12th International PhD School PLANT DEVELOPMENT

2-4 October 2019, Retzbach, Germany

www.plant-development.org

Organising committee:

Angela Hay, MPIPZ, Cologne, Germany

Arp Schnittger, University of Hamburg, Germany

Moritz Nowack, VIB, Ghent, Belgium







Table of Contents

Thanks to Sponsors	•••••	3
Programme Overview	•••••	4
Detailed Programme	•••••	6
Abstracts of Oral Presentations	•••••	10
Abstracts of Poster Presentations	•••••	30
List of participants	••••••	64

Many Thanks to our Sponsors!





Programme Overview

Programme – October 2, 2019

Arrival and lunch	12:45 – 13:45
Opening remarks	13:45 – 14:00
Session 1: Evolution of development	14:00 – 15:05
Session 2: Patterning	15:05 – 16:10
Coffee break	16:10 – 16:40
Session 3: Plant signaling I	16:40 – 17:45
Session 4: Career development & industry	17:45 – 18:15
Dinner	18:30 – 19:30
Poster session 1 (Drinks can be purchased during the poster session)	19:30 – 21:30

Programme – October 3, 2019

Breakfast	08:00 - 09:00
Session 5: Flowering and dispersal	09:10 – 10:15
Coffee break	10:15 – 10:45
Session 6: Mechanics and morphogenesis	10:45 – 11:50
Conference photo	11:50 – 12:00
Lunch	12:00 – 13:00
Session 7: Root meristem	13:10 – 14:15
Session 8: Cell biology	14:15 – 15:20
Coffee break	15:20 – 15:50

Session 9: Plant signaling II	15:50 –16:55
Poster session 2	17:00 – 18:30
(Snacks are provided, drinks can be purchased during	the poster session)
Conference dinner and wine tasting, Würzburg	18:45 – 23:00

Programme – October 4, 2019

Breakfast	08:00 - 09:00
Session 10: Reproductive development	09:10 – 10:15
Coffee break	10:15 – 10:45
Session 11: Lateral root development	10:45 – 11:50
Awards and closing remarks	11:50 – 12:00
Lunch	12:00 – 13:00
Departure	13:00

Detailed Programme

Programme – October 2, 2019				
Arrival and lunch	12:45 – 13:45			
Opening remarks	13:45 - 14:00			
Session 1: Evolution of development Chair: Elizabeta Banic	14:00 – 15:05			
• Keynote: Edwige Moyroud, SLCU Cambridge, UK "Painting by numbers: Understanding the mechanisms of pe	14:00 – 14:35 etal patterning"			
 Roisin Fattorini, University of Cambridge, UK "The Evolution and Development of Petal Spots in a S Species" Thales Leandro, São Paulo State University, BR "Fusoid cells in the grass leaf (Poaceae): a putative developmental programmed cell death process" 	14:35 – 14:50 outh African Daisy 14:50 – 15:05			
Session 2: Patterning Chair: Moutasem Omary	15:05 – 16:10			
 Keynote: Ari Pekka Mähönen, University of Helsinki, Fl "Position and regulation of stem cells in vascular cambium" 	15:05 – 15:40			
• Dan Zhang, University of Heidelberg, DE "Deciphering the role of a novel player in grass subsidiary c	15:40 – 15:55 ell development"			
• Kevin Bellande, University of Montpellier, FR "Functional study of a lectin receptor kinase, LecRK-I.9: a control for cell wall dynamics in A. thaliana"	15:55 – 16:10			
Coffee break	16:10 - 16:40			
Session 3: Plant signalling I Chair: Hannah Walter	16:40 – 17:45			
• Keynote: Michael Hothorn, University of Geneva, CH "Mechanistic insights into small molecule and peptide hormone perception by plant membrane receptor kinases"	16:40 – 17:15			
• Ana Lopez Vazquez, University of Lausanne, CH "Molecular Determinants of Phytochrome Kinase"	17:15 – 17:30			
• Ajeet Chaudhary, TUM, DE "The receptor kinase STRUBBELIG promotes the response to cellulose deficiency in Arabidopsis thaliana"	17:30 – 17:45			

Session 4: Career development and industry Chair: Angela Hay	17:45 – 18:00
• Stefan Schwarz, KWS Saat Ag, Einbeck, DE "Seeding the future with KWS - Career opportunities at a leading plant breeding company"	17:45 – 18:00
Dinner	18:30 – 19:30
Poster session 1: Odd number posters (Drinks can be purchased during the poster session)	19:30 – 21:30
Program – October 3, 2019	
Breakfast	08:00 - 09:00
Session 5: Flowering and dispersal Chair: Luise Zuehl	09:10 – 10:15
 Keynote: Naomi Nakayama, Edinburgh University, UK "Informed dispersal? Environmental responses of seeds, fruits, and diaspores" 	09:10 – 09:45
• Miguel Perez, MPIPZ, Cologne, DE "Mechanism of localized lignin deposition in explosive fruit"	09:45 – 10:00
• Francesca Giaume, University of Milan, IT "A Triple Florigen System in Rice acts in leaves and at the shoot apical meristem"	10:00 – 10:15
Coffee break	10:15 – 10:45
Session 6: Mechanics and morphogenesis Chair: Rachele Tofanelli	10:45 – 11:50
• Keynote: Arun Sampathkumar, MPIMP, Golm, DE "Mechanical forces in the regulation of cell and tissue level morphogenesis in plants"	10:45 – 11:20
• Martha Thellmann, University of Zürich, CH "Understanding lateral root formation in Arabidopsis"	11:20 – 11:35
• Methawi Chomthong, University of Cambridge, UK "Revisiting stomatal mechanics: Towards an improved definition of stomatal sensitivity"	11:35 – 11:50
Lunch	12:00 - 13:00

18:45 - 23:00

13:10 - 14:15

•	Keynote: Sabrina Sabatini, Sapienza University Rome, IT "The dynamics of root growth"	13:10 – 13:45
•	<i>Shanda Liu, MPIPZ, Cologne, DE</i> <i>"A growth-dependent framework for Arabidopsis root zonation"</i>	13:45 – 14:00
Sessi Chair	on 8: Cell biology : Oscar Sanz Mora	14:15 – 15:20
•	Keynote: Niko Geldner, University of Lausanne, CH "The SCHENGEN pathway: How plants ensure functionality barriers"	14:15 – 14:50 of their protective
•	<i>Joris Jourquin, VIB, Ghent, BE</i> <i>"Identifying the transcriptional targets of GLV peptide signaling during lateral root initiation"</i>	14:50 – 15:05
•	<i>Martina Balboni, University of Hamburg, DE</i> "ASY1 recruitment to and release from the meiotic chromosic controlled by P31 ^{COMET} "	15:05 – 15:20 ome axis is
Coffee	e break	15:20 - 15:50
Sessi Chair	on 9: Plant signalling II : Yunjing Ma	15:50 –16:55
•	Keynote: Marie-Cécile Caillaud, RDP, Lyon, FR "Membrane Lipid and Cytoskeleton Interplay at the Cortex of	15:50 – 16:25 Dividing Plant Cell"
•	Andreas Schenke, Leibniz Institute, Halle, DE "Specificities of jasmonate signalling in Arabidopsis thaliana	16:25 – 16:40 "
•	<i>Khushbu Kumari, University of Würzburg, DE</i> <i>"Female gametophyte expressed Arabidopsis lipid transfer proteins LTP2/5 provide an unprecedented link between callose homeostasis, pollen tube guidance and fertilization success"</i>	16:40 – 16:55
Posto	r session 2: Even number nosters	17·00 – 18·30

Session 7: Root meristem

Wine tasting and dinner in Würzburg

Chair: Francesca Lopez

Program – October 4, 2019

Breakfast	08:00 - 09:00
Session 10: Reproductive development	09:10 – 10:15
Chair: Ana Alarcia-Garcia	
 Keynote: Cristina Ferrándiz, University of Valencia ES "A stylish story of carpel evolution" 	09:10 – 09:45
• Heather McLaughlin, John Innes Center, UK "Investigating crosstalk between the canonical and ETT- mediated auxin signalling pathways in the Arabidopsis thaliana gynoecium"	09:45 – 10:00
• Wei Wen Chong, University of Zürich, DE "Unravelling the imprinting mechanism of MEDEA by identifying cis-elements and trans-acting factors"	10:00 – 10:15
COFFEE BREAK	10:15 – 10:45
Session 11: Lateral root development	10:45 – 11:50
Chair: Liam German	
 Keynote: Alexis Maizel, University of Heidelberg, DE "Lateral root morphogenesis in Arabidopsis: from metabolis 	10:45 – 11:20 m to cytoskeleton"
• Stefania Morales, VIB, Ghent, BE "Trehalose 6-Phosphate, SnRK1 and TOR are part of a regulatory network to create a strategy to tolerate water- limiting conditions"	11:20 – 11:35
• Annika Kortz, University of Bonn, DE "Uncovering the transcriptomic control of heterosis manifestation during lateral root initiation in maize"	11:35 – 11:50
Awards and closing remarks	11:50 – 12:00
Lunch	12:00 – 13:00
Departure	13:00

Abstracts of Oral Presentations

The Evolution and Development of Petal Spots in a South African Daisy Species

ROISIN FATTORINI, BEVERLEY GLOVER

Department of Plant Sciences, University of Cambridge, Downing Street, Cambridge. CB2 3EA

The South African daisy species Gorteria diffusa is composed of several morphotypes which differ in floral morphology. The positioning and complexity of petal spots, defined as aggregations of pigmented cells in distinct petal regions, differ between morphotypes. Phenotypic differences in floral features are associated with differential pollinator behavioural responses and, as such, this variation may influence evolutionary trajectories contributing to divergence within G. diffusa. We are investigating the molecular development of these petal spots, characterising the genes that underlie G. diffusa petal spot pigmentation. Four homologous candidate genes encoding subgroup 6 MYB transcription factors have been identified. Three of these genes are upregulated within spotted petal tissue and MYB overexpression in tobacco induces ectopic anthocyanin production. The expression patterns of anthocyanin pathway enzymes DFR and ANS indicate that they may be downstream targets of MYB proteins, and biochemical assays are being used to investigate this. Having explored the role of each MYB homologue in spot development within the focal ecotype, gene function and expression patterns will be compared between several floral phenotypes associated with different pollinator behaviours. This study system provides a powerful comparative framework to investigate how MYB homologues contribute to the intraspecific variation of a functionally relevant trait. Ultimately, this will enhance understanding of molecular evolution and diversification within G. diffusa.

Fusoid cells in the grass leaf (Poaceae): a putative developmental programmed cell death process

Thales D. Leandro, Lynn G. Clark, Vera L. Scatena

Department of Botany, São Paulo State University – UNESP, Av. 24A, 1515, Rio Claro, SP 13506-900, Brazil

Department of Ecology, Evolution, and Organismal Biology, Iowa State University, 253 Bessey Hall, 2200 Osborn Dr, Ames, IA 50010-4009, U.S.A.

Fusoid cells have been reported in seven of 12 subfamilies of Poaceae and are recognized as a synapomorphy for the grass family, as well as a prominent mesophyll feature of the early-diverging lineages (Anomochlooideae, Pharoideae, and Puelioideae) and Bambusoideae. In grass leaf blades as seen in cross section, developmental studies reveal that a fusoid cell is usually a cavity resulting from the collapse of several fusoid cells. Using a comparative approach between leaf blade and leaf sheath development, leaves of *Pharus latifolius* (Pharoideae) were studied with light and transmission electron microscopes to investigate the early development of fusoid cells. In general, early developmental stages show fusoid cells increasing in size along with leaf differentiation. In leaf blades, TEM analysis reveals several nucleoli, internal divisions with initial formation of cell plates, but also vacuoles increasing in size with remains of membranes resulting from the lysis of organelles. A similar developmental pattern is observed in leaf sheaths. These characteristics, along with the gradual collapse of fusoid cells during leaf differentiation and enlargement, strongly suggests that they go through a programmed cell death process (PCD). Molecular studies are being carried out to investigate the putative PCD process in the development of fusoid cells. (Grant 2017/10739-4, São Paulo Research Foundation - FAPESP)

Deciphering the role of a novel player in grass subsidiary cell development

Dan Zhang, Tiago D. G. Nunes, Heike Lindner, Michael T. Raissig

Centre for Organismal Studies (COS), University of Heidelberg, Im Neuenheimer Feld 230, 69120 Heidelberg, Germany

Stomata are epidermal valves that are essential for plant survival because they control the entry of carbon dioxide assimilated in photosynthesis and optimize water use efficiency. Grasses like the model system *Brachypodium distachyon* form faster and, thus, more water-use-efficient stomatal complexes by adding two intimately connected, lateral subsidiary cells (SCs) to the central guard cells. We previously discovered that the mobile transcription factor *BdMUTE* specifies subsidiary cells in *Brachypodium*. The mutant *bdmute* fails to recruit SCs and forms two-celled, eudicot-like stomata instead that are slower to open and close and fail to open as wide as four-celled wild-type stomata. Transcriptome profiling of the developmental leaf zone of both wild-type and *bdmute* discovered genes absent or strongly downregulated in the mutant that are potentially associated with subsidiary cell formation. A gene of unknown function shows strong expression levels in wild-type but very low expression in *bdmute*. CRISPR-Cas9-edited mutant lines show misoriented or even absent SC division indicating a defect in polarizing the SC precursor cell. We hypothesize that this novel player participates in SC formation and, potentially, plays a role in setting up cell polarity during SC division.

