SWISSPLANT'13

Parkhotel du Sauvage , Meiringen

30th January - 1st February 2013

www.spsw.ch - 22nd Edition

SWISSPLANT'13

TABLE OF CONTENT

Cover Picture : Haslital Tourismus

WELCOME

Two years ago, the newly created Swiss Plant Science Web (SPSW) held its first scientific meeting with «SwissPlant'11» in the Hotel du Sauvage in Meiringen. As you might know, this meeting was a sequel of sorts of the «Swiss Plant Molecular and Cell Biology Conferences», which were started in 1992 in Les Diablerets and proved to be highly popular in the scene for twenty years. «SwissPlant'11», organized by Barbara and Thomas Hohn and myself, was very successful scientifically, because it covered not only plant molecular and cell biology, but also plant ecology, biodiversity and global change, and socially because the Hotel du Sauvage proved to be a very nice meeting place. Thus, Ueli Grossniklaus, Florian Schiestl and Bernhard Schmid organized «SwissPlant'12» in the same format in the same place.

Now, with «SwissPlant'13», again in the same format and in the same place, we seem to have firmly established a new tradition. The scientific organizers from the University of Fribourg, Markus Geisler, Christian Lexer, Felix Mauch, Laurent Mène-Saffrané, Jean-Pierre Métraux, Heinz Müller-Schärer and Didier Reinhardt, have put together an attractive program, and Luca Wacker, the SPSW co-ordinator, took care of the meeting logistics and practical details. A big thank you to all the organizers!

Unfortunately, the very generous grant of the Swiss University Conference to establish the Swiss Plant Science Web, which also supported the first three editions of the SwissPlant meetings, ends in June 2013; our application for a continuation was not successful. However, we are convinced that the efforts of the SPSW during the last three years have helped to bring the community of plant scientists in Switzerland closer together, so that our new series of annual meetings will fly by itself for the years to come.

We hope that you will all enjoy science and leisure in Meiringen!

For the SPSW

Thomas Boller

ORGANIZING COMMITTEE

www.spsw.ch

Scientific Committee

Dr. Markus Geisler
Prof. Dr. Christian Lexer
Prof. Dr. Felix Mauch
Dr. Laurent Mène-Saffrané
Prof. Dr. Jean-Pierre Métraux
Prof. Dr. Heinz Müller-Schärer

Organization Committee

Swiss Plant Science Web
Dr. Luca Wacker, SPSW Coordinator

www.spsw.ch

Impressum

SWISSPLANT'13 (22th Edition of the Swiss Plant Molecular and Cell Biology Conference)
Park Hotel Sauvage, Meiringen 30th January - 1st February 2013
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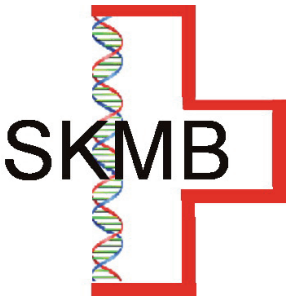

The Swiss Committee for Molecular Biology

PHILIP MORRIS INTERNATIONAL

PRACTICAL INFORMATION

Train connections
www.sbb.ch

Haslital Tourismus
www.haslital.ch

PARKHOTEL DU SAUVAGE :

PARKHOTEL
DU SAUVAGE
MEIRINGEN

The Park Hotel Sauvage is an impressive Art Nouveau hotel built in 1880 with all the updated modern comfort and pleasant atmosphere expected from a 3 *** hotel. The Park Hotel Sauvage is open year round and is situated in a beautiful park, with wonderful views of the surrounding alpine summits and pristine waterfalls. The convenient central location, only a few minutes by foot from the rail station and the mountain Post busses, makes this hotel the ideal starting point for trips and exploration of the Bernese mountain region. You will experience a memorable stay for holidays, seminars or any reason.

INTERNET CONNECTION

There is a possibility to connect to the internet in the Hotel.
Information will be available onsite.

PROGRAM SWISSPLANT'13

30th January - 1st February 2013

Meiringen

Wednesday, January 30, 2013

14:00 - 14:05		Opening remarks by the Scientific Committee
14:05 - 14:10		Opening remarks by Thomas Boller, SPSW President

Session I: Plant & light acclimation

14:10 - 14:30	Roman Ulm	Regulation of the UV-B Photoreceptor UVR8
14:30 - 14:50	Felix Kessler	The ABC of ABC1-like kinases: roles in high light acclimation and chloroplast metabolism
14:50 - 15:10	Christian Fankhauser	Sensing light direction: signal transduction during phototropism in Arabidopsis
15:10 - 15:40	*Coffee Break*	

Session II: Plants & environmental stress

15:40 - 16:00	Diana Santelia	The role of carbon metabolism in drought stress tolerance
16:00 - 16:20	Claudia Cosio	Interactions between Hg in complex aquatic environments and macrophytes
16:20 - 16:40	Kentaro K. Shimizu	Ecological transcriptome in natura: flowering genes of the perennial *Arabidopsis halleri* and the tropical tree *Shorea beccariana*
16:40 - 17:00	Irina Vaseva	Expression pattern of different dehydrin subclasses in *Trifolium repens* under drought
17:00 - 17:30	*Apéro*	

Session III: Plant-microbe interaction

17:30 - 17:50	Felix Mauch	The identification of the ABC-type transporter PDR12 as a pathogen-effector target reveals an early role of regulated ABA import in plant immunity
17:50 - 18:10	Manfred Heinlein	Plasmodesmata, macromolecular trafficking and virus movement
18:10 - 18:30	Aurélien Bailly	Soil bacteria manipulate plant growth via the prevalent indole signal
18:30 - 18:50	Didier Reinhardt	Identification of regulatory components in arbuscular mycorrhizal symbiosis
18:50 - 20:40	*Dinner*	

Session IV: Plant evolution & ecology

20:40 - 21:00	John Pannell	Explaining the males, and getting it wrong
21:00 - 21:20	Christian Lexer	Evolutionary genomics of plant adaptation and speciation
21:20 - 21:40	Dan F. B. Flynn	CO2 and N fertilization antagonizes incremental plant community functional optimization
21:40 - 22:00	Heinz Müller-Schärer	Towards understanding plant invasions
22:00	*Socializing at the Bar*	

Thursday, January 31, 2013

07:00 - 08:00 Breakfast

Session V: Plant cell transport & signaling

08:00 - 08:20	Enrico Martinoia	AtALMT9 is a malate-activated vacuolar chloride channel required for stomatal opening in Arabidopsis
08:20 - 08:40	Franck Vazquez	Post-transcriptional Regulation of Auxin Signaling Homeostasis
08:40 - 09:00	Silvia Schelbert Hofstetter	Syringolin A transport into plant cells
09:00 - 09:20	Adeline Chauvin	Long distance defense signaling in *Arabidopsis thaliana*: a key role for LOX6
09:20 - 09:40	Bruno Müller	Mechanism to generate specificity in cytokinin signaling

09:40 - 10:10 Coffee Break

Session VI: Plant development

10:10 - 10:30	Christian S. Hardtke	Impact of reduced local auxin biosynthesis on architecture and cellular anisotropy of Brachypodium distachyon roots
10:30 - 10:50	Pierre Barbier de Reuille	From data to model for 2D and 3D plant tissues. Application to early embryo development of *Arabidopsis thaliana*
10:50 - 11:10	Christiane Nawrath	Recent advances in understanding the relation between structure and function of the plant cuticle
11:10 - 11:30	Luis Lopez-Molina	The central role of the endosperm for the control of seed germination in *Arabidopsis thaliana*
11:30 - 11:50	Niko Geldner	The endodermis – how plants build their inner skin

11:50 - 17:00 Leisure time (Lunch on your own, skiing, swimming, walking ...)

