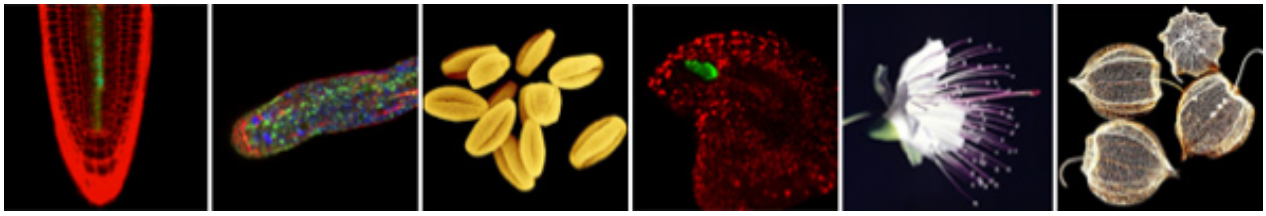

10th PhD School on Plant Development

04 – 06 October 2017

Zellingen-Retzbach, Germany

http://plant-development.org

PhD School

Welcome

Welcome to the 10th International PhD School on PLANT DEVELOPMENT

With the growing complexity of biological research projects in the last decades it has become increasingly important for scientists to communicate and collaborate across geographical and subject boundaries. Thus, young scientists not only need to be trained early in their career to present data at international meetings and discuss it with peers and leaders in the field, but also to organize chair sessions, as well as to network and identify potential collaboration partners. The PhD School on Plant Development has been set up in 2008 with the aim to train young scientists in these skills and give them a platform for communication and collaboration. Plant developmental biology is an exciting and fast moving field, which has seen many breakthroughs over the last decades, but apart from this PhD School does not have a signature meeting aimed at international graduate students and young postdocs.

Thus, the PhD School on Plant Development plays an important role within the community of plant scientists and in addition has served to increase the visibility and attractiveness of Germany for young researchers.

The International PhD School consists of ten successive sessions that are each introduced by an internationally renowned plenary speaker. Sessions cover *vegetative development* from roots to shoots, *reproductive development* from flowering to *embryogenesis*, as well as *hormone signaling*, *cell biology*, *epigenetics*, *small RNAs*, *programmed cell death*, and *synthetic biology*. It is expected that one to three PhD students/young scientists represent their research data in each of the sessions that shall be chaired by other PhD students/young scientists. It will be at their responsibility to initiate fruitful discussions and guide constructive conversations. Two poster sessions will provide extra time and informal opportunities for discussions.

The organizers are determined that this PhD school will be both scientifically rewarding and socially enjoyable, and sow a seed for further meetings and collaborations.

As organization team we are happy and proud to lead this exciting meeting into the future and to invite to the **10th International PhD School on Plant Development**. The meeting will take place **October 04-06, 2017, at Retzbach** (Benediktushöhe), close to Würzburg in Germany.

Finally, the wine tasting at the Juliusspital in Würzburg is expected to become again a cultural "highlight" of the meeting!

You are welcome to enjoy this meeting with us, both scientifically and socially!

The Organizers

Organization

Venue	**Tagungshaus Benediktushöhe** Benediktushöhe 1 97225 Zellingen-Retzbach, Germany Phone: +49 (0)9364/8098-0 Fax: +49 (0)9364/6276 E-mail: info@benediktushoehe.de
Organizers	**Markus Schmid** (University of Umeå) **Kay Schneitz** (TU München) **Rita Groß-Hardt** (University of Bremen)
Invited speakers	**Karen Alim** (MPI Göttingen) **Isabel Bäuerle** (University of Potsdam) **George Coupland** (MPI Cologne) **Thomas Dresselhaus** (University of Regensburg) **Jose Gutierrez-Marcos** (University of Warwick) **Simona Masiero** (University of Milan) **Moritz Nowack** (University of Ghent) **Ben Scheres** (University of Wageningen) **Rüdiger Simon** (University of Düsseldorf) **Miltos Tsiantis** (MPI Cologne)
Sponsors	**Gesellschaft für Entwicklungsbiologie (GfE), Cologne, Germany** **Deutsche Botanische Gesellschaft (DBG) e.V., Berlin, Germany** **Conviron Germany GmbH, Berlin, Germany**

Program – October 4, 2017

Arrival & Lunch *13:00 – 13:45*

Opening Remarks **13:45 – 14:00**

Session I: Evolutionary development **14:00 – 15:10**

Chair: Ziliang Hu

Plenary Speaker: Miltos Tsiantis (MPI Cologne) 14:00 – 14:40

"The genetic basis for diversification of leaf form:
from understanding to reconstructing"

Nicole van 't Wout Hofland (U Wageningen) 14:40 – 14:55
"The origin and evolution of vascular tissue regulators"

Roman Skokan (U Prague) 14:55 – 15:10
"Phytohormone auxin in plant algal ancestors"

Coffee break: *15:10 – 15:35*

Session II: Reproductive Development I **15:35 – 17:00**

Chair: Vilde Olsson

Plenary Speaker: George Coupland (MPI Cologne) 15:35 – 16:15

"Integration of regulatory pathways controlling floral transition at the shoot "

Savani Silva (U Utrecht) 16:15 – 16:30
"The nuts and bolts of stem elongation:
ATH1 mediates sensitivity to internal and external bolting signals"

Adriana Beatriz Arongaus (U Geneve) 16:30 – 16:45
"RUP2 is a repressor of flowering under UV-B"

Davide Marzi (U Rome) 16:45 – 17:00
"COP1 mediates light-controlled stamen growth in *Arabidopsis thaliana*"

Break: *17:00 – 17:15*

Program – October 4, 2017

Session III: Reproductive Development II **17:15 – 18:25**

Chair: Saurabh Joshi

Plenary Speaker: Thomas Dresselhaus (U Regensburg) 17:15 – 17:55

"Fertilization Mechanisms: A Comparison of Eudicots and Grasses"

Elvira Hernández-Lagana (U Montpellier) 17:55 – 18:10
"Dynamics of cell division patterns and their role on MMC fate acquisition during ovule primordium development in Arabidopsis"

Klára Aldorfová (U Prague) 18:10 – 18:25
"Characterisation of the exocyst complex SEC15 subunit in *A. thaliana"*

Thomas Thumberger (U Heidelberg, GfE) 18:25 – 18:35

"Gesellschaft für Entwicklungsbiologie - German Society of Developmental Biologists"

Dinner: ***18:35 – 19:35***

Poster Session I: **19:45 – 21:30**
(Beer and wine during and after poster session)

Program – October 5, 2017

Breakfast *07:00 – 08:30*

Session IV: Vegitative Development I: Root **08:45 – 10:10**

Chair: Vera Gorelova

Plenary Speaker: Ben Scheres (U Wageningen) 08:45 – 09:25

"An explanation of early embryo cell division planes based on microtubule dynamics"

Annika Wein (U Freiburg) 09:25 – 09:40
"Development of the root apical meristem in *Arabidopsis thaliana*"

Jorge Zamora Zaragoza (U Wageningen) 09:40 – 09:55
"Deciphering the Master's Code: Phosphorylation dependant regulation of RETINOBLASTOMA-RELATED1 protein functions."

Alja van der Schuren (U Lausanne) 09:55 – 10:10
"Molecular characterization of *Brachypodium distachyon* root development"

Coffee break: *10:10 – 10:45*

Session V: Synthetic Biology and Modelling **10:45 – 11:40**

Chair: Annika Wein

Plenary Speaker: Karen Alim (MPI Göttingen) 10:45 – 11:25

„Mechanical forces shaping morphology"

Thomas Nakel (U Bremen) 11:25 – 11:40
"Building and bypassing plant polyspermy barriers"

Lunch: *12:00 – 13:00*

Session VI: Vegitative Development II: Shoot **13:00 – 14:10**

Chair: Moritz Miebach

Plenary Speaker: Rüdiger Simon (U Düsseldorf) 13:00 – 13:40

"Dynamics of stem cell signalling pathways in plants"

Christophe Gaillochet (U Heidelberg) 13:40 – 13:55
"Control of plant cell fate transitions by transcriptional and hormonal signals"

Domenique André (U Umeá) 13:55 – 14:10
"Winter is coming – but how does a tree know that?"

Coffee break: *14:10 – 14:40*

Program – October 5, 2017

Session VII: Hormonal development **14:40 – 15:50**

Chair: Bahafid Elmehdi

Plenary Speaker: Simona Masiero (U Milan) 14:40 – 15:20

„A Fruittalk“

Vilde Olsson (U Oslo) 15:20 – 15:35
“IDA integrates the process of floral organ loss and pathogen defense responses in the abscission zone of Arabidopsis”

Jozef Lacek (U Prague) 15:35 – 15:50
“Searching for cellular determinants of auxin efflux carrier PIN-FORMED2 (PIN2) turnover regulation in Arabidopsis light- and dark-grown roots.”

