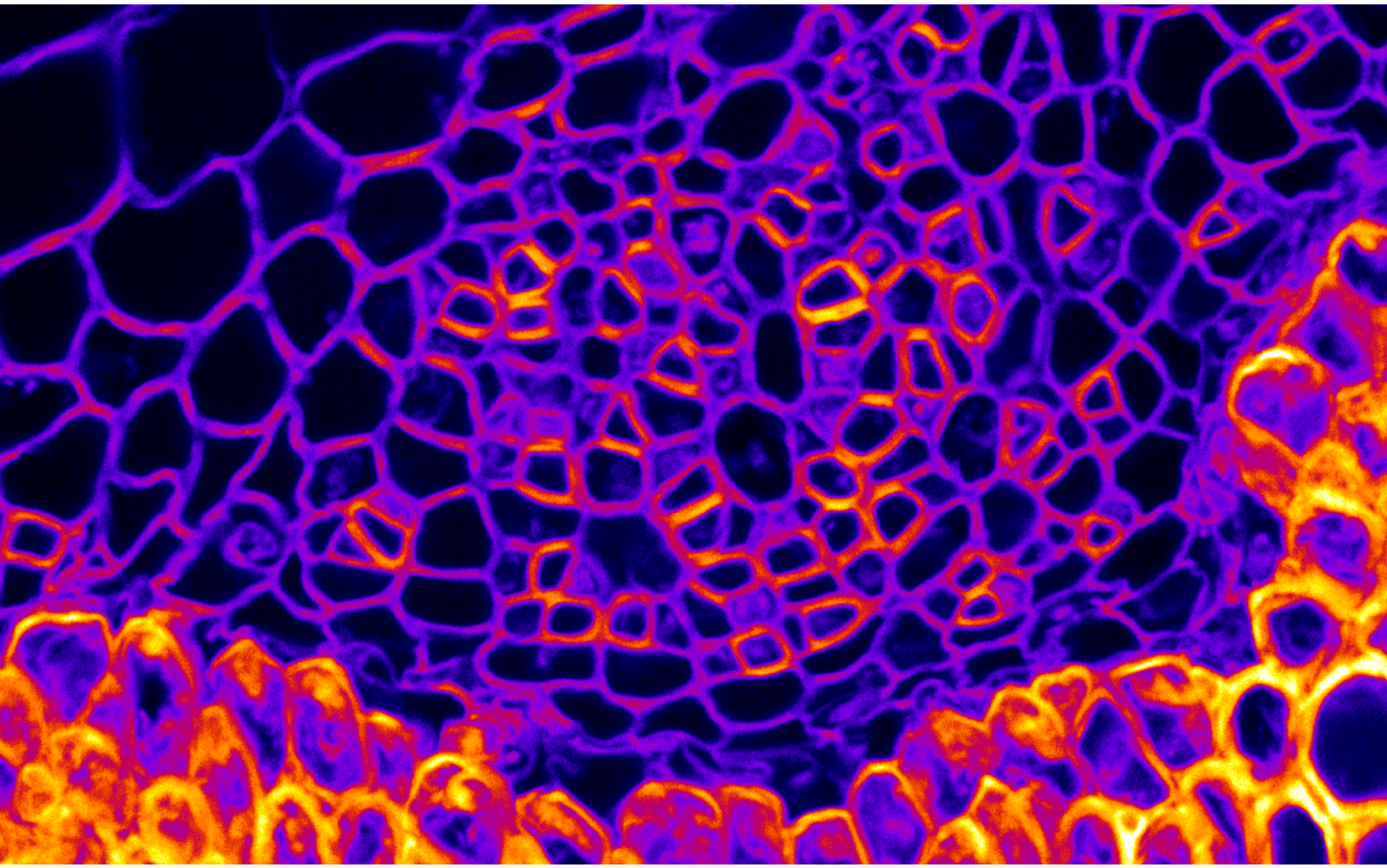
SPS Saclay Plant Sciences | université PARIS-SACLAY

SPS Summer School 2022

« Plant sugar metabolism, transport and signaling in a challenging environment »

July 3-8, 2022

Saint-Lambert-des-Bois, France



Participant's guide

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Physiologia Plantarum

Venue

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Essential information

Arrival of the participants - Sunday July 3

A bus shuttle will be organized from the RER B station «St Rémy-lès-Chevreuse» **at 4:30 PM**.
To take the bus shuttle, wait outside of the station.

Do not be late for the bus.
In case of problem, please contact the organizers.

The Summer School will begin at 5 PM.

Do not forget to bring a laptop.

Activities on site

Please note that the Summer School location is in the countryside, isolated from any town. Be sure to take with you item you could need as well as casual clothes and comfortable shoes. Moreover, there will be sport activities available so you should also bring adequate clothes.

There may be a lot of mosquitoes so you might want to bring repellent sprays.

End of the Summer School - Friday July 8

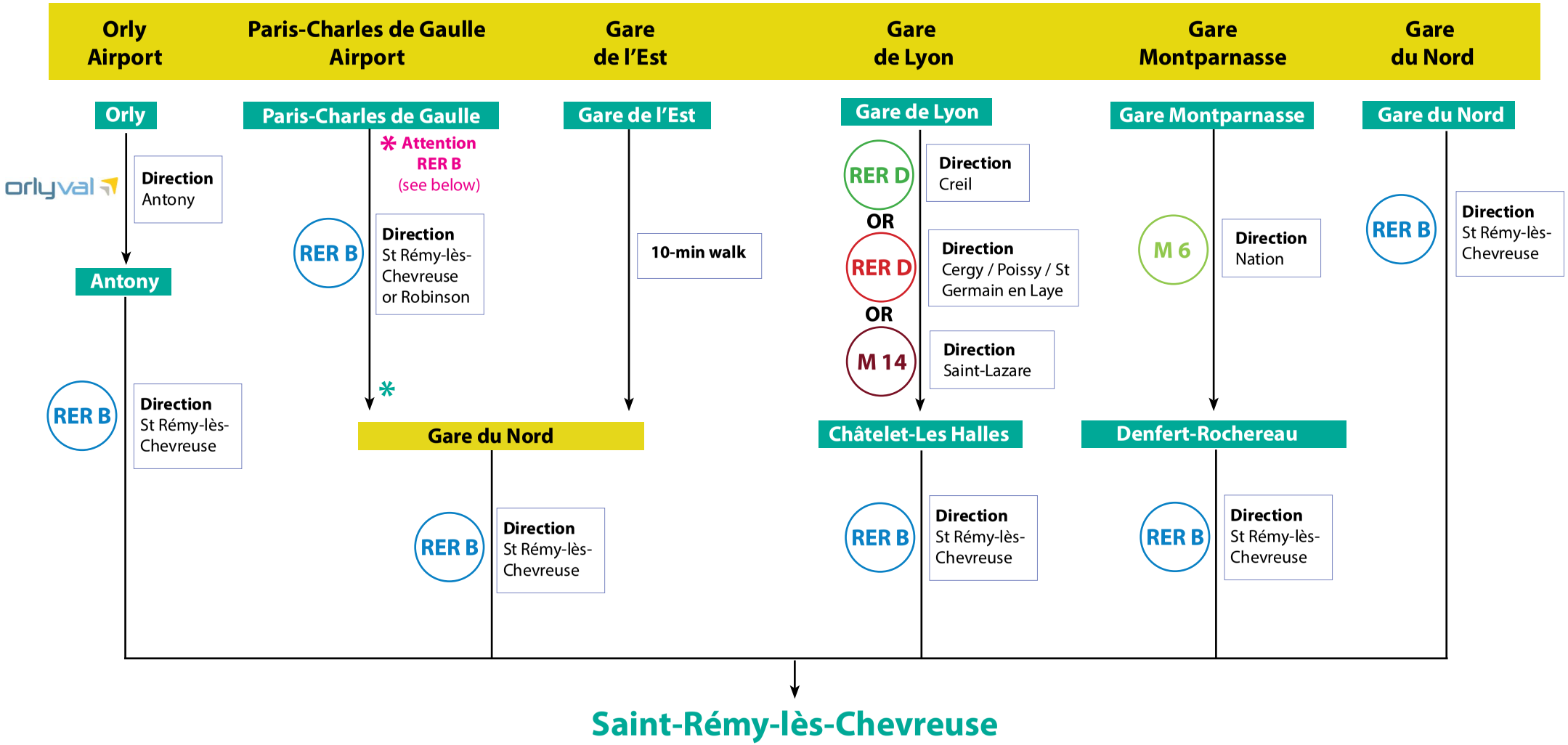
The Summer School will end around 4:30 PM.
A bus shuttle will also be organized after the Summer School to take you back to the RER B station «Saint-Rémy-les-Chevreuses». The shuttle will leave at 5 PM sharp so make sure your things are packed and ready during lunch break.

