA DENTATA PLANTS BY THE APPLICATION OF AUTOCHTHONOUS MICROORGANISMS AND COMPOST.

Elisabeth Armada* (Estación experimental del Zaídin-CSIC, Granada, Spain), Rosario Azcón (Estación Experimental del Zaidín-CSIC, Granada, Spain)

elisabeth.armada@eez.csic.es

Drought stress is one of the most important abiotic factors limiting plant growth and performance in large areas of the Southeast of Spain. Limitation of water causes a series of detrimental changes in plant nutrition and plant physiology. Specific soil microbial communities tend to develop a series of activities of great importance, in uptake of nutrients and water that improved plant growth and the quality of the soil. As result of the application of autochthonous arbuscular mycorrhizal fungi (AMF) and compost (C) increased the plant growth under natural drought conditions. The inoculation of native bacteria *Bacillus thuringiensis* (*B.t*) also was highly effective. As mechanisms of protection of *Lavandula dentata* to drought-induced dehydration (45-50% whc), the treatments applied as *B.t* or C reduced antioxidants enzymatic activities as APX, GR and proline, which indicate that these plants are suffering a lower stress. Simple *B.t* inoculation increases the content of K, and C and AMF increases P and K while the coinoculation with *B.t* and AMF promotes P, K, Ca, Mg uptake. Therefore, the interaction of plants with beneficial soil microorganisms and compost can increase drought adaptation and plant tolerance to nutrients and water shortages.

2-14

METABOLITE FINGERPRINTS AND PROLINE RESPONSE OF SOURCE AND SINK OILSEED RAPE LEAVES UNDER NITROGEN AND/OR WATER LIMITING CONDITIONS

Benjamin Albert (UMR IGEPP, Rennes-Le Rheu, France), Françoise Le Cahérec (UMR IGEPP, Rennes-Le Rheu, France), Pascal Faes (UMR IGEPP, Rennes-Le Rheu, France), Carole Deleu (UMR IGEPP, Rennes-Le Rheu, France), Marie-Françoise Niogret (UMR IGEPP, Rennes-Le Rheu, France), Solenne Berardocco (UMR IGEPP, Rennes-Le Rheu, France), Laurent Leport (UMR IGEPP, Rennes-Le Rheu, France), Alain Bouchereau* (UMR IGEPP, Rennes-Le Rheu, France)

alain.bouchereau@univ-rennes1.fr

Compared with other crops, oilseed rape (OSR) needs very high amounts of nitrogen (N) fertilisers, since this species is known to have low nitrogen use efficiency (NUE), which is mainly due to a weak N remobilisation performance from senescing source leaves to developing sink tissues and to seed filling. Moreover, in areas with temperate climate OSR is often subjected to drought stress periods that affect plant growth, development and yield.
In this context, improvement of NUE (and water use efficiency) in OSR is an actual challenge in prevision to lower N supply and limitation of water availability. Thus appreciation of N fertilization management impact on plant water stress susceptibility should be anticipated and conversely plant response to water shortage impact on NUE should be evaluated. In that way, attention about regulation of nitrogen metabolism in leaves as a major source of remobilized nutritive compounds and stress protective metabolites is of prime interest.
The goal of our work was to fingerprint (UPLC-MS ; GC-MS ; multivariate statistical analysis) leaf metabolites rank by rank at the whole plant level under different nitrogen and water regimes, to investigate at vegetative stages the metabolic impact of combined N and water shortages, and to search for biochemical attributes of N remobilisation and stress protection. Metabolic signatures of leaf development and nutritional status have been outlined. Adjustments of primary metabolism to water depletion was observed to be strongly dependent to leaf N nutritional status. Moreover a special attention has been paid to proline metabolism investigation (through proline amount and RT-Q-PCR *BnP5CS* and *BnPDH* gene transcript levels measurements) as a major syndrome of OSR response to water stress and as a putative interference to NUE under water depletion. Conversely the question was to evaluate the nitrogen limitation effect on the proline response and its corresponding impact on plant water stress tolerance.

2-15

PHOSPHOREGULATION OF THE PHOTORESPIRATORY ENZYME, GLYCOLATE OXIDASE, IN RESPONSE TO LIGHT AND CO_2 CONTENT IN ARABIDOPSIS THALIANA

Younes Dellero (Institut de Biologie des Plantes, Orsay, France), Edouard Boex-Fontvieille (Institut de Biologie des Plantes, Orsay, France), Sophie Massot (Institut de Biologie des Plantes, Orsay, France), Valérie Flesch (Institut de Biologie des Plantes, Orsay, France), Françoise Gilard (Plateforme Métabolisme-Métabolome, Orsay, France), Marlène Davanture (UMR de Génétique Végétale du Moulon, Gif-sur-Yvette, France), Michel Zivy (UMR de Génétique Végétale du Moulon, Gif-sur-Yvette, France), Guillaume Tcherkez (Institut de Biologie des Plantes, Orsay, France), Michael Hodges (Institut de Biologie des Plantes, Orsay, France), Mathieu Jossier* (Institut de Biologie des Plantes, Orsay, France)

mathieu.jossier@u-psud.fr

Photorespiration is an essential process for all oxygenic photosynthetic organisms. It allows the recycling of the carbon atoms of 2-phosphoglycolate produced by ribulose-1,5-bisphosphate carboxylase/oxygenase (Rubisco) oxygenase activity, as well as the removal of potentially toxic metabolites. Although photorespiration has been extensively described, nothing is known about the regulation of this complex cycle that takes place in chloroplasts, peroxisomes and mitochondria. A global quantitative phosphoproteomics analysis was undertaken using leaf extracts from *Arabidopsis thaliana* plants subjected to 4 h of light either at 100ppm, 380ppm or 1000ppm of CO_2 in 21% O_2 to modify the photorespiration rate (oxygenation-to-carboxylation ratio). Light-dark changes in protein phosphorylation were also analyzed at 380ppm of CO_2. Amongst the 264 phosphopeptides showing a significant change between the different conditions in their phosphorylation status (99% confidence level), phosphopeptides corresponding to only two photorespiratory enzymes were detected. One of these phosphopeptides corresponded to the peroxisomal enzyme glycolate oxidase (GOX). Mutation of the phosphorylated Thr158 residue either to an alanine or an aspartic acid (phosphorylation-mimic) modified GOX activity suggesting a potential role in photorespiratory cycle regulation. To investigate the consequence of an altered GOX activity on leaf photorespiration and primary metabolism, T-DNA insertion mutants of GOX were subjected to metabolomic analyses and our data show a strong decrease in the content of several organic acids in *gox* rosette leaves. Further work will be carried out to understand the role of GOX phosphorylation and its possible impact on plant metabolism including the Krebs cycle.

2-16

PHYSIOLOGICAL AND MORPHOLOGICAL CHARACTERIZATION OF ARABIDOPSIS TRANSPARENT TESTA 15 MUTANT SEEDS AFFECTED IN FLAVONOID METABOLISM

Isabelle Debeaujon* (Institut Jean-Pierre Bourgin, Versailles, France), Guillaume De Lagarde (Institut Jean-Pierre Bourgin, Versailles, France), Elodie Akary (Institut Jean-Pierre Bourgin, Versailles, France), Adeline Berger (Institut Jean-Pierre Bourgin, Versailles, France), Nathalie Nesi (Institut de Génétique, Environnement et Protection des Plantes, Rennes, France), Loïc Lepiniec (Institut Jean-Pierre Bourgin, Versailles, France).

isabelle.debeaujon@versailles.inra.fr

Flavonoids are polyphenolic secondary metabolites synthesized by plants during their development and in response to various biotic and abiotic stresses. They influence the agronomic and nutritional qualities of seeds. The analysis of *transparent testa (tt)* mutants of *Arabidopsis thaliana* affected in seed coat pigmentation enabled to identify many genes involved in the flavonoid biosynthetic pathway and are efficient tools to assess the roles of flavonoids in seed biology. The present study focuses on the *tt15* mutant which exhibits a pale gray-brown seed coat colour caused by the disruption of a gene encoding an UDP-glucose-sterol-glucosyltransferase. The characterization of *tt15* seeds including proanthocyanidin detection, seed coat permeability, dormancy level, germination behaviour under stress, longevity and morphological analysis of developing and mature seeds revealed that the mutation causes a range of pleiotropic phenotypes.