Coffee break: *15:50 – 16:20*

Session VIII: Stress and cell death **16:20 – 17:35**

Chair: Domenique André

Plenary Speaker: Moritz Nowack (U Ghent) 16:20 – 17:00

“Meeting the Deadline: The Molecular Regulation of Programmed Cell Death During Plant Development”

Vera Gorelova (U Ghent) 17:00 – 17:15

“Dihydrofolate reductase/thymidylate synthase fine-tunes the folate status and controls redox homeostasis in plants”

Carsten Richter (Conviron, Berlin) 17:15 – 17:35
“Importance of growth chamber conditions for uniform plant growth”

Poster Session II: **17:35 – 18:35**

Conference Dinner *18:45 – 23:00*

Program – October 6, 2017

Breakfast *07:30 – 09:00*

Session IX: Small RNAs and Epigenetics **09:00 – 10:10**

Chair: Jorge Zamora Zaragoza

Plenary Speaker: Isabel Bäuerle (U Potsdam) 09:00 – 09:40

"Chromatin regulation of heat stress memory in plants"

Moritz Miebach (U Freiburg) 09:40 – 09:55
"Spatio-temporal transcriptional control of the *Arabidopsis* root stem cell niche by chromatin modifications"

Krishna Vasant Mutanwad (U Vienna) 09:55 – 10:10

"The role of O-Glycosylation in plant developmental transitions"

Coffee break: *10:10 – 10:45*

Session X: Embryogenesis **10:45 – 11:40**

Chair: Nicole van 't Wout Hofland

Plenary Speaker: Jose Gutierrez-Marcos (U Warwick) 10:45 – 11:25

"Stress memory in plants is underpinned by epigenomic changes"

Baojian Chen (U Wageningen) 11:25 – 11:40
"A Chemical Enhancer (C1) of Arabidopsis Somatic Embryogenesis"

Speakers Award/Closing Remarks **11:40 – 12:00**

Lunch: *12:00 – 13:00*

Departure: **13:00**

Abstracts – Invited Speakers

The genetic basis for diversification of leaf form: from understanding to reconstructing

Miltos Tsiantis

Max Planck Institute for Plant Breeding Research, Cologne, Germany.

A key challenge in biology is to understand how diversity in organismal form is generated. Genetic analyses in model systems have identified key regulators that sculpt the body plans of metazoa and seed plants. However, less is known about how the action of such regulators produces particular organ shapes, or how the balance of conservation versus divergence of such form regulating pathways generated the tremendous morphological diversity of multicellular eukaryotes. One impediment to answering these questions is the relative paucity of experimental platforms where genetic tools can be utilized to unambiguously study morphogenesis and its evolution in a genome-wide, unbiased fashion. To circumvent this problem we developed the *Arabidopsis thaliana* relative *Cardamine hirsuta* into a versatile system for studying morphological evolution. We aim to understand the molecular mechanisms through which leaf morphology evolved in these species, resulting in simple, undivided leaves in *A. thaliana* and dissected leaves with distinct leaflets in *C. hirsuta.* This presentation will discuss our progress towards understanding the morphogenetic pathways that specify dissected versus entire leaf shapes and that regulate the number, position and timing of leaflet production. It will also detail how studies in *C. hirsuta* have helped understand to what degree pathways underlying morphological variation between and within species overlap.

Integration of regulatory pathways controlling floral transition at the shoot meristem

George Coupland

Max Planck Institute for Plant Breeding Research, Cologne, Germany.

Reproduction is tightly controlled in all organisms. In plants, the induction of flowering is the first step in reproduction and involves transition of the vegetative shoot meristem to an inflorescence meristem that will give rise to flowers. This transition is also controlled by environmental cues such as day length and winter temperatures that ensure reproduction occurs at the optimal time. Sensitivity to these environmental cues is regulated by endogenous mechanisms associated with the developmental age of the shoot. We use *Arabidopsis thaliana* to decipher regulatory networks controlling these responses and exploit relative species, particularly perennial *Arabis alpina*, to determine how these networks change during evolution to confer ecologically significant differences in phenotype. Recently, we showed that SQUAMOSA BINDING PROTEIN LIKE 15 (SPL15) transcription factor has a central role in this process, directly integrating signals that mediate endogenous gibberellin signaling, the age of the plant and responses to winter temperatures. The presentation will discuss how this regulatory module involving the MADS box transcription factor FLC, the SPL transcription factor SPL15 and the microRNA156 plays a crucial role in controlling the timing and duration of flowering in the perennial system. By contrast, floral induction in response to day length is controlled by a different pathway that acts through the bZIP transcription factor FD at the shoot meristem, and how the activities of SPL15 and FD are regulated to integrate the responses to day length and winter temperature will be discussed.

Abstracts – Invited Speakers

Fertilization Mechanisms: A Comparison of Eudicots and Grasses

Thomas Dresselhaus

Cell Biology and Plant Biochemistry, Biochemie-Zentrum Regensburg, University of Regensburg, 93053 Regensburg, Germany; E-Mail: thomas.dresselhaus@ur.de

After leaving their aquatic environment land plants adapted to various environmental challenges and evolved specialized reproductive structures. Originally, their sperm cells were motile and capable to swim to egg cells. This mode of fertilization is still preserved in most mosses and ferns preferring moist environments. However, flowering plants or angiosperms, which dominate in colder and dryer habitats, evolved embedded gametophytes to protect egg and immobile sperm cells. Male gametophytes or pollen grains develop in anthers, while female gametophytes or embryo sacs are produced deeply inside the maternal tissues of ovule and ovary, respectively. After release, pollen grains of angiosperms are transported by wind or animals to female flower organs of the same species. They germinate, penetrate stigmatic tissue and grow through the style towards the ovule to deliver their passive sperm cell cargo. Inside the female gametophyte double fertilization is executed resulting in embryo and endosperm formation, respectively. To avoid fertilization failures and to promote outcrossing, angiosperms have developed various mechanisms to distinguish between own and alien pollen as well as pollen of the own species. Using Arabidopsis as a eudicot and maize as a grass model, I will compare morphological differences and focus on similar and distinct molecular mechanisms associated to fertilization. These include various hurdles along the pollen tube pathway, during sperm cell release and gamete activation. The analysis of genes involved in pollen adhesion, hydration and penetration, pollen tube growth and guidance as well as double fertilization and onset of embryo development will be presented. Some genes with key functions for fertilization and egg activation in maize lack homologs in Arabidopsis, while other genes are highly polymorphic. Molecular systems to discriminate own and foreign tubes will be presented and possibilities discussed to generate a new species.

Abstracts – Invited Speakers

An explanation of early embryo cell division planes based on microtubule dynamics

Ben Scheres, Bandan Chakrabortty, Viola Willemsen, Thijs de Zeeuw, Dolf Weijers, Bela Mulder.

Wageningen University, The Netherlands

The regularity of plant cell division patterns has been approached by several geometrical rules of which the first were proposed more than a century ago. Recent molecular work has identified cortical microtubule arrays (CMAs) as important factors in the determination of cell division plane, and interaction the CMA with microtubule binding proteins has been proposed to underlie the choice of cell division plane in rapidly growing cells. It has however remained unclear what determines the division plane in slowly dividing cells such as stem cells and early embryo cells.
We show experimentally that, in the early embryo, the CMA correctly predicts cell division planes. We have built a computational framework that allows simulation of microtubule dynamics on realistic cell shapes extracted from confocal stacks using *MorphographX* software. Using this framework we can show that a combination of basic microtubule dynamics rules (growth, catastrophe, collision) plus the implementation of a microtubule stability penalty at sharp cell edges is sufficient to explain the first divisions of an auxin-insensitive embryo. Next, we simulate the effect of auxin on microtubule stability and we show that this combination of biophysical rules on microtubule behaviour and the input of auxin on stability can explain the division pattern of the wildtype embryo from the 1- to the 16-cell stage, before the occurrence of growth. We discuss how our work provides a framework for molecule-based rules on the establishment of cell division planes in plants.

Mechanical forces shaping morphology

Karen Alim

Max Planck Institute for Dynamics and Self-Organization, Göttingen, Germany.

How reproducible shapes can emerge from the collective behaviour of individual cells is a major challenge for developmental biology. The notion of reproducibility requires the existence of some level of growth coordination between individual cells. Initially, gradients of biological messengers, so called morphogens, have been proposed as source of coordination. More recently, residual mechanical stresses as they arise for example in plant cell arrangements have been shown to act as instruction signals in parallel to biochemical gradients. Here we show the active role of mechanical forces in coordinating uniform cell growth patterns at the tip of the plant shoot. We further outline how shapes and three-dimensional forms can be reproducibly generated despite the abundance of stochasticity.

Abstracts – Invited Speakers

Dynamics of stem cell signalling pathways in plants

Rüdiger Simon

Institute for Developmental Genetics and Cluster of Excellence on Plant Sciences, Heinrich-Heine University, D-40225 Düsseldorf, Germany

Organogenesis in plants is based on precise cell fate determination of undifferentiated cells, which are derived from stem cells that reside in meristems. Cell fate determination depends on the position of a cell within the plant body. A number of mechanisms provide positional information to cells or cell populations: (1) plant hormones like auxin establish gradients or local concentration maxima; (2) secreted ligands and membrane associated receptors enables cell-cell signalling between adjacent cell layers; (3) transcription factors can act non-cell autonomously to integrate cellular responses, and (4) long range communication takes place by transport of signals between cells through plasmodesmata.
Our current understanding is that these mechanisms act in a concerted manner to govern plant development. I will report on our most recent results on receptor kinase signalling pathways that control the growth and maintenance of meristems. Using a toolbox of fluorescence imaging techniques in planta, we studied the dynamics of receptor assembly into larger complexes at distinct regions of the plasmamembranes upon the perception of their specific ligands. Furthermore, we found that alterations of receptor expression levels can have profound developmental consequences. Finally, I will try to link aspects of peptide-receptor and hormonal signalling pathways.

Work in the lab was supported by CEPLAS (DFG, EXC1028) and CRC1208.