Those of you who have their flight / train a little early in the evening of July 8 might want to leave earlier and take a taxi to Saint-Rémy-les-Chevreuses (you can carpool). If so, you can ask the reception of the Centre Port-Royal to make a taxi reservation for you. But you will have to pay for the taxi and get reimbursed by your lab.

Travel instructions
> RER B station «Saint-Rémy-lès-Chevreuse»

Transports in Paris area

https://www.ratp.fr/en/
https://www.iledefrance-mobilites.fr/en

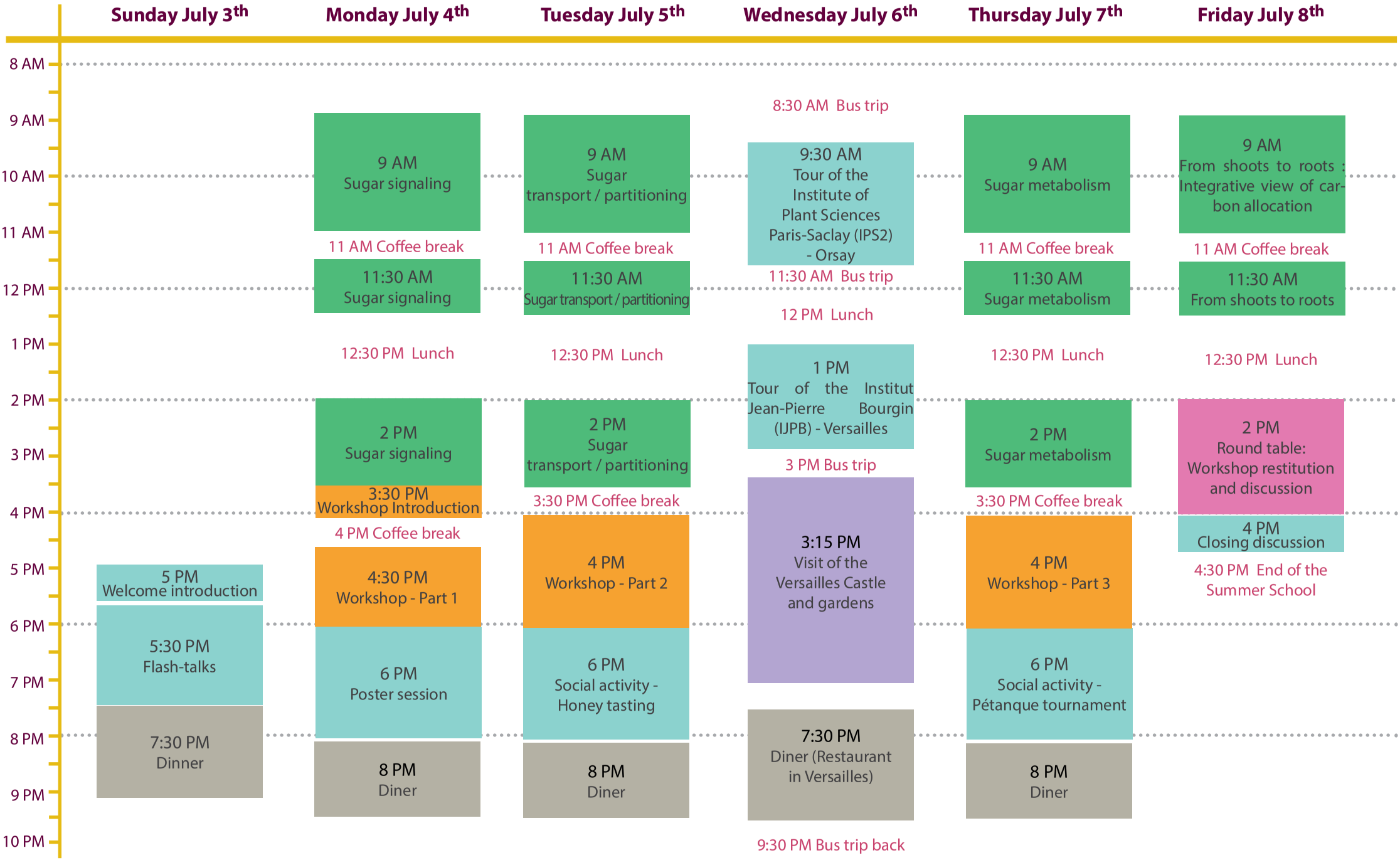


!! Warning !! Make sure the train or RER you take stops at the station you need to get off at (stops are indicated on screens or light panels).

***Attention RER B** (https://www.transilien.com/en/page-lignes/ligne-b)
On the weekend of **July 2 and 3**, modernization work will require the interruption of traffic in both directions, all day long between the stations CDG2 Airport and Gare du Nord.
>>> Direct substitution buses will be set up between «CDG2 airport» and «Stade-de-France Saint-Denis» (B5 line, ~50 min). Then, from «Stade-de-France Saint-Denis», take the RER D in direction of the south («Sud») and stop at «Gare du Nord». Finally, take the RER B as indicated above.