Functional study of a lectin receptor kinase, LecRK-I.9: a control for cell wall dynamics in *A. thaliana*

Kevin BELLANDE¹ and Hervé CANUT²

¹UMR DIADE, Université de Montpellier, IRD, Montpellier, France

²UMR LRSV, Université de Toulouse, CNRS, UPS, Castanet-Tolosan, France

Plant cell walls are dynamic structures that are continuously modified in the course of development and in response to environmental cues: cell wall proteins play a primarily role by assembling and remodelling polysaccharides, and by participating to cell signalling. A sensory complex to monitor the cell wall status should provide this coordination in an effective manner. We are interested in an Arabidopsis thaliana lectin receptor kinase (LecRK-I.9) with a Legume lectin-type extracellular domain. It is hypothesized that LecRK-I.9 is part of this cell wall surveillance system. LecRK-I.9 expression was primarily found in root tissues. LecRK-I.9 was found to be associated to Hechtian strands particularly in the cell wall anchor points. LecRK-I.9 was shown to negatively regulate the processes of adventitious and lateral root initiation and emergence. Both processes require large cell wall remodelling. Genes encoding cell wall remodelling enzymes are up-regulated and lecrk-I.9 seedlings showed modified cell walls in their polysaccharide content. Cellulose biosynthesis inhibition was employed to impair the cell wall structures. We showed that LecRK-I.9 control the JA levels during the process of ectopic lignin deposition by regulating the expression of genes encoding cell wall proteins and peptides, but also proteins for detoxifying ROS. Our results suggest that LecRK-I.9 regulates cell wall dynamics in roots by targeting JA levels, ROS homeostasis and remodelling enzymes for polysaccharides. Future prospects include relationships between cell wall composition and mineral nutrition for iron. LecRK-I.9 could be the linker to establish a physical connection between cell wall and plasma membrane.

Poster Number: 3

Understanding and manipulating functional organisation in developing lateral root primordia of *A. thaliana*

Kevin BELLANDE, Mikaël Lucas, Laurent Laplaze and Soazig Guyomarc'h

UMR DIADE, Université de Montpellier, IRD, Montpellier, France

Root branching consists in the organogenesis of new lateral root primordia that acquire a new root apical meristem [1]. Using a system biology-based approach, we intend to decipher the genetics properties that lead to control and pattern the developing lateral root primordium in *A. thaliana* [2-3] and especially, root meristem establishment [4]. Based on the analysis of the inferred topology of the gene regulatory network operating during root branching, and on the characterization of a new meristem cell identity marker expressed during lateral root primordium development, we identified candidate genes possibly involved in the root meristem stem cell niche orgnisation of the lateral root primordium.

[1] Bellande et al. (2019) Trends in Plant Science. 24(9):826-839.

[2] Voß et al. (2015) Nat. Commun. 6, 7641.

[3] Lavenus et al. (2015) Plant Cell 27, 1368–1388.

[4] Goh et al. (2016) Development. 143, 3363-71.

Molecular Determinants of Phytochrome Kinase Substrate (PKS) Proteins Controlling Phototropism

Lopez-Vazquez A $^{[1]}$, Allenbach-Petrolati L $^{[1]}$, Fankhauser C $^{[1]}$

^[1] Center for Integrative Genomics, Faculty of Biology and Medicine, University of Lausanne, Genopode Building, 1015 Lausanne, Switzerland

Phototropism is one of the most noticeable events happening in response to the light, in which the plants orient their photosynthetic organs towards the light in order to increase light capture. This process arises from an increase in growth on the shaded side of the stem resulting from asymmetric auxin accumulation. This response is mainly controlled by phototropins (phots), which are serine/threonine protein kinases belonging to the AGC family that are associated to the plasma membrane (PM) and activated in response to blue light (BL). The phototropic curvature occurs because of asymmetric phot1 activation, however how a phot1-activation gradient across the stem leads to a lateral auxin gradient remains poorly understood. BL leads to phot1 dimerization, phosphorylation and translocation to functional membrane microdomains, where the signal transduction is activated. Phots form a protein complex with two families of PM-associated proteins: NRL and PKS. In Arabidopsis thaliana the PKS family is constituted by 4 members (PKS1-4), among which PKS4 is the most important for phototropism. PKS are intrinsically disordered proteins found in all angiosperms. Despite low overall similarity this family is characterized by 6 short regions of homology that we call A to F. Here, we mutated three of these regions of PKS4 to test their requirement for phototropism and we found that two of these regions are required for PKS4 function. To address the molecular functions of these regions, we plan analyzing their importance for the phot1 protein complex formation and activation, and also their requirement for the interaction with PKS binding proteins.

The receptor kinase STRUBBELIG promotes the response to cellulose deficiency in *Arabidopsis thaliana*

Ajeet Chaudhary¹, Xia Chen¹, Jin Gao¹, Barbara Leśniewska¹, Sebastian Wolf², Corinna Dawid³, and Kay Schneitz^{1*}

¹Entwicklungsbiologie der Pflanzen, Wissenschaftszentrum Weihenstephan, Technische Universität München, Freising, Germany

²Cell wall signaling group, Centre for Organismal Studies, University of Heidelberg, Heidelberg, Germany

³Lehrstuhl für Lebensmittelchemie und molekulare Sensorik, Wissenschaftszentrum Weihenstephan, Technische Universität München, Freising, Germany

Cell wall remodeling is essential for the orchestration of cell division and growth patterns during plant development. In addition, alterations in the cell wall play a pivotal role in cellular and tissue responses to abiotic and biotic stresses. The necessary cell wall monitoring mechanisms remain poorly understood. In Arabidopsis previous data suggested that the receptor kinase STRUBBELIG (SUB) and the C2 domain protein QUIRKY (QKY) form a complex at plasmodesmata involved in the control of tissue morphogenesis. We show that SUB and QKY also promote stress gene induction, jasmonic acid production and ectopic lignin accumulation either upon exogenous application of the cellulose biosynthesis inhibitor isoxaben or in plants with a genetic defect in the cellulose synthase complex. Moreover, our data reveal that SUB signaling is required for the recovery of root growth after transient exposure to isoxaben. Genetic data further indicate that SUB promotes the isoxaben-induced cell wall stress response independently from other known receptor kinase genes mediating this response, such as THESEUS1 or MIK2. We also show that cell wall stress eventually attenuates SUB activity by a posttranscriptional mechanism. The observed downregulation of SUB correlates with the finding that treating wild-type plants with sub-lethal doses of isoxaben results in roots and floral organs that exhibit a sub-like phenocopy. Our combined data reveal a novel role for SUB signaling in the response to cell wall damage induced by a reduction in cellulose biosynthesis. Thus, we propose SUB to function in a least two distinct biological processes: the control of tissue morphogenesis and the cell wall stress response. Moreover, SUB appears to be subject to the control mechanism coordinating growth and stress responses. Finally, our data establish plasmodesmata as important organelles involved in the cell wall stress response.

Mechanism of localized lignin deposition in explosive fruit

Miguel Pérez-Antón¹, Hugo Hofhuis¹, Patrizia Kroll¹, Sabine Metzger² and Angela Hay¹

¹Max Planck Institute for Plant Breeding Research, Cologne, Germany ²Mass Spectrometry Platform, University of Cologne, Cologne, Germany

A major goal in biology is to identify the molecular basis of complex trait innovations, such as explosive seed dispersal, found in Cardamine hirsuta. The asymmetric deposition and lignification of secondary cell walls (SCWs) in endocarp b cells of the fruit valve was found to be a key morphomechanical innovation underpinning explosive dispersal. To gain insights into the mechanisms controlling this lignin patterning in C. hirsuta, we identified less lignin (lig) mutants that showed reduced lignification of the fruit valve. The *lig1* mutant is deficient in lignin deposition in endocarp b cells, resulting in a reduced seed dispersal range. By positional cloning, complementation, and the isolation of additional alleles, we demonstrate that an ortholog of the transcription factor SPL7, a central regulator of copper homeostasis, is the causal locus for the phenotype. Using ICP-MS to measure the concentration of copper, we show that SPL7 is both necessary and sufficient to regulate the concentration of copper in fruit. We find that SPL7 is expressed in endocarp b cells and lignified cells of the dehiscence zone, suggesting a local role in lignification. Three members of the lignin-polymerizing laccases, which are Cu-requiring enzymes, are expressed in endocarp b cells, where we show they precisely co-localize with lignin in the asymmetric SCWs. We propose that copper deficiency in the *lig1* fruit, due to loss of SPL7 function, reduces laccase activity in endocarp b cells, resulting in reduced lignification. This provides a testable hypothesis for the control of localized lignin deposition in explosive fruit.

A Triple Florigen System in Rice acts in leaves and at the shoot apical meristem

<u>Francesca Giaume</u>¹, Damiano Martignago^{1,3}, Micol Aldrovandi¹, Vittoria Brambilla^{1,2}, Martina Cerise¹, Fabio Fornara¹

¹ University of Milan, Department of Biosciences, 20133 Milano, Italy

² University of Milan, Department of Agricultural and Enviromental Sciences, 20133 Milano, Italy

³ Centre of Research in Agricultural Genomics -CRAG, Università Autonoma di Barcellona, Spain

E-mail contacts: francesca.giaume@unimi.it, fabio.fornara@unimi.it

The transition from a vegetative stage to a reproductive one in rice (Oryza sativa) occurs upon expression of two essential genes encoding the florigens, belonging to the phosphatidyl ethanolamine binding protein family. These are HEADING DATE 3a and RICE FLOWERING LOCUS T 1, which are transcribed and translated in the leaf and then move through the phloem until they reach the shoot apical meristem (SAM). Here they bind to a 14-3-3 protein and to a bZIP transcription factor to form the Florigen Activation Complex (FAC) that can induce conversion of the shoot apex into a panicle by promoting expression of panicle-identity genes. A transcriptional screen at the rice's SAM, identified a third phosphatidyl ethanolamine binding gene, FLOWERING LOCUS T-LIKE 1 (FT-L1), whose expression is strongly induced at the SAM during floral commitment.

I have then isolated two different mutant alleles, and their flowering time was measured under different day-length conditions, demonstrating that FT-L1 has floral promoting activity. Expression data indicated that its mRNA is mainly expressed in the SAM rather than in the leaves. Analysis of interaction patterns with FAC components further indicated that FT-L1 doesn't interact with any component of the florigen complex. Finally, detailed phenotypic analysis showed that ft-l1 mutant has panicles with a higher number of secondary branches and seeds.

My data indicate FT-L1 doesn't show the characteristics of typical florigens, and the aim of my PhD work is to define its function, its position in the flowering pathway and to discover how it regulates panicle architecture.

Session: 6, Abstract: 9

Understanding lateral root formation in Arabidopsis.

Martha Thellmann and Joop EM Vermeer

Department of Plant and Microbial Biology, University of Zürich, Switzerland

Intercellular communication is central for the development of an organism. It is mediated by biochemical gradients as well as physical forces that collectively regulate differentiation and development. Lateral root formation is a developmental process in which the integration of chemical signals and physical forces is evident. During its development, the lateral root heavily depends on spatial accommodating responses of overlaying cell layers. We have shown that pericycle cells need to swell in order to undergo formative divisions, whereas the overlying endodermis undergoes a dramatic volume loss during lateral root formation. Through manipulating SHY2-mediated auxin signaling in the endodermis, we were able to completely block cell proliferation in the pericycle. It appears that the pericycle perceives this non-accommodating endodermis as an increased resistance to its expansion growth. The pericycle-endodermis interaction now provides a unique opportunity to elucidate the molecular and cellular mechanisms underlying the interplay between cell volume regulation and mechanosensing during plant development. To that end, a large transcriptomic study has been conducted and will now help to identify candidate genes, which are orchestrating lateral root initiation and development.

Revisiting stomatal mechanics: Towards an improved definition of stomatal sensitivity

Methawi Chomthong and Howard Griffiths

Department of Plant Sciences, University of Cambridge

The dynamic responses of stomata to transient environmental conditions are considered highly relevant for maximising crop productivity in a changeable climate. Recent approaches have been framed by the concepts associated with the loss of carbon (due to a slow opening responses) or loss of additional water (due to a tardy closing responses). Whilst dynamic models help to capture such transients, we propose that consideration should also be given to the contribution that stomatal mechanics make to overall responsiveness. We have combined two traditional approaches to explore the extent that stomatal complex ultrastructure and stomatal pore geometry could contribute to improving the responsiveness of stomata.

Firstly, the "*Mechanical Advantage*" concept governs the speed of stomatal movement as a function of the relative turgor pressure in adjacent cells. Here, guard cell geometry significantly influences the rapidity of stomatal movement, and a simple equation has been developed to show that the mechanical advantage effect relates well with experimental pressure probe data.