17:00 - 18:30 Poster Session (drinks will be served)

18:30 - 20:20 Dinner

Session VII: Plant genetics

20:20 - 20:40	Bruno Studer	Targeting reproductive traits for more efficient forage grass breeding
20:40 - 21:00	Etienne Bucher	Epigenetic control of tissue and cell type specific gene expression
21:00 - 21:20	Anja Schmidt	Cell-type specific transcriptome analysis to identify genes and pathways active during apomictic reproduction
21:20 - 21:40	Michel Goldschmidt-Clermont	Nuclear control of chloroplast gene expression: why so complex?
21:40 - 21:50		Closing remarks

21:50 - open Live Music & Socializing at the hotel bar

Friday, February 1, 2013

Breakfast & Departure

Notes

1	Roman Ulm		Session I : Plants & light acclimation
2	Felix Kessler		
3	Christian Fankhauser		
4		Diana Santelia	Session II : Plants & environmental stress
5		**Claudia Cosio**	
6		**Kentaro K. Shimizu**	
7		**Irina Vaseva**	
8	Felix Mauch		Session III : Plant-microbe interaction
9	Manfred Heinlein		
10	Aurélien Bailly		
11	Didier Reinhardt		
12		John Pannell	Session IV : Plant evolution & ecology
13		Christian Lexer	
14		Dan F. B Flynn	
15		Heinz Müller-Schärer	
16	Enrico Martinoia		Session V : Plant cell transport & signaling
17	Franck Vazquez		
18	Silvia Schelbert Hofstetter		
19	Adeline Chauvin		
20	Bruno Müller		
21		Christian S. Hardtke	Session VI : Plant development
22		Pierre Barbier de Reuille	
23		Christiane Nawrath	
24		Luis Lopez-Molina	
25		Niko Geldner	
26	Bruno Studer		Session VII : Plant genetics
27	Etienne Bucher		
28	Anja Schmidt		
29	Michel Goldschmidt-Clermont		

Roman Ulm
Felix Kessler
Christian Fankhauser

SESSION 1

Plants & light acclimation

Regulation of the UV-B Photoreceptor UVR8

Marc Heijde, **Roman Ulm**

Department of Botany and Plant Biology, University of Geneva

Survival of plants in sunlight requires UV-protective responses. Plants respond to UV-B radiation with a coordinated photomorphogenic response that allows acclimation to this environmental stress factor. The UV-B photoreceptor UVR8 is highly specific and sensitive in perceiving UV-B radiation. However, an elevated UV-B response is associated with dwarf growth, indicating the importance of balancing UV-B–specific signaling. We will present data on how REPRESSOR OF UV-B PHOTOMORPHOGENESIS 1 (RUP1) and RUP2 function as crucial negative regulators of UVR8 action, providing new insight into the mechanism of UV-B photoreceptor regulation.

Felix Kessler

The ABC of ABC1-like kinases: roles in high light acclimation and chloroplast metabolism

Felix Kessler, Jacopo Martinis, Sergiu Valimareanu

Institut of Biology, University of Neuchâtel

The ABC1/ADCK/UbiB kinase family members in bacteria and mitochondria of yeast and humans, regulate ubiquinone synthesis and their mutation results in severe respiration defects. Little is known about plant ABC1-like kinases. It is particularly intriguing that in plants ABC1-like kinases are present not only in mitochondria but also at plastoglobule lipid droplets in chloroplasts.

The Arabidopsis ABC1k1 homolog corresponds to PGR6 (proton gradient regulation 6), the corresponding mutant showing reduced Fv/Fm, ETR (electron transport rate) and NPQ (non-photochemical quenching) after exposure to high light intensities. Strikingly however, abc1k1 plants are able to acclimate to high light, concurrently with a recovery of ETR and NPQ. We here demonstrate using non-targeted lipidomics that the effects of the abc1k1 mutation on the photosynthesis may be attributed mostly to changes in xanthophyll but also prenylquinone metabolism. Plants lacking ABC1k1 are defective in the production of lutein, tocopherol (Vitamin E), plastochromanol-8 (a plastoquinone-derived lipid antioxidant) and in the Redox recycling of tocopherol (Vitamin E). The effects on plastochromanol-8 and tocopherol Redox recycling indicate that ABC1k1 regulates prenylquinone metabolism at the level of the tocopherol cyclase VTE1. We demonstrate that ABC1k1-dependent regulation may be due to direct phosphorylation of VTE1 by ABC1k1.

Our results also suggest that ABC1K1 has other targets and acts as an important coordinator of photosynthesis and metabolism during acclimation to high light.

Christian Fankhauser

Sensing light direction: signal transduction during phototropism in Arabidopsis

Christian Fankhauser[1], Tobias Preuten[1], Emilie Demarsy[1], Tim Hohm[2,3] and Paolo Schumacher[1]

[1]Center for Integrative Genomics, University of Lausanne
[2]Department of Medical Genetics, University of Lausanne
[3]Swiss Institute of Bioinformatics, Lausanne

Phototropin blue-light receptors (phot1 and phot2 in Arabidopsis) activate a range of light regulated responses, including phototropism, leaf movements, stomatal opening, leaf expansion, and chloroplast movements. Those responses generally serve to optimize photosynthesis and allow the plant to adapt to changing light environments. Phototropins are light-regulated protein kinases that are broadly expressed and present at the plasma membrane in the dark. Blue light induces their protein kinase activity and leads to internalization of a fraction of the photoreceptor. In order to understand the steps leading from photoreceptor activation to asymmetric hypocotyl growth leading to phototropism we are addresing the following questions : (1) Where does phot1 perceive the light signal that activates phototropism? Does the photoreceptor act cell autonomously, or does the response involve transportation of a signal from the site of light perception to the site of action? (2) Is light-induced phot1 translocation from the plasma membrane to the cytosol a mechanism of desensitization or is its transport into the cytosol essential for signaling? (3) What are the substrates of phot1 kinase activity and how does phosphorylation regulate these targets. I will discuss our latest findings on these questions.

Notes

Notes

Diana Santelia
Claudia Cosio
Kentaro K. Shimizu
Irina Vaseva

SESSION 2

Plants & environmental stress

The role of carbon metabolism in drought stress tolerance

Diana Santelia

Institute of Plant Biology, University of Zurich

A major factor limiting the productive potential of crops is the availability of water. The incidence of drought stress in agriculture is expected to increase with climate change, leading to greater agricultural losses at a time of increasing world population.