A Fruittalk

Chiara Mizzotti, Bianca Galliani, Lisa Rotasperti, **Simona Masiero**

Dipartimento di Bioscienze Universita' degli studi di Milano, Italy

Given the fundamental nature of both the dietary and biological significance of fruit, molecular dissection of fruit growth and maturation has considerable interest. The yield and quality factors associated with fruits are of key importance to agricultural production, future improvements of fruit characteristics will rely on the comprehension of the mechanisms controlling fruit development and maturation.
Here we report our efforts to shed light into the molecular networks controlling fruit development in the model plant Arabidopsis thaliana. In order to identify genes whose products control Arabidopsis fruit development and maturation, a transcriptome analysis by RNA-deep-sequencing has been performed, comparing wild-type (WT) siliques, devoid of seeds, at 3, 6, 9 and 12 DPA, thus covering all the phases of silique development and maturation. The bioinformatics and statistical analysis of the data led to the identification of about thousand genes differentially expressed between early and late stage of silique development. Among all the different clusters, we are currently exploring the role of the NAC transcription factors. Our data strongly indicate that NAC proteins participate to the homeostasis of gibberellins and other hormones. Auxins, gibberellins, cytokinins, abscisic acid, and ethylene have been implicated at various stages of fruit development, to better understand their role in arabidopsis siliques growth and maturation we have measured GA and ABA content at 3, 6 9 and 12 DPA Arabidopsis fruits.

Abstracts – Invited Speakers

Meeting the Deadline: The Molecular Regulation of Programmed Cell Death During Plant Development

Moritz Nowack

VIB, Center for Plant Systems, University of Ghent, Belgium

Programmed cell death (PCD) is a fundamental cellular process leading to the actively organized suicide of a cell. An abundance of cell death processes occur throughout plant development and during the plant's responses to biotic and abiotic stresses. Evidence suggests that the correct initiation and execution of PCD is crucial for plant growth, reproduction, and fitness. In comparison to some types of animal PCD such as apoptosis, still only little is known about the molecular regulation of PCD in plants.

We investigate the molecular regulation of cell death processes during vegetative and generative plant development. We have taken a meta-analysis approach on an extensive compendium of cell death related publicly available transcriptome data sets. Our results suggest that there is little conservation between environmentally induced cell death (ePCD) and developmentally controlled cell death (dPCD) processes on the gene regulation level. However, there exists a core of genes that are commonly upregulated between different established and putative dPCD types. Taking these putative dPCD marker genes as a lead, we have established a set of dPCD model systems to investigate both conserved as well as distinct molecular pathways controlling dPCD in plants.

PCD still being a frontier of plant developmental research, we are focusing on tackling unresolved fundamental questions about the regulation of developmental PCD in plants: How do cells acquire PCD competency during differentiation? Which cell physiological signals trigger PCD execution at just the right moment? What are the actual mechanisms that disrupt the vital functions of a plant cell? We are obtaining answers to these questions through a comprehensive strategy combining cell-type specific transcriptomics, forward and reverse genetics, advanced live-cell imaging, biochemistry, cell physiological approaches, and computational modeling.

Fundamental research on plant PCD will not only enable us to understand cell death as a central biological principle, but also create new leads to tap the so far underexploited potential of PCD in agricultural applications.

Importance of growth chamber conditions for uniform plant growth

Carsten Richter

Conviron Germany GmbH, Berlin, Germany

Generation of comparative data e.g. for different cultivars, genotypes, or different experimental treatments very often requires growth of plants in uniform environments. Plants in growth chambers need highly specific conditions e.g. for evaporation of water, heat dissipation of irradiant energy, and gas exchange to achieve normal and consistent growth and physiological performance. Meeting these demands requires a growth room design very different e.g. from ordinary cooling rooms. Design factors crucial for achieving homogeneous and consistent plant growth as well as examples of plants showing non-uniform growth will be presented and discussed.

Abstracts – Invited Speakers

Chromatin regulation of heat stress memory in plants

Isabel Bäuerle

Institut für Biochemie und Biologie, Universität Potsdam, Potsdam-Golm, Germany

In nature, plants often encounter chronic or recurring stressful conditions. An increasing number of observations suggest that plants can remember a past exposure to stress in order to be better prepared for a future stress incident. My lab studies heat stress memory in plants as a model case for environmental stress memory. Seedlings acquire thermotolerance through a heat treatment at sublethal temperatures (priming heat stress (HS)) that enables them to survive an otherwise lethal HS. This thermotolerance is actively maintained for several days as indicated by the existence of mutants which are able to establish thermotolerance, but fail to maintain it.

We are currently exploring the molecular basis of HS memory in the model plant Arabidopsis thaliana. We have found that HS induces sustained histone methylation at HS memory-related loci that outlasts the transcriptional activity of these loci and hence marks them as recently transcriptionally active. At certain loci, this is correlated with hyper-induction of gene expression upon a recurring HS, - a signature readout of transcriptional memory. In a forward genetics approach, we have found that regulation of nucleosome occupancy is also required for sustained activation of memory gene expression. Sustained low nucleosome occupancy is mediated by the highly conserved FORGETTER1 protein, which is the orthologue of metazoan strawberry notch, through interaction with chromatin remodeling proteins.

In summary, the physiologically defined phenomenon of HS memory has a molecular equivalent in the transcriptional memory and associated changes in chromatin structure.

Stress memory in plants is underpinned by epigenomic changes

Jose Gutierrez-Marcos

Life Sciences, University of Warwick, UK

Eukaryotes are able to effectively adapt to both long term and short-term adverse environmental conditions through either genetic or epigenetic changes/response mechanisms. While the latter response is thought to be heritable, the precise mechanisms by which this occurs remains unknown. My group has found that plants develop a short-term stress memory underpinned by targeted epigenetic changes at labile genomic regions. These acquired epigenetic marks are associated with conditional heritable adaptive phenotypic stress responses but are not transmitted equally through parental sexual lineages. We have found the plant germline undergoes epigenetic reprogramming to remove these marks.

Abstracts – Oral Presentations

Session I: Evolutionary Biology

The origin and evolution of vascular tissue regulators

Nicole van 't Wout Hofland[1], Thomas Laux[2]

[1] Wageningen University and Research, The Netherlands
[2] BIOSS Centre for Biological Signalling Studies, Faculty of Biology, Albert-Ludwigs-University Freiburg, Freiburg, Germany

The development of vascular tissues is considered as one of the major steps in the evolution of land plants, allowing the transport of water, nutrients, hormones and other regulators in larger structures. Recently, TARGET OF MONOPTEROS5 (TMO5) and its homologs were shown to interact with LONESOME HIGHWAY (LHW) and its homologs. These TMO5/LHW heterodimers are bHLH transcription factors and control the number of cell files in the vascular tissues by triggering periclinal (longitudinal) cell divisions through local production of the phytohormone cytokinin (CK). Here, we investigate to what extent the recruitment of TMO5 and LHW has contributed to complexity of the vascular tissue over evolution.
First, an extensive bioinformatical analysis was performed, using PLAZA, phytozome and the 1kp database, in order to identify possible orthologs of TMO5 and LHW in plant species throughout the plant kingdom. We found orthologs of TMO5 in non-vascular plants like the liverwort *Marchantia polymorpha* (Mp). In contrast, LHW orthologs were solely identified in vascular plants. LHW could not be found back in plant species diverging before *Equisetum diffusum (horsetail)*. Ergo, LHW seems to be less conserved than TMO5. Next, we performed complementation studies in which we used the previously identified orthologs to rescue an *Arabidopsis tmo5* double mutant or a *lhw* single mutant, which have a monarch phenotype. Rescue was evaluated by reversion of the monarch root architecture back to the wild-type diarch structure. These results supported our initial bioinformatical findings.
Intriguingly, protein-protein interaction studies (BiFC and FRET-FLIM), showed that MpTMO5 was able to interact with the closest LHW ortholog of *Marchantia polymorpha* (pre-MpLHW). This suggests a co-evolution of TMO5 and LHW. Additionally, we observed that MpTMO5, as well as AtTMO5, were capable of homodimerization, suggesting TMO5 may have an additional function.
In this study we showed that the evolution of the TMO5/LHW dimer is correlated to the rise of vascular tissue in plants. Functions of preTMO5 and preLHW are yet to be determined.

Phytohormone auxin in plant algal ancestors

Roman Skokan[1,2], Stanislav Vosolsobě[1], Jan Petrášek[1,2]

[1] Department of Experimental Plant Biology, Faculty of Science, Charles University, 128 44 Prague 2, Czech Republic
[2] Institute of Experimental Botany, ASCR, 165 02 Prague 6, Czech Republic

Developmental regulation by plant hormone auxin has been amply described in land plants. The so called PIN-FORMED (PIN) family transporter proteins determine the direction of auxin flow in plant tissues and help establish local concentration maxima, which trigger a differential developmental response on cellular level. The evolutionary origin of auxin transport and other mechanisms of action have been shown genome-wise to lie in "charophyte" green algae, the freshwater ancestors of land plants.
For the first time we characterize experimentally an algal PIN homolog and bring evidence to its functionality and specificity as an auxin transporter. Due to growing available sequence information, we have also revisited the issue of PIN evolution and made some interesting observations, as well as supporting some earlier claims. Finally, we bring new insights regarding algal growth reaction to auxins, providing hints to its possible physiological role. Thus, we are slowly picking up leads in solving the mystery of ancient auxin action.