Secondly, the "Bent Beam" framework suggests that larger stomata can adopt a higher dynamic range of apertures whilst experiencing lower shear stress than smaller stomata. A model has been developed to compare data from stomata comprising dumbbell- and kidney-shaped guard cells. The results are consistent with the contrasting opening mechanics and the interplay with specialised adjacent epidermal cells (known as subsidiary cells) and their associated reciprocal turgor pressure adjustment processes (hereafter defined as "solute coupling").

By unifying Mechanical Advantage and Bent Beam concepts into a new stomatal mechanics framework, a generalised dynamic model of stomatal sensitivity has been developed. Furthermore, we suggest that recent steady-state and dynamic stomatal models will be improved by incorporating stomatal mechanics, as well as associated osmotic and biophysical drivers of stomatal sensitivity. Ultimately, this opinion piece emphasises the significance of stomatal mechanics as part of the holistic view of stomatal responsiveness (Bertolino *et al.*, 2019; Yi *et al.*, 2019).

A growth dependent framework for Arabidopsis root zonation

Shanda Liu¹, Milad Adibi¹, Raffaele Dello Ioio², Richard Smith¹, Miltos Tsiantis¹

¹Department of Comparative Development and Genetics, Max Planck Institute for Plant Breeding Research, Cologne, Germany

²Department of Biology and Biotechnology "Charles Darwin" BBCD, Sapienza University of Rome, Rome, Italy

As a model organism for root study, Arabidopsis root is widely used for investigating root development, patterning and growth. The classical root longitudinal zonation pattern divides Arabidopsis root tip into three zones: the meristem, the elongation zone, and the differentiation zone. However, there is no general consensus about the boundaries between these zones, meaning that data reported by different groups regarding root zonation effect are not fully comparable. To describe root zonation and identify boundaries in a more objective way, we developed a growth dependent approach to define root zonation pattern by combining a microfluidic device, confocal live imaging, and MorphoGraphX image processing software. This framework can help to measure cellular growth distribution along the longitudinal axis, which provides a quantitative and dynamic way for defining root zonation. On the other hand, the microfluidic device within the system allows to test cellular growth and root zonation dynamics at high spatial and temporal resolution in response to different hormones, which will give us a better understanding of the cellular and molecular mechanisms that underlie root zonation organization.

Identifying the transcriptional targets of GLV peptide signaling during lateral root initiation

Joris Jourquin^{1,2}, Ana Fernandez^{1,2}, Boris Parizot^{1,2}, Nick Vangheluwe^{1,2}, Ke Xu^{1,2} and Tom Beeckman^{1,2}

¹ VIB-UGent Center for Plant Systems Biology, VIB, B-9052 Ghent, Belgium

² Department of Plant Biotechnology and Bioinformatics, Ghent University, B-9052 Ghent, Belgium

Communication between neighbouring cells is a prerequisite for proper cell patterning during organ development. Signaling peptides play a major role as molecular messengers in numerous plant developmental processes, including lateral root (LR) development. Two members of the GOLVEN peptide family in Arabidopsis, GLV6 and GLV10, are expessed in lateral root founder cells (LRFCs) at the onset of LR initiation. While GLV6 overexpression disrupts the essential asymmetry of the first anticlinal cell division, thereby inhibiting further LR organogenesis, higher order glv mutants show an increase in LR initiation events, suggesting that GLV6/10 peptides function as regulators of asymmetric cell divisions in the pericycle. Previous work in our lab identified RGI1 and RGI5 as the main transmembrane receptors responsible for GLV peptide perception during LR initiation and found MPK6 as a component of the downstream phosphorylation cascade. However, the targets of this pathway, which are responsible for the GLV-induced effects, have yet to be discovered. To understand how GLV peptides regulate early LRFC divisions, we characterized the transcriptional response triggered upon GLV6 perception, specifically during LR initiation. Using RNA-sequencing, several putative GLV6 target genes were identified, including some well-known players in LR development as well as genes for which a role in this process has not yet been described. The ongoing characterization of these genes will allow us to incorporate the GLV signaling pathway into the current models of LR development and has the potential to result in the identification of new genes with an important role in this process.

ASY1 recruitment to and release from the meiotic chromosome axis is controlled by P31^{COMET}

Martina Balboni¹, Chao Yang¹, Shinichiro Komaki^{1,2} and Arp Schnittger¹

¹ Department of Developmental Biology, University of Hamburg, 22609 Hamburg, Germany

² Nara Institute of Science and Technology, Graduate School of Biological Sciences, 8916-5 Takayama, Ikoma, Nara 630-0192, Japan

The chromosome axis plays a key role in chromosome organization and recombination during meiosis. An important component of the chromosome axis is the HORMA domain protein ASY1, known as Hop1 in yeast and mammals. In early meiosis, ASY1 is loaded onto chromosomes. During zygotene, when the chromosome axes of each homolog pair starts to be integrated into the synaptonemal complex (SC) as lateral element, ASY1 is depleted and the transverse filaments are deposited to reach full synapsis between the homologous chromosomes by the end of pachytene. While the regulated assembly of HORMA domain protein complexes has been extensively characterized, the mechanisms underlying their disassembly are still largely unknown. Here, we have identified P31^{COMET}, well known for its function in the mitotic spindle assembly check-point (SAC), as a central regulator of the meiotic HORMA domain protein ASY1. We find that P31^{COMET} directly interacts with ASY1 and recruits the AAA+ ATPase PCH2, an ATP hydrolase able to promote conformational changes in target proteins, to ASY1. PCH2 then disassembles the HORMA domain-closure motif complexes with P31^{COMET} being an indispensable adaptor of PCH2 for HORMA recognition and remodeling. Thus, a similar molecular module appears to regulate the disassembly of HORMA domain proteins during SAC function and the formation of the SC. Furthermore, we find that ASY1 is not efficiently targeted to the nuclei of meiocytes in the absence of P31^{COMET}. This indicates an early role of PCH2 and P31^{COMET} in promoting axis formation possibly by making ASY1 mobile and/or available for axis formation.

Specificities of jasmonate signalling in Arabidopsis thaliana

Andreas Schenke, Stefan Mielke, Adina Schulze, Marlene Zimmer, Debora Gasperini

Department of Molecular Signal Processing, Leibniz Institute of Plant Biochemistry, Halle, Germany

The phytohormone Jasmonate-Isoleucine (JA) regulates a wide range of physiological events from abiotic and biotic stresses to plant growth and reproductive development. Although the molecular mechanisms of JA signalling are well characterized, it remains unclear how can a single molecule regulate the wide-range of processes observed for JA-Ile responses. Part of this specificity may rely upon the JASMONATE-ZIM DOMAIN (JAZ) protein family. Under basal conditions JAZ proteins inhibit the activity of JA-dependent transcription factors (TFs), and hence, the expression of JA responsive genes. When JA-Ile levels rise, JAZ repressors interact with the co-receptor CORONATINE INSENSITIVE 1 (COI1), resulting in JAZ degradation and the liberation of TFs to initiate JA responses. Arabidopsis thaliana encodes for 13 different JAZ proteins, most of them exhibiting different splicing variants. Thus, JAZ protein diversity could be a pivotal parameter for modulating JA signalling. We therefore aim to determine the specific roles of individual JAZ proteins in the regulation of JA signalling by investigating their protein expression, their degradation in different cell types, their knock-out phenotypes, as well as JAZ-COI1 binding affinities. Our preliminary data revealed cell-specific expression patterns of different JAZ transcriptional and translational reporters, supporting the hypothesis that different JAZ proteins modulate specific JA responses. Furthermore, knock-out mutants in specific JAZ genes displayed root growth phenotypes under basal conditions and in response to JA treatment. Findings from this project will increase our basic understanding of JA signalling and potentially uncover cellspecific defence strategies that could be eventually used in breeding programs for crop improvement.

Session: 9, Abstract: 15, Poster Number: 12

Female gametophyte expressed *Arabidopsis* lipid transfer proteins LTP2/5 provide an unprecedented link between callose homeostasis, pollen tube guidance and fertilization success.

Khushbu Kumari¹, Meng Zhao², Rosalia Deeken¹, Dirk Becker^{1,#}

¹ Institute for Molecular Plant Physiology and Biophysics, University of Würzburg, Julius-von-Sachs Platz 2, 97082, Würzburg, Germany.

² Life science college, Shanxi Normal University, Linfen, Shanxi Province, China

[#] to whom correspondence should be addressed (dirk.becker@uni-wuerzburg.de)

The fertilization process of flowering plants (angiosperms) is highly complex and demands an intricate communication between cells of the two haploid gametophytes involving the polar growing pollen tube (carrying the two non-motile sperm cells) and the ovule (hosting the egg cell/synergid cells) to provide for successful reproduction. The polar path of the pollen tube towards the target female gamete is guided by different signaling molecules, including sugars, amino acids and peptides. Depending on the plant species several lines of evidence have also suggested potential roles for non-specific lipid transfer proteins (nsLTPs) LTPs during pollen germination or pollen-tube guidance. Although *Arabidopsis thaliana* has 49 *nsLTP* genes, several of which are involved in plant immunity and cell-to-cell communication, the role of most members of this family during fertilization is unknown.

Hypothesizing that extracellular LTPs play an important role during fertilization, here we have investigated the role of selected, female gametophyte expressed nsLTPs from *Arabidopsis thaliana*. Using reporter-based transcriptional and translational fusions we determined in planta the temporal and spatial expression patterns together with protein localizations for LTP2, 3, 4, 5 6, and 12. While all selected LTPs where expressed among others in the stylar region, they split into two categories with respect to protein localization – either plasma membrane or cell wall. Focusing on extracellular LTPs, we show that CRISPR/Cas9-mediated knock-out of LTP2/LTP5 results in significantly reduced fertilization success. Cell biological analyses revealed that the *ltp2/5* double mutant was impaired in pollen tube guidance towards the ovules and that this phenotype correlated with aberrant callose depositions in the micropylar region during ovule development.

Investigating crosstalk between the canonical and ETTmediated auxin signalling pathways in the *Arabidopsis thaliana* gynoecium.

Heather McLaughlin, Lars Østergaard

Department of Crop Genetics, John Innes Centre, Norwich, UK

In the canonical model of auxin signalling, transcription factors known as Auxin Response Factors (ARFs) are bound by AUX/IAA repressors in the absence of auxin, preventing the modulation of auxin responsive genes. When the concentration of auxin rises, AUX/IAA repressors interact with TIR1/AFB auxin receptor complexes and are subsequently ubiquitinated and degraded, allowing ARFs to regulate gene expression. Auxin sensitivity in this system is conveyed through the binding of AUX/IAA repressors to ARFs in the absence of auxin, however, many ARFs have a limited capacity to interact with these repressors, and an atypical ARF known as ETTIN (ETT) lacks the C-terminal PB1 domain required for this interaction. ETT interacts with a wide range of transcription factors and chromatin modifying proteins in an auxin sensitive manner, and it has recently been shown that ETT target genes are upregulated in response to auxin independently of the canonical pathway. Auxin homeostasis is tightly regulated throughout plant growth and development, and so crosstalk between the canonical and ETT-mediated auxin signalling pathways is likely, although no mechanisms have been established to date. ARFs are modular transcription factors containing an N-terminal DNA Binding Domain (DBD), flanked by two Dimerization Domains (DDs) which allow ARF homodimerization and the binding of Auxin Responsive Elements in the promoters of downstream targets. The DBD and DDs are highly conserved even between divergent ARFs, and we hypothesize that crosstalk may occur between the canonical and ETT-mediated auxin signalling pathways through ETT: ARF heterodimerization via the DBD.

Unravelling the imprinting mechanism of *MEDEA* by identifying *cis*-elements and *trans*-acting factors

Wei Wen Chong, Ueli Grossniklaus

Department of Plant and Microbial Biology, University of Zürich, Zollikerstrasse 107, CH-8008 Zürich

Genomic imprinting is a type of epigenetic gene regulation that occurs in flowering plants and mammals and results in allele-specific expression depending on the parent of origin. In *Arabidopsis*, DNA methylation and histone modifications by *Polycomb* Repressive Complex 2 (PRC2) have been implicated in the regulation of genomic imprinting but many imprinted genes are regulated by other factors. Wöhrmann et al. (2012) identified an imprinting control region (ICR) localised in the 250-bp *cis*-regulatory region of the maternally expressed imprinted gene *MEDEA* (*MEA*). This ICR alone is sufficient to mediate *MEA* transcriptional activation and imprinted expression, but this activity neither depends on DNA methylation nor PRC2, implicating other, yet unknown factors. This first aim of this project is to identify the exact sequence of the ICR in the 250-bp *MEA* promoter fragment using directed CRISPR/Cas9 mutagenesis. We will screen for mutants with a 50% *mea*-like seed abortion phenotype, indicating a mutation in the *MEA*-ICR. The second aim of this project is to identify novel factors that regulate *MEA* imprinted expression by binding to the *MEA*-ICR. Candidates have previously been isolated in a yeast 1-hybrid screen and electrophoretic mobility shift assays will be used to determine whether the candidate protein interacts with the 250-bp region *in vitro*. Subsequently, the binding of promising candidates will be checked *in vivo* using chromatin immunoprecipitation assays.