***If you arrive earlier than July 2 :** some trains from the CDG airport go to Saint-Rémy-lès-Chevreuse with no need to transfer at Gare du Nord

Planning at a glance



Program

Sunday July 3

5 PM – 5:30 PM: Summer School introduction

5:30 PM – 7:30 PM: Flash-talks of the participants' research

7:30 PM: Diner

Monday July 4

9 AM – 11 AM: Sugar signaling

John Lunn (Max Planck Institute of Molecular Plant Physiology - Potsdam-Golm, Germany)
"Trehalose 6-phosphate signalling in plants" - *Trehalose 6-phosphate (Tre6P) has a dual function as a signal and homeostatic regulator of sucrose levels in plants. In source leaves, Tre6P regulates supply of sucrose with demand, while in sink organs it links growth and development to sucrose availability, in part via regulation of SNF1-RELATED KINASE 1. Progress towards understanding the molecular mechanisms underlying the nexus between sucrose and Tre6P will be discussed.*

Christine Horlow (Institut Jean-Pierre Bourgin – Versailles, France)
"Signalling and sensing: the role of Class II TREHALOSE 6-PHOSPHATE SYNTHASE genes in suppression of eskimo1 dwarfism phenotype" - *In Arabidopsis, eskimo1 mutant is due to a mutation in XOAT1 gene (Xylan O-acetyl transferase 1). Its mutation leads to abnormal xylan acetylation resulting in collapsed xylem. This cell wall defect reduces water conductivity and generates constitutive plant stress leading to a dwarfism phenotype. This phenotype is supressed by a mutation in Class II TREHALOSE 6-PHOSPHATE SYNTHASE 7 (TPS 7) gene. The two closest homologous class II genes of TPS7, TPS5 and TPS6, were also investigated for esk1 dwarfism suppression. The results will be presented here.*

11 AM – 11 :30 AM: Coffee break

11 :30 AM – 12:30 PM: Sugar signaling

Christian Meyer / Anne-Sophie Leprince (Institut Jean-Pierre Bourgin – Versailles, France)
"Sugars and the TOR kinase signalling pathway: reciprocal and complex relations" - *The conserved and important TOR (Target of Rapamycin) kinase is closely associated with sugar signals as well as sugar metabolism, including starch and polysaccharides. We will present the current knowledge on the regulation of the TOR signalling pathway by sugar signals. This regulatory processes control growth and development but also the overall N and C metabolisms as well as nutrient transport and recycling.*

12:30 PM – 2 PM: Lunch

2 PM – 3 PM: Sugar signaling

Michel Vincentz (Centro de Biologia Molecular e Engenharia Genética - CBMEG - Campinas, Brazil)
"Energy management: a SnRK1-bZIP63-Circadian clock perspective" - *Management of energy resources is fundamental to the fitness of an organism. Adjustment to energy limitation imposed by environmental changes that limit sugar production by photosynthesis involves transcriptional reprogramming induced by the SnRK1-mediated activation of bZIP63 Transcription Factor. The SnrK1-bZIP63 regulatory module interacts with the Circadian Oscillator to define the expression profile of key energy-related genes.*

3 PM – 3:30 PM: Sugar signaling - Discussion with the invited speakers

3:30 PM – 4 PM: Workshop introduction

4 PM – 4:30 PM: Coffee break

4:30 PM – 6 PM: Workshop - Part 1

6 PM – 8 PM: Poster session

8 PM: Diner

Tuesday July 5

9 AM – 11 AM: Sugar transport/partitioning

Evelyne Téoulé (Institut Jean-Pierre Bourgin – Versailles, France)
"Freezing in a warming world: what role(s) for sugars?" - *According to specialized researchers, the climate on Earth is becoming more and more warm. Nevertheless, this modification will probably occur with unexpected cold episodes. So, the identification of new targets for plant breeding allowing the plants to support spring freezing for example could be of major importance in the future. The genetic approaches developed to achieve this objective and also the growing importance of sugars in cold tolerance will be discussed here.*

Nathalie Pourtau (Écologie et Biologie des Interactions - Poitiers, France)
"Variation of carbon source-sink relationship in plants" - *Source-to-sink transport of sucrose is one of the major determinants of plant growth which, depends exclusively on the fine-tuned import of sugar from the aerial part through the phloem. Sucrose produced in leaves by photosynthesis is exported to sink organs like roots, flowers, and siliques through the phloem and involves two types of transporters: AtSUCs and AtSWEETs transporters.*

11 AM – 11 :30 AM: Coffee break

11 :30 AM – 12:30 PM: Sugar transport/partitioning

Ekkehard Neuhaus (TU Kaiserslautern, Germany)
"Regulation and impact of vacuolar and chloroplast sugar transporters" - *Vacuoles represent the central storage compartment in plant cells and chloroplasts represent cellular hubs for acclimation to challenging conditions. In both compartments we find several sugar transport proteins with different properties in respect to substrate specificity and mode of transport. I will demonstrate on selected carriers how transport activity can be regulated and how selected transporters influence plant yield and their tolerance against abiotic stress stimuli. Analyses on corresponding mutants with altered carrier activity allow to broaden our understanding how tolerance against low temperatures is achieved and might lead to novel concepts to improve properties of crop plants.*

12:30 PM – 2 PM: Lunch

2 PM – 3 PM: Sugar transport/partitioning

Rozenn Le Hir (Institut Jean-Pierre Bourgin – Versailles, France)
"From plant development to response to the environment: a SWEET story" - *From their discovery 10 years ago, the study of the SWEET sugar transporters family has focused a lot of attention and lead to further illustrate the central role of sugar transport in all steps of plant growth, development and response to biotic and abiotic environment. In this talk I will present the current knowledge on the SWEET family with a special emphasis on their role in plant response to a changing environment.*

3 PM – 3:30 PM: Sugar transport/partitioning - Discussion with the invited speakers

3:30 PM – 4 PM: Coffee break

4 PM – 6 PM: Workshop - Part 2

6 PM – 8 PM: Social activities - Honey tasting

8 PM: Diner

Wednesday July 6

8:30 AM – 9:30 AM: Bus trip to Gif-sur-Yvette

9:30 AM – 11:30 AM: Tour of the Institute of Plant Sciences Paris-Saclay (IPS2, member of the SPS network)

The IPS2 aims at better understanding the molecular and genetic mechanisms controlling plant growth and their regulation by endogenous and exogenous signals of biotic and abiotic origins. Analysis of these mechanisms is conducted in an integrated manner at cellular, organ and whole plant levels. IPS2 applies multidisciplinary approaches (combining genomics, molecular and cellular biology, bioinformatics, biochemistry, genetics, physiology) and develops tools (including bioinformatics and modelling) required to provide more predictive knowledge and facilitate «translational» research between model species and crops.

11:30 AM – 12 PM: Bus trip to Versailles

12 PM – 1 PM: Lunch at the INRAE cafeteria

1 PM – 3 PM: Tour of the Institut Jean-Pierre Bourgin (IIJPB, member of the SPS network)

Hosted on the INRAE site of Versailles, IJPB is one of the largest European research centers in the field of plant biology. As a joint research unit under the supervision of INRAE and AgroParisTech, it brings together a unique set of resources and skills in biology, chemistry and mathematics dedicated to plant research. The place is steeped in history. In 1973, Jean-Pierre Bourgin took the direction of the INRA laboratory of Cell Biology in Versailles and transformed this laboratory into a pole recognized worldwide in the field of plant cellular and molecular genetics.