In response to water deficit, plants rapidly degrade transitory starch in the leaf to soluble sugars (mainly glucose and maltose), lowering the osmotic potential of the cell to avoiding dehydration and protecting membranes, enzymes and other structures from damage and denaturation. We have obtained solid evidence that the Arabidopsis β-amylase 1 (BAM1), which releases maltose from the exposed nonreducing ends of glucan chains in starch, is a key enzyme of the stress-induced degradation of starch in the light. *BAM1* gene expression is strongly induced in response to acute osmotic stress, and *bam1* mutants show significantly reduced starch degradation and maltose accumulation in response to stress compared to wild type.

Additionally, *bam1* mutants show increased starch content specifically in guard cell chloroplasts and reduced stomatal opening compared to wild type. Since in guard cells storage and mobilization of starch follows an opposite rhythm with respect to mesophyll cells, BAM1 may play a role in starch breakdown in guard cells under light to release the precursors for malate and sucrose to sustain stomatal opening. Stomata are essential to the control of water balance during transitory period of drought stress. Interestingly, we found that *bam1* mutants are more tolerant to drought stress compared to wild type, revealing an unprecedent role of starch metabolism in guard cells in drought stress tolerance.

Interactions between Hg in complex aquatic environments and macrophytes

Claudia Cosio

Institute F.-A. Forel, University of Geneva

Proteins in every cellular compartment and in diverse metabolic pathways require metals or metal-cofactors for function. As a consequence, elements like Zn, Cu, Fe are essential nutrients for all organisms. But metal metabolism is subject to tight homeostatic control because of their reactivity and therefore toxicity. However these systems can also transport non-essential and toxic trace elements and affect global cycling of metals in the environment. My group works on direct and indirect interactions of plants with metals, including i) trace element metabolism, ii) plant's responses to combined environmental abiotic stresses, and iii) plant's influence on metal cycling.

In connection with the UNEP Global Mercury (Hg) Partnership, we addressed several aspects of interaction between Hg and macrophytes, including Hg cycles in water column and sediments, Hg methylation, bioaccumulation, and biomagnification in the food chain. Data will be illustrated with our findings regarding *Elodea nuttallii*, namely a high Hg and methyl-Hg accumulation and tolerance in the field and the laboratory, a change of microbial communities associated with rhizospheric sediments, an increase of MeHg proportion in rhizospheric sediments, a mobilization of MeHg from the sediments to the shoots, a carrier mediated internalization of both inorganic Hg and MeHg and evidences of trophic transfer of Hg through these plants. Results highlight the need for an increased understanding and consideration of the role of macrophytes in the functioning of Hg biogeochemical cycle in aquatic environment and in risk assessment of this highly toxic metal as shallow water represent a significant surface of our planet. Based on the current work concerning interaction of Hg with macrophytes, we believe that the definition of a global Hg policy must take in consideration the shallow water ecosystems in the UNEP monitoring and assessment mercury programs. We also propose to apply this fundamental knowledge to develop new bioassays in order to prevent possible risks to the environment and human health.

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Kentaro K. Shimizu

Ecological transcriptome in natura: flowering genes of the perennial *Arabidopsis halleri* and the tropical tree *Shorea beccariana*

Kentaro K. Shimizu[1], Masaki Kobayashi[1], Yayoi Takeuchi[1], Kenta Tanaka[2], Tomonori Kume[3], Bibian Diway[4]

[1]Institute of Evolutionary Biology and Environmental Studies, University of Zurich
[2]University of Tsukuba
[3]National Taiwan University
[4]Botanical Research Centre Semenggoh

The time at which flowering occurs has a considerable effect on plants' reproductive success. Molecular and genetic studies using a few model plants in controlled laboratory conditions have revealed that they have a complex gene network that regulates flowering in response to both environmental and endogenous cues. However, recent studies have suggested that molecular regulation of flowering in complex natural environments can differ from that observed in constant laboratory conditions. We have shown that *FLC* gene of *Arabidopsis halleri* functions as the memory of the temperature of past six weeks in a naturally fluctuating environment.

Community-level mass flowering, known as general flowering, which occurs in Southeast Asia at supra-annual irregular intervals, is considered a particularly spectacular phenomenon in tropical ecology. Although several proximate factors inducing general flowering have been proposed, the hypotheses have incorporated little empirical data on the developmental and physiological processes. We conducted an "ecological transcriptome" study of a mass flowering species, *Shorea beccariana*, comparing meteorological data with genome-wide expression patterns obtained using next-generation sequencing. Among the 284 flowering-related genes identified, the homologs of a floral pathway integrator, SbFT, and a floral repressor, SbSVP, showed dramatic transcriptional changes before flowering, and their flowering functions were confirmed using transgenic Arabidopsis thaliana. Expression in drought-responsive and sucrose-induced genes also changed before flowering. All these expression changes occurred when the flowering-inducing level of drought was reached, as estimated using data from the preceding 10 years. These genome-wide expression data support the hypothesis that drought is a trigger for general flowering.

Expression pattern of different dehydrin subclasses in *Trifolium repens* under drought

Irina Vaseva, Urs Feller

Institute of Plant Sciences and Oeschger Centre for Climate Change Research (OCCR), University of Bern
Institute of Plant Physiology and Genetics, Bulgarian Academy of Sciences

Irina Vaseva

The accumulation of dehydrins (known as LEA group 2) is part of the mechanisms protecting plants from protein denaturation under adverse environments. They are also involved in developmental processes, such as late embryogenesis, which require stabilization of macromolecules. LEA group 2 proteins are highly hydrophilic and remain stable under denaturating conditions. Their molecules contain one or more lysine-rich motifs called K-segment. Some dehydrins may also have a consensus Y-segment (DEYGNP) near the N terminus region or a serine-rich tract (the S-segment) that can be modified by phosphorylation. The number and order of the Y-, S- and K-segments define five different DHN sub-classes: Y_nSK_n, SK_n, K_n, Y_nK_n and K_nS.

Expression analyses of different dehydrin classes in *T. repens* confirmed their distinc roles in drought stress response and vegetative development, demonstrating some specific characteristic features for legumes. The acidic SK dehydrin exhibited constant expression in leaves and roots. Developmental processes did not affect its expression profile. It showed relative unresponsiveness towards water deprivation in the aboveground parts, while its transcript levels increased in roots. Neutral YSK-type dehydrin showed transient age-dependend changes in leaves and its expression increased most significantly in the youngest developing leaves and roots under drought. KS, another neutral dehydrin, was characterised with expression of at least tree splice forms, co-exsisting in plant tissues during the early vegetation. Some of them were inhibited by drought treatment and their content, especilally in the older leaves, was not detectable. Transcripts of YK dehydrin, which has been characterised as a specific type for legumes, were underrepresented in all plant parts during the vegetative development but the severe drought provoked strong increase of YK gene expression. The obtained expression profiles suggest unique roles of the different dehydrin classes in plant development under normal and stress conditions.