Trehalose 6-Phosphate, SnRK1 and TOR are part of a regulatory network to create a strategy to tolerate water-limiting conditions

Stefania Morales ^{1,2,3,4}, Patrick Van Dijck ^{3,4}, Tom Beeckman ^{1,2}

¹ Ghent University, Department of Plant Biotechnology and Bioinformatics, Belgium

- ² VIB Center for Plant Systems Biology, Belgium
- ³ Laboratory of Molecular Cell Biology, KU Leuven, Belgium
- ⁴ VIB Center for Microbiology, Belgium

Plants have diverse adaptive strategies to tolerate water-limiting conditions, like the closing of stomata in response to decreasing water potential to avoid water loss and the increment of root length to search for deeper water resources in the soil. Trehalose 6-Phosphate (T6P) has been proposed to act as a plant growth-regulator and was shown to increase stress tolerance. Preliminary data from our laboratories suggest a role for T6P in stomatal conductance and root growth. Therefore, our main goal is study T6P's role in lateral root development and stomatal conductance and the putative connection with the sensor kinases SnRK1 and TOR. Through the study of trehalose metabolism gain-of-function or loss-of-function mutants, and by treatments with T6P and the T6P analog (DNMB-T6P) we established that the increase of T6P in planta promotes lateral root formation. Moreover, plants adapting to adverse environmental conditions, can activate protein kinase complexes like SnRK1 and TOR. Interestingly, there are evidences that SnRK1 activity is inhibited by T6P and different publications showed a link between SnRK1 and TOR in diverse developmental processes. We found that lateral root development is promoted in low SnRK1 activity conditions and high TOR activity conditions. In addition, expression analyses of SnRK1-target genes corroborated that T6P is able to inhibit SnRK1 activity in planta. We are studying TOR activity to find a direct link with this growth regulator. We will perform metabolic profiling, stomatal conductance measurements and phospho-proteomics to generate a comprehensive model that can explain the adaptive strategies to tolerate drought in Arabidopsis.

Uncovering the transcriptomic control of heterosis manifestation during lateral root initiation in maize

Annika Kortz, Peng Yu, Frank Hochholdinger

INRES Crop Functional Genomics, University of Bonn, Bonn, Germany

Heterosis is the superior performance of hybrid offspring compared to the average of its parents. In plants, heterosis is observed for yield and plant height, but also for traits of the root systems. In young maize seedlings, lateral root density (LRD) displays the highest degree of heterosis among studied root traits. LRD has an impact on nutrient and water acquisition efficiency. The transcriptomic regulation underlying the heterosis manifestation of LRD in maize remains largely unknown. In this study we compare recombinant inbred lines with contrasting LRD and their backcrosses into the founder lines Mo17 and B73. While RNA-Seq of total root samples enables the identification of active genes, important candidate genes for lateral root formation are potentially masked by expression patterns of tissues not involved in the lateral root development. The application of laser capture microdissection of a specific cell type in combination with RNA-Seq allows studying transcriptomic changes related to heterosis manifestation in pericycle cells which give rise to lateral roots. This strategy enables the identification of heterosis-associated gene expression patterns such as single parent expression in specific cell types. By combining the determination of auxin concentrations, DNA content and transcriptome activity, gene regulation network analysis will facilitate the identification of candidate genes involved in lateral root initiation in hybrids, which could help to optimize the maize root system for future climatic challenges.

Abstracts of Poster Presentations

Subsidiary Cells: the right-hand of guard cells for improved stomatal movements in grasses

Tiago D. G. Nunes, Dan Zhang, Heike Lindner, Michael T. Raissig

Centre for Organismal Studies (COS), University of Heidelberg, Im Neuenheimer Feld 230, 69120 Heidelberg, Germany

Stomata are cellular gateways on leaves that allow gas-exchange and balance carbon dioxide uptake and water loss. Grasses, such as the important food crops maize and wheat, present physiological improved stomata with four cells contrasting the typical two guard-cell morphology in most of plants. Grasses recruit two lateral subsidiary cells (SCs), which is triggered by the mobile bHLH transcription factor BdMUTE in the model grass Brachypodium distachyon. The Arabidopsis orthologue AtMUTE is non-mobile and specifies guard mother cell identity instead. It is unknown if BdMUTE mobility is required for SCs recruitment nor which motifs within MUTE are responsible for BdMUTE mobility and functionality. Generally, MUTE orthologues are well conserved but some differences between the grass and non-grass peptides were identified particularly within MUTE-specific and SMF domains. To identify required domains or motifs for grass-specific function and mobility, domain swaps between BdMUTE and AtMUTE will be performed and reporter chimeric proteins expressed. To study the contribution of SCs to stomatal gas exchange efficiency in grasses, a transcriptome analysis between wt and bdmute mutant mature leaves (with and without subsidiary cells respectively) identified differentially expressed candidate genes potentially associated with SCs function. Reverse genetic approaches (CRISPR-based gene editing and use of available mutant collections), reporters and/or overexpression constructs combined with infrared gas-exchange analysis are used to determine the candidate genes' role in SC function and contribution to stomatal physiology. We hope to better understand how SCs form and contribute to improved gas exchange efficiency and plant performance observed in the grass family.

The role of DEVIL peptides in controlling cellular proliferation and plant morphogenesis

Ana Alarcia-García, Elena Perpiñan, Priscilla Rossetto, Amparo Primo, Cristina Ferrándiz

Instituto de Biología Molecular y Celular de Plantas, Consejo Superior de Investigaciones Científicas—Universidad Politécnica de Valencia, 46022 Valencia, Spain.

DEVIL (DVL) peptides have been identified in Arabidopsis. These are small size and unknown function peptides, encoded by a gene family of 24 members and are located in the plasma membrane. The *DVL* genes were characterized a decade ago by their overexpression phenotypes, which dramatically affect fruit morphology (Wen *et al.* 2004; Narita *et al.* 2004; Guo *et al.* 2015). However, the loss-of-function mutants do not have any obvious morphological phenotype, so the biological function of these peptides is essentially unknown.

In the lab we have carried out different studies to characterize how DVL peptides interact with the plasma membrane, how are they expressed and accumulate, identify other proteins that interact physically with these peptides, and whether the combination of multiple loss-of-function mutants affect development. Our preliminary results suggest that DVL peptides appear to be mobile signals not integrated in the plasma membrane, and to participate in the control of cell cycle and cell division processes. We are currently extending our work in these directions, generating combinations of multiple mutants in different *DVL* genes with high sequence similarity (*dvl8 dvl11 rtfl9, dvl8 dvl11 rtfl11,* and *dvl8 dvl11 rtfl9 rtfl11*). These combinations do not show obvious loss of function phenotypes, thus supporting the idea of high gene redundancy among members of *DVL* family (Wen *et al.* 2004), and that higher order combinations must be required to observe a morphological phenotype. Therefore, we have also used CRISPR/Cas9 technology (Wang *et al.* 2015) to mutate additional *DVL* genes to include in this study.

In addition, to confirm and better understand the differences in spatial distribution of mRNA and peptides for DVL1 and DVL8, we have generated pDVLxDVLx:GUS or pDVLx::DVLx:GFP and promoter:reporter lines to try to determine whether this differential distribution is due to peptide active transport. In addition, we are starting to functionally validate the possible protein interactions uncovered in a yeast-2-hybrid screening. These and other preliminary results will be presented, along with possible future directions.

Narita NN, Moore S, Horiguchi G, Kubo M, Demura T, Fukuda H, Goodrich J, and Tsukaya H. (2004) Overexpression of a novel small peptide Rotundifolia4 decreases cell proliferation and alters leaf shape in Arabidopsis thaliana. Plant J. 38, 699-713.

Wang Z, Xing H, Dong L, Zhang H, Han C, Wang X and Chen Q. (2015) Egg cell-specific promoter-controlled CRISPR/Cas9 efficiently generates homozygous mutants for multiple target genes in Arabidopsis in a single generation. Genome Biology 16,144.

Wen J, Lease KA and Walker, J.C. (2004) DVL, a novel class of small polypeptides: overexpression alters Arabidopsis development. Plant J, 37, 668-677.

Guo P, Yoshimura A, Ishikawa N, Yamaguchi T, Guo Y, and Tsukaya H. (2015) Comparative analysis of the RTFL peptide family on the control of plant organogenesis. J. Plant Res. 128, 497-510.

The SnRK1-C/S₁-bZIP network links energy homeostasis to lateral root development

Prathibha Muralidhara, Christoph Weiste, Wolfgang Droege-Laser

University of Würzburg, Department of Pharmaceutical Biology, Julius-von-Sachs Platz 2, 97082 Würzburg

Maintaining energy homeostasis is of crucial importance for all living organisms. This is especially important for plants, as they encounter many environmental challenges due to their sessile lifestyle. Therefore, plants modulate and fine-tune their growth and metabolism depending on the available resources. Here we report that evolutionarily conserved SUCROSE NON-FERMENTING1 RELATED KINASES1 and their downstream C/S_1 basic leucine zipper (bZIP) transcription factors control root architecture and specifically lateral root (LR) development under energy limiting conditions. To disclose the molecular mechanism of SnRK1-C/S1-bZIP signaling in modulating LR development, we employ reverse genetics, confocal microscopy and high-throughput sequencing platforms. Upon shortterm unexpected darkness, extended night or low light-treatments, wild-type seedlings display increased LR density, which is diminished in snrk1 and group C bZIP loss-of-function lines. Accordingly, bZIP and SnRK1 expression was shown to be specifically localized to defined stages of LR development. Differential Interference Contrast (DIC) imaging of wild-type and confocal imaging of the lateral root primordia marker GATA23:GFP, disclosed an increased number of LR primordia upon energy limiting conditions. As identified by ChIPseq (Chromatin immunoprecipitation sequencing), group C bZIPs bind promoters of several target genes, known to be central in LR initiation. Overall, our work proposes a novel role of the low-energy activated SnRK1-C/S1-bZIP network in modulating LR development.

CRISPR-CAS9 MUTAGENESIS UNRAVELS THE ROLE OF 45S RDNA GENES IN *A. THALIANA* DEVELOPMENT

<u>Francesca Lopez¹</u>, Antoine Fort^{1,2}, James Friel¹, Aline Probst³, Frédéric Pontvianne⁴, Ronan Sulpice² and Charles Spillane¹

¹ Genetics and Biotechnology Laboratory, Plant and AgriBiosciences Research Centre (PABC), School of Natural Sciences, National University of Ireland Galway, Ireland

² Systems Biology Laboratory, Plant and AgriBiosciences Research Centre (PABC), School of Natural Sciences, National University of Ireland Galway, Ireland

³ GReD, Universite Clermont Auvergne, CNRS, INSERM, BP 38, 63001 Clermont–Ferrand, France

⁴ CNRS, Laboratoire Génome et Développement des Plantes (LGDP), Perpignan, France; Université de Perpignan Via Domitia, LGDP, Perpignan, France

Although 45s rDNA genes are ubiquitous to all eukaryotes, the number of repeats and their chromosomal locations are highly polymorphic across different phyla. The role of transcriptionally active and inactive 45s copies has been amply characterized in cellular biology by numerous studies linking loss of 45s copies to abnormal proliferation and cancer (Xu et al., 2017), and also to genome integrity and senescence (Obayashi and Kobayashi, 2014).

In plants however, it hasn't yet been possible to formulate a direct link between variation in 45s copy number and their function in development. Previous studies employed the Chromatin Assembly Factor-1 (*CAF-1*) mutants which are knock-out alleles of histone chaperones *FASCIATA 1/2*, which lead to a drastic reduction in rDNA copy number (Kaya et al., 2001; Mozgova et al., 2010), however it has not yet been possible to draw a causative association between the phenotypes observed and the reduction of copy number. Here, we show how CRISPR-Cas9 mutagenesis has been employed to generate *A. thaliana* plants with a stably inherited reduction of up to 80% of 45s rDNA genes. We report that decrease of 45s copies is linked with aberrant seedling and seed development, which is also associated with deregulated mechanisms of cell proliferation and meristematic activity.

Currently we are investigating potential chromatin rearrangements which may have been caused by the drastic reduction of rDNA Copy Number, as well as mechanisms that regulate transcription of rDNA genes which in plants remain uncharacterised.

Cell-Division Plane Orientation During Symmetry Establishment

Billy Tasker-Brown, Laila Moubayidin and Lars Østergaard

John Innes Centre, Norwich Research Park, Norwich, NR4 7UH, United Kingdom

During *Arabidosis* gynoecium development, the apical part undergoes an unusual symmetry transition from being bilaterally symmetric to form the radially symmetric style. Failure to establish radial symmetry in the style seriously compromises fertility. Two transcription factors, SPATULA (SPT) and INDEHISCENT (IND) are centrally important in mediating this transition and they do so both by tight regulation of auxin distribution and the expression of genes involved in cell-cycle control. Using the *Arabidopsis* gynoecium as a model system, my research project is focussed on understanding the role of cell-division plane orientation during organogenesis. I hypothesise that transverse anticlinal cell division in a pool of auxin-accumulating cells that form in the medial part of the gynoecium apex is required for this bilateral-to-radial symmetry transition. My results show that in a *spt* mutant background, which lacks the apical-medial auxin foci, division plane is misoriented compared to wild type.