2-17

PROVING THE ROLE OF TACBF14 AND TACBF15 TRANSCRIPTION FACTORS IN THE COLD ACCLIMATION PROCESS AND IN FROST TOLERANCE BY TRANSFORMATION OF WHEAT AND BARLEY

Alexandra Soltész* (Agricultural Institute, Centre for Agricultural Research, Hungarian Academy of Sciences, Martonvásár, Hungary), Mark Smedley (John Innes Centre, Norwich, United Kingdom), Caroline Sparks (Rothamsted Research, Harpenden, Hertfordshire, United Kingdom), Gábor Galiba (Agricultural Institute, Centre for Agricultural Research, Hungarian Academy of Sciences, Martonvásár, Hungary), Huw D Jones (Rothamsted Research, Harpenden, Hertfordshire, United Kingdom), Wendy Harwood (John Innes Centre, Norwich, United Kingdom), Attila Vágújfalvi (Agricultural Institute, Centre for Agricultural Research, Hungarian Academy of Sciences, Martonvásár, Hungary)

soltesz.alexandra@agrar.mta.hu

Drought and frost are the two most important abiotic stresses which limit crop production in temperate zones. The enhancement of winter hardiness is one of the essential tasks facing breeders of winter cereals. For this reason, the examination of those regulatory genes involved in the cold-acclimation processes is of central importance. *CBF* (C-repeat Binding Factor) genes are among the most studied regulators in the plant kingdom, coding for transcription factors involved in cold and drought stress response. Our previous work and several further studies have highlighted that two CBF transcription factors (CBF14 and CBF15) have an outstanding role in the development of frost tolerance in wheat. The aim of our work was to confirm their function directly by transformation of economically important cereals. *TaCBF14* and *TaCBF15* genes were isolated from winter wheat (*Triticum aestivum* L. Cheyenne) and then transformed into spring barley (*Hordeum vulgare* L. Golden Promise) and spring wheat ('Cadenza'). Transgenic lines exhibited moderate retarded development, slower growth and minor late flowering compared to the wild type, with enhanced transcript level of a gibberellin catabolic gene. The frost tolerance of all the CBF over-expressing lines has been tested. Two different types of frost tests were applied; plants were hardened at low temperature before freezing, or plants were subjected to frost without a hardening period. Our analysis showed, that *TaCBF14* and *TaCBF15* transgenes improve the frost tolerance to such an extent that the best transgenic lines were able to survive freezing temperatures several degrees lower than that which proved lethal for the wild type. The freezing test was evaluated by the injury of the photosystem II, by examining the level of cell damage, and also by studying the level of plant survival. The transgene expression and several cold-regulated downstream genes were studied.

2-18

RESPONSES OF MYCORRHIZAL TOMATO PLANTS TO FLOODING STRESS

Mónica Calvo-Polanco* (Estación Experimental del Zaidín, CSIC, Granada, Spain), Angel María Zamarreño (Timac Agro España SA, Navarra, Spain), Sonia Molina (Estación Experimental del Zaidín, CSIC, Granada, Spain), José María García-Mina (Timac Agro España SA, Navarra, Spain) and Ricardo Aroca (Estación Experimental del Zaidín, CSIC, Granada, Spain)

monica.calvo@eez.csic.es

Flooding creates hypoxic conditions around the roots affecting gas exchange, hormonal balance and water relations. Stomata closure is among the initial symptoms of oxygen deficiency in the root zone due to the reduced ability of the root system to conduct water. The reduction of root hydraulic conductivity in hypoxic plants has been linked to the inhibition of aquaporin mediated water transport. However, the role of plant hormones in the adaptation of plants to low oxygen conditions is less studied and yet not well understood. Arbuscular mycorrhizal fungi are symbiotic microorganisms that have been shown to play a major role in plant tolerance to water deficit under several abiotic and biotic stresses through their effect on plant aquaporins. With fungal mycelia containing their own aquaporins, the role of arbuscular mycorrhizal fungi in plant aquaporin function and regulation is still unknown and may be one of the main contributions to plant tolerance. Also, the role of mycorrhizal fungi under flooding conditions has not been addressed yet.We examined the effects of flooding in non-mycorrhizal wild type and never ripe tomato (*Solanum lycopersicum*) plants and plants inoculated with *Rhizophagus irregularis* for seven days. Mycorrhizal and non-mycorrhizal flooded plants showed a reduction in stomatal conductance, while root hydraulic conductivity increased in the presence of mycorrhizal fungi subjected to water logging both in wild type and never ripe plants. In wild type mycorrhizal plants, the increase on root hydraulic conductivity was attributed to the increase of the expression of plant aquaporins *SlPIP1;7* and *SlPIP2;3*, and the fungal aquaporin *GintAQP1*. There was also an increase on the abundance of phosphorylated PIP2 proteins at serine 280, as well as root IAA, while root ethylene decreased compared with non-mycorrhizal plants. In the case of never ripe plants, a higher root hydraulic conductivity in plants inoculated with *R. irregularis* was not related with the aquaporin expression of any of the plant or fungal aquaporins studied, although it could be related with an increase of root IAA and Salicylic Acid. In conclusion, wild type and never ripe tomato plants have different strategies to cope with the lack of oxygen within the roots and the presence of mycorrhizal fungi helps maintaining a fine regulation of water uptake.

2-19

ROLE OF PI3K (PHOSPHATIDYLINOSITOL 3-KINASE) AND TOR (TARGET OF RAPAMYCIN) SIGNALLING PATHWAYS ON PROLINE METABOLISM IN RESPONSE TO SALT STRESS

Anne-Sophie Leprince* (UPMC, Paris, France), Nelly Magalhaes (UPMC, Paris et IJPB, Versailles, France), Maxime Szambien (UPMC, Paris et IJPB, Versailles, France), Alexandra Thielland (UPMC, Paris et IJPB, Versailles, France), Delphine De Vos (UPMC, Paris, France), Gilles Clément (IJPB, Versailles, France), Christian Meyer (IJPB, Versailles, France) and Arnould Savouré (UPMC, Paris, France)

anne-sophie.leprince@upmc.fr

Plants exposed to drought and salinity constraints have developed various strategies such as accumulation of osmolytes in order to challenge water loss. Proline is considered as an important osmolyte, which accumulation may participate to osmotic adjustment. Proline accumulation under either water or salt constraint results from a balance between its biosynthesis and its catabolism. In order to understand how proline metabolism is regulated under salt constraint, we focused our studies on TOR (Target Of Rapamycin) and PI3K (PhosphatidylInositol 3-Kinase) signalling pathways. These proteins are important regulatory elements in eukaryotic organisms, controlling various metabolic and developmental processes. The TOR kinase forms a complex with the LST8/GbetaL and RAPTOR proteins (TORC1). The activity of TOR complex is induced under favourable conditions while nutrient limitations or stresses modify its activity. In animals TORC1 can be regulated by a class-III PI3K, which catalyses the formation of PI3P (PhosphatidylInositol-3-Phosphate) from PI. In plants, both TOR and PI3K seem to be involved in plant responses to salt and water constraints. Indeed we show that both LY294002, a PI3K inhibitor, and *raptor* mutations cause a decrease in proline accumulation under salt treatment. Consistently, this lower accumulation of proline is correlated with an accumulation of transcripts and proteins of ProDH1 (Proline Dehydrogenase 1, the key-enzyme of proline catabolism). Metabolomic data suggest that the metabolism of other osmolytes, such as galactinol and raffinose, could also be regulated by these two kinases. Taken together, these data suggest that the PI3K and TOR signalling pathways could be interconnected, influencing the response of plants to salt stress.

2-20

SALT STRESS SIGNALLING: SHEDDING FURTHER LIGHT ON THE SOS NETWORK

Kerstin Duscha*, Maria Müller and H. Ekkehard Neuhaus (University of Kaiserslautern, Germany)

kduscha@rhrk.uni-kl.de

As salinity causes a severe decrease in growth rate and yield plants have developed different mechanisms to cope especially with the predominant overabundance of sodium: to maintain Na^+ homeostasis they either extrude sodium ions from the cell or they compartmentalize sodium into the vacuole. These processes are achieved by Na^+/H^+ antiporters catalyzing sodium movement against the respective electrochemical gradient. There are three different families of these antiporters known in plants: the plastidial IT/NhaD family, the intracellular IC-NHE/NHX family and the NhaP/SOS1 family, whose members are localized to the plasma membrane.

The highly specific Na^+/H^+ exchanger SOS1 from *Arabidopsis thaliana* consists of 12 transmembrane domains followed by a remarkably long hydrophilic C-terminal extension facing the cytosol. A major lack of understanding concerns the necessity and function of this C-terminal tail as it is absent in all members of the other transporter families although the N-terminal domains are conserved. Thus, pull-down assays should help to identify potential binding partners interacting with the C-terminal region. In addition, qualitative vacuole proteome studies of wild type as well as *sos* mutant plants are carried out in order to decipher tonoplast proteins involved in salt stress response.

