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Abstracts

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https://ijpb.versailles.inrae.fr/en/research-teams/branching-control-in-plants/presentation

Genetic and molecular mechanisms involved in rhizospheric plant-plant interactions

Our lab has been working for several years on strigolactones, as a plant hormone, but also as allelopathic compounds released in the rhizosphere by the roots. The release of these molecules leads to the stimulation of parasitic seed germination (Striga and Orobanche). Our work on strigolactone perception mechanism by Phelipanche ramosa, allows us today to elaborate strategies against this pest.

More recently, we have started a project aiming to identify new allelopathic compounds exuded by roots and the genes involved in their biosynthesis. For this, we use multidisciplinary approaches combining biology/genomics and analytical chemistry tools. Our first work on the plant model Arabidopsis thaliana, allowed us to identify two families of molecules potentially involved in plant-plant interactions. This presentation will be an opportunity to present our latest progress on these 2 research projects which aim to better understand the communications between plants within the rhizosphere.

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Insights from "Grafting biology"

Plant grafting has been an important technique in agriculture to propagate clones and to obtain benefits of certain rootstocks. However, graft-incompatibility has limited the technique. Recently, we found that *Nicotiana* species of Solanaceae show the ability to graft with distantly related plant species beyond the family. Graft adhesion with diverse angiosperms by *Nicotiana* species was probably facilitated by the secretion of a subclade of ß-1,4-glucanases. The capability of interfamily grafting was also found in the model Orobanchaceae hemiparasitic plant, *Phtheirospermum japonicum*, which naturally invades to the tissues of host plants of different families. Transcriptome analysis indicated that the same clade of ß-1,4-glucanase plays an important role in plant parasitism. Thus, grafting biology have opened new insights on plant systems to cure the wound tissues, such as sealing damaged tissues, reallocation of energy resources, cell division, tissue adhesion, tissue differentiation and vascular connection for systemic communication.

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The role of cell layers during petunia petal development

Flowering plants' aerial organs are all organized in independent cell layers, derived from the meristematic L1, L2 and L3 layers. How these layers differentiate during organ development, and coordinate their growth to build up an organ with a functional and reproducible shape, stands unresolved. Here, we use the model species petunia that makes fused petals organized in a basal tube and distal pigmented limbs. We have isolated layer-specific mutants in the MADS-box gene *PhDEF*, encoding a major petal identity regulator. Strikingly, these flowers present very different phenotypes: flowers expressing *PhDEF* in their epidermis form proper limbs but their tube hardly grows, while flowers expressing *PhDEF* in their mesophyll form a proper tube but their limbs are very reduced and unpigmented. This reveals that the petunia petal has a modular architecture, whose development is driven by distinct cell layers. I will present the detailed characterization of these phenotypes and our recent results using this unique material, as well as the multiple questions that they raise on the role of cell layers in petal development.

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CLE peptide as a positive regulator of plant stem cell identity

Overall morphology of shoot systems relies on the activity of shoot apical meristems at the tips of plant bodies. Each meristem maintains a pool of stem cells at the center, surrounded by differentiating daughter cells. In the flowering plant *Arabidopsis thaliana*, the stem cell population is maintained at a constant size by the canonical CLV3-WUS feedback loop. To understand the evolutionary origin of this cell signaling system, we study MpCLE2, the sole CLV3 ortholog in the liverwort *Marchantia polymorpha*. We show that treatment with MpCLE2 peptide resulted in the accumulation of undifferentiated cells, marked by MpYUC2 expression, in the apical meristem. Removal of MpCLE2 peptide resulted in supernumerary branching from the accumulated cells, demonstrating that MpCLE2 positively regulates stem cell identity in *M. polymorpha*, in sharp contrast to the stem cell-limiting activity of CLV3 in flowering plants. We are now trying to elucidate the mechanisms of the inversion of the CLE/CLV3 peptide activity on stem cells during the evolution of land plants.

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Physiology and immunity of *Brassicaceae* hydathodes upon infection by bacterial pathogens

Hydathodes are leaf organs present in all vascular plants where they mediate guttation. These organs provide a facilitated access to plant inner tissues including xylem vessels. Hydathodes are thus preferred infection sites for several bacterial vascular pathogens such as *Xanthomonas campestris* pv. *campestris* (*Xcc*), the causal agent of black rot disease in *Brassicaceae*. Our goal is to unravel the physiology and immunity of hydathodes in Arabidopsis and cauliflower using metabolomic and transcriptomic approaches in healthy and infected hydathodes. Latest results will be presented.