Cell-division plane orientation is determined by specific regulatory events, such as positioning of the preprophase band and the spindle. Spindle formation during mitosis is organised by a class of AURORA kinases, which interact with microtubule-associated proteins of the TPX2 family. I have found that SPT and IND directly regulate the expression of an uncharacterised homologue of TPX2, TPXL8. Similar to TPX2, TPXL8 interacts with AURORA kinases offering a mechanism by which SPT and IND influences spindle formation and thereby cell division plane orientation. My continued work is focussed on unravelling the molecular details of the role of the TPXL8/AURORA module in gynoecium symmetry establishment.

Forward genetic screen as a tool to uncover genes governing plant meiosis

Darya Volkava¹, Karel Řiha¹

¹CEITEC, Brno, Czech Republic

Forward genetic screen is a powerful experimental method allowing to identify genes causing phenotype of interest. This method is becoming very popular again thanks to the progress in whole genome sequencing technologies that facilitate identification of causative mutations. In contrast to T-DNA insertional mutagenesis that causes gene disruptions, chemical mutagenesis used in forward genetic screens produces much wider range of mutations and, hence, higher diversity of phenotypes. This bring a new dimension in gene characterization efforts.

We use this technique to identify new genes governing cell fate transitions during germ-line differentiation. We are particularly interested in understanding mechanism by which non-sense mediated RNA decay factor, SMG7, regulates meiotic exit in *Arabidopsis thaliana*. We set up a suppressor screen in which we look for mutations that rescue infertility of SMG7 deficient plants. We have obtained at least 10 suppressor lines and we currently perform mapping in combination with whole genome sequencing to identify causative mutations. We expect that identified mutations will reveal new genes involved in regulation of meiotic progression.

This work was supported by the Ministry of Education, Youth and Sports of the Czech Republic, European Regional Development Fund-Project "REMAP" (No. CZ.02.1.01/0.0/0.0/15_003/0000479).

Flowers on Alert: GLRs based long-distance Ca²⁺ waves in *Arabidopsis* inflorescence

¹Matteo Grenzi, ¹Fabrizio Gandolfo Doccula, ¹Ambra Selene Parmagnani, ¹Roberto Quadri, ¹Andrea Alfieri, ¹Laura Luoni, ¹Maria Crisitina ¹Bonza and ¹Alex Costa

¹Department of Biosciences, University of Milan, Via G. Celoria 26, 20133 Milan. e-mail: <u>matteo.grenzi@unimi.it</u>

In plants the communication of locally perceived environmental stimuli over long distance it is crucial for development and growth optimization. In *Arabidopsis* rosette, wounding induces the expression of jasmonate responsive genes in unwounded leaves. Recent studies suggest this defense response is promoted by Glutamate-Receptor-Likes (GLRs) channels which mediate leaf-to-leaf electrical signals and Ca²⁺ waves propagation.

Here, performing live imaging of cytosolic Ca^{2+} dynamics in *Arabidopsis* inflorescence, we show that rosette leaf burning promotes the generation of a rapid long-distance Ca^{2+} wave travelling to floral tissues. While the suppression of GLR3.3 almost abolishes the burning-induced Ca^{2+} wave, preliminary results show that the ablation of GLR3.7 results in its exacerbation. Similarly, these two GLR isoforms regulate the aminoacids-induced Ca^{2+} increses in root tip cells. In tobacco leaves, GLR3.3 and GLR3.7 colocalize in the ER and possibly in the PM. We hypothesize that these two isoforms form a heteromeric channel in which the subunit composition can define the heteromer functional properties.

Ongoing experiments are focusing on GLR3.3 and GLR3.7 expression pattern and their functional interaction by using homologous and heterologous expression systems.

Positional clues in petal patterning: the role of auxin and flavonoids

S. GATTI, V. TRAVAGLIA, G. ALSTON, M. YEO, E. MOYROUD

The Sainsbury Laboratory Cambridge University (SLCU), University of Cambridge, 47 Bateman Street CB2 1LR, Cambridge, UK

Petal patterns are an attractive and important feature of the corolla, often acting as a key clue for pollinators' attraction and as a shield against UV radiations. The establishment and maintenance of those complex patterns raise interesting developmental questions: how can differences in cell pigmentation, cell shape and cell cuticle ornamentation combine in an organised fashion within the same organ to produce an elaborate pattern? The Venice mallow (*Hibiscus trionum*) flower is an ideal model to investigate one of the most frequent type of patterns, the bullseye, where the centre of the flower contrasts sharply with its periphery. Bullseye petals are characterized by having two significantly different regions along the proximo-distal axis of the epidermis (the proximal base and the distal top), separated by a third border region (boundary region).

My project aims at finding the positional clues that lead cells to adopt specific fates depending on their respective location, creating a reproducible pattern across the petal epidermis. Here, I will present the preliminary data I have obtained while assessing the role of two possible players: 1) the hormone auxin, an essential molecule that controls almost every known plant developmental processes, and 2) the secondary metabolites flavonoids, very abundant in petals, known to modulate auxin transport and cell shape.

How is Jasmonate biosynthesis initiated in plant plastids?

Yunjing Ma, Marlene Zimmer, Debora Gasperini

Department of Molecular Signal Processing, Leibniz Institute of Plant Biochemistry, Halle, Germany

The bioactive Jasmonic Acid Isoleucine conjugate (JA-IIe) is a lipid-derived phytohormone that is widespread across higher plants. This signaling molecule protects plants against biotic and abiotic challenges and plays essential roles in reproductive development. In recent years, the genetic components involved in JA-Ile biosynthesis, perception and signaling have been uncovered, while the molecular mechanism(s) responsible for activating biosynthetic enzymes are still unknown. It is generally accepted that JA-Ile biosynthesis relies upon the activation of pre-existing plastidial enzymes of the 13-LIPOXYGENASE family, which contain a PLAT (polycystin-1, lipoxygenase, Alpha-Toxin) domain which putatively mediates membrane associations and/or calcium (Ca²⁺) binding. We therefore aim to characterize the activation of plastidial JA biosynthesis enzymes following mechanical wounding in the model plant Arabidopsis. To this end, we are combining live cell imaging with transmission electron microscopy of several 13-LOX constructs to follow their associations with plastidial membranes, as well as determining the role of Ca²⁺ in 13-LOX activation in vitro and in vivo. Furthermore, we will investigate whether newly identified plastidial cation channels impact JA-Ile production through genetic, transcriptional and hormone analysis. Collectively, our efforts are geared towards understanding cytosol-plastid dynamics regulating the activation of JA-Ile biosynthesis, with profound implications for plant stress acclimation and fertility.

Robustness of bract suppression during floral transition in *Arabidopsis thaliana*

S. Dieudonné, F. Besnard

Reproduction et Développement des Plantes laboratory, UMR 5667, ENS Lyon, CNRS, INRA, INRIA

Continuously during their development, plants produce a basic developmental unit, the phytomer. Phytomers are composed of an internode and an axillary meristem subtended by a leaf. The abrupt transition from vegetative to reproductive state remodels the composition of the phytomers. In most Brassicaceæ, but not the closely related Cleomaceæ, the reproductive phytomer loses its leaf, also called bract, producing a naked lateral floral meristem. However, the robustness of this remodeling can be disturbed in some conditions leading to the emergence of some chimeric phytomers at the transition, with flowers subtended by bract. It has been hypothesized that such "bract-flowers" result from a conversion of branches into flowers hence keeping its cauline leaf. In a natural accession of Arabidopsis thaliana, Tsu-0, the first few reproductive phytomers are chimeric, regardless of the environmental condition. Moreover, these "bract-flowers" do not results from a conversion of branches into flowers. Tsu-0 genetic background enables us to explore the genetic basis of such chimeric phytomers during floral transition. This will be performed by a comparative genomic analysis of F2 segregant populations coupled with a transcriptomic analysis over floral transition. By precise and functional imaging at the floral transition, we will investigate how the genetic variations identified modify plant development to generate such chimeric phytomers. This study will provide a better understanding on what controls the robustness of developmental toggle switches associated with floral transition, and a possible insight into the mechanism of bract loss and its evolutionary origins in Brassicaceæ.

Abstract: 30; Poster Number: 25

RAV mechanism in floral represion in abiotic stress

Osnato, Michela; Cota, Nacho; Cereijo, Unai; Pelaz, Soraya

Centre for Research in Agricultural Genomics, CSIC-IRTA-UAB-UB, Campus UAB, Bellaterra (Cerdanyola del Vallès) 08193 Barcelona, Spain

Floral transition is a process of major relevance for plants. Flowering out of time decreases seed production, negatively affecting their biological fitness. Plants cannot escape environmental challenges; therefore, they must adjust their physiology to give response and adapt to suboptimal conditions. Plants sense multiple external and internal factors in leaves and integrate them through complex genetic networks to promote floral induction at most accurate moment.

TEMPRANILLO 1 (TEM1) and TEM2 have been identified as repressors of flower induction. TEM1 and TEM2 act mainly over the expression of the florigen FLOWERING LOCUS T (FT) and gibberellins (GA), which make them pivotal in the control of flowering. These genes belong to RAV transcription factor family, which is characterized by the presence of two DNA binding domains and counts with six members in Arabidopsis.

RAV genes are present in all vegetal species, and physically interact with genes related to abiotic and biotic stress response. Moreover, downregulation of TEM genes has been reported in drought stress rice.

After different tem mutants were challenged to salt and drought stresses, they presented a higher resistance to these abiotic stresses than their wild type counterparts. These results allow us to hypothesize a putative role of TEM genes in regulating the timing of reproductive organs development in response to various stresses, and that this character could be conserved along evolution.

Regulation of LMI1 and WOX3 in leaf base development

Hannah Walter, Miltos Tsiantis

Max Planck Institute for Plant Breeding Research, Carl-von-Linne-Weg 10, D-50829 Cologne

Stipules are lateral appendages of leaves and studying their development presents exciting opportunities to study the regulation of organ proportions¹. Stipule development is regulated by the HD-ZIP class I transcription factor LATE MERISTEM IDENTITY1 (LMI1) and Imi1 mutants display overgrown stipules in Arabidopsis thaliana². This mechanism is conserved in Brassicaceae as Cardamine hirsuta plants which lack LMI1, also develop fully dissected leaves replacing the stipules¹. Wild type stipules in A. thaliana originate from 1-5 epidermal founder cells whereas stipules in Imi1 mutants initiate from larger founder cell numbers $(7-12)^1$. Additionally, it was shown that the transcriptional regulator WUSCHEL-related homeobox/PRESSED FLOWER (WOX3/PRS) is involved in lateral organ formation, by recruitment of founder cells from lateral domains of the shoot apical meristem and that prs mutants lack stipules^{3 4}. Since both transcriptional regulators seem to have an antagonistic effect, we propose to test whether they act in the same pathway to modulate stipule development. This is further indicated by double mutants of *Imi1* and *prs/wox3* which do not display overgrown stipules¹. Because LMI1 negatively regulates mitosis by activating WEE1 and WOX3 was shown to regulate cell division in lateral regions of P3 – P4 leaf primordia⁵, one possibility is that both LMI1 and WOX3 influence stipule development by antagonistically modulating cell cycle regulators. These interactions might be part of a broadly acting regulatory mechanism that causes organ primordium cells to exit from a proliferative state and progress to endoreduplication and differentiation.

3. Matsumoto N, Okada K. A homeobox gene, *Genes Dev.* 2001:3355-3364. doi:10.1101/gad.931001.from

4. Shimizu R, Ji J, Kelsey E, Ohtsu K, Schnable PS, Scanlon MJ. Tissue Specificity and Evolution of Meristematic WOX3 Function. *Plant Physiol.* 2009;149(2):841-850. doi:10.1104/pp.108.130765

5. Nakata M, Matsumoto N, Tsugeki R, Rikirsch E, Laux T, Okada K. Roles of the Middle Domain–Specific WUSCHEL-RELATED HOMEOBOX Genes in Early Development of Leaves in Arabidopsis. *Plant Cell*. 2012;24(2):519-535. doi:10.1105/tpc.111.092858

^{1.} Vuolo F, Kierzkowski D, Runions A, et al. LMI1 homeodomain protein regulates organ proportions by spatial modulation of endoreduplication. *Genes Dev.* 2018;32(21-22):1361-1366. doi:10.1101/gad.318212.118

^{2.} Saddic LA. The LEAFY target LMI1 is a meristem identity regulator and acts together with LEAFY to regulate expression of CAULIFLOWER. *Development*. 2006;133(9):1673-1682. doi:10.1242/dev.02331

Early patterning of adventitious roots initiation in tomato (Solanum lycoperiscum)

Moutasem Omary, Idan Efroni

The Hebrew University of Jerusalem, Israel

Plant organs are produced by the activity of specialized regions of patterning and growth known as meristems. While the two first meristems are laid down during embryogenesis, meristems are formed *de novo* throughout the life of the plant in order to allow response to environment conditions or recovery from severe damage. Post embryogenesis, root meristems are formed either as lateral roots (LR), initiated from prepatterned root pericycle cells, or as adventitious roots (AR), arising from a variety of non-prepatterned tissues. LR initiation has been extensively studied, but how do AR emerge, what are the early developmental processes that allow the transition from non-meristematic to meristematic tissue has remained unknown.