Notes

Felix Mauch
Manfred Heinlein
Aurélien Bailly
Didier Reinhardt

SESSION 3

Plant-microbe interaction

The identification of the ABC-type transporter PDR12 as a pathogen-effector target reveals an early role of regulated ABA import in plant immunity

Lorelise Branciard, Michael Stumpe, Bangjun Wang, Markus Geissler and **Felix Mauch**

Department of Biology, University of Fribourg

Felix Mauch

To counteract plant immunity pathogens have evolved effector proteins that are delivered into host cells to enhance pathogen virulence by subverting specific host immune responses. The identification of host effector targets offers a chance to look at plant defenses from a pathogens perspective. Our aim was to identify the host target of the RxLR5 effector of the Arabidopsis pathogen *Phyophthora brassicae*. *PbRxLR5* encodes a small protein with no sequence homology to any other protein except highly conserved RxLR5 orthologs in other Phytophthora species. RxLR5 raised our interest because gain-of-function RxLR5 Arabidopsis plants show enhanced disease susceptibility to *P. brassicae* that correlates with a defect in the formation of structural cell wall barriers consisting of the β-1,3-glucan polymer callose. Protein-protein interaction screens revealed that RxLR5 physically interacts with the plasma-membrane localized ABC transporter PDR12/ABCG40 that functions as an importer of the stress hormone abscisic acid (ABA). Agroinfiltration experiments and analysis of transcript accumulation of the ABA-marker gene ABR1 in response to ABA or pathogen challenge confirm that RxLR5 is an inhibitor of the ABA transport capacity of PDR12/ABCG40. Hence, the identification of PDR12 as the host target of RxLR5 establishes PDR12 catalyzed cellular ABA uptake as a crucial early immune response.

Manfred Heinlein

Plasmodesmata, macromolecular trafficking and virus movement

Manfred Heinlein

Institue of Botany, University of Basel

Plant development depends on intercellular communication through cytoplasmic cell wall channels known as plasmodesmata (PD) and involves the cell-to-cell trafficking of macromolecules. *Tobacco mosaic virus* (TMV) and other viruses use this route to spread their genomes and cause systemic infection. The spread of TMV RNA (vRNA) depends on virus-encoded movement protein (MP) and occurs in a non-encapsidated form, likely through exploitation of the cellular RNA transport machinery. We have developed *in vivo* tools to investigate transport processes in virus movement at the protein and RNA level, by which we gain evidence for a role of mobile RNA particles that target PD through interactions with membranes and the cytoskeleton. Our studies also focus on the role of small RNAs in virus movement. Interestingly, TMV interacts with the silencing host response in different ways. Whereas the viral 126k/183k replicase acts as a suppressor of RNA silencing, the MP promotes the spread of silencing. Since virus infection produces a unique population of virus and host-derived small RNAs, we are interested to understand whether these small RNAs may play a role in a viral strategy to influence host cell susceptibility with the help of MP. Our aim is to link the cell biology of virus movement to a better understanding of virus-induced defense and signaling responses and thus to attain a more complete picture of mechanisms involved in compatible virus:host interactions. These studies also provide important insights into plant intercellular communication mechanisms as well as leads for the development of antiviral strategies in crops.

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Soil bacteria manipulate plant growth via the prevalent indole signal

Aurélien Bailly, Amber Gardiner, Rita Baumgartner, Ulrike Groenhagen, Stefan Schulz, Leo Eberl and Laure Weisskopf

Institute for Plant Biology, Microbiology, University of Zurich
Institut für Organische Chemie, Technische Universität Braunschweig

Effective establishment of plants in soil depends on the initiation of a dense root architecture providing essential nutrients to efficiently grow aerial organs and achieve full development. In its natural environment the plant rhizosphere hosts microbial populations, thus describing a dynamic ecosystem where rapid and targeted communication is an asset for survival. Recently, microbial volatile organic compounds (VOCs) were shown to promote plant growth but attempts to identify effective molecules failed to describe a clear mechanism of action. In a screen for VOCs-mediated Arabidopsis growth-promotion, candidate molecules were extracted from the complex volatile blends emitted by bacteria using GC-MS. We isolated indole as a potent plant-growth modulator. Indole increases Arabidopsis secondary root network by interfering with the auxin machinery. Indole is increasingly recognized as a major signal in bacteria-bacteria interactions. It is involved in a wide range of bacteriological processes such as plasmid maintenance, biofilm formation, virulence or antibiotic resistance. Moreover, indole has been implicated as mediator of inter-kingdom interactions, for instance in attraction of pollinators through floral scent or in reduction of inflammation in human intestinal epithelial cells by the commensal gut microflora. However, the interference with auxin-controlled plant physiology through production of a volatile signal represents a new mechanism by which bacteria manipulate the plant hormonal balance to their own advantage.

Identification of regulatory components in arbuscular mycorrhizal symbiosis

Laure Bapaume, Srividya Velagapudi, Mélanie Rich, Martine Schorderet, **Didier Reinhardt**

Department of Biology, University of Fribourg

Most plants improve their mineral nutrition by hosting in their roots arbuscular mycorrhizal (AM) fungi, which provide phosphate and other mineral nutrients in exchange for sugar. AM symbiosis is controlled by the plant through the action of a suite of genes (SYM genes) which constitute the symbiosis signaling pathway (SYM pathway). In legumes, the SYM pathway is involved both, in AM symbiosis and in root nodule symbiosis. Up- and downstream of the SYM pathway, the components required for recognition of AM fungi, and the components required for intracellular accommodation remain largely elusive.

To identify such components, we take two routes: forward genetic screens on transposon-mutagenized populations of petunia, and large scale genome comparisons in order to identify genes conserved only in species competent to engage in AM symbiosis. The latter strategy is based on the finding that the known SYM genes are conserved in monocots and dicots, but absent from the non-symbiotic species *Arabidopsis thaliana* and *A. lyrata* as well as *Brassica* spec. A systematic genomic analysis will provide us with a short list of candidate genes that will be functionally studied by reverse genetics. This approach has recently become facilitated by the generation of large numbers of flanking sequences in populations of transposon-mutagenized plants, which allows for straight-forward identification of insertion mutants in petunia. This strategy has the potential to identify symbiosis-related members of gene families which have escaped forward genetic screens due to redundancy.

Notes

SESSION 4

Plant evolution & ecology

John Pannell

Christian Lexer

Dan F. B Flynn

Heinz Müller-Schärer

Explaining the males, and getting it wrong

John Pannell

Department of Ecology and Evolution, University of Lausanne

Male-sterility mutations are common in natural plant populations, and often occur at high frequencies. In contrast, female-sterility mutations in otherwise hermaphroditic populations are almost unknown. In this presentation, I will explain why this is so. I will also introduce work we have been conducting on two species in the Oleaceae (olive family) in which female-sterility mutations have nevertheless risen to high frequency: the Mediterranean plants *Phillyrea angustifolia* and *Fraxinus ornus*. In both cases, unusual explanations appear to be required to explain the unusual. This is a salutary tale of the potential impact of deep homology, limits to imagination, and mistakes made when adopting accepted protocols.