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Control of root growth direction through indole-3-butyric acid uptake mediated by NPF7.3/NRT1.5 in Arabidopsis

An ability to precept and respond to gravity, so-called gravitropism, is largely known to be a fundamental function for ensuring the optimal development of terrestrial plants. Plant hormone auxin serves as a signal molecule, and its uneven distribution in developing tissues, which triggers gravitropic responses, are established by the polar transport of indole-3-acetic acid (IAA) that is the most abundant naturally occurring auxin. Here, we propose that cellular uptake of indole-3-butyric acid (IBA), one of the precursors of IAA, contributes to the formation of auxin-uneven distribution in a root tip as well as IAA transport and that this step is mediated by one family protein of the Nitrate transporter1/Peptide transporter Family (NPF), NPF7.3/NRT1.5 in Arabidopsis. Mutations in the NPF7.3/NRT1.5 gene inhibited root gravity responses inducible by auxin, consequently resulting in the disrupted growth direction of Arabidopsis roots. Direct transport assay using yeast cells showed that IBA is preferable to IAA as a transport substrate of NPF7.3/NRT1.5. Our findings shed light on the physiological significance of endogenous IBA and its uptake mediated by NPF7.3/NRT1.5 in root development in Arabidopsis.

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Live single cell transcriptional dynamics via RNA labelling during the phosphate response in plants

Plants are sessile organisms constantly adapting to ambient fluctuations through spatial and temporal transcriptional responses. We implemented the MS2-MCP system and combined it with microfluidics to visualize transcriptional regulation in living Arabidopsis plants. This enabled quantitative measurements of the transcriptional activity of single loci in single cells, real time and changing environmental conditions. Using phosphate responsive genes as model, we found that active genes displayed high transcription initiation rates (~3s) and frequently clustered together in endoreplicated cells. We observed gene bursting and large allelic differences in single cells, revealing that at steady-state, intrinsic noise dominated extrinsic variations. Moreover, we established that transcriptional repression triggered in roots by phosphate, a crucial macronutrient limiting plant development, occurred with fast kinetics (~minutes) and striking heterogeneity between neighboring cells. Access to single cell RNA polymerase II dynamics within live plants will benefit future studies of signaling processes.

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Coordination mechanisms to transport phosphate in marine diatoms

Marine diatoms drive global elemental cycles as a major oceanic primary producer. Dissolved inorganic P (Pi) is assimilated by photoautotrophs such as diatoms in the ocean, and a part of which is incorporated into ocean food web and transported to the land by activities of birds and anadromous fishes. Because of the non-volatility of Pi, photoautotrophs have an important role to transfer Pi to generate counter gravity movement. However, the molecular mechanisms of Pi assimilation in marine diatoms are still unclear. Uptake systems of Pi were characterized in two kinds of diatom, *Phaeodactylum tricornutum* and *Thalassiosira pseudonana*. Pi uptake rate of whole cell was increased in cells acclimated from Pi repletion to Pi limitation (-Pi), and the activity reached to the maximum in -Pi about 6 days. In the same transfer treatment to -Pi, external alkaline phosphatase activities were increasing after about 2 days of lag time and the activity keep increasing linearly over more than 7 days in both species. These suggest that Pi transportation across plasma membrane constitutes an initial rapid acclimation, which followed by a relatively slow and sustained a coordination system to hydrolyte external organic phosphate in marine diatoms.

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Epigenetic regulation of heat memory in *Arabidopsis*

Acclimation to high temperature increases plants' tolerance of subsequent lethal high temperatures. Although epigenetic regulation of plant gene expression is well studied, how plants maintain a memory of environmental changes over time remains unclear. I have found that JUMONJI (JMJ) proteins, demethylases involved in histone H3 lysine 27 trimethylation (H3K27me3), are necessary for *Arabidopsis thaliana* heat acclimation. Acclimation induces sustained H3K27me3 demethylation at *HEAT SHOCK PROTEIN22* (*HSP22*) and *HSP17.6C* loci by JMJs, poising the *HSP* genes for subsequent activation. Upon sensing heat after a 3-day interval, JMJs directly reactivate these *HSP* genes. Finally, *jmj* mutants fail to maintain heat memory under fluctuating field temperature conditions. In this short talk, I will present an epigenetic memory mechanism involving histone demethylases for environmental adaptation of field plants.