3 PM – 3:15 PM: Bus trip to the Versailles Castle

3:15 PM – 7 PM: Tour of the Versailles Castle and its gardens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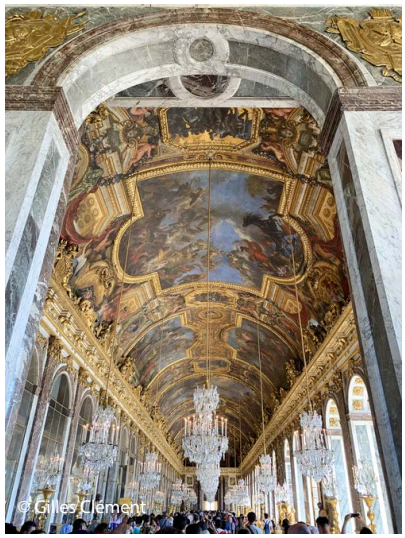
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7:30 PM - 9 PM: Diner at a the restaurant « Le Bœuf à la mode «, 4, rue au Pain 78000 Versailles

9:30 PM: Bus trip back to the Centre Port-Royal

Thursday July 7

9 AM – 11 AM: Sugar metabolism

Benjamin Pommerrenig (TU Kaiserslautern, Germany)
"Protective role of sugars under abiotic stress" - *Plants accumulate sugars in response to various stress stimuli to mitigate impact of challenging environmental conditions. The talk will highlight how photosynthesis and sugar metabolism and transport are affected by cold and drought stress and highlight the role of different sugars including glucose, sucrose, and raffinose and sugar alcohols for protection of cellular metabolism. A focus will be set on vacuolar sugar accumulation.*

Sylvie Dinant (Institut Jean-Pierre Bourgin – Versailles, France)
"Sucrose metabolism: gateway to specific carbon use and efficient source-to-sink carbon allocation" - *Sucrose metabolism plays key roles in development, abiotic stress responses, and plant–microbe interactions, by generating a range of sugars as metabolites to fuel growth and synthesis of essential cellular compounds and as signals molecules. Sucrose metabolic enzymes play diverse roles in source and sink organs, and in the vascular tissues, indicating a coupling between sucrose metabolism, sugar allocation and sugar signaling. In this presentation, I will outline recent progress on engineering sugar metabolism and transport for high yield and acclimation to stresses.*

11 AM – 11 :30 AM: Coffee break

11 :30 AM – 12:30 PM: Sugar metabolism

Bertrand Gakière (Institute of Plant Sciences Paris-Saclay - Gif-sur-Yvette, France)
"Metabolic interactions between sugar metabolism and pyridine nucleotides in plants" - *The pyridine nucleotides nicotinamide adenine dinucleotide NAD(H) and nicotinamide adenine dinucleotide phosphate NADP(H) are both energy transmitters and signal molecules. In photosynthetic organisms, they are involved in the synthesis of sugars and their metabolism, and thus underpin the harmonious development and productivity of plants while allowing plants to acclimatize to environmental variables (biotic and abiotic stress). Sugars are themselves involved in these mechanisms, establishing a cross-talk between these two metabolisms.*

12:30 PM – 2 PM: Lunch

2 PM – 3 PM: Sugar metabolism

Fabien Chardon (Institut Jean-Pierre Bourgin – Versailles, France)
"The two faces of the primary metabolism - when Janus invites Saturn" - *Carbon and nitrogen metabolism are the two sides of the same coin. From the uptake and assimilation to the final nutrient recycling and remobilization, each step in the efficiency of carbon use is interconnected with nitrogen use efficiency in plants. However, despite this intrinsic co-regulation, we can identify some specific molecular signatures of environmental stress in response to each elemental limitation.*

3 PM – 3:30 PM: Sugar metabolism - Discussion with the invited speakers

3:30 PM – 4 PM: Coffee break

4 PM – 6 PM: Workshop - Part 3

6 PM – 8 PM: Social activities - Pétanque tournament

8 PM: Diner

Friday July 8

9 AM – 11 AM: From shoots to roots: integrative view of carbon allocation

Yves Gibon (Biologie du Fruit et Pathologie - Villenave d'Ornon, France)
"Multilevel approach to the dynamics of fruit development and ripening" - *An approach combining experimentation and modelling, taking into account the dynamic aspects of fruit growth and ripening, allows us to better understand how metabolism participates in the development of a plant organ.*

Olivier Loudet (Institut Jean-Pierre Bourgin – Versailles, France)
"Natural variation for Arabidopsis response to stress and stress combination: complex interactions!" - *It is now quite popular to use naturally-occuring variation contained in Arabidopsis thaliana accessions as the source of quantitative genomics and/or systems genetics approaches, combined with all sorts of fine-scale phenotyping. We are exploiting our unique high-troughput phenotyping robot (the Phenoscope) together with diverse omics, to decompose the dynamic growth response to the abiotic environment and ultimately integrate this variation into a plant-scale model. This contributes to drawing a more general picture as to how and where in the pathways adaptation is shaping natural variation.*

11 AM – 11 :30 AM: Coffee break

11 :30 AM – 12:30 PM: From shoots to roots: integrative view of carbon allocation

Frédéric Rees (ECOSYS - Thiverval-Grignon, France)
"Simulating the dynamics of sugars within the root systems: consequences on plant-soil interactions" - *The activity of roots largely depends on the allocation of photoassimilates (e.g. sucrose) from the shoots. Carbon management and exchange between different parts of the root system could thus explain a significant part of plant's plasticity in terms of root growth, root architecture and rhizodeposition. In this presentation, we will introduce a new plant model able to describe the fate of sugars within a 3D root system, and use it to understand what may drive the dynamics of carbon in the rhizosphere.*

12:30 PM – 2 PM: Lunch

2 PM – 4 PM: Workshop - Restitution and discussion

4 PM – 4:30 PM: Closing discussion

4:30 PM: End of the Summer School

Abstracts

The vacuolar sugar porter SFP1 is a novel element in the senescence program of Arabidopsis leaves

Jintao Cheng[1], Meerim Arystanbek Kyzy[2], Benjamin Pommerrenig[2] and Ekkehard Neuhaus[2]

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The genome of vascular plants contains about 60 isoforms of monosaccharide transporters, comprising several subgroups (Pommerrenig et al., 2018). Among these subgroups we find the Early Response to Dehydration Like (ERDL) family, covering in total 19 isoforms. Most of these ERDL carrier proteins have so far not been characterized on the molecular level. The two putative carriers, Sugar-porter Family Protein 1 and 2 (SFP1 and 2) belong to this ERDL family (Pommerrenig et al., 2018). For SFP1 it was shown that its gene is senescence induced (Quirino et al., 2001). However, any further information on SFP1 and its corresponding gene is lacking.

With help of a SFP1-GFP fusion protein we demonstrated that this transporter locates to the vacu-olar membrane. After recombinant expression in baker´s yeast cells and use of the fluorescing sugar analogs 2-(*N*-(7-nitrobenz-2-oxa-1,3-diazol-4-yl)amino)-2-deoxyglucose (2-NBDG, green fluorescing) and esculin (blue fluorescing) we raised evidence that SFP1 transports both, glucose and sucrose in counter exchange to vacuolar protons. The SFP1 promotor contains binding sites for ABA-responsive element binding-type transcription factors (ABREs). In fact, we demonstrated by exploiting the power of SFP1$_{Prom}$::GUS mutants that the *SFP1* gene is positively ABA responsive, which is in line with the marked upregulation of *SFP1* gene expression during late senescence or under conditions of extended darkness. Surprisingly, the typical symptoms of senescence as loss of pigmentation or activation of the expression of senescence associated genes (*SAGs*) is less prominent in *sfp1* mutants. Senescent leaves from *sfp1* mutants contain less sugars and produce smaller seeds, which contain less lipids than present in corresponding wild types.

In summary of these observations, we present a molecular scheme to set SFP1, as a first sugar porter, into the scenario of a controlled senescence program in Arabidopsis.