2-21

STRICT REGULATION OF A PLASTIDIC NA^+/H^+-ANTIPORTER IS IMPORTANT FOR ARABIDOPSIS SALT-STRESS TOLERANCE

Maria Müller[1], H.-Henning Kunz[2], Julian Schroeder[2], Grant Kemp[3], Howard Young[3] and H. Ekkehard Neuhaus[1]

[1] Plant Physiology, University of Kaiserslautern,
[2] UC San Diego, Division of Biological Sciences Cell and Developmental Biology Section & Ctr for Mol. Genetic
[3]University of Alberta, Department of Biochemistry

mariamue@rhrk.uni-kl.de

High salt concentrations in soil impose osmotic and ionic stress on plants and thus limit their growth and yield. Therefore, plants have evolved mechanisms to avoid deleterious cytosolic Na^+ concentrations, including active Na^+ extrusion out of the cell and intracellular compartmentalization - mainly to the vacuole. Na+/H+ antiporters in the plasma membrane (SOS1 family) and the tonoplast (NHX family) have been characterized in detail so far. It is discussed, whether mitochondria and plastids (NhaD family) contribute to a certain extent to the overall subcellular compartmentalization of sodium.
NhD1 from *Arabidopsis thaliana* locates to the inner envelope membrane of plastids, as shown by GFP analysis. Expression studies reveal its involvement into salt stress response. Complementation of an *E. coli* knock-out strain (lacking the main bacterial Na^+/H^+ antiporters) and reconstitution of NhD1 into proteoliposomes, followed by uptake studies indicate the function of NhD1 as a Na^+/H^+ transporter. Analysis of *nhd1* knock-out and overexpressor lines reveal an increased salt sensitivity of these *Arabidopsis* mutants, displayed by a reduced fresh weight production and lower chlorophyll content. Furthermore, mutants show diminished photosynthetic activity and increased plastidic sodium levels indicating the importance of a strict regulation of NhD1 expression upon salt stress.

2-22

SUCROSE PROMOTES AXILLARY BUD OUTGROWTH IN ROSA HYBRIDA AND PLAYS A SIGNAL ROLE DURING THIS PROCESS

François Barbier* (IRHS Angers), Jessica Bertheloot (IRHS Angers), Thomas Péron (IRHS Angers), Maria-Dolores Perez-Garcia (IRHS Angers) and Soulaiman Sakr (IRHS Angers).

IRHS : Institut de Recherche en Horticulture et Semences (INRA, Agrocampus-Ouest, Université d'Angers), Angers, France.

francois.barbier@agrocampus-ouest.fr

Shoot branching is a developmental process by which axillary buds are released from dormancy and develop into new axes. Bud outgrowth is largely affected by a number of environmental factors such as temperature, air and soil humidity, gravitropism and light, confering thus plasticity to the plant development. Control of bud outgrowth is thereby a key mechanism in the establishment of plant architecture in response to environment. Sugars, whose levels in plant are also highly dependent on the environment, have recently been shown to be implicated during bud outgrowth in *Rosa hybrida*. To test the impact of different sucrose levels on bud outgrowth, excised buds have been *in vitro* cultivated on MS media containing different sucrose concentrations. We then tested the impact of different non-metabolizable sucrose analogs to put in evidence a potential sucrose-signaling pathway in this process. Our results revealed that sucrose levels modulated bud outgrowth and that this dissacharide could also play a signal role during this event. Moreover, increasing sucrose supply to *in vitro* cultivated buds released the inhibitory effect of auxin on bud outgrowth, putting for the first time in evidence an antagonsim between this nutrient and this hormone. Further analysis revealed that the polarization of the auxin transport between bud and stem, which is a prerequisite to allow bud to grow out, is a target of the antagonism between sucrose and auxin. This work proposes a model that integrates sucrose as an endogenous signal in the complex network that regulates bud outgrowth in response to environment.

2-23

THE N-END RULE PATHWAY OF TARGETED PROTEOLYSIS: NITRIC OXIDE AS A MEDIATOR OF TRANSCRIPTION FACTOR DEGRADATION?

Bulut Hamali, Prabhavathi Talloji, Andreas Bachmair* (Dept. of Biochemistry and Cell Biology, Center for Molecular Biology, Max F. Perutz Laboratories, University of Vienna, Vienna, Austria)

andreas.bachmair@univie.ac.at

The N-end rule pathway of ubiquitin- and proteasome-dependent protein turnover targets proteins for rapid proteolytic destruction that have an amino acid with large side chain at the amino terminus. Mutants in this pathway have defects in their response to anoxia (as occurs during root flooding) and in germination, and display a delayed senescence phenotype. A group of ERF transcription factors with Met-Cys amino terminus are substrates of the N-end rule pathway. After Met removal, the exposed Cys can be oxidized to Cysteic acid. This side chain enlargement results in degradation of the oxidized transcription factor via the N-end rule. In animals, similar oxidation steps require an S-nitroso intermediate, linking the N-end rule pathway to nitric oxide signaling. To explore possible NO-dependence of Cys oxidation in plants, we study degradation of a GUS model substrate with amino-terminal Cys residue. Furthermore, we have collected mutations in all known components of the N-end rule pathway, to assess the contribution of specific genes to the reaction. The mutants also allow us to explore functions of the N-end rule pathway that are independent of nitric oxide.

2-24

UBIQUITIN-DEPENDENT ENDOCYTOSIS AND POLAR RECYCLING OF IRT1 TRANSPORTER CONTROLS PLANT METAL NUTRITION

Grégory Vert (Institut des Sciences du Végétal, CNRS UPR2355, Gif-sur-Yvette, France)

gregory.vert@isv.cnrs-gif.fr

The model plant Arabidopsis takes up iron from the soil using the IRON-REGULATED TRANSPORTER 1 (IRT1) high-affinity iron transporter at the root surface. Sophisticated regulatory mechanisms allow plants to tightly control the levels of IRT1, ensuring optimal absorption of essential but toxic iron, as well as other metals transported by IRT1. We recently uncovered that IRT1 protein localizes to the early endosomes, but rapidly cycles with the cell surface to mediate iron and metal uptake. The dynamic behavior of IRT1 is under the control of monoubiquitin-dependent endocytosis and is independent of iron nutrition. Limiting the pool of IRT1 at the plasma membrane appears as a critical protective mechanism to limit the absorption of readily available metals over iron. This scenario is further confirmed by the metal dependency of IRT1 ubiquitin-dependent endocytosis. IRT1 is found at the plasma membrane upon metal depletion and harbors lateral polarity in root epidermal cells. We identified a factor controlling the localization of IRT1 at the cell surface and its polarity. Interestingly, plants displaying non-polar IRT1 localization show defects in iron homeostasis, hence pointing to the crucial role of polarity in driving the radial transport of iron in plant roots.

SESSION 3:

BIODIVERSITY AND NATURAL VARIATION

3-01

BIODIVERSITY AND ADAPTATION OF PLANTS TO ENVIRONMENTAL STRESS: RESPONSE TO CADMIUM IN THE MODEL PLANT *MEDICAGO TRUNCATULA*

Sondes Rahoui (*F.S.B, Université de Carthage*, Tunisie & ECOLAB UMR 5245, INP-ENSAT, Toulouse, France), Ivana Todorovic (ECOLAB UMR 5245, INP-ENSAT, Toulouse, France), Godratollah Saeidi (Isfahan University of Technology, Isfahan, Iran), Cécile Ben* (ECOLAB UMR 5245, INP-ENSAT, Toulouse, France), Camille Dumat (ECOLAB UMR 5245, INP-ENSAT, Toulouse, France), Xiangyang Hu (Kunming Institute of Botany CAS, Kunming, Chine), Martina Rickauer (ECOLAB UMR 5245, INP-ENSAT, Toulouse, France), *Ezzedine El Ferjani* (*F.S.B, Université de Carthage*, Tunisie), Laurent Gentzbittel (ECOLAB UMR 5245, INP-ENSAT, Toulouse, France)

ben@ensat.fr

Anthropogenic activity has contributed widely to soil pollution with Cadmium (Cd), a highly toxic heavy metal. Our study aims at characterizing the biodiversity of the response to this pollutant in the model legume *Medicago truncatula*.

Thirty lines of *M. truncatula* originating from contrasted pedoclimatic regions around the Mediterranean sea were studied for their response to cadmium. Cd treatment of seedlings inhibited root growth in a genotype-specific manner. Comparison of root growth in control and polluted conditions suggests that Cd tolerance has a cost.

Six lines were studied in detail. The ratio of root growth (Cd-treated/control) was used to identify tolerant and susceptible lines. Cadmium and mineral nutrients as well as carbohydrate and amino acid contents were measured in seedlings treated with Cd. For all accessions, Cd was accumulated preferentially in the root but part was transferred to the shoot. The distribution of other ions from the seed reserve was affected by Cd-treatment in a genotype-dependent manner. Solute leakage into the germination medium indicates a failure in reserve mobilization after pollutant exposure. This response, which is correlated with delay in radicle growth, is associated with sugar transporter alteration, membrane integrity impairment and redox status damages as evidenced by high malondialdehyde and hydrogen peroxide levels in Cd-poisoned tissues. Microscopic studies of Cd impact on root cytology showed that ROS and phenolic compounds were accumulated. Expression of related defense genes (e.g. SOD, POX, glutathione metabolism) was also studied.