Abstracts – Oral Presentations

Session II: Reproductive Development I

The nuts and bolts of stem elongation: ATH1 mediates sensitivity to internal and external bolting signals

Savani Silva[1], Sjef Smeekens[1] and Marcel Proveniers[1]

[1] Molecular Plant Physiology, Department of Biology, Utrecht University, The Netherlands (S.S.Silva@uu.nl)

Premature bolting in crops such as lettuce, cabbage and sugar beet can cause significant yield losses in agriculture. Although bolting often coincides with the development of flowering, bolting and flowering are distinct processes. Floral primordia are generated from the SAM while stem elongation depends on the function of the rib meristem and rib zone. Little is known about the regulation of bolting, which is surprising as it is a fundamental developmental process in rosette plants. Here, we show that the TALE homeodomain transcription factor ATH1 is a key negative regulator of bolting in *Arabidopsis thaliana*. Overexpression of *ATH1* can completely repress bolting, but flowering is unaffected. In *ath1* mutants, internode elongation occurs between rosette leaves, a process that could be viewed as "ectopic" or "vegetative" bolting. During the vegetative phase the rib zone and rib meristem, remain inactive in wild type plants, but *ath1* seedlings already show strong elongation in the rib zone. Rosette internode elongation in *ath1* is affected by temperature, light quality, auxin, brassinosteroids and gibberellin. This phenotype can also be induced in the boundary gene mutants *bop1 bop2* and *lob,* which have previously been shown to function in the same regulatory pathway as ATH1. Rosette internode elongation cannot be induced in wild type plants, suggesting that ATH1 mediates sensitivity to both internal and external bolting signals during vegetative growth, possibly by blocking a more general regulatory module of cell elongation.

RUP2 is a repressor of flowering under UV-B

Adriana Beatriz Arongaus[1], Song Chen[1], Nina Glöckner[2], Klaus Harter[2] and Roman Ulm[1]

[1] Department of Botany and Plant Biology, University of Geneva, Switzerland
[2] Zentrum für Molekularbiologie der Pflanzen (ZMBP), Universität Tübingen, Germany

Time of flowering is a key trait in plant biology, with great impact on ecological and evolutionary processes. Precise timing of flowering relies on both endogenous and environmental signals. Sunlight, perceived by specific photoreceptors, represents a major environmental cue regulating the transition to flowering. Despite the fact that plants are exposed to ultraviolet-B radiation (UV-B) in natural environments, it is unknown whether UVR8 (UV RESISTANCE LOCUS 8) photoreceptor-mediated UV-B signaling affects photoperiodic regulation of flowering. We will present our latest findings showing that RUP2 / EFO2 (REPRESSOR OF UV-B PHOTOMORPHOGENESIS 2 / EARLY FLOWERING BY OVEREXPRESSION 2) is a crucial repressor of flowering specifically in the presence of UV-B. *rup2* mutants show a striking early flowering under short day (SD) conditions supplemented with UV-B but flower identical to wild type in the absence of UV-B. The early flowering of *rup2* correlates with upregulation of *FT* (*FLOWERING LOCUS T*) gene expression, whereas *CO* (*CONSTANS*) transcript levels are not altered. Moreover, the early flowering phenotype is dependent on FT and CO, as well as the UVR8 photoreceptor. We will also provide evidence suggesting that RUP2 activity directly affects the core of photoperiodic flowering time regulation. This work uncovers an important repressor of early flowering in SD supplemented with UV-B in the facultative long-day plant *Arabidopsis thaliana*

Abstracts – Oral Presentations

COP1 mediates light-controlled stamen growth in *Arabidopsis thaliana*

Davide Marzi[1], Nadia Napoli[1,2], Erica Spaziani[1], Minami Matsui[3], Giovanni Mele[2], Paolo Costantino[1], Giovanna Serino[1] and Maura Cardarelli[2]

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For successful reproduction of self-pollinating plants, stamens and pistils must develop and elongate in a coordinated fashion. Light, already known to play a central role in determining flowering time, could also modulate the growth of stamens and pistils.
We show here that COP1 (CONSTITUTIVE PHOTOMORPHOGENIC 1), a well characterized repressor of light-mediated responses, regulates stamen length, since *cop1* mutant alleles have a reduction in overall stamen growth from pre- to post-flower opening. In seedlings, COP1 promotes hypocotyl elongation through the targeted degradation of transcription factors such as HY5, HYH, PIL1 and HFR1. These transcription factors repress hypocotyl growth also by downregulating the expression of AUX/IAA19, a negative regulator of auxin signaling. Our results indicate that a similar regulatory module could control stamen elongation as well. Kinetic analyses of stamen growth reveal that HY5, HYH and HFR1 (but not PIL1) regulate stamen elongation, as their respective mutants have increased stamen length. Transcriptomic and quantitative PCR analyses indicate that, similarly to seedlings, *AUX/IAA19* is underexpressed in *cop1-4* stamens and overexpressed in *hy5 hyh* stamens. In addition, *cop1-4* stamens display higher expression levels of light-induced genes.
Taken together, our results highlight a novel role for COP1 in promoting stamen elongation, possibly linking light perception and auxin response during the development of flower organs and suggest that light could control stamen and hypocotyl growth through a shared set of signaling components. We are currently evaluating this hypothesis using a genetic and molecular approach.

Session III: Reproductive Development II

Dynamics of cell division patterns and their role on MMC fate acquisition during ovule primordium development in Arabidopsis

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The ovule primordium of *Arabidopsis* is the site where the onset of the female germline precursor or Megaspore Mother Cell (MMC) takes place. The MMC is developed from a single somatic subepidermal cell at the tip of the ovule. How the ovule primordium grows and achieves its shape while integrating the differentiation of the MMC remains poorly understood. Some genetic and epigenetic factors that partially regulate the number of MMCs formed within the ovule primordium have been identified. Yet, we still lack of a better understanding of how these regulators act in synergy with architectural cues to coordinate this process. Our recent findings show that, rather than cell volume, cell proliferation is the main parameter contributing to the growth of ovule primordium tissue surrounding the MMC. Furthermore, disturbances in cell growth and cell divisions during early ovule morphogenesis lead to ectopic MMCs and abnormal definition of MMC fate. To elucidate how cell division dynamics drives ovule primordia patterning and to investigate its influence in MMC fate and plasticity, we are currently generating high-resolution 3D static and live-imaging datasets of developing ovule primordia expressing fluorescent markers of mitotic events. Our perspectives include testing the hypothesis that specific ovule domains have a distinct influence in MMC establishment, by generating domain-targeted constructs to affect cell division parameters, combined with mutants displaying a lack or an ectopic MMCs formation.

Abstracts – Oral Presentations

Characterization of the exocyt complex SEC15 subunit in *A. thaliana*

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Morphogenesis, cell growth such as pollen tube growth, delivery of various cargoes, signalling and wide range of other processes is highly dependent on secretory path. Exocytosis, the final step of secretion is established by the interplay between the cytoskeleton and secretory pathway. The exocyst complex was described to function during exocytosis tethering secretory vesicles to the plasma membrane. SEC15 is a conventional exocyst subunit present in all eukaryotes that mediates interaction between the rest of the exocyst complex and the secretory vesicle. Here we present an insight into the role of the SEC15b exocyst subunit in plants.
We have characterized *sec15b Arabidopsis* mutant, which displays phenotype similar to the other exocyst mutants and includes decreased elongation of etiolated hypocotyls, altered formation of seed coat mucilage, and defective pollen tube growth. These features were analysed also during *sec15a* and *sec15b* complementation tests with SEC15A and SEC15B proteins. The sporophytic defects of *sec15b* mutant were complemented by both, SEC15A and SEC15B that appear to act redundantly in this case. In contrast, the gametophytic defect of *sec15a* mutant was fully complemented by SEC15A but not SEC15B, which potentially implicates functional diversification of these isoforms. Moreover, we have elucidated that SEC15B interacts directly with RAB GTPases in a GTP dependent manner. Taken these data together, the function of SEC15B appears to be evolutionary conserved, but does not completely complement *sec15a* mutation in male gametophyte.

Session IV: Vegetative Development I: Root

Development of the root apical meristem in *Arabidopsis thaliana*

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Plants form new organs from pluripotent stem cells throughout their lives and under changing environmental situations. Consequently, the maintenance of stem cells is crucial for plant survival. Although morphological changes of the Arabidopsis root apical meristem during ageing of the root were described 15 years ago, the behaviour of the genetic network regulating root meristem activity remains elusive. Knock-out mutants of *WOX5* (*WUSCHEL-RELATED HOMEOBOX 5*) are highly unorganized and mirror the phenotype of four-week-old roots, which display loss of stereotypic patterning and change from indeterminate to determinate growth. We therefore addressed whether WOX5 activity decreases with increasing root age. Furthermore, we asked whether the expression of other known root regulators, such as SCR (SCARECROW), SHR (SHORT-ROOT) or PLT (PLETHORA), alter their expression during ageing. Here I report the expression patterns of transcriptional and translational reporters of these genes during ageing. Additionally, morphological changes of the root apical meristem of wild type and *wox5-1* are compared using confocal microscopy and cell type markers.

Abstracts – Oral Presentations

Deciphering the Master's Code: Phosphorylation dependant regulation of RETINOBLASTOMA-RELATED1 protein functions.

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Development of multicellular organisms rely on the proper spatio-temporal balance of cell division, growth and differentiation. Since plant cells cannot move, this balance is particularly responsive to environment. RETINOBLASTOMA-RELATED1 (RBR1), has been shown to play an essential role integrating internal and environmental cues to make plant development adaptable to living conditions
Processes regulated by RBR1 in Arabidopsis thaliana include: Stem Cell (SC) maintenance and SC asymmetric cell division, DNA damage response and cell death, and immunity against pathogens. Thus, RBR is a master regulator of development that coordinates growth, stress and defense responses but, how is the master regulator regulated itself? And what are the molecular mechanisms allowing a single protein control so many different processes?
RBR interacts with and regulates Transcription Factors (TF) required for cell fate decisions. RBR-TF interactions are in turn controlled by RBR phosphorylation on different sites. An emerging paradigm on phosphorylation-dependent RBR regulation hypothesizes the existence of a so-called "phosphorylation code": specific combinations of phospho-sites control specific RBR-TFs interactions, allowing RBR to spatio-temporally coordinate cell fates with clockwork precision in a context dependent manner.
By systematically mutating RBR phosphosites in various combinations, mutant complementation experiments show that RBR-linked phenotypes could be separated. Moreover, Y2H screenings suggest that RBR interacts differentially with TF depending on its phosphorylation status. Our results strongly support the phosphorylation code hypothesis and constitute a big first step to decipher the master's code.