References:

Pommerrenig B, Ludewig F, Cvetkovic J, Trentmann O, Klemens PAW, Neuhaus HE (2018) In concert: orchestrated changes in carbohydrate homeostasis are critical for plant abiotic stress tolerance. Plant, Cell Physiol. 59: 1290-1299

Quirino BF, Reiter W-D, Amasino RD (2001) One of two tandem Arabidopsis genes homologous to monosaccharide transporters is senescence-associated. Plant Mol. Biol. 46: 447-457

Regulation of leaf primary metabolism and sink/source relationships during acclimation to drought stress in Brassicaceae (PRIMABRA)

Mathieu Aubert[1], Solenne Berardocco[1,2], Nathalie Marnet[2], Alain Bouchereau[1,2], Younès Dellero[1,2]

1 Institute for Genetics, Environment and Plant Protection (IGEPP), National Research Institute for Agriculture, Food and Environment (INRAE), Université Rennes, Institut Agro, Le Rheu, France
2 Metabolic Profiling and Metabolomic Platform (P2M2), Institute for Genetics, Environment and Plant Protection (IGEPP), National Research Institute for Agriculture, Food and Environment (INRAE), French institute of cider productions (IFPC), Le Rheu, France

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Oilseed Rape (OSR) is the second most cultivated oleaginous in the world, with important applications for human and animal nutrition (seed oil and seed cake). However, its grain yield is threatened by climate change, which will increase the duration and the intensity of drought stress periods. Indeed, plant acclimation to water shortage induces a rapid stomatal closure in order to reduce water losses by evapotranspiration. This has important consequences for the maintenance of plant growth, leaf primary metabolism and sink/source relationships. Depending on their sink/source status and developmental stage, OSR leaves either go into an accelerated senescence (old leaves) to remobilize trophic resources or maintain part of their metabolic activity through accumulation of osmoprotective compounds (young leaves). However, these ultimate metabolic adjustments require coordinated regulations of metabolic fluxes within plant primary metabolism, which remained to be dissected. To investigate these regulations in young and old leaves of OSR, we will subject plants to a progressive water shortage of 18 days and will combine physiological measurements of water status and photosynthetic parameters with metabolomic fingerprints of leaf primary metabolism and 13C-metabolic flux analysis. Overall, our first results validated the experimental setup for drought stress application to OSR. We also identified leaf ranks having contrasted responses according to the evolution of their relative water content, chlorophyll index and proline content.

Tulip reproduction and the role of sink-source dynamics

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Tulip (*Tulipa gesneriana*) is undisputedly of ornamental value and economical importance. Like other geophytes, tulip plants yearly alternate a dormant bulb stage with a stalk stage which bears a flower and leaves. It derives that sink-source dynamics are at the base of this behavior, ultimately favoring the formation of above- or below ground organs. Currently, the molecular regulation of these major developmental events in tulip is unknown. Extensive literature on PHOSPHATIDYLETHANOLAMINE-BINDING PROTEINS (PEBPs) in a multitude of plant species has converged in highlighting their central role in the regulation of developmental events such as sexual and vegetative reproduction, storage organ formation, branching and dormancy. Therefore, our study aims to unravel the role of tulip PEBPs during major events of the adult tulip lifecycle: flower induction, bulb formation and dormancy release.
Due to their mobile nature, PEBPs might not only regulate the single processes, but also mediate their cross-talk, which necessarily involves energy sensing and sugar transport. For this purpose, detailed investigation of the currently identified PEBPs was performed, including *in silico* profiling, gene expression analyses, protein-protein interaction screenings and heterologous functional studies.
Our latest results strongly point at *TgFT1* as the bulb inducer, while the role of florigen could be shared between the leaf expressed *TgFT2* and the meristem-expressed *TgFT4A*. Interestingly, emerging tulip leaves show the highest *FT* expression, suggesting a possible involvement of these genes in unfolding and growth of above ground organs. Although this points to PEBPs as mediators of sink-source regulation, further experimental evidence is needed to unravel their involvement and exact functioning in this process.

Regulation of fruit set by light quality: A case study on pepper

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Fruit set is a crucial plant developmental process, determining yield in many crops. Pepper (*Capsicum annuum* L.) is a crop with poor fruit set as typically about two-third of all flowers abort even in protected cultivation. Weeks with good fruit set are alternated with weeks with poor fruit set, resulting in a flushing pattern in fruit set and yield. Abortion of flowers and fruit in peppers is an active process involving the formation of an abscission layer. So far, the effect of light spectrum on this process has hardly been investigated. Opportunities for detailed investigations of light quality effects on fruit set have strongly increased because of the development of narrow band LED lighting. We aim to improve our understanding on how fruit set can be manipulated by light spectrum, by studying the physiological and molecular mechanisms. Red: far-red ratio of the light is a crucial environmental signal to plants. We have observed improved fruit set in pepper at high red: far-red ratio. Auxin export from flowers and young fruit is necessary for fruit set to prevent the formation of an abscission layer. A higher light integral, thus higher amount of assimilates, improves fruit set. So, we will investigate the role of auxin and sugar in this effect with a series of climate room experiments. The outcomes will be meaningful for lighting optimization in the cultivation of fruit bearing crops.

Unravelling molecular mechanisms underlying the superior performance of grass stomata

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In higher plants, stomata are found on most aerial parts. Thanks to the guard-cell-pair structure, stomatal pores are adjustable in response to exogenous and endogenous signals, thereby controlling the gas exchange between the plants and the ambient environment. By balancing the trade-off between photosynthetic CO_2 assimilation and transpirational water loss, stomata determine plant water use efficiency, which is among the essential traits of crops to adapt to the changing environment.

Interestingly, the dumbbell-shaped stomata, found only in grass (Poaceae) family species, show superior responsiveness to fluctuating environmental conditions compared to their kidney-shaped counterparts, which are commonly found in other land plants[1]. Understanding the molecular mechanisms underlying this difference will be one of the first fundamental steps towards improving plant water use efficiency.

Very recent studies revealed the essential roles of Arabidopsis guard cell starch metabolism and sugar transporters in the speed of stomatal movement[2]. These findings have not yet been demonstrated in the unique dumbbell-shaped stomata of Poaceae. The patterns of carbohydrate metabolism and transportation in grass guard cells are still under-investigated. Hence, my project aims to elucidate whether starch and sugars play a role in the superior performance of grass stomata. The project compares starch and sugar metabolic pathways between dumbbell-shaped stomata and their kidney-shaped counterparts.

References:

[1] Lawson, T. and Vialet-Chabrand, S. (2019) Speedy stomata, photosynthesis and plant water use efficiency. *New Phytol.* 221, 93–98

[2] Flütsch, S. *et al.* (2020) Guard cell starch degradation yields glucose for rapid stomatal opening in Arabidopsis. *Plant Cell* 32, 2325–2344

Starvation-induced transcriptional control: metabolic signals, their transmission via the central metabolic kinase SnRK1 and downstream gene regulation

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Plants use sunlight to synthesize energy-rich carbohydrates, which provide energy reservoirs during the night and support growth and development. The central metabolic kinase SnF1-RELATED PROTEIN KINASE1 (SnRK1) orchestrates plant's catabolic functions when energy is limited via activation of alternative energy-conserving pathways. In this respect, SnRK1-dependent phosphorylation of the downstream group C/S1 bZIP (basic leucine Zipper) transcription factors (TFs) has been well-established. Moreover, SnRK1 performs as a crucial integrator of resources and growth during normal growth and development. Here, we are using extended night treatments as an experimental starvation system in Arabidopsis leaves leading to a rapid nuclear activation of SnRK1 kinase activity within less than 30 min. We aim at understanding the metabolic signals (e.g. sugars like sucrose, glucose and Trehalose-6-Phosphate) or regulatory cues (e.g. clock) which control nuclear SnRK1 activity and its impact on gene regulation. Studying the frequently used *ASPARAGINE-SYNTHETHASE* (*ASN1*) marker gene, we analyzed the impact of the SnRK1 catalytic subunit on nuclear transcription factor - cis-element interplay. This prototypic study will gain deeper insights into the mechanisms involved in transcriptional control by energy limiting conditions.