These results provide first indications of mechanisms that allow the plant to tolerate Cd pollution. Understanding the response of plants in polluted natural and peri-urban areas should help to develop varieties that might be used for restoration of contaminated soils through phytoremediation. Legume plants will have the additional advantage of improving the soil's nitrogen status.

3-02

DECODING THE COMPLEXITY OF QUANTITATIVE NATURAL VARIATION AND RESPONSE TO DROUGHT STRESS IN *ARADIDOPSIS THALIANA*

Mathieu Hanemian* (Institut Jean-Pierre Bourgin, Versailles, France), Elodie Marchadier (Institut Jean-Pierre Bourgin, Versailles, France), Parham Haddadi (Institut Jean-Pierre Bourgin, Versailles, France), Christos Bazakos (Institut Jean-Pierre Bourgin, Versailles, France), Sébastien Tisné (CIRAD, Montpellier, France), Liên Bach (Université Montpellier 2, Montpellier, France), Francisco Cubillos (Institut Jean-Pierre Bourgin, Versailles, France) & Olivier Loudet (Institut Jean-Pierre Bourgin, Versailles, France).

mathieu.hanemian@versailles.inra.fr

Most phenotypic traits of agronomical, ecological and economical interest are of quantitative nature, revealing the complex interplay of multiple genetic factors contributing to phenotypic variation, as well as their interaction with the environment. To date as an alternative to generating laboratory-induced mutants, it is relatively popular to use naturally-occurring variation among genetically distant *Arabidopsis thaliana* accessions as the source of quantitative genomics approaches, designed to map quantitative trait loci (QTLs) and try and resolve them at the gene level. Furthermore the (relatively few) success of the QTL studies has often been because of the use of quantitative phenotyping, as opposed to the qualitative scales often used in typical mutant screens. Therefore the objective of our work is to apply genome-wide quantitative molecular genetics to both, a very integrative and classical quantitative trait (shoot growth) and a molecular trait *a priori* more directly linked to the source of variation (gene expression under cis-regulation), in both cases studied in interaction with drought stress. To this aim a combination of our unique high-troughput phenotyping robot (the *Phenoscope*), RNA-seq, fine-mapping, complementation approaches and association genetics is used to pinpoint a significant number of medium- or small-effect QTLs and expression QTLs (eQTLs) to the gene level and identify causative polymorphisms and the molecular variation controlling natural diversity. Thus, exploiting these approaches in large-scale and highly reproducible conditions, thanks to the Phenoscope, should allow to resolve enough QTLs to start drawing a more general picture as to how genetic diversity shapes phenotypic variation. Towards decoding the genetic architecture of plant growth response to drought stress, in order to understand how Arabidopsis adapts to its environment, recent results will be presented to illustrate our strategies and research.

3-03

IDENTIFICATION OF *VICIA SATIVA* L. SUBSPECIES AND ASSESSMENT OF THEIR PHYLOGENETIC RELATIONSHIP BASED ON AGRO MORPHOLOGICAL TRAITS AND RAPD MARKERS

Aouida Marwa* (Laboratory of Legumes,Borj Cédria,Tunisia), Taamali Wael (Laboratory of Extremophil Plants, Borj Cédria,Tunisia), Abid Ghassen (Laboratory of Legumes, Borj Cédria, Tunisia) , Abderrazek Smaoui (Laboratory of Extremophil Plants, Borj Cédria,Tunisia), Badri Yazid (Laboratory of Legumes, Borj Cédria,Tunisia) and Jebara Moez (Laboratory of Legumes Borj Cédria, Tunisia)

aouidamarw@gmail.com / marwaouida@hotmail.fr

Common vetch (Vicia sativa L.) is used as a cover crop, green manure, pasture, silage, hay and grain for livestock feed. Its high dry matter and nitrogen accumulation make it an excellent winter leguminous cover crop in annual vegetable rotations. Several works showed that *Vicia sativa* is taxonomically complicated since morphological distinctions between their subspecies are not well expressed but little efforts were done to describe relationship between its subspecies.
This work aims to identify and to assess phylogenetic relationship among Tunisian *Vicia sativa* subspecies using seventeen agro morphological traits and 7 RAPD markers. We identified three subspecies: *sativa*, *macrocarpa* and *nigra,* and three potentially distinct subspecies I1, I2, I3 still unknown.
According to AMOVA most of the genetic variation was found within subspecies (80%), and only 20% of it between subspecies . Subspecies differed in terms of their internal genetic variation, with Nei's gene diversity ranging from 0,0046 to 0,0102. In addition agro-morphological traits related to the plant growth and plant fertility were measured. Genetic matrices calculated on the basis of 17 agro morphological traits and RAPD markers .Both of dendrograms based on the unweighted pair-group method with arithmetic averages (UPGMA) generated by RAPD markers and based on Euclidean distances for agro morphological traits showed three main clusters. The first included *V.sativa ssp sativa*, *V. sativa ssp macrocarpa*, *V.sativa ssp niga* and the unknown subspecies *V.sativa ssp I1*. I1 clustered close to V*icia sativa ssp* in group 1 while *V.sativa ssp I2* and *V.sativa ssp I3* clustered far from the other subspecies and each formed a separate group. The Mantel test performed on the basis of genetic distances generated by the two types of markers showed no correlation between them.

3-04

INTERSPECIFIC VARIABILITY OF CARBON ISOTOPE COMPOSITION IN THE CARTHAMUS GENUS AND INTRASPECIFIC VARIABILITY IN *CARTHAMUS TINCTORIUS* SPECIES: EFFECTS ON THE YIELD IN A MEDIERANNEEN CLIMATE

Imene MIHOUB* (USTHB, Bab Ezzouar, Algérie), Jaleh Ghashghaie (Univiversité Paris-XI, Orsay,France), Thierry Robert (Univiversité Paris-XI, Orsay, France), Fatiha Aïd (USTHB, Bab Ezzouar, Algérie)

mihimene14@yahoo.fr

The carthamus genus, mainly growing in arid and semi arid areas, is highly tolerant to water stress. *Carthamus tinctorius* (safflower) is the only cultivated species of this genus. The carthamus is an oleaginous species, highly regarded for its oil quality. The variability of the water-use efficiency (WUE) in the carthamus genus has not yet been investigated. It is well known that the carbon isotope composition ($\delta^{13}C$) provides a good prediction of the WUE. The present work aimed to study the interspecific variability of $\delta^{13}C$ in some species of the carthamus genus as well as the intraspecific variability in safflower. Analysis of $\delta^{13}C$ of the leaf bulk organic matter focused on four species, including three wild species (*Carthamus lanatus Carthamus pinnatus and Carthamus caeruleus*), widespread in the Mediterranean region and the cultivated species (*Carthamus tinctorius*). Fifty safflower accessions from different geographical origins were also analysed for carbon isotope composition of leaf bulk organic matter as well as for their yield. Interspecific and intraspecific variability in $\delta^{13}C$ showed amplitudes of 3.35 ‰ and 3.85 ‰, respectively. The strong differentiation observed between accessions for $\delta^{13}C$ values compared to the low intra-accession variability, suggests the existence of an inter-accession genetic variability of this character, the heritability of which needs to be evaluated. Grain yields were shown to be in general relatively low due probably to the high temperatures at the flowering period. However, a strong variability in the grain yield was observed between accessions (0.3 to 13.8qt/ha), the difference being highly significant among accessions ($p < 0.001$). No correlation was observed between yield and $\delta^{13}C$, suggesting that the observed drought, especially during the flowering period, induce a partial closure of the stomata which permit a reduction in loss water but maintaining the level of photosynthetic activity.

3-05

NATURAL VARIATION OF RPF PROTEINS IN THEIR AFFINITY FOR RNA TARGETS

Catherine Colas des Francs-Small* (Centre of Excellence in Plant Energy Biology, University of Western Australia, Crawley 6009 Western Australia), Stefan Binder (Molekulare Botanik, Universitât Ulm, 89069 Ulm, Germany), Ian Small (Centre of Excellence in Plant Energy Biology, University of Western Australia, Crawley 6009 Western Australia)

Catherine.colasdesfrancs-small@uwa.edu.au

PPR proteins are essential for the expression of organelle genes as they are involved in all stages of transcript processing. The recent discovery of a 'code' describing how PPR proteins recognize their target RNAs (Barkan et al., 2012) is a major breakthrough as it allows us to predict binding sites. The code was discovered thanks to the very high variability in the amino acids that determine binding specificity exhibited by one small clade of PPR proteins. This clade includes most of the known 'restorer of fertility' proteins able suppress expression of mitochondrial genes associated with cytoplasmic male sterility.