John Pannell

Evolutionary genomics of plant adaptation and speciation

Christian Lexer, Dorothea Lindtke, Celine Caseys, Camille Christe, Kai Stölting

Unit of Ecology & Evolution, Department of Biology, University of Fribourg

Understanding the processes that facilitate the origin, functioning and maintenance of biological diversity is of great interest to the fundamental and applied life sciences. Our group`s research interests revolve around the use of novel laboratory and computational tools for studying the genomics of adaptation, speciation, and of traits involved in range shifts in plants. To achieve these goals, we make use of Northern hemisphere 'model' taxa for which extensive genomic and biological resources are available, such as *Populus spp.*, and we have initiated the transfer of knowledge gained from this work to other plant radiations in highly structured and species-rich environments ('biodiversity hotspots'). In this talk, I will highlight our recent work on the ecological and evolutionary genomics of adaptation and speciation in *Populus spp*.

Christian Lexer

CO_2 and N fertilization antagonizes incremental plant community functional optimization

Dan F.B. Flynn, Nico Eisenhauer, Christopher M. Clark, Bradley Butterfield, Peter B. Reich

University of Zurich, Friedrich-Schiller-University Jena, Global Change Research Program, US EPA, Northern Arizona University , University of Minnesota

Plant communities are considered to assemble via non-random processes, with functional diversity of assemblages increasing due to past and present interactions between species. The mutual exclusion among functionally similar species and likely to an overall 'functional optimization' of the community therefore results. However, anthropogenic global change agents may alter this process. Nitrogen addition reduces plant diversity in terrestrial ecosystems due to competitive exclusion through competition for soil nutrients, while CO_2 effects are less conclusive, but have recently been shown to counterbalance the negative effect of N deposition. Here, we use realized species richness and functional diversity of long-term grassland experiment simultaneously manipulating plant diversity, atmospheric CO_2 concentrations and soil N availability to test if elevated CO_2 concentrations and N deposition influence the functional optimization of randomly assembled plant communities. We found that (i) plant species richness to decrease over time, although (ii) functional diversity increases over time due to a decrease in abundance of functionally redundant species and an increase in functionally dissimilar species, and (iii) N deposition to limit the degree of functional optimization of plant assemblages, while CO_2 counterbalances this effect.

Dan F. B Flynn

Towards understanding plant invasions

Heinz Müller-Schärer

Department of Biology, Unit Ecology & Evolution, Universtiy of Fribourg

Biological invasions still remain an enigma to ecologists and evolutionary biologists, but research on invasives have led to major advances in ecology and evolution. I will present a holistic approach to better understand invasions by combining (i) historical data to reconstruct the spatio-temporal invasion routes, (ii) niche modeling to follow potential changes in niche limits across these invasion routes, and (iii) experimental data on performance of the invader across multiple environments and for populations from both the native and introduced range. I will illustrate this for the European native and highly invasive *Centaurea stoebe* (Asteraceae), which experienced an exceptionally high shift in cytotype frequency and climatic niche during its invasion into North America. Both diploid (EU2x) and tetraploid (EU4x) cytotypes occur in Europe, but only tetraploids have been recorded so far in North America (NA4x). In the talk, main emphasis will be given to disentangle pre-adaptation (through differences in traits and plasticity of EU2x vs. EU4x) from post introduction evolution (EU4x vs. NA4x) to explain differences in the spatio-temporal dynamics of the observed range expansions and invasion routes, using our extensive experimental data. I will conclude by outlining the strengths and limitations of this novel multi-forked approach. Further study systems will then be briefly described.

Heinz Müller-Schärer

Notes

SESSION 5

Plant cell transport & signaling

Enrico Martinoia
Franck Vazquez
Silvia Schelbert Hofstetter
Adeline Chauvin
Bruno Müller

AtALMT9 is a malate-activated vacuolar chloride channel required for stomatal opening in Arabidopsis

Alexis De Angeli, Jingbo Zhang, Stefan Meyer, **Enrico Martinoia**

Institute of Plant Biology, University of Zurich

Water deficit strongly affects crop productivity. Plants control water loss and CO_2 uptake by regulating the aperture of the stomatal pores within the leaf epidermis. Stomata aperture is regulated by the two guard-cells forming the pore and change their size in response to ion uptake and release. While our knowledge about potassium and chloride fluxes across the plasma membrane of guard cells is advanced, little is known about fluxes across the vacuolar membrane. Here we present the molecular identification of the long-sought-after vacuolar chloride channel. AtALMT9 is a chloride channel activated by physiological concentrations of cytosolic malate. Single channel measurements demonstrated that this activation is due to a malate-dependent increase in the channel open probability. Arabidopsis thaliana atalmt9 knock-out mutants exhibited impaired stomatal opening and wilted more slowly than the wild type. Our findings show that AtALMT9 is a vacuolar chloride channel playing a major role in controlling stomata aperture.

Enrico Martinoia

Post-transcriptional Regulation of Auxin Signaling Homeostasis

David Windels, Myriam Ebneter, Nora Buri and **Franck Vazquez**

Institute of Botany, University of Basel

RNA silencing encompasses a wide set of recently discovered RNA-dependent regulatory mechanisms that act as a major bandmaster to coordinate the expression, protection, stability, and inheritance of eukaryotic genomes. Our work has recently provided pioneer insights into the regulation of Auxin signaling homeostasis and its impact on plant development. We showed that, during leaf development, the expression of *TIR1/AFB2* Auxin Receptor (*TAAR*) genes and are regulated by the miRNA miR393 and by a specialized secondary siRNA network, which we termed siTAARs, to fine-tune Auxin signaling homeostasis. We have also now identified an additional layer in the regulation of *TAAR* transcripts that involves RNA decay, and that appears as a major regulator of Auxin signaling homeostasis and plant development. I will present these most recent data and highlight the future directions of our work that aims to understand the biological role of the simultaneous regulation of *TAARs* by RNA silencing and RNA decay.

Franck Vazquez

Syringolin A transport into plant cells

Silvia Schelbert Hofstetter, Robert Dudler

Institute of Plant Biology, University of Zurich

Syringolin A is a virulence factor secreted by the plant pathogen Pseudomonas syringae pv. syringae that irreversibly inhibits the eukaryotic proteasome by novel mechanism (1). Syringolin A is a hydrophilic molecule readily soluble in water, consisting of a tripeptide and is negatively charged at the slightly acidic pH values found in the cell wall compartment. Thus, syringolin A is not expected to diffuse through membranes. This implies that transporters are involved in the syringolin uptake to reach its proteasome target. Although it is shown in rice cells that syringolin A is able to enter the cytoplasm of plant cells very efficiently (2), it is still an open question how syringolin A is taken up by plant cells.

Due to the chemical character of syringolin A, as well as due preliminary experiments indicating that peptides competitively inhibit syringolin A uptake, we consider transport by peptide transport systems. This project aims to elucidate the Arabidopsis syringolin A uptake transporter using a yeast system. Thereby, we functionally express members of the Arabidopsis *PTR* and *OPT* gene families in yeast to study their syringolin A uptake ability on a plate assay.