In order to study the genetic mechanisms that regulate AR meristems initiation in a natural context, we have employed the tomato (*Solanum lycoperiscum*). Tomato generate a large number of stemborne adventitious roots (SBR) along high accessible stems without the need for external hormonal or mechanical stimuli, making it a powerful system to study AR initiation.

Characterization of SBR initiation using hormonal marker lines revealed that tomato SBR appear to initiate from phloem-associated cells, the first step being a co-activation of response to the plant hormones auxin and cytokinin in a population of ~30 cells. As the meristem proliferates, the signalling domains for the two hormones separate to form a distal auxin and proximal cytokinin domain. Remarkably, this developmental dynamics phenocopied meristem initiation process during embryogenesis and root regeneration.

To better understand the early stages of meristem formation, we performed single-cell mRNA-Seq of meristem primordia at the 20-30 cell stage. This revealed that the early primordium is composed of several cell types, characterized by the expression of specific *LATERAL ORGAN BOUNDARY (LBD)* genes. I generate CRISPR/Cas9 tomato mutants in single and multiple LBDs selected based on their expression pattern in the single-cell transcriptome. My preliminary results show that mutants in specific LBD genes cause disruption to the SBR initiation process, the most severe being a complete lack of SBR, even after wounding stimuli. Strikingly, this effect was specific to the SBR, highlighting the shared and unique developmental programs acting during root meristem initiation in different developmental contexts.

Abstract: 33; Poster Number: 28

Role of TEMPRANILLO genes in Regulating Flowering

Paula Suarez-Lopez, Michela Osnato, Poonam Nebhnani, Soraya Pelaz

Centre for Research in Agricultural Genomics, Spain

The period in which a plant transitions from juvenile to adult phase and therefore flowering can be induced, is a crucial determining factor in its degree of reproductive success. Plants initiate this process based on various internal and external signals. The regulatory network then perceives these signals and activates or represses floral induction.

TEM (TEMPRANILLO) genes are transcription factors identified by our lab in Arabidopsis thaliana. TEM's are involved in delay of flowering by repression of FT (Flowering Locus T, a signal for flowering initiation) by binding to a region near its transcription start site and blocking transcription. Our main goal is to dissect the mechanisms of TEM activity in controlling flowering time and understanding how these proteins reprogram plant development for environmental adaptation.

Yeast 2-hybrid experiments revealed that basic Helix Loop Helix transcription factors MYC2 and MYC4 are TEM interactors. MYC2 also binds to the *FT* promoter (same region where TEM binds) during jasmonic acid mediated flowering inhibition. Other related and similar functions between TEM and MYC led us to hypothesize that these transcription factors may work as a complex in repressing *FT*, hence hindering flowering. MYC/TEM interactions with a few other factors also suggested that the complex might recruit histone modification enzymes to suppress expression of target genes. This could be of great relevance in unveiling the role of TEM's, in complex with MYC, in the molecular mechanisms underlying flowering.

Abstract: 34; Poster Number: 29

Regulatory specificity of cambium stem cell signaling

Min-Hao Chiang¹, Vadir Lopez¹, Dongbo Shi¹, Klaus Brackmann², Jiyan Qi¹, Virginie Jouannet¹, Thomas Greb^{1*}

¹Centre for Organismal Studies (COS), Heidelberg University, Germany ²Department of Cell and Molecular Biology, Molecular Systems Biology, Uppsala University, Sweden *corresponding author, email: <u>thomas.greb@cos.uni-heidelberg.de</u>

Cambium stem cells are a group of undifferentiated cells located in the plant stem. The cell division and differentiation activity of cambium cell is the original source for plant lateral growth which include the formation of xylem and phloem cell, thus for the production of terrestrial biomass and for wood formation. In order to maintain its long-term proliferation activity and well-organized tissue formation ability, cambium cell requires delicate cell-cell communication with surrounding tissues. Previous studies have shown that cambium cell is maintained by a network of interacting signal pathway including hormones such as auxin, cytokinin and gibberellins, and small peptides molecule. However, the spatio-temporal organization of cambium stem cell communication with surrounding tissues has not yet been investigated. Especially, it was found recently that the cambium can be further subdivided into the proximal cambium and distal cambium and be characterized by specific gene activity such as PXY and SMXL5. Therefore, in this project, I want to investigate the unique features of the cambium and focus on two important hormonal signaling pathways fundamental for cambium activity: auxin and cytokinin signaling. To locally interrupted hormone signaling, negative regulators of auxin and cytokinin signaling were specific expressed in the proximal cambium or distal cambium in a Dexamethasone-inducible manner. Histological analysis and gene expression profile were performed to study the effect after hormone signaling interruption. Overall, I identified that the proximal cambium for auxin signaling and distal cambium for cytokinin are essential site for cambium regulation.

Quantitative analysis of cellular growth patterns in the *Arabidopsis* ovule

Athul R Vijayan, Rachele Tofanelli and Kay Schneitz

Plant Developmental Biology, Technical University of Munich, Freising, Germany

The cellular and molecular mechanisms underlying tissue morphogenesis in plants remain poorly understood. The Arabidopsis ovule, with its small size and characteristic curvature, represents an interesting model to address this topic. The ovule is the major female reproductive organ of higher plants and will eventually give rise to the seed after fertilization. Development of the Arabidopsis thaliana ovule has been qualitatively well described in past years and a staging system has been proposed that is based on the appearance of characteristic morphological features within the organ (1). As soon as the ovule arises as a finger-like protrusion from the placental tissue of the gynoecium, three elements are recognized along the proximal-distal (PD) axis: the funiculus, chalaza and nucellus. At maturity, the ovule exhibits a distinct 180° curvature (anatropy). Although genetic and molecular mechanisms controlling the outgrowth and patterning of the ovule are known in detail, a quantitative analysis of the cellular growth patterns that contribute to the final three-dimensional shape of the ovule is still lacking (2). Our approach for the extraction of quantitative information of ovule growth consists of 3D image acquisition of fixed samples at full cellular resolution using confocal microscopy (3). The obtained z-stacks are processed using a convolutional neural network and cells are segmented and analyzed using MorphographX (4). From the 3D computational reconstruction of 150 ovules, covering all developmental stages, we have obtained information on cell number, cell size and shape, and spatial distribution of cell divisions. Additionally, cell type annotation has been performed using 3D cell atlas tools. The 3D digital models allow to explore the contribution of the different tissues to the overall growth of the organ. We will discuss our present findings. Our quantitative cellular analysis represents a novel approach in the study of ovule morphogenesis and will lay the foundation for further investigations aimed at understanding how molecular, cellular, and mechanical mechanisms are translated into final ovule shape.

Abstract: 36; Poster Number: 31

A new role of the embryo patterning gene WOX9 in regulating the root meristem

Ramsankar Chandrasekar, Ning Zhang, Thomas Laux

Signalling Research Centres BIOSS and CIBSS, Faculty of Biology, University of Freiburg, Schaenzlestrasse 1, 79104 Freiburg, Germany

The WUSCHEL RELATED HOMEOBOX (WOX) genes WOX8 and WOX9 play a central role in partitioning basal and apical cell fates during early embryogenesis and in enabling auxin transport during early apical-basal patterning of the embryo (Breuninger et al., 2008). Studying their function, I found that WOX9 also affects the size of the root meristem. In my future PhD work, I will address the molecular basis of WOX9 in embryo development and root meristem regulation. As a first goal, I will study how WOX9 can be integrated into known regulatory pathways, such as those defined of HD-ZIP III genes and cytokinin function.

Breuninger, H., Rikirsch, E., Hermann, M., Ueda, M., & Laux, T. (2008). Differential expression of WOX genes mediates apical-basal axis formation in the Arabidopsis embryo. *Developmental cell*, *14*(6), 867-876.

Role of reproductive sink size (RSS) as a driver for resource allocation in barley

Isaia Vardanega, Rüdiger Simon

Department of Developmental Genetics, Heinrich-Heine University, D-40225 Düsseldorf, Germany

Seed number and viability, together with the appropriate environment, determine a plant's fitness. Barley (*Hordeum vulgare*) is the 4th most abundant crop plant in the world, for this reason the number of seed that a plant can produce play an important role on the agricultural production. It is known that the initiation and development of seeds requires extensive resource allocation over prolonged time periods. However, it is not yet known whether the total number of seeds that can be generated by a model or crop plant is source-limited or sink-controlled, i.e. by the strength of the sink tissues to attract nutrient allocation to the developing seeds. The impact of resource availability on reproductive growth and seed formation has been studied in great detail, however, the role of reproductive sink size as a driver for resource allocation and photosynthetic activity remains not investigated. The aim of this project is to manipulate meristem functions and RSS by genome editing starting from barley homologs of Arabidopsis CLAVATA pathway genes involved in the well-studied pathway controlling the development and size of the meristem in Arabidopsis. Futhermore, we want to investigate the role of sink strength in resource allocation and seeds production, using FRET based sensors and other cell biology toolboxes that allow direct measurements of metabolites and signalling pathway activities in living plants.

Abstract: 38; Poster Number: 33

Searching candidate genes acting downstream of NTT

David Díaz Ramírez¹, Ricardo Chávez Montes², Stefan de Folter², Nayelli Marsch Martínez^{1,3}

¹ Department of Biotechnology and Biochemistry, Center for Research and Advanced Studies of the National Polytechnic Institute, km 9.6 Libramiento Norte, Carretera Irapuato-León, Irapuato, Guanajuato, Mexico

² Advanced Genomics Unit (Langebio), Center for Research and Advanced Studies of the National Polytechnic Institute, km 9.6 Libramiento Norte, Carretera Irapuato-León, Irapuato, Guanajuato, Mexico

³Corresponding author e-mail <u>nayelli.marsch@cinvestav.mx</u>

NTT is a zinc finger transcription factor that has the capacity to alter developmental programs when overexpressed. Moreover, the loss of NTT function, alone or in combination of closely related genes, leads to developmental defects in pistils and roots. The latter, reported to be very severe, highlight an essential role for this kind of transcription factors in specific tissues. Knowing genes that act downstream of NTT can be very useful to understand more about its role in developmental programs. We are exploring downstream candidate genes. One was previously identified using strategies such as yeast-one-hybrid assays. Others were recently identified by analyzing the co-expression networks of NTT and one if its interactors (STK), leading also to the description of new altered phenotypes of one of them. In order to find more genes acting downstream, some of which may be related to hormonal pathways, we performed an RNA-seq analyses using a line where NTT activity can be induced. We collected different tissues from induced and not induced plants and obtained differentially expressed genes. We performed enrichment analyses and found response to hormone to be one of the most enriched categories. Currently, we are analyzing the data to select and confirm the changes of expression to afterwards perform crosses with mutants and markers of these selected genes to better understand and clarify their genetic interactions.

Investigating transcriptional regulation of cell wall synthesis during floral morphogenesis

Benno Stüwe, Dijun Chen, Lazar Novaković, Alfie Hao, Tony Bacic, Kim Johnson, Kerstin Kaufmann, , Arun Sampathkumar

Max Planck Institute of Molecular Plant Physiology, 14476 Potsdam, Germany

One of the central questions in biology is to understand the growth and form of cells, organs, and organisms. Although much is known about the gene regulatory networks influencing morphogenesis in plants, integrated understanding of how molecular networks operate at multiple scales to control overall geometry is missing. We address this problem in flowers, which develop at the outer flanks of the apical meristem and transform from a simple convex surface to a highly convoluted structure, with organs emerging sequentially in a centripetal fashion. As plant cells are encased by a rigid yet flexible cell wall, the overall mechanical status of the cell wall acts as a bottle neck in regulating growth rates. Further the structural organization of the cell wall specifically the alignment of its stiffest polymer the cellulose microfibrils and its interaction with other cell wall polysaccharides influences growth direction. Using interdisciplinary approaches my PhD project aims to address the contribution of the cell wall in regulation of floral morphogenesis. Cell wall linkage analysis data suggests that the abundance of the different cell wall polysaccharides is similar to that of the apical meristem but different from leaf tissue. In addition, drastic changes in polysaccharide levels were not observed between Stage 1 to Stage 8 floral primordia. We have further identified a core set of cell wall glycosyltransferases that operated during floral morphogenesis. Finally, analysis of publically available ChIPseq data shows putative binding of several transcription factors involved in organ specification to regulatory regions of cell wall glycosyltransferase genes.

A molecular hub regulating root stem cell replenishment in *Arabidopsis*

Vivien I. Strotmann, Yvonne Stahl

Institute for Developmental Genetics, Heinrich Heine University, Düsseldorf

The root system of higher plants plays an important role in plants life cycle, since it provides the access to water and nutrients and anchorage in the soil. The root is formed by a group of four slowly dividing, pluripotent stem cells from which most of the plant body structures arise. Loss of stem cells due to differentiation or different stresses e.g. genotoxicity, the root's stem cell pool needs to be replenished by coordinated divisions of the quiescent center (QC) cells.

One of the most important questions in root development is how stem cell homeostasis is regulated in the root in response to different external and internal clues. A multitude of different factors such as phytohormones and transcriptional regulators are known to play important roles in regulating QC divisions, and therefore stem cell replenishment. Still, most of the interconnections of the many different factors are largely unknown. The aim of this project is to examine a newly emerging potential hub linking developmental transcriptional regulators involved in stem cell replenishment, such as WOX5 and PLTs, with known regulators in response to BRs and genomic stresses, e.g. BRAVO and BES1.