Analysis of the activity of fructosyltransferases identified in *Agave tequilana* using *Arabidopsis thaliana* as a heterologous model

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Fructans are fructose polymers synthesized from sucrose in around 15% of angiosperms and are considered as dynamic molecules thought to be involved in stress tolerance and signaling in addition to carbohydrate storage. The synthesis of these polymers is carried out by fructosyltransferases (FTs) which in monocotyledonous plants, involves at least 3 distinct enzymatic activities and combinations of FTs with distinct activities to produce a diversity of fructan molecules in different plant species. *Agave tequilana* is capable of storing high amounts of fructans (agavins) that are characterized by complex structures and a high degree of polymerization. However, extensive RNAseq analysis in *A. tequilana* has identified transcripts encoding 1-SST, 6G-FFT and 1-FFT type FTs but no definitive evidence for 6-SFT encoding transcripts, leading to the hypothesis that Agave FT enzymes may be able to carry out dual activities. To functionally characterize the predicted *A. tequilana* FTs, cDNAs encoding a 1-SST and two isoforms of 6G-FFT enzymes were expressed in *Arabidopsis thaliana,* a non-fructan accumulating plant, under the CaMV35S promoter. The proteins were tagged with a yellow fluorescent protein in order to immunoprecipitate the proteins to characterize the posttranslational modifications by expressing them in *Nicotiana benthamiana*. Analysis of carbohydrate profiles will be carried out to assess the synthesis of fructan molecules in transformed plants, and these plants will go under water stress conditions to confirm the biological function of fructans. Agave FTs were experimentally confirmed to be glycoproteins, and mass spectrometry analyzes will be carried out to identify modified amino acids and their glycosyl moieties.

Sugar signalling in the transcriptional regulations of shoot branching

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Plant architecture is a key determiner of crop yield. Sucrose, the main transported plant sugar, is suggested to be the first activator of axillary bud outgrowth, which leads to highly branched or bushy plants. I am using a combination of genomics, physiological, and molecular tools to determine interactions between strigolactones and sugar signalling in Arabidopsis.

The transcription factor, basic leucine zipper 11 (bZIP11) and trehalose 6-phosphate (Tre6P), a hormone-like metabolite, are both sucrose sensors in plants, and both influence plant shoot architecture. bZIP11 signals low sucrose availability in plants, is strongly repressed by high sucrose, and inhibits axillary bud outgrowth. Tre6P signals high sucrose availability and promotes bud outgrowth. There is some evidence that both bZIP11 and Tre6P negatively regulate each other. I am using transgenic Arabidopsis plants harbouring inducible systems to investigate genetic responses to both bZIP11 and Tre6P. I have surveyed the chromatin landscape following 45-minute induction of bZIP11 translocation into the nucleus in Arabidopsis protoplasts, using ATAC-seq.

The findings so far, shed light on how bZIP11 represses axillary bud outgrowth. Over 2000 genes are made differentially accessible by the induction of bZIP11. Strigolactone perception gene *MORE AXILLARY GROWTH 2* (*MAX2*) was identified as one of these genes, showing chromatin being differentially accessible upstream of the coding region. bZIP11-induced opening of the chromatin around *MAX2* also coincides with enhanced *MAX2* gene expression. Other putative targets of bZIP11 include *Trehalose 6-phosphate phosphatase* (*TPP*) genes. It appears that bZIP11 might negatively regulate Tre6P by enhancing its dephosphorylation. I have begun to investigate the transcriptional response to the induction of Tre6P and would like to determine if Tre6P has negative feedback on the bZIP11 protein activity.

Branched malto-oligosaccharides cause spontaneous starch granule initiation in *A. thaliana* chloroplasts

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Plants store starch in their chloroplasts to use as source of energy during the night. A mature Arabidopsis chloroplast cross-section holds about 4-7 starch granules which grow during the day and shrink at night. Granules are initiated as chloroplasts grow and divide, creating a balance between chloroplast size and granule number.

Starch granule initiation is a controlled process during which oligosaccharides present in the stroma are selected by Protein Targeting To Starch (PTST) 2/3 proteins and extended by the Starch Synthase IV (SSIV) protein. The extended oligosaccharides can serve as a substrate for branching enzymes and other starch synthases, and eventually crystallize, forming a granule initial, that is presumably resistant to breakdown. The oligosaccharides used for starch granule initiation may result from de-novo synthesis, or from starch breakdown.

When the two debranching enzymes Isoamylase 3 (ISA3) and Limit Dextrinase (LDA) are missing, branched oligosaccharides accumulate and large numbers of small granules are observed. We hypothesized that this over-initiation effect could be caused by the accumulation of branched oligosaccharides. To test this, I investigated the triple mutant *isa3 lda amy3*, which is also deficient in Alpha-Amylase 3 – the enzyme that can release branched oligosaccharides from starch. Compared to *isa3 lda*, the triple mutant has increased accumulation of starch, but no accumulation of branched oligosaccharides, and far fewer granules per chloroplast. This indicates branched oligosaccharides are indeed responsible for granule over-initiation in *isa3 lda*.

To determine whether branched oligosaccharides are substrates for the known granule initiation system involving PTST2/3 and SSIV, or if they bypass it entirely, I am examining plants deficient in ISA3, LDA, and SSIV. Preliminary data show that despite the absence of SSIV, over-initiation still occurs. This points towards branched oligosaccharides being used as granule initials without involving the presently described granule initiation system. These findings give insight into how starch granules are initiated and established, and how this process can be influenced to initiate more.

Group S1 bZIP Transcription Factors Control Axillary Branching by Steering Carbon Allocation in *Arabidopsis thaliana*

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As sessile organisms, plants must respond to energy-limiting conditions by adjusting resource mobilization and allocation. The Snf1-RELATED PROTEIN KINASE 1 (SnRK1) has been found to be involved in these responses, acting as a master regulator for a plethora of downstream targets. Among those, the group C and S1 basic leucine zipper (bZIP) transcription factors have been implicated in a variety of developmental and metabolic adaptations, reviewed in [2].
Most studies conducted in this field utilize harsh stress conditions or overexpression approaches, as single bZIP knockouts often show no significant effects due to partial redundancy. Using CRISPR, we generated a wide range of novel loss-of-function combinations. We observed specific and highly reproducible phenotypes in sink organs without affecting overall plant growth, showing the systems importance in plant development even under non-stressed conditions.
The agronomically important trait of axillary branching is tightly linked to carbon availability [3]. While branching speed was increased in bZIP mutants, seed size was reduced, indicative of a change in resource allocation subsequently confirmed by sugar measurements. In line with the promoters of several sugar transporters containing common bZIP binding sites, we found a strong reduction in gene expression of those potential downstream targets. Unravelling the mechanistic details of these interactions will grant insights into the complex regulatory network controlling resource distribution between competing sink tissues.