In Arabidopsis, this clade has about 20 members, primarily clustered on chromosome 1, several of which have been shown to be RNA processing factors (RPFs). This gene cluster is well known as a major site for tasiRNA generation. Preliminary data suggests that several of these genes are extraordinarily variable between Arabidopsis accessions. Variation in the N-terminal half of *RPF4* leads to dramatic changes in predicted RNA binding specificity that correlate with observed accession-specific mitochondrial RNA patterns. We are examining the binding specificity of RPF4 proteins from Ler, Col-0 and Ws by in vitro RNA electrophoretic mobility shift assays (REMSA). The use of different fluorescent labels on mixed RNA oligonucleotides allows the quantification of the binding affinities of one protein for various targets. We are also expressing the RPF4 proteins in various backgrounds to test their effect on organelle gene expression. The reasons for the variability of these genes within *A. thaliana* isn't clear; they may be relics left from an outcrossing ancestor where they could have been selected for to counteract mitochondrial CMS genes.

3-06

PHENOTYPING NATURAL VARIATION OF ARABIDOPSIS THALIANA

Marcus Jansen*, Silvia Braun, Fabio Fiorani (Jülich Plant Phenotyping Centre, IBG-2 Plant Science, Forschungszentrum Jülich GmbH, 52425 Jülich, Germany)

m.jansen@fz-juelich.de

Automated non-invasive phenotyping revealed a broad spectrum of phenotypic variability of natural Arabidopsis isolates grown under controlled conditions. Studying natural variation of Arabidopsis, it is well known that vegetative rosette stages exhibit a large range of variability in size and growth habitus depending on the respective ecotype and geographic conditions. Concomitantly, a high genetic diversity has been found among natural isolates and turned out to be a valuable genetic resource for basic and applied science. However, phenotypic comparison of accessions across habitats carries uncertainty due to multiple differences in environmental factors. A tool for direct phenotypic comparisons of different ecotypes is cultivating them together in one location under controlled environmental conditions. Excluding influences deriving from different habitats, phenotypic inter-ecotype variability is fully generated by genomic differences. This allows linking phenotype data with genome properties.

The Arabidopsis 1001 Genomes sequencing project provides an important genetic resource for natural diversity. One of its aims is linking genome sequence variation to phenotypic diversity. We have assessed phenotypic variability in shoot growth, morphology, and quantum yield of 80 accessions out of the 1001 genomes collection grown at controlled conditions in a common climate chamber. Non-invasive growth measurements were carried out using an automated GROWSCREEN FLUORO phenotyping platform (Jansen et al. 2009). Performance of accessions was ranked based on growth and biomass data. At applied non-stressing growth conditions plants produced leaf area sizes varying by a factor of 2.8. Accessions were further characterized using chlorophyll fluorescence as well as morphological factors. Along with rosette area variability, rosettes were variable in compactness. This resulted in an accession-specific ratio of size and compactness. Ecotypes from same geographic origins shared similarity in this ratio, but there were strong overlaps between geographic groups. Thus, grouping by macro-geographic origins does not take into account small-scale adaptations to local conditions driving growth characteristics.

3-07

AFLP MARKER-BASED IDENTIFICATION AND GENETIC RELATIONSHIPS OF OLIVE CULTIVARS IN THE REGION OF HBEBSA "NORTH WEST OF TUNISIA"

Mnasri Rahmani Sameh [1] *, Saddoud Debbabi Olfa[1] and Ferchichi Ali [2]

1: National Gene Bank of Tunisia
2: National Institute of Agronomy of Tunisia

mnasrisameh@yahoo.fr

In the region of Hbebsa, little is known about the olive germplasm, and even though there is an important olive biodiversity, studies about characterization and evaluation are scarce. The aim of this work was to make a molecular characterization by the use of amplified fragment length polymorphism (AFLP) markers. A total of 13 olive varieties were genotyped using nine different EcoRI–MseI AFLP primer combinations. Auto radiographs revealed 201 polymorphic markers in a total of 409 detected fragments. A set of redundant marker patterns was identified and deleted from the binary data matrix; data analysis demonstrated a high degree of polymorphism with an average of 49%. In a dendrogram calculated by the Jaccard's clustering technique, two major groups were identified, separating the cultivars "Chemlali" and "Chetoui" from the other eleven varieties. Three subgroups have been revealed in the second main group. The groups and subgroups identified on the genetic basis are closely related to the genetic biodiversity of the studied cultivars. The analysis of AFLP profiles found in our set of olive cultivars showed a wide genetic diversity among olive germplasm in the region of Hbebsa. These findings were of interest to protect the specimens studied varieties.

SESSION 4:

TRANSLATIONAL BIOLOGY

4-01

MYB118, A TRANSCRIPTION FACTOR CONTROLLING STORAGE COMPOUNDS PARTITIONING IN THE SEED OF ARABIDOPSIS

Guillaume Barthole * (Institut Jean-Pierre Bourgin, INRA Versailles, France), Loïc Lepiniec (Institut Jean-Pierre Bourgin, INRA Versailles, France), Sébastien Baud. (Institut Jean-Pierre Bourgin, INRA Versailles, France)

Guillaume.barthole@versailles.inra.fr

During the maturation phase, Arabidopsis seeds accumulate mainly two types of storage compounds, namely triacylglycerols (TAGs) and seed storage proteins (SSP). They account for nearly 60% of the mature seed dry weight. TAG and SSP mostly accumulate in the embryo, although both are also found in the endosperm. The biosynthetic networks underlying the accumulation of these storage compounds are well characterized. However, their regulation remains poorly understood. In the laboratory, several screening procedures were carried out to isolate new transcription factors involved in the regulation of seed maturation. AtMYB118 was thus identified as a candidate putatively involved in this regulatory process. MYB118 is a seed-specific transcription factor. A fine and comprehensive characterization of *MYB118* expression pattern showed a peak of expression 10 days after flowering, mostly in the endosperm. The analyses of mRNA accumulation and activity of the *MYB118* promoter in *leafy cotyledon 2* (*lec2)* mutant seeds and in an inducible *LEC2* overexpressing line, showed *MYB118* expression to be positively regulated by this master regulator of seed maturation. Biochemical analysis of *myb118-1* seeds showed that TAG and SSP content were doubled in the endosperm and decreased in the embryo of this mutant compared to the wild type. A transcriptomic analysis of *myb118* mutant seeds pointed out some putative targets, the deregulation of which could explain this phenotype. Interestingly, among them is *LEC2*. The mRNA accumulation of which goes up in *myb118-1* seeds. Therefore, we investigated this feedback regulatory loop involving *LEC2* and *MYB118* and its potential effect on storage compound balance between embryo and endosperm.

SESSION 6:

EPIGENETICS

6-01

A LONG NON-CODING RNA INVOLVED IN ROOT DEVELOPMENTAL PLASTICITY

Natali Romero* (CNRS Institut des Sciences du Végétal, Gif-sur-Yvette, France), Federico Ariel (CNRS Institut des Sciences du Végétal, Gif-sur-Yvette, France), Florian Bardou (CNRS Institut des Sciences du Végétal, Gif-sur-Yvette, France), and Martin Crespi (CNRS Institut des Sciences du Végétal, Gif-sur-Yvette, France)

romero@isv.cnrs-gif.fr

Non protein coding RNAs (npcRNAs) are emerging as key players in the regulation of varied important cellular processes. Long npcRNA represent a class of riboregulators, which either act directly in this long form or are processed into shorter small si/miRNAs, leading to mRNA cleavage, translational repression or epigenetic DNA/chromatin modification of their targets. Hence, long npcRNAs are now known to confer a degree of temporal and spatial specificity not possible with proteins. In our Lab, a bioinformatic approach served to identify 76 *Arabidopsis thaliana* npcRNAs including several siRNA precursors and antisense RNAs. Abiotic stress, such as phosphate starvation, drought or salt stress altered the accumulation of many of these npcRNAs. The expression pattern of a set of long npcRNAs in response to phytohormones and environmental stresses suggested a link with root growth and development. One of these long npcRNAs is the npc351. Interestingly, auxin treatment in roots may lead to a decrease of npc351 expression concomitant with an increase in transcript levels of its neighboring gene, the *LOB DOMAIN-CONTAINING PROTEIN 40* (*LBD40*). Overexpression of *NPC351* results in increased lateral root density. Data from siRNA distribution and DNA methylation indicate that the *NPC351* locus is hypermethylated and accumulate 24 nt siRNAs. Furthermore, ChIP-qPCR experiments suggest that the over accumulating npc351 lines have a modified chromatin status on its locus due to changes in histone modifications. In parallel, our Lab showed a direct implication of this npcRNA in the regulation of alternative splicing of specific target genes related to auxin responses. Our results suggest an implication of npc351 in the auxin control of lateral root development.