Silvia Schelbert Hofstetter

(1) Groll, M., Schellenberg, B., Bachmann, A. S., Archer, C. R., Huber, R., Powell, T. K., Lindow, S., Kaiser, M., and Dudler, R. (2008). A plant pathogen virulence factor inhibits the eukaryotic proteasome by a novel mechanism. Nature 452, 755-758.

(2) Hassa, P., Granado, J., Freydl, E., Waspi, U., and Dudler, R. (2000). Syringolin-mediated activation of the Pir7b esterase gene in rice cells is suppressed by phosphatase inhibitors. Mol. Plant-Microbe Interact. 13, 342-346.

Long distance defense signaling in Arabidopsis thaliana: a key role for LOX6

Adeline Chauvin[1,2], Daniela Caldelari[1], Jean-Luc Wolfender[2] and Edward E. Farmer[1]

[1]Plant Molecular Biology, University of Lausanne
[2]Pharmacy and Phytochemistry, University of Geneva

Mechanical wounding activates the production of jasmonate both in and near the wound and in distal connected leaves. The jasmonate pathway is initiated in plastids by 13-lipoxygenases (13-LOXs), enzymes that oxygenate tri-unsaturated fatty acid to provide the first precursors for jasmonate which controls defense against both invertebrates and vertebrates. Four 13-LOXs have the potential to initiate the jasmonate synthesis. A recent study described a role for LOX2 in leaf defense against herbivory while LOX3 and LOX4 are known to be essential for male fertility. The biological role for LOX6 is unknown. In this present study we identified a role of LOX6 in the defense of young leaves near the apical meristem in response to herbivory. Interestingly, in bioassay comparing the WT and *lox6A*, *Spodoptera littoralis* weight gain was not affected. Indeed, when all 13-LOXs except LOX6 were down-regulated (in the lox2-1 lox3B lox4A mutant) larvae preferred older leaves to plant centre, whereas they fed in an inverse manner on the quadruple mutant (*lox2-1 lox 3B lox 4A lox6A*). Concomitant with these observations the expression pattern of 13-LOX6 was stronger in plant centre and mainly localized in a specific cell type in the vasculature. All evidence suggests that, through long distance signalling, herbivores activate JA synthesis through LOX6 in the region of the apical meristem.

Adeline Chauvin

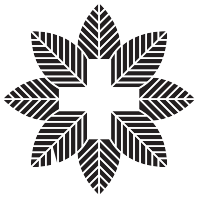
Mechanism to generate specificity in cytokinin signaling

Bruno Müller, Deborah Tavor-Deslex, Evelyne Zürcher

Institute of Plant Biology, University of Zurich

Cytokinins are classic plant hormones that orchestrate plant growth, development, and physiology. They affect the transcription profile of target cells by activating a multistep phosphorelay network. To identify the signaling domains in planta, we have constructed a synthetic reporter. Using this tool, we uncovered a role of cytokinin in specifying the embryonic root meristem in Arabidopsis, where it antagonistically interacts with auxin signaling. I will report the identification of cytokinin target genes, and present a case study how plants specifically compensate a steady mutant, but not an induced mutant of the same gene pair.

Tight control of the cytokinin signaling domains suggests the existence of a dedicated transport system operating at the tissue level. We have identified genes from the family of purine permeases that exert positive or negative roles on cytokinin signaling, respectively. Purine permeases acting positively are expressed as target genes, establishing a positive feedback loop, while a negatively acting purine permease is expressed complementary to the cytokinin output. Thus, I propose that these genes could be involved in controlling the cytokinin signaling domains *in planta*.



Bruno Müller

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SESSION 6

Plant development

Christian S. Hardtke
Pierre Barbier de Reuille
Christiane Nawrath
Luis Lopez-Molina
Niko Geldner

Impact of reduced local auxin biosynthesis on architecture and cellular anisotropy of *Brachypodium distachyon* roots

David Pacheco--Villalobos, Martial Sankar & **Christian S. Hardtke**

Department of Plant Molecular Biology, University of Lausanne

Auxin biosynthesis via indole--3--pyruvic acid (IPA) is essential for root formation and growth in the dicotyledon *Arabidopsis thaliana* (Arabidopsis) and requires the redundant *TRYPTOPHAN AMINOTRANSFERASE OF ARABIDOPSIS 1 (TAA1)* and T*AA1-- RELATED (TAR)* genes. Consequently, treatment of Arabidopsis with L--kynerunine, a specific inhibitor of TAA1/TAR enzymes, induces a short root phenotype. In this study, we isolated a T--DNA tagged hypomorphic mutant in the monocotyledon *Brachypodium distachyon* (Brachypodium) *TAR2--LIKE* gene (*BdTAR2L*). This promoter T--DNA insertion severely down--regulates *BdTAR2L* in both root and shoot system, suggesting that tryptophan aminotransferase activity is reduced in the mutant, which thus represents a hypomorphic Bdtar2l allele (*Bdtar2l^{hypo}*). Surprisingly however, *Bdtar2l^{hypo}* mutants only display a mild shoot phenotype in leaf expansion and a counterintuitive root system phenotype with dramatically longer and more frequently branched seminal roots. While radial patterning and meristem size are normal in *Bdtar2l^{hypo}* roots, mature cells are thinner and more elongated and therefore more anisotropic. Enhanced root cell elongation and anisotropy are also observed in another *BdTAR2L* mutant allele, in which the gene transcript is nearly undetectable. However, compared to *Bdtar2l^{hypo}* mutants, this quasi--null mutant displays stronger shoot phenotypes and an eventually smaller root meristem. The *Bdtar2l^{hypo}* phenotype can be mimicked by L--kynerunine treatment of wild type and contrasts with the short root phenotypes described for Arabidopsis IPA pathway mutants. Unlike its Arabidopsis homolog, *BdTAR2L* expression is only mildly ethylene--responsive. Moreover, *Bdtar2l^{hypo}* roots are insensitive to the ethylene biosynthesis inhibitor AVG and restored to wild type by the ethylene precursor ACC. Complementary then, the *Bdtar2l^{hypo}* root phenotype is also observed upon AVG application to wild type. In summary, our data suggest that auxin levels are supra-- optimal for cell elongation in Brachypodium roots. This effect appears to be ethylene-- mediated, suggesting an inverted regulatory relation between the two hormones as compared to Arabidopsis roots.

Christian S. Hardtke

From data to model for 2D and 3D plant tissues. Application to early embryo development of *Arabidopsis thaliana*

Pierre Barbier de Reuille[1], Lane Brendan[2], Saiko Yoshida[3], Dolf Weijers[4], Przemyslaw Prusinkiewicz[2], Richard Smith[1]

[1]Institute of Plant Sciences, University of Bern
[2]Department of Computer Science, University of Calgary
[3]Department of Biological Sciences, University of Tokyo
[4]Laboratory of Biochemistry, WU Agrotechnology & Food Sciences

When modelling spatial processes, it is convenient to work with mock geometries such as grids or simple shapes. While this can be very valuable in helping us to understand the processes involved it is equally important to ensure the models are consistent with the biology, and do not depend on the simplicity of the initial shapes. As we strive to improve the biological relevance of our models we are faced with three main issues: how to observe and analyse spatial biological processes? How to produce a spatial or growing model? How to use the biological observations (including cell shapes and growth patterns) in our models?