Molecular characterization of *Brachypodium distachyon* root development

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The *Poaceae* family of grasses is economically important and used worldwide for consumption. Amongst others it includes the crops sugarcane (*Saccharum spp*), maize (*Zea mays L.*) and rice (*Oryza sativa*). In contradiction to the commonly researched model plant *Arabidopsis thaliana*, these are all monocots. Recently it was discovered that the auxin-ethylene crosstalk in monocots might differ substantially from dicots[1]. In order to increase the knowledge on these differences, my PhD focuses on a model plant for monocots: *Brachypodium distachyon*.
Homologs of genes with importance in root development for *Arabidopsis*, were chosen for further research in *Brachypodium*. Mutants in these genes were obtained from an existing library of T-DNA insertion lines or by the production of CRISPR-Cas lines. After obtaining preliminary results, a mutant in AUXIN RESISTANT 1 (AUX1) was chosen for research in more detail. In Arabidopsis, mutations in the auxin influx carrier AUX1 lead to problems in gravitropic responses in the root[2]. Apart from a-gravitotropism, *Bdaux1* additionally displays significantly longer and thinner roots then wildtype. Cells elongate more, however less cells per cell type are formed. Shoot development is affected as well, since plants are sterile and remain small. Sterility is likely caused by non-synchronous maturation of pollen and gynoecium. These preliminary results seem to confirm that there are conceptual differences between *Arabidopsis thaliana* and *Brachypodium distachyon*. More research is being performed in order to determine the underlying mechanisms. This includes auxin analysis, localization studies, the production of CRISPR-Cas knockouts of BdAUX1 and complementations in both species.

1 Pacheco-Villalobos D. et al. **Disturbed local auxin homeostasis enhances cellular anisotropy and reveals alternative wiring of auxin-ethylene crosstalk in Brachypodium distachyon seminal roots**. *PLoS Genet* 2013.

2 Peret B et al. **AUX/LAX genes encode a family of auxin influx transporters that perform distinct functions during Arabidopsis development**. *Plant Cell* 2012

Abstracts – Oral Presentations

Session V: Synthetic Biology

Building and bypassing plant polyspermy barriers

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The ultimate goal for the survival of all species on earth is to reproduce. Flowering plants are internally fertilized and utilize pollen tubes for sperm transport. While initially an armada of pollen tubes set out to target the ovules, an individual ovule is typically only conquered by a single pollen tube. Supernumerary pollen tube attraction is prevented by a pollen tube block, which is mounted after fertilization (1-3). Key to the establishment of the pollen tube block is the degeneration of pollen tube attracting synergid cells, a process that requires Polycomb Repressive Complex 2 and an ethylene response cascade (3, 4). Here, we provide insights into some of the mechanisms underlying the establishment of the pollen tube block and discuss the developmental consequences associated with defective polyspermy barriers.

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Session VI: Vegetative Development II: Shoot

Control of plant cell fate transitions by transcriptional and hormonal signals

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Plant meristems carry pools of continuously active stem cells, whose activity is controlled by developmental and environmental signals. After stem cell division, daughter cells that exit the stem cell domain acquire transit amplifying cell identity before they are incorporated into organs and differentiate. In this study, we used an integrated approach to elucidate the role of *HECATE* (*HEC*) genes in regulating developmental trajectories of shoot stem cells in *Arabidopsis thaliana*. Our work reveals that *HEC* function stabilizes cell fate in distinct zones of the shoot meristem thereby controlling the spatio-temporal dynamics of stem cell differentiation. Importantly, this activity is concomitant with the local modulation of cellular responses to cytokinin and auxin, two key phytohormones regulating cell behaviour. Mechanistically, we show that HEC factors transcriptionally control and physically interact with MONOPTEROS (MP), a key regulator of auxin signalling, thereby modulating the autocatalytic stabilization of auxin signalling.

Abstracts – Oral Presentations

Winter is coming – but how does a tree know that?

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Changing environments pose a challenge to perennial plants, especially the harsh conditions of winter. These changes need to be anticipated and properly prepared for in order to survive. Here we show that in *Populus* trees these mechanisms are at least in part regulated by two orthologs of *FLOWERING LOCUS T* (*FT*) – one in response to photoperiod and one in response to temperature.
In the boreal regions, the two most drastic environmental changes are happening in spring and autumn with a quick rise or fall in temperatures and day length. Plants need to find a balance between maximising their growth period and avoiding potential frost damage. Thus both, the initiation and the cessation of growth, are tightly regulated. *FT1* is expressed during winter in vegetative and reproductive buds. In spring, warm temperatures repress *FT1* and long days promote growth through a *CONSTANS/FT2* regulon similar to the one in Arabidopsis. Short days in autumn cause *FT2* expression and subsequently growth to cease. So far, the individual roles of *FT1* and *FT2* in bud break and flowering are unknown, neither is the mechanism timing the short-day induced growth cessation well understood.
In this project we aim to identify their individual functions by using CRSIPR/Cas9 generated knockout mutants of FT1 and FT2, respectively, to study the trees' phenology. This approach is combined with gene expression analyses and studies of natural variation among Swedish aspen.

Session VII: Hormonal Control of Development

IDA integrates the process of floral organ loss and pathogen defense responses in the abscission zone of Arabidopsis

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Plants use small peptide ligands and plasma-membrane receptor proteins to regulate important developmental processes and to respond to both biotic and abiotic stresses. The small peptide ligand INFLORESCENCE DEFICIENT IN ABCISSION (IDA) regulates floral organ abscission in Arabidopsis thaliana through the receptor-like kinases (RLK) HAESA (HAE) and HAESA-LIKE 2 (HSL2) which act redundantly to regulate the cell separation process leading to abscission. Here we suggest that IDA, in addition to its developmental role, as well has a role in regulating a FLS2 dependent innate immune response in the abscission zone. As with flg22, IDA induces a receptor dependent rapid release of cytosolic Ca2+ and an extracellular ROS-burst. In plant cells, Ca^{2+} transduces hormonal and environmental signals to diverse Ca^{2+} sensors and relays them to downstream target effectors. Ca^{2+} has been implicated in multiple defense- and developmental responses and links between defense, hormonal signaling and Ca^{2+} have been shown. Additional common components of the IDA and flg22 induced signaling pathways are the use of SERKs as co-receptors and downstream MAPK cascades giving the possibility for crosstalk between the intracellular responses. Interestingly, exogenously application of flg22 enhances IDA expression in the abscission zone. Also, we suspect that important defense genes in the flg22 induced response are upregulated in response to IDA. Taken together we suspects IDA to be an enhancer for the FLS2 induced pathogen defense pathway giving increased protection of exposed cells after the abscission process.

Abstracts – Oral Presentations

Searching for cellular determinants of auxin efflux carrier PIN-FORMED2 (PIN2) turnover regulation in Arabidopsis light- and dark-grown roots.

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The sessile life style of plants forces them to adapt constantly to changing environmental conditions. From subtle events at cellular level till obvious changes of plant architecture, phytohormones interact in a complex network to ensure proper plant growth. The *Arabidopsis thaliana* root is a well-studied model system for various environmental adaptations and hormonal crosstalk. Auxin modulates plant architecture during development and adaptation processes, in a concentration-dependent manner. Auxin gradients are generated by active directional cell-to-cell auxin transport through plasma membrane located auxin import and export carriers, whose abundance is tightly regulated. Among others, it was shown that proper root growth is associated with the regulation of endocytic trafficking of the auxin efflux carrier PIN2. Cellular components as the cytoskeleton or the lytic vacuole are important players in maintaining auxin carrier turnover between the PM and their pathway for degradation via the lytic vacuole. We observed that PIN2::PIN2:mCherry intracellular distribution differs in roots of seedlings that were exposed to light, i.e. grown on vertical plates with 16/8 L/D period, and exposed to dark, i.e. grown on vertical plates with 16/8 L/D period in D-Root system. Therefore, by using crosses of PIN2-mCherry and PIN2-GFP marker lines with selected mutants affected in actin and vacuolar dynamics we now follow actin cytoskeleton assembly and vacuolar morphology under different light growth conditions, and the resulting impact on PIN2 turnover and abundance.

Abstracts – Oral Presentations

Session VIII: Stress and Cell death

Dihydrofolate reductase/thymidylate synthase fine-tunes the folate status and controls redox homeostasis in plants

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Folates, also known as B9 vitamins, are essential cofactors in one-carbon (C1) metabolism. Since the C1 transfer reactions are involved in synthesis of proteins, nucleic acids, vitamin B5 and other important biomolecules, folates are absolutely essential for all living organisms. Disturbances to folate biosynthesis result in severe repression of plant growth and development. Although the biochemical processes wherein various folate derivatives take part are well-known, our understanding of the physiological roles of folates and folate pathway enzymes in plants is far from being advanced. Our work is focused on investigating the roles of folates in C1 metabolism in plants.