Effects of changing nitrate concentrations on intercellular communication in *Medicago truncatula* roots.

Liam German, Rocio Gaudioso-Pedraza, Yoselin Benitez-Alfonso

University of Leeds, Faculty of Biological Sciences, Sheila Gosden Studentship

Plasmodesmata (PD) are structures connecting the cytoplasm of neighbouring plant cells and required for intercellular (symplastic) communication. They can transport RNAs, proteins and hormones that coordinate cell, tissues and organ responses to external stimuli such as changes in nutrient availability, environmental stresses and microbes. Our lab recently reported that changes in symplastic communication alter the number of nitrogen-fixing nodules initiated when the model legume *Medicago truncatula* interacts with the soil borne bacteria *Sinorhizobium meliloti*. Now we are using confocal microscopy and symplastic reporter lines to explore how changes in nitrate availability influences this response. We are also beginning to identify and characterise the role of PD proteins in this mechanism. Through further dissection of the regulatory mechanisms at play during nodulation we aim to better understand the establishment and maintenance of plant-microbe symbiosis and its links to symplastic transport in plants. This work has important implications for crop management strategies, particularly in optimising nutrient supplementation and the maintenance of soil quality.

A possible role for DNA methylation in regulation of temperature-dependent flowering

Suze Blom^{1,2}, Froukje van der Wal^{1,2}, Gerco C. Angenent^{1,2}, Richard G.H. Immink^{1,2}

¹Laboratory of Molecular Biology, Wageningen University, Wageningen, The Netherlands ²Biocience, Wageningen University and Research, Wageningen, The Netherlands

Like other plants, Arabidopsis thaliana senses its environments to adjust its flowering time to the local temperature. Many genes have previously been identified to play a part in this temperature-sensitive response but underlying molecular mechanisms haven't been fully resolved. Increasing evidence suggests that epigenetic marks, like histone proteins and DNA methylation, are important for plants to sense and adapt to their environment. DNA methylation has been linked to regulation of flowering time, e.g. at the FLOWERING WAGENINGEN (FWA) locus, or at INCREASE IN BONSAI METHYLATION 1 (IBM1). However, it is unknown whether DNA methylation also affects ambient temperaturedependent flowering. In this study, the link between DNA methylation and temperature-sensitive flowering in Arabidopsis was investigated. For this purpose, the flowering time of multiple DNA methylation mutants was studied at two different temperatures, 16°C and 25°C. A T-DNA mutant of DECREASE IN DNA METHYLATION 1 (ddm1-10), was found to have an altered temperature-sensitivity response. The *ddm1-10* mutant bolted simultaneously with Col-0 at 25°C, but initiated bolting significantly later than Col-0 at the lower temperature. Thus, a genome-wide decrease in CG, CHH and CHG methylation causes a hypersensitive temperature response of flowering time. This suggests that the control of DNA methylation is important for integration of environmental signals into developmental decisions and adaptation of reproduction strategy to the environment. Further research will have to show which kind of DNA methylation is important and which specific loci are affected by this epigenetic process, to elucidate the causal and underlining molecular pathway.

Exploring the catabolism of inositol pyrophosphates and its impact on signalling pathways in plants

Laurent F., Couto D., Hothorn M.

University of Geneva, Switzerland

The inositol pyrophosphate (PP-InsPs) molecules are involved in many cellular processes in yeast and animal cells. It has also recently been shown that this family of molecules could be involved in the regulation of phosphate homeostasis in plants, but their roles in other plant processes have not been studied in detail. Studying PP-InsPs and their catabolic pathway could be the key to understand their regulation and putative involvements in other signalling pathways in plants. I focused on the catabolism of PP-InsPs by two phosphatase families. Two phosphatase families have been found to interact with PP-InsPs by a proteomics approach in the Hothorn lab. These two promising families are the NUcleoside Diphosphates linked to moiesty-X (NUDIX) hydrolase family and the Plant and Fungi Atypical Dual-Specificity Phosphatase (PFA-DSP) family. Therefore, I monitored the enzymatic activity of DSP3 and NUD17. I found that these enzymes specifically hydrolyze 5-IP₇ and have a higher turnover number than the yeast homologue SIW14. In addition, genetic tools have been created in order to study these proteins *in planta*. Lines expressing GFP-NUD17 showed a localization of the protein GFP-NUD17 in the nucleus and in the cytosol. Other lines expressing FLAG-DSP2 have a dwarf development phenotype.

Spatiotemporal dynamics of the *Arabidopsis* shoot stem cell niche at organ initiation

Henrik Åhl^{1,2}, Weibing Yang¹, Aram Gurzadyan¹, Benoit Landrein¹, Henrik Jönsson^{1,2}

¹Sainsbury Laboratory, Univertsity of Cambridge, Cambridge, United Kingdom

² Department of Applied Mathematics, University of Cambridge, Cambridge, United Kingdom

Virtually all aerial development of plants originates in the shoot, in a region called the shoot apical meristem (SAM). Due to the inherent need to adapt to various environments, stresses, and modes of development, regulation of the stem cell niche must be robustly regulated. Unfortunately, most of the current understanding is limited to the static picture, and little is understood of the dynamical aspects of the transition between two modes of development, e.g. between vegetative and floral development.

In order to address this gap, we have developed a protocol for elucidating the intricacies of dynamical regulation for the main stem cell regulators, WUSCHEL (WUS) and CLAVATA-3 (CLV3). By growing plants on the auxin transport inhibitor NPA, we are able to induce a developmental state without flower-development. A transient change in the treatment is then used to investigate the differential regulation that occurs when the very first floral primordia are initiated. Through the combination of state-of-the-art confocal microscopy, as well as mathematical modelling and novel quantification techniques, we are able to intricately link experiments and theory.

Our preliminary results show that the WUS expression pattern appears to be dynamically regulated by the process of organ initiation. Interestingly, we do not observe a similar effect in CLV3, which contrasts with the current understanding of CLV3-WUS interplay. We believe that these results are indicative of fundamental aspects of plant stem cell regulation that impact plant development broadly.

Characterization of the meiotic cyclin SOLO DANCERS (SDS) in maize

Óscar Sanz Mora, Reinhold Brettschneider, Arp Schnittger

University of Hamburg

Meiosis, in contrast to mitosis, halves the nuclear DNA content. This is important for sexual reproduction as it maintains genome size in the offspring. Furthermore, meiosis is key for genetic variation due to recombination between the homologous (parental) chromosomes and a new assortment of chromosome set. Similar to mitosis, meiosis is also regulated by cyclin dependent kinases, which function in conjunction with cyclin co-factors. One of the meiosis specific cyclins is called SOLO DANCERS (SDS) and has previously been described in A. thaliana. There, SDS is expressed specifically in male meiotic cells where it functions in homolog synapsis and recombination. While putative SDS homologs in Zea mays have already been discovered, their actual biochemical function in the cell cycle remains unknown. Here, we present our recent experiments to characterize the structure and function of the putative orthologues ZmSDS16 and ZmSDS57. A double mutant was generated in the maize inbred line A188. Conversely, heterologous experiments were conducted with ZmSDS57 in Arabidopsis. Preliminary data lead to the idea that the introns of ZmSDS57 have an important role for the expression levels in Arabidopsis. Furthermore, biochemical experiments were started to address the activity of ZmSDS57, foremost to assess its function as a CDK co-factor.

Quantitative analysis of cellular growth patterns in integument morphogenesis and ovule curvature in *Arabidopsis thaliana*

Rachele Tofanelli, Athul R Vijayan, and Kay Schneitz

Plant Developmental Biology, Technical University of Munich, Freising, Germany

How the Gestalt of a plant tissue is generated remains an open question in plant developmental biology. Ovule curvature represents a unique phenomenon in plant tissue morphogenesis and invites interesting questions regarding the cellular, molecular, and mechanical mechanisms involved in the formation of this characteristic shape. The ovule of Arabidopsis thaliana represents a nice model system to address this question. At maturity, the ovule exhibits a distinctive curved shape such that the micropyle is placed next to the base of the funiculus, resulting in a 180° bending (anatropy). A detailed qualitative description of ovule development in Arabidopsis thaliana has been established, as has been a staging system based on discrete morphological features (1). Ovules originate from the placental tissue of the gynoecium as protrusions and eventually three morphologically recognizable pattern elements are recognized along the proximal-distal (PD) axis: the funiculus, chalaza, and nucellus. A. thaliana carries bitegmic ovules as the chalaza originates two integuments, the precursors of the seed coat. It has been postulated that differential growth of the outer integument contributes to curvature (1). Comparative evolutionary studies suggested that the presence of an outer integument is crucial for anatropy in bitegmic ovules (2). Moreover, genetics provided additional evidence on the central role of the outer integument, through the identification of mutants carrying ovules with a defect in outer integument development (3). We have performed a quantitative analysis of the cellular growth patterns of the integuments in wild-type and mutants at different developmental stages. Our pipeline consisted of 3D image acquisition of fixed samples at cellular resolution followed by image processing and analysis in MorphographX (4). The results indicate that the outer layer of the outer integument is a central regulator of ovule curvature. Furthermore, they validate our quantitative approach as a promising strategy to explore the cellular mechanisms underlying ovule curvature.

⁽¹⁾ Schneitz, K., Hülskamp, M., & Pruitt, R. E. (1995). Wild-type ovule development in Arabidopsis thaliana: a light microscope study of cleared whole-mount tissue. The Plant Journal, 7(5), 731–749.

⁽²⁾ Endress, P. K. (2011). Angiosperm ovules: diversity, development, evolution. Annals of Botany, 107(9), 1465-1489.

⁽³⁾ Chaudhary, A., Gao, J., & Schneitz, K. (2018). The genetic control of ovule development. Reference Module in Life Sciences. doi: 10.1016/B978-0-12-809633-8.20737-1.

⁽⁴⁾ Barbier de Reuille, P., Routier-Kierzkowska, A.L., Kierzkowski, D., Bassel, G.W., Schupbach, T., Tauriello, G., Bajpai, N., Strauss, S., Weber, A., Kiss, A., et al. (2015). MorphoGraphX: A platform for quantifying morphogenesis in 4D. eLife 4, 05864

Characterization of Global Prolifferative Arrest (GPA) process in Arabidopsis Thaliana and Pisum sativum

Eduardo Burillo-Richart, Vicente Balanzà, Irene Martínez-Fernández, Cristina Ferrándiz.

Instituto de Biología Molecular y Celular de Plantas, Consejo Superior de Investigaciones Científicas—Universidad Politécnica de Valencia, 46022 Valencia, Spain

A big proportion of the economically important crops belong to the group of monocarpic plants. In these, after the production of a certain number of fruits, the activity of the Shoot Apical Meristem (SAM) and of the meristems of the secondary axes stop in a coordinated way, leading to the cessation of flower production. This phenomenon is called Global Prolifferative Arrest (GPA)¹ and it has been interpreted as a mechanism to optimize the channeling of available resources towards seed production. Also, it has been described in numerous species, so it can be assumed that it is a general phenomenon in monocarpic plants.^{2, 3, 4}

Studies in the model species *Arabidopsis thaliana* have allowed the identificaton of factors that affect GPA timing. One of the most important is the production of fruits and seeds: sterile mutants produce more flowers than fertile ones ^{4, 5}. In addition, we have recently proposed a genetic pathway that controls GPA: *FRUITFULL (FUL)* and the miR172 control the levels of *APETALA2 (AP2)* and AP2-LIKE genes in the SAM, which finally modulate SAM arrest⁶. Currently, our research is focused on determining in more detail the specific contribution of the different AP2-LIKE genes to GPA control. Finally, recent studies working with the legume species *Pisum sativum* (pea) indicate that the mechanisms that control GPA are very similar to those of *Arabidopsis thaliana*, and we are currently working in the determination of the differences and similarities that exist in this process between both species.⁷

- 1. Hensel, Nelson, Richmond, Bleecker. The fate of inflorescence meristems is controlled by developing fruits in Arabidopsis. *Plant Physiology* **106**, 863-876 (1994).
- 2. Lockhart, Gottschall. Fruit-induced & apical senescence in Pisum sativum. Plant Physiol 36, 389-398 (1961).
- 3. Murneek. Effects of Correlation between Vegetative and Reproductive Functions in the Tomato. *Plant Physiol* **1**, 3-56 57 (1926).
- 4. Nooden, Singh, Letham. Correlation of xylem sap cytokinin levels with monocarpic senescence in soybean. *Plant Physiol* **93**, 33-39 (1990).
- 5. Landrein, et al. Nitrate modulates stem cell dynamics in Arabidopsis shoot meristems through cytokinins. PNAS **115**, 1382-1387 (2018).
- 6. Balanza, Martínez-Fernández, Sato, Yanofsky, Kauffman, Angenent, Bemer, Ferrandiz. Genetic control of meristem arrest and life span in Arabidopsis by a novel FRUITFULL-APETALA2 pathway. *Nat Commun*:565, (2018).
- 7. Martínez Fernández. La ruta FUL-AP2 y la longevidad de los meristemos en Arabidopsis thaliana. Bases moleculares y potencial biotecnológico. *Tesis doctoral*. Universitat Politécnica de València. (2017).