We have designed methods and software to allow better integration of biological data and models. MorphoGraphX is a software we designed to visualise, segment and analyse 3D confocal images. Originally developed for the study of the shoot apical meristem epidermis, we have extended it to allow for full 3D segmentation and data processing. In the end, we can extract 2D and 3D cell meshes, that can then be used as initial condition for models. In parallel, we are developing modelling methods both for 2D (with VVe) or 3D (with 3D Cell Complexes) cell tissues. Both environments rely on the mathematical notion of cell complexes(do you need to explain this?) which are designed to decouple the exact shape from the modelling itself. As such, it is possible to use either simple, canonical shapes ?, or realistic shapes extracted from an image processing software, such as MorphoGraphX. This enables biological data to be integrated into an initially simple geometric model.

We have successfully applied our methods to the modelling of roots [1], leaves [2] and fly wing disc [3] in 2D. We are now using the method for the study of cell division in the early Arabidopsis embryo. Our method offers two major advantages. Firstly, the MorphoGraphX software enables us to visualise and quantify the cell divisions in 3D. Secondly we can model the divisions on the actual cell geometry. Starting from the apical cell, we are modelling the first rounds of division in the early embryo. Modelling the apical cell as a truncated sphere, and assuming cells divide using the shortest wall going through their centre, our model recreate accurately the three first round of division. However, after a careful study using MorphoGraphX, it became clear that, not only the first cell is not spherical, but the first rounds of division are not the shortest possible walls going through the centre. I will present how we combined these software to create an accurate model of the first rounds of division in the early embryo of Arabidopsis thaliana.

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[2] Robinson, A; Barbier de Reuille, P; Chan, J; Bergmann, D; Prusinkiewicz, P and Coen E. Generation of Spatial Patterns Through Cell Polarity Switching, Science (2011), Vol 333, 1436-1440.

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Pierre Barbier de Reuille

Recent advances in understanding the relatio between structure and function of the plant cuticle

Christiane Nawrath[1], Sylwester Mazurek[1], Imène Garroum[1], Guillaume Fabre[1], Jean Daraspe[2], Antonio Mucciolo[2], Bruno Humbel[2]

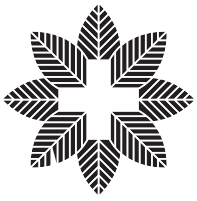
[1]Department of Plant Molecular Biology, University of Lausanne
[2]Electron Microscopy Facility, University of Lausanne

A hydrophobic extracellular matrix layer termed cuticle covers the epidermis of several plant organs, such as leaves, fruits and stems in primary growth stage. The cuticle plays important roles in protecting the plant against biotic and abiotic aggressions as well as during plant development. The major structural component of the cuticle is the polyester cutin formed by oxygenated fattyacids, glycerol and minor amounts of phenolic acids.

Characterization of the *permeable cuticle1* (*pec1*) mutant in Arabidopsis and the *eibi* mutant in barley lead to the identification of an essential role of the homologous full--size ABCG transporters of the Pleiotropic Drug Resistance family, AtABCG32 and HvABCG31, respectively, in cutin formation (Bessire et al, Plant Cell, 2011; Chen et al, PNAS, 2011).

Studies aimed on the elucidation of the molecules that are transported by AtABCG32 lead to new insights in the formation of the cutin polyester in leaves and flowers as well as the structure and functions of the cuticle in Arabidopsis.

Christiane Nawrath



The central role of the endosperm for the control of seed germination in Arabidopsis thaliana

Keun Pyo Lee[1], Urszula Piskurewicz[1], Solenne Carat Dumont[2], Richard Chappuis[1], Christian Fankhauser[3] and **Luis Lopez-Molina[1]**

[1]Department of Botany and Plant Biology, University of Geneva
[2]Bioinformatics & Biostatistics Core Facility, EPFL School of Life Sciences
[3]Center for Integrative Genomics, University of Lausanne

The Arabidopsis mature seed is genetically equipped to detect surrounding physical parameters relevant for the optimal growth of the seedling. Parameters such as seed age (i.e. fresh vs ripened seed), light quality (e.g. canopy light vs sunlight), water quality (e.g. salty vs fresh water) or temperature are detected by the plant in order to allow or prevent germination. Germination arrest protects the plant by preventing the developmental transition towards the highly fragile young seedling state. This necessitates the accumulation of the phytohormone abscisic acid (ABA), which also maintains the protective genetic programs characteristic of late embryogenesis such as those conferring osmotolerance. The endosperm is a triploid single cell tissue layer surrounding the embryo. It is becoming increasingly evident that the task of controlling germination is "delocalized" towards the endosperm whereas the role of the embryo is secondary. This conclusion was reached after developing a seed coat bedding assay, where embryonic growth is monitored under the influence of an underlying layer of dissected endosperm. This assay allows dissecting genetically how the endosperm influences embryonic growth. We will review the evidence showing that the endosperm monitors all the relevant physical parameters in order to control the release of ABA towards the embryo where it determines the germination potential of the seed. Furthermore, we will present evidence that the endosperm also mediates parental-specific control of seed germination. The presentation will focus on the remarkable developmental switch mediated by phytochromes A and B whereby, early upon seed imbibition, canopy light prevents phyB-dependent germination whereas, later on, it stimulates phyA-dependent germination.

Luis Lopez-Molina

Lee KP, Piskurewicz U, Turečková V, Carat S, Chappuis R, Strnad M, Fankhauser C, Lopez-Molina L. Spatially and genetically distinct control of seed germination by phytochromes A and B. Genes Dev. 2012 Sep 1;26(17):1984-96.

Lee KP, Piskurewicz U, Turecková V, Strnad M, Lopez-Molina L. PNAS 2010 Nov 2;107(44):19108-13. A seed coat bedding assay shows that RGL2-dependent release of abscisic acid by the endosperm controls embryo growth in Arabidopsis dormant seeds.