SESSION 7:

HORMONES

7-01

AN UNIDENTIFIED SA-CONJUGATE IS SPECIFICALLY ACCUMULATED IN THE ROOTS OF THE ACCESSION BUR-0, BUT IS NOT ASSOCIATED WITH THE MAJOR QTL OF RESISTANCE TO CLUBROOT IN ARABIDOPSIS

Antoine Gravot* (UMR1349 IGEPP-Université Rennes 1, Rennes, France), Nathalie Marnet (UR117 CBFL, Rennes, France), Severine Lemarié (UMR1349 IGEPP-INRA, Rennes, France), Christine Lariagon (UMR1349 IGEPP-INRA, Rennes, France), Régine Delourme (UMR1349 IGEPP-INRA, Rennes, France), and Maria Manzanares-Dauleux (UMR1349 IGEPP-Agrocampus Ouest, Rennes, France)

antoine.gravot@univ-rennes1.fr

In Arabidopsis, the accession Bur-0 exhibits several contrasted traits compared to the canonical genotype Col-0 and has been therefore used for several quantitative genetic studies. In the present work, we aimed to study the possible role of phytohormones in the expression of quantitative partial resistance to clubroot in Bur-0. Hormone profiling was performed on inoculated and non-inoculated roots from Bur-0 and Col-0, using a high resolution chromatographic method (UPLC-TQD). In Bur-0, a MRM procedure to quantify salicylic acid led to the detection of a second peak of high intensity. This unknown compound eluted very close to salicylic acid, and exhibited identical mass transitions. Surprisingly, this compound was never detected in Col-0 in any of our experimental conditions. Additional investigations suggested that this analyte would correspond to a SA-conjugate (but not to any of the previously reported glucosides), whose source fragmentation leads to similar transitions as SA. This compound could easily be confounded with SA when using conventional chromatographic procedures, possibly leading to substantial overestimations, depending on Arabidopsis accessions. We speculated that the specific accumulation of this compound in the roots of Bur-0 could also be related to the partial resistance to clubroot. To test this hypothesis, we used two near-isogenic lines (Heterogeneous Inbred Lines from the Bur-0xCol-0 progeny) differing from each other only for a small genomic fragment in the region of the major QTL controlling clubroot resistance. These two genotypes had contrasted clubroot symptoms, confirming the effect of the Bur-0 resistance allele at this QTL. However, both lines displayed identical accumulation of the SA-conjugate. Our results suggest that the specific accumulation of an unknown SA-conjugate in the roots of Bur-0, is not genetically related to the major clubroot resistance QTL.

7-02

AtNSRs PROTEINS LINK ALTERNATIVE SPLICING AND THE ACTION OF NON CODING RNAs

Florian Bardou* (ISV-CNRS, France), Federico Ariel (ISV-CNRS, France), Philippe Laporte (ISV-CNRS-France), Craig Simpson (SCRI, Scotland), John Brown (SCRI, Scotland), Martin Crespi (ISV-CNRS, France)

Florian.Bardou@isv.cnrs-gif.fr

In eukaryotes several RNA binding proteins (RBPs) act on mRNA at various levels from splicing to translation. Recently a large number of non-protein coding RNAs (npcRNAs) have been identified in eukaryotes and shown to integrate into a variety of ribonucleoproteins (RNP) to control posttranscriptional gene expression. We have identified a plant Nuclear-Speckle RBP (or NSR) constitutively expressed that interacts with an npcRNA, ENOD40 that accumulates during lateral root and nodule formation in legumes. This NSR is relocalised into a cytoplasmic RNP in the ENOD40-expressing cells (Campalans et al., 2004). Two AtNSR homologs exist in Arabidopsis thaliana, named AtNSRa and AtNSRb that also localise in nuclear speckles together with certain splicing-related proteins. The AtNSRb gene is regulated by auxin whereas AtNSRa is constitutive. Single Atnsr mutants do not show any phenotype but root growth of double mutants is partially insensitive to auxin, suggesting a redundant function of these proteins in roots. We have analysed the splicing profile of 288 genes in wt and Atnsr roots in response to auxin in Arabidopsis. From these 288 differentially spliced genes, 180 change their splicing profile in response to auxin and 133 required AtNSR proteins for this change. These results correlate with the partial auxin insensitive phenotype of the double mutant lines. In order to validate the interaction of NSRs with mRNA and npcRNAs, we have co-immunoprecipitated NSRs with at least 5 alternatively spliced mRNAs and the npcRNA351 (RIP approach). Interestingly, this npcRNA351 partner of AtNSRs can modify the alternative splicing pattern of the AtNSRs-dependant targets. We propose that NSRs may link alternative splicing and the action of non-coding RNA during plant growth and development.

7-03

B'θ REGULATORY SUBUNIT OF PROTEIN PHOSPHATASE 2A IS INVOLVED IN PEROXISOMAL FATTY ACID β-OXIDATION, FLOWERING, AND PLANT INNATE IMMUNITY

Amr Kataya*, Behzad Heidari, and Cathrine Lillo (Centre for Organelle Research, Faculty of Science and Technology, University of Stavanger, Stavanger, Norway)

amr.kataya@uis.no

Protein phosphatase 2A (PP2A) is a serine/threonine-specific phosphatase. PP2A holoenzymes are heterotrimeric complexes comprising a catalytic subunit (C), a scaffolding subunit (A), and a regulatory subunit (B). The B subunits are responsible for substrate specificity and localization of the holoenzyme complex and are classified into B55, B', and B'' non-related families. In our previous studies, the *Arabidopsis thaliana* member of B' family "B'θ" was found to be localized in peroxisomes. Peroxisomes are subcellular organelles that are traditionally known to be involved in processes like photorespiration, fatty acid β-oxidation, detoxification of active oxygen species, and recently in plant innate immunity. The regulation of peroxisomal metabolism by reversible phosphorylation has not been conclusively demonstrated yet. Several protein kinases were found in peroxisomes, but no protein phosphatases were associated with peroxisomes except B'θ subunit of PP2A complex. Here we describe the isolation of T-DNA knockout mutant of the gene encoding B'θ and the application of phenotypic analyses to better study the function of PP2A-B'θ complex in peroxisomes and plant development. The *Arabidopsis b'θ*_b01 homozygous T-DNA mutant seedlings were developmentally arrested when germinated without supplemental sucrose, suggesting defects in fatty acid β-oxidation. Indole-3-butyric acid (IBA) and the synthetic auxin 2,4-dichlorophenoxybutyric acid (2,4 DB) are converted to the bioactive auxins IAA and 2,4 D, respectively, in a mechanism that involves peroxisomal β-oxidation. The *b'θ*_b01 mutants showed weak but statistically significant resistance to both IBA and 2,4 DB when compared with the wild type. In addition, preliminary data from total lipid analysis of the mutant show that eicosenoic acid (20:1), a convenient reporter of TAG breakdown, is not degraded in the early phases of seedling development and supports the suggestion of the defect in fatty acid β-oxidation in the mutant. Interestingly, the *b'θ*_b01 mutant showed different phenotypes as delayed flowering including up-regulation of *FLC* expression. Moreover, the mutant showed noticeable resistance to the infection by *Pseudomonas syringae* (*Pst DC3000*). Interesting but still preliminary data shows that the mutant is more resistant in case of basal thermotolerance and high salt concentrations. This running study is suggesting potential roles for PP2A-B'θ complex in peroxisomes, plant development, and biotic and abiotic stresses. The protein complex of PP2A-B'θ function in ROS metabolism and defense mechanisms, and the underlying signal transduction cascades will be further studied by proteome and biochemical approaches.

7-04

CHARACTERIZATION OF A RECEPTOR KINASE CONTROLLING *MEDICAGO TRUNCATULA* ROOT SYSTEM ARCHITECTURE

Emeline Huault* (Institut des Sciences du Végétal, CNRS, Gif-sur-Yvette, France), Carole Laffont (Institut des Sciences du Végétal, CNRS, Gif-sur-Yvette, France), Kyrankumar S. Mysore (Noble foundation, Ardmore, USA), Henri de Larambergue (INRA, Dijon, France), Gérard Duc (INRA, Dijon, France), Pascal Ratet (Institut des Sciences du Végétal, CNRS, Gif-sur-Yvette, France) and Florian Frugier (Institut des Sciences du Végétal, CNRS, Gif-sur-Yvette, France)

huault@isv.cnrs-gif.fr

The plant root system architecture is crucial for plants to adapt to environmental conditions, such as nutrient availability and stress conditions. Plants can either modulate their root growth rate and the number of lateral organs formed, depending on environmental conditions. In legumes, two types of root-derived organs can be initiated: lateral roots and nitrogen-fixing nodules. To explore regulatory mechanisms controlling legume root system architecture, we used a forward genetic approach in the *Medicago truncatula* model to identify insertional mutants affected in root development. Among the different mutants identified, we characterized plants showing a *compact root architecture* phenotype (*cra*), consisting of a highly branched root system. Cloning of the gene involved revealed the involvement of a Receptor Kinase controlling lateral root formation in *M. truncatula*.