Improving the industrial and agricultural potential of arbuscular mycorrhiza through the modulation of the carbon status in tomato

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Arbuscular mycorrhizal fungi (AMF) are mutualistic symbionts of a wide range of plants, including tomato. AMF supply their hosts with nutrients and receive sugars and lipids in return. They can offer a sustainable solution for nutrient deficiencies and drought in tomato cultivation during ongoing climate change. Plant-derived sugars are also essential for tomato fruit development and quality. Hence, a fine-tuned nutrient exchange at the sites of plant-AMF interactions, i.e. arbusculated root cells, is essential to obtain benefits for both AMF and tomato. My project aims to engineer the sugar flux specifically at these sites to optimize mycorrhization without affecting tomato fruit development and quality and to enhance AMF spore production. I will first select tomato sugar transporters and sensors specifically expressed in arbusculated root cells using LCM-qRT-PCR. To elucidate their role during mycorrhization, arbusculated cell-specific transgenic composite tomato plants will be generated for each of the candidates using state-of-the-art CRISPR-TSKO technology. Next, I will study the molecular mode-of-action of five candidates that affect mycorrhization using a multidisciplinary approach. In the last stage of the project, we will collaborate with INRAE-Bordeaux to unravel how an altered sugar flux in arbusculated cells regulates tomato growth and fruit development using stable transgenic tomato lines. This project will lead to more sustainable agricultural practices.

Carbohydrate transport dynamics during root tip regeneration

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Plant roots are formed by proliferation and patterning activity occurring at the root apical meristem and the stem cell niche within it. Remarkably, the complete removal of the stem cell niche triggers its rapid reformation from a well-defined regeneration zone in the stump. Removal of the shoot system severely inhibits root regeneration, suggesting it is regulated by shoot-derived cues. While myriads of molecules are transported from the shoot, I show that sucrose is the key shoot-derived molecule required for regeneration and that its availability at the regeneration zone controls the decision of whether or not to regenerate. However, how sucrose is transported and recruited by the regeneration zone is unclear.

During normal growth, sucrose is transported through the phloem and is unloaded symplastically at the root meristem phloem unloading zone. Paradoxically, I show that the regeneration zone becomes symplastically isolated during regeneration, limiting this mode of transport. Preliminary results suggest that following wounding, the decapitated root tip transiently shifts to an apoplastic sucrose transport route in order to bypass the symplastic block. I am currently mapping the spatio-temporal expression patterns of the different apoplastic transport route components in order to study how these are coordinated during this temporary shift. My aim is to combine genetic, imagining and modelling approaches in order to understand how plants mediate their resource reallocation to allow tissue repair following damage.

Roles of sugar transporters during the competition for sugars at the Grape/fungus interface

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In a context in which alternative methods of control against cryptogamic diseases are increasingly expected, there is an urgent need of better understanding the plant/pathogen dialogue. Fungi, because of their heterotrophic dependency on carbon, take sugars as a source of energy from plants tissues. In grapevine, the necrotrophic fungus *Botrytis cinerea*, causing Gray mold disease, engenders qualitative and quantitative losses on grapes. During my PhD project, I explore Grape/fungus interactions to investigate the mechanisms set up by the plant to deprive fungus of sugars. Particularly, I study the role of a candidate gene for which the closest homolog in *Arabidopsis thaliana* has been demonstrated to be involved in the basal resistance against *B. cinerea*. Furthermore, I investigate the genetic variability from different varieties with contrasted tolerance levels to *B. cinerea*. Through this genetic variability, I would like to highlight correlations between sugar transporter expression and tolerance levels to the fungus. Finally, transcriptomic study from this different varieties will bring new candidates to consider for improving Grape to fight efficiently against pathogens. Results from this project will help to better understand the dialogue between plant and fungi, but it will also bring determinants for the creation of resistant varieties against cryptogamic diseases.

Optimized carbon fluxes for improved yields and water stress tolerance in peas

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Seed development and seed filling rely on carbon fluxes and partitioning toward seeds. Sugar transporters are key molecular actors mediating this long-distance transport between source organs and sink organs, including seeds. In this study, we focus on pea crop (*Pisum sativum*), an agroecological model legume cultivated for its high nutritional seed value. We applied a 40% of field capacity water stress during early flowering of the pea plant, thereby inducing major changes in carbon fluxes and decreasing seed yield. By comparing two pea varieties widely grown in Europe, we also observed different seed abortion rates and water use strategies. Then, we targeted main sugar transporters families, SUT (sucrose transporters), MST (monosaccharide transporters) and SWEET (sugar will eventually be exported transporters), mediating carbon allocation from source leaves to seed sinks. This study aims to characterize key transporters unloading carbon into seeds and differentially expressed during drought. In parallel, we also conducted a transcriptomic approach in carbon source (leaf) and sink organs (seed and root) to uncover genome-wide expression changes during drought stress. Altogether, our integration of phenotypic, molecular, and genomic approaches, will further improve our understanding of drought tolerance and nutrient filling in seed crops.

Regulation of sugarcane aerenchyma formation and sugar accumulation mediated by sugar sensing and signaling

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Aerenchyma is a structure characterized by large intercellular spaces filled with gases. In sugarcane roots it occurs constitutively, and studies related to the changes that occur during its formation can add knowledge about the structure of the cell wall. During aerenchyma formation, the ethylene-responsive transcription factor ScRAV1 negatively regulates the action of an endopolygalacturonase, a pectinase that acts on the middle lamella of root cortex cells. Evidences indicate that the regulation of cell wall biosynthesis and degradation processes can be controlled by the available carbon status in the cell, providing a regulation mediated by the sugar signaling process. Thus, the objective of this work is to elucidate the participation of sugar sensors in the formation of aerenchyma in sugarcane roots, in order to understand the relationship between carbon flux in plants that leads to cell wall production. In this way, transgenic plants that overexpress *ScRAV1* were evaluated for aerenchyma formation, structural and non-structural carbohydrate contents, and carbon assimilation capacity, in addition to the expression of target genes related to sugar signaling. The increase in the expression of *ScRAV1* led to a delay in the formation of aerenchyma in roots, in addition to an increase in the accumulation of sugars and an advance in the maturation of the plants. This dataset raises important questions about the sugar-mediated signaling mechanisms that affect cell wall modulation, and consequently, their necessary interconnections for aerenchyma formation.

RNA-seq analysis reveals sugar-related genes association to drought tolerance in pepper plants

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Among abiotic stresses affecting crops, water scarcity rises as a major problem in many parts of the world, causing important losses in productivity every year (Ashraf, 2010). Grafting, meaning the fusion between the root and the shoot from different plants, emerges as a sustainable and eco-friendly technique that can be used to face this threat, particularly in Solanaceae species, usually cultivated in areas suffering from water starvation (Fullana- Pericàs et al., 2020; Gisbert-Mullor et al., 2020, Rouphael et al., 2018). Little has been published about the processes underlying grafted plants tolerance to water stress, especially in pepper plants. This work aimed to unravel tolerance mechanisms by studying the transcriptomic profile of two pepper rootstocks: NIBER®, designed to cope with water stress, and A10, sensitive to water stress. Plants were exposed to 4% polyethylene glycol in hydroponic culture for 5h (T1) and 24h (T2) to induce water stress and RNA-sequencing was performed with root samples. Differences among rootstocks in the transcriptomic profile for genes related to sugar transport (SWEETs), synthesis (GolS) and modifications (RFS) were found, mainly for galactinol and raffinose family of oligosaccharides (RFOs). These results suggest the implication of sugars that can function as osmoprotectants in the response to water stress, dealing to a differential response of tolerance or sensitivity. Therefore, they can be employed as biomarkers to select tolerant rootstocks and use them in grafting under water stress conditions.

References:

Ashraf, M. (2010). Inducing drought tolerance in plants: Recent advances. Biotechnology Advances, 28, 169-183.