Our aim is to study the DHFR-TS (dihydrofolate reductase - thymidylate synthase) gene family that implements the penultimate step in the folate biosynthesis pathway in Arabidopsis. The Arabidopsis DHFR-TS family is comprised of three enzymes (THY1, THY2, THY3) that couple folate biosynthesis (DHFR activity) with DNA synthesis (TS activity). We show that one of the DHFR-TS family members (THY3) functions as an inhibitor of its homologs, thus regulating folate abundance in plant cells. Our work draws a parallel with a recently discovered role of folate metabolism in animals in the modulation of cellular redox state by contributing to NADPH production, thus allowing a plant to cope with oxidative stress.

Furthermore, our work underscores the notion that folate abundance affects plant development and reveals its regulation through adjustment of DHFR activity.

Abstracts – Oral Presentations

Session IX: Small RNAs and Epigenetics

Spatio-temporal transcriptional control of the *Arabidopsis* root stem cell niche by chromatin modifications

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Plants exhibit lifelong formation of new tissues and organs that is enabled by pluripotent stem cells in the plant meristems. In the root stem cell niche, the columella stem cells (CSCs) are maintained by the transcriptional repressor WOX5, which is specifically expressed in the organizer cells of the quiescent centre (QC) from where it moves to the CSCs. WOX5 acts by recruiting TOPLESS (TPL) and the HISTONE DEACETYLASE 19 (HDA19), thus modifying the epigenetic status of its target genes. Since WOX5 is not only important in the maintenance of CSCs, but also can reprogram already differentiated columella cells (CCs) into induced columella stem cells (iCSCs), it enables us to obtain sufficient cell numbers to investigate the epigenetic signature of plant stem cells. This will be done by Chromatin immunoprecipitation (ChIP) from nuclei purified by Fluorescent Activated Nuclei Sorting (FANS). In a second step, we will adopt ChIP protocols to small cell numbers. This shall enable us to compare the chromatin signature between specific cell types of the root columella and to explore the dynamic changes of cell fate transitions from an undifferentiated cell towards a differentiated.

The role of O-Glycosylation in plant developmental transitions

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Plants undergo several developmental transitions in the course of their life cycle, marked by specific morphological changes such as altered leaf morphology, the formation of trichomes and the onset of flowering. In Arabidopsis, mutants in the O-fucosyltransferase SPINDLY (SPY) show an accelerated transition from the juvenile to the adult phase, as well as early flowering. O-fucosylation is a posttranslational modification and closly connected to O-GlcNAcylation, where either a single N-acetyl-β-D glucosamine (GlcNAc) or fucose is O-linked to side chains of serine- or threonine residues in nuclear and cytoplasmic proteins. In the current working model O-fucosylation and O-GlcNAcylation compete for the same targets, but affect protein-protein interaction of these targets in different ways. It has been suggested that O-GlcNAcylation may integrate stress and nutrient availability to basic cellular processes, as the availability of UDP-GlcNAc reflects the overall state of nutrition. On the other hand, the timing of developmental transitions in plants is regulated by the amount of available sugar accumulating during plant growth. This process is mediated by balancing the ratio between miRNA156 and miRNA172, therefore regulating the downstream miRNA156 target genes, a family of SPL-transcription factors. Given the observed early transition phenotypes in *spy*-mutants, this project aims to investigate a potential role of SPY in developmental transitions, focusing on the juvenile to adult phase change and the regulation of flowering time. These experiments will establish if SPY regulates developmental transitions via the miRNA156 pathway and/or SPL transcription factors, potentially in response to the accumulation of sugar during plant growth.

Abstracts – Oral Presentations

Session X: Embryogenesis

A Chemical Enhancer (C1) of Arabidopsis Somatic Embryogenesis

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Somatic embryogenesis (SE), in which embryos are derived from vegetative cells of the plant, is generally induced by exposing explants to exogenous growth regulators. SE can be induced by 2,4-D in arabidopsis from different explants with varying efficiency. We used a chemical genomics approach to identify genetic factors that enhance 2,4-D mediated SE from germinating seeds in arabidopsis. Screening of the LATCA library identified several enhancers, of which 4-chloro-N-methyl-N-(2-methylphenyl) benzenesulfonamide, named C1, was the most powerful, increasing the number of responding explants from 20% in the control to 70% after C1-treatment. The timing and location of somatic embryo formation did not appear to be different between C1 treatment and control. Moreover, microarray analysis showed that a short C1 treatment induced changes in gene expression that were also observed after 2,4-D treatment, including the expression of many genes involved in seed specific ABA pathways. Together this data suggests that C1 and 2,4-D impinge on the same developmental pathway. Here we report on the role of the ABA receptor PYL10 in 2,4-D/C1-mediated somatic embryogenesis.

Abstracts – Poster

Session I: Evolutionary Development

P-01 Robustness of floral development: of evolutionary importance?

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Robustness is a ubiquitously observed property of organisms and is a fundamental feature of complex evolvable systems. It can be described as the persistence of a trait under perturbations. In theory, a system must be able to maintain a high degree of robustness for it to function in erratic environments. Sometimes the apparent high degree of robustness is hard to explain. A good example of this is the floral structure of the Brassicaceae (mustard family). This family of angiosperms is composed of 4000 species and many of these have flowers composed of 4 petals, 4 sepals, 6 stamens and two carpels. Since the Brassicaceae either are predominately selfing or have general pollinators there appears to be no obvious explanation for this high level of conservation of the floral groundplan.
The aim of this project is to test whether robustness was selected for during evolution using mutation accumulation (MA) lines. We also target candidate genes that may play a role in floral robustness using the CRISPR-Cas9 method of genome editing. Preliminary results indicate that the genetic robustness of organ length is not as strong as the robustness of floral organ number. Preliminary data also indicate that there may be a gradient of robustness in floral organ number across the different organ types. Maintaining a robust number for some organ types might thus be more critical than for others.

P-02 How does *CUC1* regulate leaf shape and diversity

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Here we investigate the function of *CUC1*(*CUP SHAPED COTYLEDON1*) in the diversification of leaf forms between simple leafed Arabidopsis thaliana and compound leafed Cardamine hirsuta. *CUC* transcription factors are conserved regulators in leaf margin dissection and leaflet formation. *ChCUC1*,*ChCUC2* and *ChCUC3* function redundantly and they are required for the leaflet formation in *C. hirsuta*. Recently we discovered that *ChCUC1* has species specific expression in leaves in *C.hirsuta*. Moreover, interspecies gene transfer of *ChCUC1* allele into *A.thaliana* is sufficient to increase leaf complexity. On this basis we hypothesize that redeployment of *ChCUC1* in leaves result in formation of leaflets instead of serrations. However, the mechanism underlying *ChCUC1* regulating cell division, cell polarity and thus leaf marginal patterning remains largely unknown. To address this issue, we employed unbiased approaches including Chip-seq and RNA-seq to identify the downstream regulating genes of *ChCUC1*. We have identified the flavin-binding monooxygenase family protein YUCCA4 is the direct target of *ChCUC1*. This infers that *ChCUC1* contribute to the leaflet formation by promoting local auxin biosynthesis and modifying the PIN1 and auxin-related gene regulatory network.

Abstracts – Poster

Session II: Reproductive Development I

P-03 Bioactive molecules in the control of flowering time

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Flowering time is one of the most relevant traits influencing crop productivity and yield. The identification of natural or synthetic compounds bioactive in the control of floral induction is of great interest. Identified compounds could allow us to fine-tune flowering time in crops, adapting it to the most favourable environmental conditions. Also, the characterization of the mechanism of action for each molecule will provide us with novel information on the regulation of flowering time.
In order to identify bioactive compounds in the control of flowering time we have undertaken two different approaches. First, we have carried out a chemical genomics screening to identify small molecules with potential to control the expression of the florigen, *FLOWERING LOCUS T* (*FT*), in Arabidopsis. We have used transgenic plants expressing the *β-glucuronidase* gene (*GUS*) under the control of the *FT* promoter. Positive hits from the screening are currently being confirmed by secondary screenings to choose the most promising molecules.
Our second approach is oriented towards the characterization of endogenous molecules contributing to the control of flowering. We will characterize metabolic profiles of leaf and shoot apical meristem tissues during floral transition. We will compare metabolite changes between wild type and mutant samples defective in photoperiod perception (*constans* and *ft* mutants) after a short day to long day shift, treatment that induces flowering in the wild type.
In summary, we are analysing floral induction by means of chemical biology and metabolic profiling to identify candidate synthetic and endogenous molecules with potential to control flowering time.

P-04 Interaction between SVP and FLM isoforms have an antagonistic effect on ambient temperature-responsive flowering

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SHORT VEGETATIVE PHASE (SVP), a MADS-box protein that acts as a transcriptional repressor, is a main component of ambient temperature-responsive flowering. SVP associates with other transcription factors to form a high-order regulatory complex. The most well-known co-factor of SVP is FLOWERING LOCUS M (FLM), which produces various splicing variants, depending on temperature changes. We previously revealed that SVP protein is unstable at high temperatures, leading to less abundant repressor complex with FLM-β, which is the pivotal splicing isoform and actively expressed at low temperatures.
Here, we further investigated the temperature sensing mechanism modulated by SVP and FLM proteins. We found that SVP ubiquitination was remarkably increased at high temperatures. We identified a putative E3 ligase that is involved in SVP protein degradation. Consistent with its role, its knock-out mutants showed delayed flowering time at high temperatures. We also found that SVP translocation varied depending on both temperature and the presence of FLM isoforms. SVP localized in the nucleus at low temperatures, whereas abundant SVP were found in the cytoplasmic region at high temperatures. Interestingly, SVP localization was limited to the cytoplasm in the absence of FLM and the nuclear localization of SVP was recovered by co-expression of SVP with FLM-β. In contrast, over-expression of FLM-δ accelerated SVP degradation at high temperatures. Taken together, we suggested that the interaction with FLM-β is important for SVP nuclear translocation at low temperatures and the association with FLM-δ makes SVP unstable at high temperatures.