7-05

CYTOKININ SIGNALING THROUGH DIFFERENT SIGNALING PATHWAYS CONTROL VARIOUS *MEDICAGO TRUNCATULA* ROOT ENVIRONMENTAL RESPONSES

Stéphane Boivin* (Institut des Sciences du Végétal, CNRS, Gif-sur-Yvette, France), Carole Laffont (Institut des Sciences du Végétal, CNRS, Gif-sur-Yvette, France), Mathias Brault (Institut des Sciences du Végétal, CNRS, Gif-sur-Yvette, France), Thomas Rey (Université de Toulouse, CNRS, Castanet-Tolosan, France), Mara Novero (Università degli Studi di Torino, Torino, Italy), Kirankumar S. Mysore (Noble foundation, Ardmore, USA), Jiangqi Wen (Noble foundation, Ardmore, USA), Paola Bonfante (Università degli Studi di Torino, Torino, Italy), Christophe Jacquet (Université de Toulouse, CNRS, Castanet-Tolosan, France) and Florian Frugier (Institut des Sciences du Végétal, CNRS, Gif-sur-Yvette, France)

Stephane.Boivin@isv.cnrs-gif.fr

Plants can adapt to changing soil environmental conditions by controlling their root system architecture to optimize nutrients uptake and limit exposure to stress. Depending on environmental conditions, legumes are able to develop two types of organs on their roots, lateral root and nitrogen-fixing nodules. In this later case, organogenesis results from the interaction with symbiotic Rhizobium bacteria. In the *Medicago truncatula* model legume, cytokinins perceived by the CHK1/CRE1 (CHASE Histidine Kinase/ Cytokinin Response 1) receptor are necessary and sufficient to induce nodule organogenesis, whereas this pathway negatively controls lateral root formation. In addition, cytokinins are involved in biotic and abiotic stress responses. Analysis of root responses to various environmental conditions of the different cytokinin receptors (CHK2, CHK3 and CHK4) has been undertaken, to identify and better understand their functional specificities and redundancies.

7-06

GENOTYPE-DEPENDENT JASMONATE AND SALICYLATE REGULATIONS DURING THE RESPONSE TO CLUBROOT IN *ARABIDOPSIS THALIANA*

Severine Lemarié* (UMR1349 IGEPP-INRA, Rennes, France), Christine Lariagon (UMR1349 IGEPP-INRA, Rennes, France), Nathalie Marnet (UR117 CBFL, Rennes, France), Jocelyne Lemoine (UMR1349 IGEPP-INRA, Rennes, France), Mélanie Jubault (UMR1349 IGEPP-Agrocampus Ouest, Rennes, France), Régine Delourme (UMR1349 IGEPP-INRA, Rennes, France), Maria Manzanares-Dauleux (UMR1349 IGEPP-Agrocampus Ouest, Rennes, France) and Antoine Gravot (UMR1349 IGEPP-Université Rennes 1, Rennes, France)

severine.lemarie@rennes.inra.fr

Clubroot, a root disease affecting *Brassicaceae,* is caused by the obligate biotrophic protist *Plasmodiophora brassicae*. The infection triggers hyperplasia and hypertrophy of root cortex cells, leading to the formation of root galls in several weeks. Auxin and cytokinin signaling pathways has been reported to play a prominent role in this process. Furthermore, previous works on *Arabidopsis thaliana* have also highlighted a role of the *JAR1*-dependent jasmonate signalling pathway in the attenuation of clubroot symptoms in the susceptible accession Col-0. However, the role of 'classical' defence hormone signalling remains unclear in this pathosystem. In this work, we compared the involvement of JA and SA signalling during the clubroot response, between two accessions Bur-0 (partially resistant to clubroot) and Col-0, and between two pathogen isolates eH (medium aggressiveness) and e2 (highly aggressive). Jasmonate and salicylate contents were analysed in root tissues at different time-points along 3-weeks after inoculation. Jasmonate levels were found to be induced in the susceptible accession Col-0 when using the eH isolate, but not with e2. In Col-0, the salicylate content was found depleted in infected roots, for both isolates. By contrast, the salicylate content was found transitory accumulated before the last steps of the infection kinetics in the partially resistant Bur-0, whatever the pathogen isolate. In Bur-0, jasmonate content was only slightly regulated by clubroot. These results suggest a role for SA-signalling in the partial resistance of Bur-0, and the possible importance of JA-signalling in expression of isolate aggressiveness. These hypotheses can now be the matter of further investigations using functional genomics and quantitative genetics.

7-07

GIBBERELLINS CONTROL ROOT GROWTH AND NODULATION IN *MEDICAGO TRUNCATULA*

Camille Fonouni-Farde* (Institut de des Sciences du Végétal – CNRS Gif sur Yvette, France), Anouck Diet (Institut de des Sciences du Végétal – CNRS Gif sur Yvette, France, Université Paris Diderot-Paris 7, France) and Florian Frugier (Institut de des Sciences du Végétal – CNRS Gif sur Yvette, France).

camille.fonouni-farde@isv.cnrs-gif.fr

The adaptation of root system architecture to environmental constraints is a major agricultural trait. Legumes, in addition to root branching through lateral roots, can develop symbiotic interactions with soil bacteria of the Rhizobiacae family to form another secondary root organ, the nitrogen-fixing nodule. Nodule formation is regulated by several plants hormones including cytokinins which are necessary and sufficient to activate nodule organogenesis. Gibberellins (GAs) are plant growth regulators associated with several plant growth and development processes, such as seed germination, stem elongation, flowering and fruit development. GAs are also involved in pea and *Lotus japonicus* nodulation with opposite behaviours, since GAs have respectively a positive and a negative role.
We therefore aim to understand how gibberellins and their signaling pathway interfere with root and symbiotic nodule development in the model legume *M. truncatula*.
Exogenous application of GAs negatively control lateral root formation and root growth. Confocal microscopy experiments reveal that meristems of root treated with GAs are shorter and display a reduced number of cells. Concerning the nodulation process, *M. truncatula* plants treated with GAs exhibit a reduced number of nodules. Expression of *NIN* and *ERN1* symbiotic genes was reduced upon GA treatment. These results suggest that GAs play a negative role in the regulation of nodule organogenesis. Activation of the GA signaling pathway using an RNAi construct targeting three *DELLA* genes, encoding for negative transcriptional regulators of the GA pathway, led to a decrease in *M. truncatula* nodule formation. We are also currently analyzing the cross-talk between GAs and cytokinins in the regulation of the nodulation process using a mutant impaired in the cytokinin receptor CRE1.

7-08

THE CYTOKININ SIGNALING IN THE REGULATION OF THE AMMONIA ASSIMILATION OF THE CEREALS.

Murat Gilmanov*(Institute molecular biology and biochemistry, Almaty, Kazakhstan), Saniam Ibragimova (Institute molecular biology and biochemistry, Almaty, Kazakhstan), Arailym Esebbaeva (Institute molecular biology and biochemistry, Almaty, Kazakhstan), Adlet Esmambetov (Al-Faraby's Kazakh national university department biology and biotechnology, Almaty, Kazakhstan)

baltakay@mail.ru

It's well known that cytokinin plays main role in crop production however the cytokinin signaling mechanisms of the regulation of the ammonia assimilation of the cereals are stayed unstudied, whereas this process determines the protein productivity of the plants. Our long time investigation let to discover the very powerful cytokinin secondary (CSH) hormone which related to fusicoccin compounds. CSH was isolated from germinated wheat seeds by hydrophobic chromatography and by reverse phase chromatography. CSH is formed in response to cytokinin action. Then CSH is turn bound with plasmatic membrane receptor that leads to the activation of Ca^{+2} ATPase. As result the concentration of cytosolic Ca^{+2} is increased. This Ca^{+2}ions are turn bound with molecular of glutamate dehydrogenase (GDh) which related to Ca^{+2} sensor proteins. The last step of the cytokinin signaling is the phosphorylation of the Ca^{+2} bound GDh. That lets to activation of NADPH – GDh. This enzyme has very high affinity to ammonia K_M = 0,8 mkM that much better than glutamine synthetase (GS). It was shown by us in the ripening wheat ears the 75 % more than 45% the assimilation of ammonia carry out by NADPH – GDh whereas GS-GOAT road plays secondary role in this process. Thus the cytokinin signaling switch on the process of the assimilation of ammonia by activation NADPH-GDh by increasing of the concentration of the cytosolic Ca^{+2} ions and by phosphorylation precursor of the GDh molecular.