Fullana-Pericàs, M., Conesa, M., Pérez-Alfocea, F. and Galmés, J. (2020). The influence of grafting on crops' photosynthetic performance. Plant Science, 295, 110250.

Gisbert-Mullor, R.; Pascual-Seva, N.; Martínez-Gimeno, M.A.; López-Serrano, L.; Badal Marín, E.; Pérez-Pérez, J.G.; Bonet, L.; Padilla, Y.G.; Calatayud, Á.; Pascual, B.; López-Galarza, S. (2020)

Altering microbe-induced stomatal closure to restrict foodborne diseases of leafy greens

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Stomata are tiny pores on the leaf surface. Despite their key role in regulating gas exchange, stomata also represent a natural entry point for bacteria. However, plants can sense the presence of pathogens and rapidly close the stomata to reduce their entrance. In this scenario, sugars represent an interesting subject of study. Not only sugars are the main substrate for pathogens to survive, but also sugar homeostasis is known to influence stomatal opening and closure. The role of sugars in plant immunity is currently a hot topic in the phytopathology field, and every year new roles for sugars in the immunity mechanisms are being discovered. Nevertheless, the link between sugar metabolism in guard cells, stomatal closure and the signalling cascade triggered by the perception of pathogens, has not yet been investigated. This doctoral project aims to study how changes in sugar metabolism in guard cells may be linked to pathogen-triggered stomatal closure, exploiting the model plant Arabidopsis thaliana and the interaction with the biotrophic bacteria Pseudomonas syringe pv. Tomato (*Pto*). Since fast stomatal closure triggered by sensing of pathogen invasion is an efficient way to reduce pathogen proliferation in the plant tissue, genetic manipulation of this defence mechanism can lead to more resistant crops. Given the urge of finding targets for improving food safety, genetically modified lettuce for increased stomatal immunity will be evaluated for the internalization of the human pathogen Salmonella enterica (*Se*).

Wounds as a path for SWEET sucrose transporter-dependent kresek infection of rice roots

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Bacterial leaf blight (BLB) of rice caused by *Xanthomonas oryzae* pv. *oryzae* diminishes rice yields by up to 70 % annually. The most destructive form of BLB occurs on seedlings and leads to the complete wilting of plants, called *kresek* (translated: "sound of dead leaves"). Due to the observation that *kresek* often occurs after transplantation, we speculate that wounds at the root interface serve as gate for *Xoo* invasion. We use translational SWEETGUS reporter lines and confocal microscopy of fluorescent *Xoo* to understand the virulence mechanism of *Xoo* in the root and the alteration of sugar transport after infection.

Limitation of etioplast gene-expression induces skoto-morphogenesis reprogramming

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Seedlings of terrestrial plants can develop in the dark when seeds are covered by soil, following a unique developmental program (skotomorphogenesis: nongreen cotyledons, long hypocotyl and apical hook). They present the "triple response" features (hypocotyl shortening/thickening, hook curvature exaggeration (twist)) in the presence of a compact soil overlay, strengthening the seedlings towards soil friction.

During skotomorphogensis, mitochondria are the major organelles involved in ATP synthesis. We recently identified the involvement of mitochondria in *Arabidopsis* seedling skotomorphogenesis where it was found that the transcriptional/respiratory dysfunctional *rpoTmp* seedlings show apical twist/hypocotyl shortening, and a significant mitochondrial stress. The phenotypic induction also requires the ANAC017-dependent mitochondrial retrograde pathway and alternative oxidase (AOX) activity.

During growth in darkness, also etioplasts contribute to ATP synthesis by a process called etio-respiration. Because of the energetic activity of etioplasts and the various similarities and interactions between mitochondria and plastids, we investigated the potential role of plastids in skotomorphogenesis. We found that etiolated seedlings treated with rifampicin or spectinomycin, targeting the transcription and translation of etioplast, have a pronounced apical twist. We found a substantial elevation of the O2 consumption capacity of the mitochondrial AOX enzymes. These findings strongly suggest that the plastid is involved in the regulation of skotomorphogensis and point to the existence of a possible interaction between plastids and mitochondria, prompting us to choose metabolomics to uncover metabolic cross-talk between plastids and mitochondria.

Uncovering the Wintertime Function of a Plant Sugar-binding F-box Protein

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Plants anticipate seasonal changes by measuring the relative length of day and night, or photoperiod. Photoperiod changes are a consistent signal that denotes time of year and can be measured to adjust physiology in preparation for unpredictable seasonal fluctuations, such as temperature and water availability. Photoperiod measurements allow plants to enact biological processes and regulate genes that are appropriate for yearly variations in the environment. We have used bioinformatics to identify a group of winter photoperiod-induced genes in Arabidopsis. We have observed that one "winter gene," PHLOEM PROTEIN 2-A13 (PP2-A13), is critical for plant health in short day winter-like photoperiods. PP2-A13 is a lectin containing F-box protein that may be involved in glycoprotein degradation. Using the PP2-A13 promoter driving luciferase as a real-time photoperiod reporter, we begin to identify the genetic and cellular drivers of winter photoperiodism. Our work reveals a mechanism that relies on coincidence between photosynthesis mediated light sensing and rhythmic starch metabolism. This photoperiod measuring mechanism enables plants to enact biological and developmental processes that are crucial for survival in different seasons. Additionally, we are beginning to understand the cellular function and sugar binding capabilities of PP2-A13 through biochemistry, mass spectrometry, and genetics.

Using 13C isotope tracer to study how sink limitation affects *Betula pendula* sugar translocation

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In plants, photoassimilates are mainly produced by leaf mesophyll, and translocated through phloem. The driving force of phloem flow is usually seen as the osmotic gradiant, generated by sugar loading at the source and unloading at the sink. However, mechanistic understanding of loading-transport-unloading processes and the influence of carbon sink strength on phloem transport remains particularly understudied.

We aim to understand the effects of sink limitation on canopy gas-exchange and phloem transport by first applying 4°C cooling treatment to birch seedling (*Betula pendula*) root systems, to decrease sink activity. On top of which, we use 13C isotope pulse labelling to trace sugar transport. To monitor the 13C enrichment in different tissues and of different positions, we implemented isotope and gas concentration analyzer in-situ, and δ13C isotope-ratio mass spectrometry ex-situ. Photosynthesis assimilation rate was also measured on canopy leaves, along with osmotic potential.

By tracing 13CO2 of stem bark respiration and leaf/phloem sap 13C-sugar, our initial results show a typical pattern of the after-pulse 13C enrichment: a 13CO2 peak at stem usually comes 1-2 days after pulse; compared with control seedlings, the 13CO2 peak comes 18-30 hours later on the soil-cooled seedlings, indicating a slower carbon transport velocity under sink limitation. Leaf gas exchange results show that the assimilation rate of control seedling leaves are significantly higher compared with soil-cooled seedlings, suggesting that weakened sinks might debilitate photoassimilation, possibly by slowing down the unloading process.

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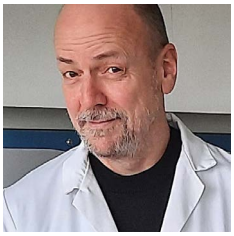

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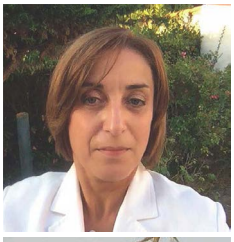

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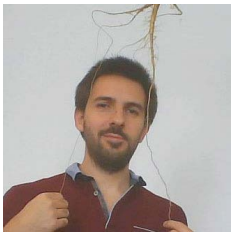

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