Abstracts – Poster

Session III: Reproductive Development II

P-05 Analysis of SUB-dependent signal transduction in *Arabidopsis*

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STRUBBELIG (SUB) is an atypical receptor-like kinase localized at plasma membrane and plasmodesmata, which mediate tissue morphogenesis in *Arabidopsis thaliana. sub* mutants exhibit defects in the initiation and outgrowth of ovule intertegument, alteration of floral shape, stem morphology, leaf shape, root hair patterning and reduced plant height. In previous works was found that the RLK-encoding genes *ANGUSTIFOLIA, QUIRKY* and *ZERZAUST* contribute to the SUB-dependent signal, since plants with recessive mutation in these three genes, exhibited a *sub*-like phenotype. This work aims to the identification and characterisation of suppressor genes of *SUB* through a genetic screening, consisting of the finding of recessive mutation which cause the rescue of the wild-type phenotype in *sub-1* mutants and a mapping-by-sequencing analysis of outcrossed *sub-1* and *sub-9* mutants. Then, my work focus on the molecular and phenotypic characterisation of the cellulose-biosynthesis inhibition in *sub* mutants

Session IV: Vegetative Development II: Shoot

P-06 How organ primordia are generated at the shoot apical meristem: Repression of KNOX homeobox genes through the JAGGED LATERAL ORGANS (JLO)/ASYMMETRIC LEAVES complex

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Growth of the shoot is controlled by the shoot apical meristem (SAM), which gives rise to organs such as the leaves and flowers. Once primordia have been initiated, the separation between lateral organs and the meristem is established by the creation of a functional boundary domain with low rates of cell proliferation. In the SAM the initiation and development of the lateral organ requires repression of class I KNOTTED1 LIKE HOMEOBOX (KNOX) genes. KNOX genes promote stem cell activity by maintaining the stem cell self-renewal. It was shown previously by Chromatin Immunoprecipitation (ChIP) experiments that the repression of KNOX genes is regulated by the ASYMMETRIC LEAVES complex. The transcription factors ASYMMETRIC LEAVES 2 (AS2) together with ASYMMETRIC LEAVES1 (AS1) recruits the polycomb-repressive complex 2 (PRC2) to establish repression of KNOX genes in organs. The JAGGED LATERAL ORGANS (JLO) gene, a member of the LATERAL ORGAN BOUNDARY DOMAIN (LBD) gene family, is required for organ development and the establishment of morphological meristem-to-organ boundaries. JLO was shown to physically interact with the ASYMMETRIC LEAVES complex, and to form hetero-trimeric protein complexes with AS1 and AS2 both in vitro and in vivo. Furthermore, loss of JLO function causes a developmental arrest at embryonic or early seedling stages due to misregulation of auxin efflux carriers. We will now investigate when and where during shoot apical meristem development JLO/AS complexes are forming, and how these complexes regulate KNOX gene expression in space and time. For that, I have generated fluorescent reporter lines for AS1, AS2 and JLO, in order to follow complex formation using in vivo FRET/FLIM. Details on the approach I have taken, and first results will be presented.

Abstracts – Poster

P-07 Regulation via miR394b-LCR pathway in shoot apical meristem maintenance

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In *Arabidopsis thaliana* all the above-ground organs are initiated from a small group of stem cells in the shoot apical meristem (SAM). The SAM can be divided into three clonal cell layers (L1-L3) and several functional zones. The plant homeobox gene *WUSCHEL* is expressed in a small group of L3 layer cells, named the Organizing Center (OC). Its protein can move into the stem cells and activate the expression of the CLV3 gene. WUS and CLV3 form a negative feedback loop, which is one part of shoot meristem self-regulation.
miR394 promotes WUS function in stem cell maintenance by guiding mRNA cleavage of the F-box protein LCR. The MIR394B gene is expressed initially in the whole embryo during the early stages but from the late globular stage, its expression will be gradually become restricted to the L1.
My goal is to reveal how MIR394B expression is regulated. To find possible regulators of MIR394B, my strategy is to analyze promoter truncation and to perform a yeast one hybrid screen. First, we constructed promoter deletion constructs of different lengths and used them to drive a 3xNLS-DsRed construct. For comparison all deletion constructs are to be observed in tandem with the full miR394B promoter driving YFP. The expression of all constructs will be analyzed to identify cis elements necessary for the correct expression pattern of MIR394B. These sequences I will use to do the yeast one hybrid screen for candidate transcription factors that can regulate MIR394B expression.

P-08 Cryptochromes: negative regulators of phototropic response in green Arabidopsis seedlings

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Phototropism is an adaptive growth response, enabling plants to optimize light capture. In flowering plants, the phototropic response is primarily regulated by unidirectional blue light gradient, sensed by phototropins (phot1 and phot2 in Arabidopsis), a family of blue light photoreceptors. The blue light activation of phototropins triggers the formation of the auxin gradient that lead an hypocotyl asymmetric growth. However, also other family of photoreceptors, such as phytochromes and cryptochromes (sensing red/far red and blue light), are involved in asymmetrical hypocotyl growth during phototropic response. Recently, it was shown that the changing in red/far red ratio, perceived by photoreceptor phyB, affects the phototropic response of Arabidopsis green seedlings. Now we demonstrate that also the changing in environmental blue light affects phototropism in green seedlings, and we propose cryptochromes as negative regulators of this growth response. In fact, in open environments (high blue light) the phototropic response is inhibited by cryptochromes. In contrast, in limiting blue light condition, green seedlings show a robust phototropic response. We also identified the PHYTOCHROME INTERACTING FACTORs (PIF) as positive regulators of this process and we show a clear epistatic relationship between *cry1* and *pif* mutants.
This work clarify how the changing in environmental blue light, sensed by cryptochromes-PIFs pathway, regulates the phototropic response in green Arabidopsis seedlings.

Abstracts – Poster

P-09 A conserved carbon-starvation response underlies bud dormancy in woody and herbaceous species

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Shoots are formed from axillary meristems and buds, whose activity is controlled by endogenous and environmental signals that modulate growth and development. These signals are translated into a local response, in meristems and buds, of growth or quiescence. Although some key genes involved in the onset of bud latency have been identified, the gene regulatory networks (GRN) controlled by these genes are not yet well defined. Moreover, it has not been determined whether bud dormancy induced by environmental cues, such as low red to far-red light ratio, shares genetic mechanisms with bud dormancy induced by other causes, such as apical dominance or a short-day photoperiod. We have reanalyzed public transcriptomic datasets that compare quiescent and active axillary buds of Arabidopsis with datasets of axillary buds of the woody species Vitis vinifera (grapevine) and Populus tremula x Populus alba (poplar) during the bud growth-to-dormancy transition. Our aim was to identify potentially common GRN induced during the process that leads to para-, eco- and endodormancy in buds. In Arabidopsis buds entering eco- or paradormancy, we identified four induced, interrelated GRN that correspond to a carbon (C) starvation syndrome, typical of tissues undergoing C depletion. This response is also detectable in poplar and grapevine buds before and during the transition to dormancy. In all eukaryotes, C limiting conditions are coupled to growth arrest and latency. Thus, bud dormancy might in part be a consequence of the underlying C-starvation syndrome triggered by environmental and endogenous cues that signal conditions unfavorable for sustained shoot growth.

Abstracts – Poster

Session V: Synthetic Biology

P-10 Measuring the protrusive forces of pollen tubes

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Gamete fusion is key to reproductive success. In many animals, egg cells are challenged by huge amounts of sperm cells to increase the likeliness of an egg becoming fertilized. By contrast, flowering plants produce immotile sperm, which develop in pollen. Upon deposition of pollen on the stigma, pollen grains germinate and grow into long tubular structures, which target the egg-containing ovules. The amount of pollen tubes largely exceeds the number of ovules and the ability to grow fast and to get to the ovule first critically influences reproductive success of the sperm delivered. We have established a home-built force microscopic setup to make pollen tube growth accessible. This technology enabled us to determine the protrusion forces of this remarkably fast growing structure. Additionally, with Atomic Force Microscopy (AFM), we were able to determine stiffness and topography of the pollen tube.

P-11 Revealing the Importance of Transctription Kinetics in Plant Development

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Functional gene expression is crucial for plant development, by which plants transform from zygotes into multi-cellular living forms. RNA polymerase II (Pol II) plays an irreplaceable role in ensuring function gene expression by conducting the transcription of all genes and coordinating their co-transcriptional mRNA processing events (i.e. splicing). Recently, A kinetic model of transcription offers a better way to understand the connection between transcription and co-transcriptional events. However, obtaining direct support for the biological significance of the kinetic model is often challenging due to pleiotropic effects of perturbing transcription. The transcription elongation speed is a core parameter in the kinetic model of transcription, which is tightly linked to co-transcriptional events. To address the biological significance of transcription speed in plant development we are focusing on the transcription kinetics of Pol II in model plant *Arabidopsis*. We have generated plant lines harboring speed-altering point mutations in the Pol II enzyme, which have been reported in budding yeast to be able to change transcription speed. Our kinetic *Arabidopsis* mutants illustrate that accelerating and decelerating transcription speed both have profound biological implications on plant architecture, life-span and fitness. This provides strong biological support for the current kinetic model of Pol II transcription. In molecular level, wide-ranging pre-mRNA processing changes have been detected in our kinetic Pol II mutants, which has a huge impact on functional gene expression. In summary, our research illustrates the physiological and molecular significance of the Pol II transcription speed.