7-09

THE MEDIATOR OF THE CYTOKININ AS POWERFUL BIOREGULATOR FOR INVEROMENTAL TECHNOLOGIES

Adil Yerezhepov (Al-Farabi Kazakh National University, department biology and biotechnology, Almaty, Kazakhstan)

Adil.Erezhepov@kaznu.kz, Adil.Yerezhepov@mail.ru

Global warming, dramatically forests reducing, deserts expanding and strong increasing the area of saline lands requires the creation of new innovative technologies for improving of the environmental conditions. The most promising for this are the development and application of effective bioregulators. Bioregulators are acted at very low concentration but they cause the strong positive changes in metabolism of the plants that let to increasing of the productivity and tolerance to stress conditions. In this aim we are developed the method of the purification of the mediator of the cytokinin (MS) from wheat by adsorption chromatography on nanostructured carbon sorbent. The purified MS shows very high physiological activity. It acts at the nano and micro molar quantity whereas all phytohormones act at millimolar quantity. So the treatment of the seeds of the wheat and barley by MS strong increases the tolerance of this seedlings 2 % sodium chloride. So in this salt all control seedlings died whereas MS treated 71% seedlings survived. Also MS increases tolerance of the seedlings to low and high temperature stresses. Especially promising the applications MS for increasing of the productivity of the crop plants. The preserving treatment of the seeds of wheat and winter wheat and winter rye by MS lets to the increase of the productivity of thus crops on 30-40% and they are ripened on half month earlier than control plants (without preserving treatments by MS). So we propose the new powerful bioregulator for increasing of the plants to the stress conditions and for the increasing of the productivity of the plants.

Gala dinner

Castle of Vaux-le-Vicomte

© Olga Khomitsevich

The Vaux le Vicomte estate forms a unique whole that spans nearly 500 hectares. It is testament to an era and the fruit of the bold vision of Nicolas Fouquet, Superintendent of Finances under young King Louis XIV. Discover its fascinating history, from plots against the royal court to the outstanding achievements of iconic 17th century artists.
The art of French gardens is an expertise that is appreciated across the world. To design the gardens of the Vaux le Vicomte estate, one of the most representative of the era, Fouquet called upon André le Nôtre, a master in the art.
Vaux le Vicomte is the pioneer of French garden art. The perfect harmony between garden and architecture results from the collaboration of a trio of geniuses brought together by Fouquet: Le Nôtre the landscape gardener, Le Vau the architect and Le Brun the painter-decorator.
(Source: Official Website http://www.vaux-le-vicomte.com/en/)

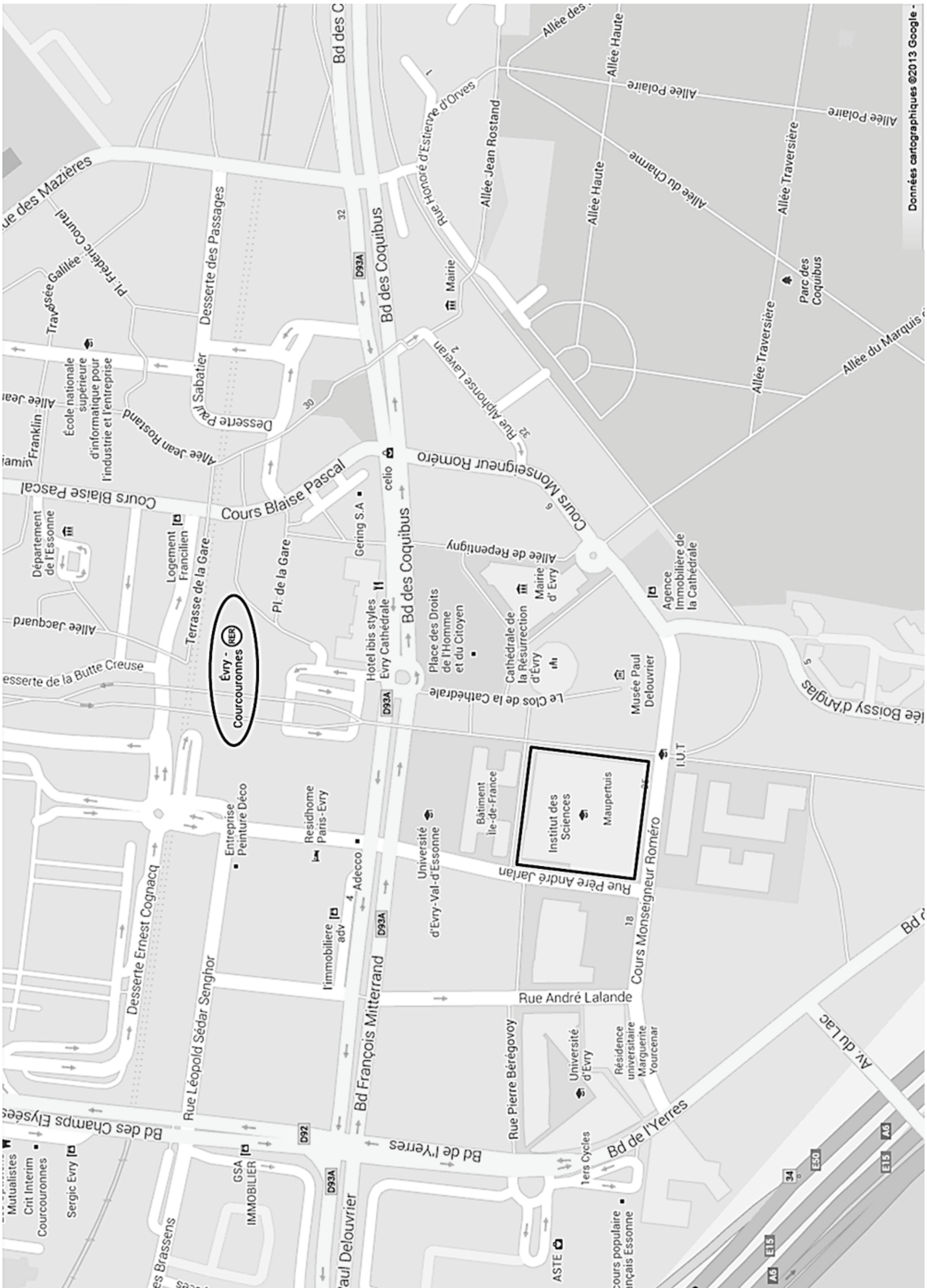
Schedule

6:00 PM Departure from Evry

7:00 PM Diner (~ 1h30)
8:30 PM Visit of the Castle (~ 1h30)
10:00 PM Walk in the gardens (~ 30min)
11:00 PM Fireworks (~ 5 min) and Visit of the “Equipages” Museum (~ 30min)

11:30 – 11:45 PM: Departure from Vaux-le-Vicomte (Arrival in Evry ~ midnight)

Map



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Organizers

Heribert HIRT
Unité de Recherches en Génomique Végétale
Rue Gaston Crémieux, CP 5708
91057 Evry Cedex
Tel: +33 (0)1 60 87 45 08
hirt@evry.inra.fr

Martin CRESPI
CNRS, Avenue de la terrasse Bât. 23
F-91198 Gif-sur-Yvette Cedex
Tel: +33 (0)1 69 82 37 03
martin.crespi@isv.cnrs-gif.fr

Michael HODGES
Institut de Biologie des Plantes
Université Paris-Sud CNRS, Bat 630
91405 Orsay Cedex
Tel: +33 (0)1 69 15 33 35
michael.hodges@u-psud.fr

Herman HÖFTE
IJPB, UMR 1318 INRA-AgroParisTech
INRA, centre Versailles-Grignon
Route de ST Cyr (RD10)
78026 Versailles Cedex
Tel: +33 (0)1 30 83 33 90
herman.hofte@versailles.inra.fr

Annie MARION-POLL
IJPB, UMR 1318 INRA-AgroParisTech
INRA, centre Versailles-Grignon
Route de ST Cyr (RD10)
78026 Versailles Cedex
Tel: +33 (0)1 30 83 30 68
annie.marion-poll@versailles.inra.fr

Marie-Jeanne SELLIER
Manager of the Saclay Plant Sciences LabEx
IJPB, UMR 1318 INRA-AgroParisTech
INRA, centre Versailles-Grignon
Route de ST Cyr (RD10)
78026 Versailles Cedex
Tel: +33 (0)1 30 83 30 82
marie-jeanne.sellier@versailles.inra.fr

+33 (0)6 63 03 62 25

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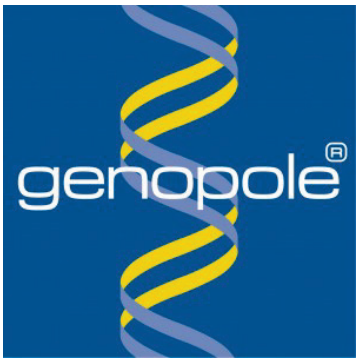

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SPS@versailles.inra.fr
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