Characterizing the CYCB1 group and its potential role in microtubule organization

Mariana Romeiro Motta, Arp Schnittger

University of Hamburg

Mitotic progression is mainly controlled by the action of Cyclin-dependent kinases (CDKs) in complex with their cyclin counterparts. Compared to metazoans, there is rather limited available functional information on the cyclin families in plants. It is known that all three plant cyclin groups A, B and D are involved in the regulation of mitotic progression. However, plant cyclin families contain a large number of members, which makes functional studies more complex. Here, we present a report on the function of the B1-type cyclins in Arabidopsis thaliana. We show CYCB1;2 is of higher importance in plant development, acting together with other CYCB1 members in a tissue-dependent manner. More strikingly, double cycb1;1 cycb1;2 mutants have a general delayed development with disorganized and shorter roots and shoots. When studied in detail, these mutants reveal defects in microtubule organization, defective spindles and longer mitotic divisions. We have identified GIP1 (GCP3-INTERACTING PROTEIN 1A) as a potential substrate and confirmed in vitro phosphorylation by CYCB1;1, CYCB1;2 and CYCB1;4 together with CDKB2;2. Localization studies of GIP1 in a cycb1;1 cycb1;2 double mutant give further evidence of the potential regulation of GIP1 by the CYCB1 family. With this work, we expect to shed light on the function of the B1-class cyclins in plant development and increase our understanding of the connection between the cell cycle and major regulators of microtubule organization during mitosis.

Protein interactions of HD-Zip class I transcription factors shaping leaves in Brassicaceae

Luise Zühl, Francesco Vuolo, Miltos Tsiantis

Max Planck Institute for Plant Breeding Research, Carl-von-Linné-Weg 10, 50829 Cologne, Germany

lzuehl@mpipz.mpg.de

We are studying leaf shape as a model to understand how morphological diversity arises in angiosperms. In this context we are investigating developmental processes leading to generation of simple leaves in *Arabidopsis thaliana versus* complex leaves in its close relative *Cardamine hirsuta*. Previously our group identified the paralogous HD-Zip class I homeobox transcription factors LATE MERISTEM IDENTITY 1 (LMI1) REDUCED COMPLEXITY (RCO) as important regulators of development of distinct leaf shapes in these two species and other Brassicaceae ^{1,2}. Both act as local growth repressors ²⁻⁴, but knowledge about co-factors and downstream components of underlying regulatory processes is limited. Therefore, regulatory hierarchies through which these proteins influence leaf shape remain poorly understood. With my PhD project I aim to clarify these hierarchies and mainly focus on the predicted interaction partners of *A. thaliana* LMI1 and *C. hirsuta* RCO. To identify interactors, I will apply affinity purification coupled with mass spectrometry approaches and subsequent targeted interaction analyses. Results will also be compared to preliminary data from a yeast-two-hybrid screen, hinting towards interaction of AtLMI1 with members of other transcription factor families. Finally, I hope to analyse the role of identified interactions for leaf shape development and their conservation in Brassicaceae.

- 1. Saddic, L.A. *et al.* The LEAFY target LMI1 is a meristem identity regulator and acts together with LEAFY to regulate expression of CAULIFLOWER. *Development* **133**, 1673-82 (2006).
- 2. Vlad, D. *et al.* Leaf shape evolution through duplication, regulatory diversification, and loss of a homeobox gene. *Science* **343**, 780-3 (2014).
- Vuolo, F. *et al.* Coupled enhancer and coding sequence evolution of a homeobox gene shaped leaf diversity. *Genes & Development* 30, 2370-2375 (2016).
- 4. Vuolo, F. *et al.* LMI1 homeodomain protein regulates organ proportions by spatial modulation of endoreduplication. *Genes & Development* **32**, 1361-1366 (2018).

Modulating recombination frequency to stabilize tetraploid meiosis

Stephan Portheine¹, Stefan Hoth¹ and Arp Schnittger¹

¹Universität Hamburg

Polyploids are widespread and polyploidization is a driving force behind generating new variation and thus adaptation. Polyploidy also occurs more frequently under various environmental stresses and alters stress response on a transcriptional and physiological level. Agriculturally polyploids are of interest, as increased ploidy level is typically associated with larger crops and higher yield. Yet, polyploidization, even when resulting in a balanced chromosome number, comes with many challenges and de novo polyploids go through a strong bottleneck. This is largely due to negative effects on meiotic outcome. Additional chromosome copies in *de novo* polyploids lead to multivalents in meiosis I, which do not segregate into balanced meiotic products. Established polyploids usually have a diploid like meiosis where chromosomes form bivalents. The main molecular mechanism underlying this is control of cross-over number, as multivalents are dependent on the formation of chiasmata between the multivalent chromosomes. We hypothesize that reduced cross-over number alone, up to down to one obligate cross-over, is enough and sufficient to balance de novo polyploid meiosis. Increased cross-over number would then have the opposite effect, with a stronger reduction of the number of balanced meiotic outcomes. To test this we will assess the number of chromosomes in microspores of de novo tetraploid anti recombination pathway mutants (RecQ4A/RecQ4B, FancM and Figl1) and double mutants compared to tetraploid Colombia-0. To do so we utilize an established centromeric marker (CENH3-GFP) under a microspore specific promotor. As a measure of meiotic fidelity the distribution of chromosomes in the microspores will be taken. We expect this distribution to flatten in the mutants at equal pace with their described increase in cross-over number.

The function of LATE MERISTEM IDENTITY 1 homolog in Marchantia polymorpha

Elizabeta Banić^{1,3}, Saiko Yoshida¹, Remco Mentink¹, Tom Dierschke², Sabine Zachgo², Miltos Tsiantis¹

¹Department of Comparative Development and Genetics, Max Planck Institute for Plant Breeding Research, Cologne, Germany

²Botany Department, School of Biology and Chemistry, Osnabrück University, Osnabrück, Germany

³University of Cologne, Cologne, Germany

Polyploids are widespread and polyploidization is a driving force behind generating new variation and thus adaptation. Polyploidy also occurs more frequently under various environmental stresses and alters stress response on a transcriptional and physiological level. Agriculturally polyploids are of interest, as increased ploidy level is typically associated with larger crops and higher yield. Yet, polyploidization, even when resulting in a balanced chromosome number, comes with many challenges and de novo polyploids go through a strong bottleneck. This is largely due to negative effects on meiotic outcome. Additional chromosome copies in *de novo* polyploids lead to multivalents in meiosis I, which do not segregate into balanced meiotic products. Established polyploids usually have a diploid like meiosis where chromosomes form bivalents. The main molecular mechanism underlying this is control of cross-over number, as multivalents are dependent on the formation of chiasmata between the multivalent chromosomes. We hypothesize that reduced cross-over number alone, up to down to one obligate cross-over, is enough and sufficient to balance *de novo* polyploid meiosis. Increased cross-over number would then have the opposite effect, with a stronger reduction of the number of balanced meiotic outcomes. To test this we will assess the number of chromosomes in microspores of de novo tetraploid anti recombination pathway mutants (RecQ4A/RecQ4B, FancM and Figl1) and double mutants compared to tetraploid Colombia-0. To do so we utilize an established centromeric marker (CENH3-GFP) under a microspore specific promotor. As a measure of meiotic fidelity the distribution of chromosomes in the microspores will be taken. We expect this distribution to flatten in the mutants at equal pace with their described increase in cross-over number.

Role of MYB and bHLH transcription factors in bullseye pattern evolution

Alice Fairnie, Edwige Moyroud

The Sainsbury Laboratory Cambridge University (SLCU), University of Cambridge, 47 Bateman Street CB2 1LR, Cambridge, UK

The flowers of angiosperms often display a diversity of spotted, striped, venation and bullseye patterns on their petals. These visual signals are thought to fulfil abiotic and biotic functions. Patterns, for example, can act as visual signals to attract and guide animal pollinators. Research identifying the molecular and developmental mechanisms underlying pattern formation are rare. I study the development, evolution and function of the bullseye pattern in Hibiscus trionum. To create the H. trionum bullseye pattern epidermal cells in the proximal and distal petal vary in their pigmentation, shape and cuticle structure. The project focussed on development of the pigmentation and cell shape patterns and is divided into three parts: (1) identifying regulatory mechanisms controlling bullseye pattern formation including upstream pre-pattern elements; (2) characterising and comparing natural varieties that differ in the bullseye patterns to understand the developmental and molecular changes accounting for these morphological differences; (3) testing pollinator preferences to dissect the contribution of the different elements of the pattern and the importance of the bullseye geometry in plant-pollinator communication. Classic molecular and biochemical techniques have been used to understand parts one and two of the project. A transcription factor, bHLH130, has been isolated from H. trionum. bHLH130 does not show preferential expression in the distal petal before pattern emergence and is not thought to to regulate development of the bullseye. Their function is currently being investigated in planta using a transgenic approach. Morphological and chemical differences in the bullseyes of three Hibiscus varieties were also characterised. The three varieties vary in their pigmentation pattern but not in their cell shape or cuticle pattern

Participant List

last name	first name	affiliation	page
Åhl	Henrik	SLCU, Cambridge, GB	55
Alarcia-García	Ana	IBMCP (UPV-CSIC), Valencia, ES	32
Balboni	Martina	University of Hamburg, DE	23
Banic	Elizabeta	MPIPZ, Köln, DE	62
Bellande	Kevin	University of Montpellier, FR	14
Blom	Suze	University of Wageningen, NL	53
Burillo-Richart	Eduardo	IBMCP (UPV-CSIC), ES	58
Caillaud	Marie-Cecile	ENS, Lyon, FR	8
Cereijo	Unai	CRAG, Barcelona, ES	41
Chandrasekar	Ramsankar	University of Freiburg, DE	47
Chaudhary	Ajeet	TUM, Freising, DE	16
Chiang	Min-Hao	COS, University of Heidelberg, DE	45
Chomthong	Methawi	University of Cambridge, GB	20
Chong	Wei Wen	University of Zurich, CH	27
Diaz	David	CINVESTAV, Irapuato, MX	49
Dieudonné	Sana	ENS, Lyon, FR	40
Fairnie	Alice	SLCU, Cambridge, GB	63
Fattorini	Roisin	University of Cambridge, GB	11
Ferrándiz	Cristina	University of Valencia, ES	9, 32, 58
Gatti	Stefano	SLCU, Cambridge, GB	38
Geldner	Niko	University of Lausanne, CH	8
German	Liam	University of Leeds, GB	52
Giaume	Francesca	University of Milan, IT	18
Grenzi	Matteo	University of Milan, IT	37
Hay	Angela	MPIPZ, Köln, DE	17
Hothorn	Michael	University of Geneva, CH	6, 54
Jourquin	Joris	VIB-UGent, Ghent, BE	22
Kortz	Annika	INRES, University of Bonn, DE	29
Kumari	Khushbu	University of Würzburg, DE	25
Laurent	Florian	University of Geneva, CH	54
Leandro	Thales D.	UNESP, Rio Claro , BR	12
Liu	Shanda	MPIPZ, Köln, DE	21
Lopez	Francesca	NUI, Galway, IE	34
Lopez Vazquez	Ana	University of Lausanne, CH	15
Ма	Yunjing	IPB, Halle, DE	39
Mähönen	Ari Pekka	University of Helsinki, FI	6
Maizel	Alexis	University of Heidelberg, DE	9
McLaughlin	Heather	JIC, Norwich, GB	26
Morales	Stefania	University of Ghent, BE	28
Moyroud	Edwige	SLCU, Cambridge, GB	6, 38, 63

Muralidhara	Prathibha	University of Würzburg, DE	33
Nakayama	Naomi	University of Edinburgh, GB	7
Nebhnani	Poonam	CRAG, Barcelona, ES	44
Omary	Moutasem	HUJI, Jerusalem, IL	43
Perez	Miguel	MPIPZ, Köln, DE	17
Portheine	Stephan	University of Hamburg, DE	61
Romeiro Motta	Mariana	University of Hamburg, DE	59
Sabatini	Sabrina	University of Rome, IT	8
Sampathkumar	Arun	MPIMP, Golm, DE	7, 50
Sanz Mora	Oscar	University of Hamburg, DE	56
Schenke	Andreas	IPB, Halle, DE	24
Schnittger	Arp	University of Hamburg, DE	23, 56, 59, 61
Schwarz	Stefan	KWS Saat AG, Einbeck, DE	7
Strotmann	Vivian	HHU, Düsseldorf, DE	51
Stüwe	Benno	MPIMP, Potsdam, DE	51
Tasker-Brown	Billy	JIC, Norwich, GB	35
Thellmann	Martha	University of Zurich, CH	19
Tiago	Nunes	COS, University of Heidelberg, DE	13, 31
Tofanelli	Rachele	TUM, Freising, DE	46, 57
Vardanega	Isaia	HHU, Düsseldorf, DE	48
Vijayan	Athul	TUM, Freising, DE	46, 57
Volkava	Darya	CEITEC, Brno, CZ	36
Walter	Hannah	MPIPZ, Köln, DE	42
Zhang	Dan	University of Heidelberg, DE	13, 31
Zühl	Luise	MPIPZ, Köln, DE	60