Abstracts – Poster

Session VII: Hormonal Control of Development

P-12 TOR as an integrator of metabolism and environmental signals controls hormonal networks in the plant stem cell niche

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The molecular mechanisms of how plants integrate internal and external developmental signals remain enigmatic. Two major determinants of plant development are the nutritional status of the plant and the light regime, both are sensed, transmitted and integrated in the regulation of meristem activity. Depending on the environment, this information modulates growth patterning within the stem cell populations and changes plant development and morphology. Our work links sensing of plant metabolism and the environment with the expression of the stem cell maintenance factor WUSCHEL (WUS). Furthermore, we show that WUS expression is dependent on an interplay between “Target of Rapamycin” (TOR) and signaling cascades of the plant hormone cytokinin. Together, these factors span an integrative network for the regulation of plant development. We present biochemical and genetic evidence for an intimate connection between signaling relays of the plant hormone cytokinin and activation of the TOR in the shoot apical meristem (SAM).

P-13 Identification of mutant candidates defective in auxin mediated inhibition of endocytosis.

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Auxin is a small signaling molecule that through its accumulation patterns acts as a trigger of multiple processes in plant development. Its differential distribution across the plant is an outcome of long transport through the phloem and notably through cell-to-cell transport that is, among other molecular players, regulated by polarly localized auxin efflux carriers (PIN proteins) which enable the directional auxin flow between cells.

This asymmetric PM localization of PINs within cells is maintained by the action of endocytic machinery. PIN endocytosis can be visualized utilizing BFA (Brefeldin A) that leads to internalization of those transporters into so-called ‘BFA compartments’. Conversely, auxin inhibits endocytosis retaining PINs on the PM thus resulting in inhibition of BFA body formation. Those observations are well documented in literature however; detailed understanding of the inhibitory effect of auxin on endocytosis is still lacking.

Striving to answer this question we identified mutants resistant to auxin effect on endocytosis. Here we present one of the selected candidates showing clear BFA bodies formation after short treatment with media containing both NAA and BFA. We also show an initial analysis of cellular and morphological phenotypes of this mutant.

Abstracts – Poster

P-14 Characterization of the mutants displaying auxin related defects in intracellular trafficking.

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The asymmetric distribution of the hormone auxin controls cell elongation, axis formation and organ differentiation as well as plant tropisms. Auxin directional flow and the resulting maxima are formed largely by the action of polarly localized efflux carriers named PIN proteins. They are constantly cycling between the plasma membrane (PM) and endosomal compartments. Auxin has been shown to inhibit endocytosis thus promoting retention of PINs on the PM consequently enhancing its own transport.

To further characterize the mechanism by which auxin regulates internalization of PM cargos, we employed a pharmacological approach to interfere with intracellular trafficking of PIN proteins. We used Brefeldin A (BFA), an inhibitor of PIN recycling from endosomes to the PM causing their acummulation in the BFA-compartments. This BFA–induced internalization is efficiently inhibited by auxin.

From a forward genetic screen, in which EMS-mutagenized seedlings were co-treated with BFA and auxin, we have selected candidates resistant to auxin effect on vesicular trafficking. Here we present the cellular and seedling phenotypes of one such mutant. We hope that the characterization of this mutant candidate will broaden the knowledge of the mechanism by which auxin inhibits endocytosis.

Session VIII: Stress and Cell death

P-15 *CIPK9*: a calcineurin B-like protein-interacting protein kinase from *Nitraria tangutorum*, confers the salt resistance in *Arabidopsis*

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CBL-interacting protein kinases (*CIPKs*) play essential roles in plant response to abiotic stresses. In order to better understand salt tolerance, we cloned and analyzed the *NtCIPK9* gene from the halophyte *Nitraria tangutorum*.

1. Phylogenetic analysis of NtCIPK9 shows that *NtCIPK9* is in a sister clade together with the *Arabidopsis AtCIPK9* gene that is thought to localize to the plasma membrane.

2. *NtCIPK9*-overexpressing Arabidopsis plants showed a higher seed germination rate, longer root length and higher salt tolerance than wild type seedlings.

3. The *NtCIPK9* gene showed a higher expression level in the root of *Nitraria tangutorum* under normal growth conditions, whereas after 500 mM NaCl treatment, a higher expression level was found in the blade.

4. Overexpression of *NtCIPK9* could enhance the expression level of *AtHKT1* (K^+ transporter) 4-fold and improve the content of K^+ in transgenic seedlings after 150mM NaCl treatment for 24 hours.

Thus, we conclude that *NtCIPK9* increases transgenic plants salt tolerance and mitigates damage associated with salt stress by regulating expression of genes controlling ion homeostasis.

Abstracts – Poster

Session IX: Small RNAs and Epigenetics

P-16 *De novo* DNA methylation in mature seeds as a mechanism to regulate dormancy in response to cold temperature during seed set

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Primary dormancy is the absence of a viable seed to germinate in spite of favourable conditions. It is a complex property of the seed that is genetically determined and influenced by environmental cues, such as temperature. It has been described that, when plants are exposed to cold temperatures during seed set, their progeny will display increased dormancy levels. Despite the identification of some molecular actors involved in this phenomenon, the molecular mechanism controlling how seeds can remember that the parental plant grew in cold temperatures remains elusive.
One hypothesis is that cold temperatures could induce some epigenetic modifications, such as DNA methylation, in living seed's tissues (in the embryo or endosperm) and this epigenetic modifications could function as a mark for memory.
Here we try to understand the relationship between *de novo* DNA methylation and seed dormancy induced by cold maturation using the *ALLANTOINASE (ALN*) locus as reference. *ALN* has been previously shown to be imprinted in the endosperm and having an expression level correlating with dormancy depth. Therefore, it is a suitable locus for our analysis regarding seed dormancy and cold temperature.

P-17 Regulatory network interactions in tomato fruits ripening

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Fruit ripening involves changes of biochemical and physiological processes. In tomato, transcription factors (TF) NON-RIPENING (NOR), COLORLESS NON-RIPENING (CNR), and RIPENING INHIBITOR (RIN), which were discovered through mutations affecting their function, together with the gaseous hormone ethylene control ripening. The connections between this regulatory network and its downstream effectors modulating colour, texture and flavour are still relatively poorly understood. Most of the TFs mentioned above have a positive regulatory effect on ripening effectors and provide feed-forward regulation for other major regulators as well as for themselves. Fruit ripening is also accompanied by changes in DNA methylation levels and possibly chromatin remodelling. Transcription factors may regulate downstream effector genes directly through promoter binding and activating or inhibiting a gene's transcription or indirectly, through the activation or inhibition of ethylene production with its own downstream regulatory circuit. We want to probe the ripening regulatory network interactions by introducing imbalances in regulator levels through the use of heterozygous lines and knock- out lines by using CRISPR targeting binding sites in promoters. We proved *Cnr* mutant is not dominant as described before by crossing as the phenotype of F1 generation is intermediated between WT and *Cnr* homozygous, but *Cnr* indeed affects other mutants a lot, resulting in similar phenotype with *Cnr* homozygous in double heterozygous mutants *Cnr* x *rin* and *nor* x *Cnr*.

Abstracts – Poster

Session X: Embryogenesis

P-18 Assembly, localization and translational control of *WUS*-/*WOX*-containing mRNPs in *A. thaliana*

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Plant reproduction, embryogenesis and development are regulated by a complex signaling network. *WUSCHEL*-related homeobox (*WOX*) superfamily members are key regulators of such developmental processes. This gene family is founded by *WUSCHEL* (*WUS*) and consists of 15 members, which are essential for stem cell maintenance in shoot apical meristem. Other members such as *WOX2*, *WOX8* and *WOX9* are essential for pro-embryo development. And *WOX5* is essential for root meristem formation.
Temporal and spatial control of mRNA translation is a precise mechanism to control the presence of gene products. This mechanism is found in all kingdoms of life and appears to be essential for asymmetric cell division, cell fate determination and many other developmental processes. In contrast to animals and fungi, to date little is known about mRNPs localization and translational control in plants. Using fluorescent whole-mount *in situ* hybridization (F-WISH) we could show that WOX2/5/8 and WUS mRNAs accumulate in globular structures. These globular structures indicate specific mRNPs formation, which might be essential for localization and translational control. I currently try to identify response elements for mRNP association using deletion and synthetic derivatives in F-WISH experiments. To characterize mRNP composition I perform RNA pull down experiments using specific RNA-tag or labeled antisense probes.

P-19 Dose-Dependent BABY BOOM Function

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AINTEGUMENTA-LIKE (AIL) transcription factors are key regulators of cell proliferation and meristem identity that can induce pluripotency (organogenesis) and totipotency (embryogenesis) when overexpressed. It was previously suggested that the PLT2 protein regulates root meristem size and maintenance through a protein concentration gradient, with high, intermediate and low AIL concentrations instructing stem cell fate, cell division and differentiation, respectively. Here we show that the AIL protein BABY BOOM (BBM) promotes cell proliferation in a dose-dependent manner, with a high dose inducing somatic embryogenesis, a medium dose inducing shoot organogenesis, and a low dose inducing cell dedifferentiation. PLT2 also directs the same dose-dependent overexpression phenotypes as BBM. These data suggest that AIL protein dose drives the developmental fate of regenerating tissues. We are determining the molecular basis for this dose-dependent regeneration by identifying and characterizing the genes that are directly regulated at different BBM doses. The results of these experiments will be presented.

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