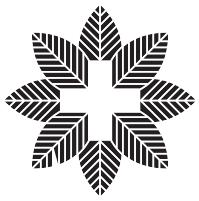
The endodermis – how plants build their inner skin

Niko Geldner

Department of Plant Molecular Biology, University of Lausanne

The endodermis is an invariable barrier within the root of vascular plants. Its barrier function is mediated by the Casparian strips, ring-like hydrophobic cell wall thickenings that are coordinated between cells and form a supracellular network within the apoplast. Casparian Strips effectively blocks passage of nutrients and pathogens through the extracellular space, while still allowing for signal perception and nutrient uptake to take place - very much resembling the dual, protective/uptake function of polarised gut epithelia in animals. The molecular players and mechanisms that underlie this intricately structured cell layer have remained obscure. Recently, we have described the formation of polar plasma membrane sub-domains in the endodermis and identified a new family of proteins that localize to the plasma membrane where the Casparian strips will form and are necessary for its formation. We also could demonstrate that the Casparian strip itself is an essentially lignin-based structure. Based on forward and reverse genetic screens, we have now identified a number of important players in Casparian strip formation and I will report on our latest results and models of how the endodermis achieves the formation of a strictly localized, lignified cell wall impregnation. The mutants that specifically interfere with Casparian strip formation also allow for the first time to study the consequences of a lack of this apoplastic diffusion barrier for plant nutrition, defense and stress resistance. The phenotypes we observe in our mutants turn out to be comparatively modest and specific, which is at first sight inconsistent with the textbook views of the endodermis as a very broadly acting, generic diffusion barrier. I will report on our latest physiological characterizations of these mutants and discuss how this might alter our current models of root function.

Niko Geldner



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SESSION 7

Plant genetics

Bruno Studer
Etienne Bucher
Anja Schmidt
Michel Goldschmidt-Clermont

Targeting reproductive traits for more efficient forage grass breeding

Bruno Studer

Institute of Agricultural Sciences, Forage Crop Genetics, ETH Zurich

Perennial ryegrass (*Lolium perenne* L.) is one of the most important grassland species of temperate regions worldwide. Due to its allogamous nature, perennial ryegrass has – so far – been improved as open pollinated populations or synthetic varieties.

Using novel grass breeding schemes to increase our efficiency to exploit the genetically available heterosis may have the potential to improve the genetic gain for agronomically important traits such as biomass yield. However, current attempts to produce classical hybrids in forage crops have major limitations: Firstly, inbreeding is hampered by an efficient self-incompatibility (SI) system promoting cross pollination. Secondly, plant performance and fertility is affected by a severe inbreeding depression that is characteristic of allogamous species. Moreover, tools to control pollination for efficient hybrid seed production are currently not available.

Combining research activities for traits involved in plant reproduction such as the two-locus SI system, fertility restoring self-compatibility (SC), cytoplasmic male sterility (CMS) and doubled haploid (DH) induction will pave the way for a shift towards hybrid breeding in forage grass species. Starting with an overview about the latest scientific achievements for these traits in perennial ryegrass (which serves as a diploid model for other major grass species), we will evaluate the prospects of using SI, SC, CMS and DH as breeding tools for efficient forage grass improvement.

Arias, A., Studer, B., Frei, U., Lübberstedt, T. (2011) Prospects for hybrid breeding in bioenergy grasses. BioEnergy Research 5: 1-10

Bruno Studer

Epigenetic control of tissue and cell type specific gene expression

Etinne Bucher

Institue of Botany, Uiversity of Basel

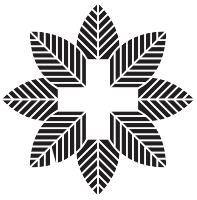
In plants, and in eukaryotes in general, epigenetic control of gene expression is very often caused by the presence of a transposable element (TE) within the promoter region of a gene. Transcription of TE is repressed by the presence of epigenetic marks, such as DNA and histone methylation. These repressive marks can then also affect expression of nearby endogenous genes. In Arabidopsis, thousands of genes in are associated with transposons, potentially putting them under epigenetic control. We are interested in better understanding how TEs affect nearby gene expression and more speciDically how TEs affect tissue speciDic gene expression. So far little is known about how epigenetic states are set up and maintained in differentiated plant organs (such as roots, leafs, Dlowers and stems), individual tissues and cell lineages.In order to study in these processes we set up a transgenic Arabidopsis GFP reporter line based on an epigenetically controlled promoter. The transgene contains an endogenous promoter that includes the long terminal repeat (LTR) of a TE. Interestingly the GFP reporter line shows a very high tissue speciDicity. GFP expression can almost exclusively be observed in two cell rows in the dehiscence zone of the siliques (fruits). Since DNA methylation is an important factor in repressing TEs, we then wanted to test whether tissue speciDicity of the reporter gene could be modiDied by changing DNA methylation levels. The main players in setting up de novo DNA methylation in plants are Pol IV and Pol V. When we introgressed the nrpd1 and nrpe1 mutations, affected in Pol IV and Pol V respectively, into our reporter line we found that tissue speciDicity of GFP expression changed. We not only observed GFP expression in the siliques, but now also in the stem. This shows that the epigenetic code contributes to the tissue speciDicity of gene expression.As a next step we wanted to identify novel epigenetic factors, involved in this process. For this purpose we mutagenized the reporter line and screened for novel expression patterns. To our surprise we found many new expression patterns in these mutants. We found plants with Dluorescent Dlowers, meristems, veins, leaf margins and others. We are currently focusing on three promising mutants showing release of GFP expression in the meristem, leaf margin and veins. The point mutations are currently being identiDied by high throughput sequencing and classical genetic mapping.

Etienne Bucher

Cell-type specific transcriptome analysis to identify genes and pathways active during apomictic reproduction

Anja Schmidt, Marc Schmid, Manuel Waller, Daniela Guthörl, Christian Sailer, and Ueli Grossniklaus

Institute of Plant Biology, University of Zurich & Zurich-Basel Plant Science Center, Developmental Genetics



Apomixis, the asexual reproduction of plants through seeds, leads to the formation of clonal offspring. Although apomixis occurs in over 400 plant species, it is rarely represented in crop species. Engineering of apomixis into crop plants has a great potential for plant breeding and seed production, as any complex genotype could be preserved. To date the limited knowledge about the molecular genetic basis underlying apomixis hampers its agronomical application.

Sexual reproduction and apomixis are closely interrelated. However, during apomictic as compared to sexual reproduction, key steps are altered: The megaspore mother cell (MMC) circumvents meiosis (apomeiosis) to give rise to an unreduced embryo sac, and an unfertilized egg cell initiates embryo development parthenogenetically.

In order to identify genes and pathways of potential importance for apomictic reproduction, we are analyzing the cell type specific transcriptomes of the apomictic MMC and egg cell. Transcriptome analysis of the corresponding cell types has previously been described for the sexual model plant Arabidopsis (1, 2). To compare cell type specific profiles between both reproductive modes, we are using *Boechera gunnisoniana*, a closely related apomictic species as a model plant. Interestingly, gene ontology analysis of genes expressed both in the sexual and apomictic MMCs suggests a conservation of a number of functions, likely of general importance for initiation of the germline lineage. In addition, functional classification of genes expressed in one of the cell types only in sexual Arabidopsis or apomicitc Boechera suggests a differential activity of a number of pathways, including cell cycle activity, hormonal pathways, signal transduction, and epigenetic regulation. In summary, our study gives new insights into the transcriptional basis underlying apomixis.

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Anja Schmidt

Nuclear control of chloroplast gene expression: why so complex?

Karen Loizeau, Damien Douchi, Linnka Legendre, Jean-David Rochaix and **Michel Goldschmidt-Clermont**

Department of Molecular Biology and Department of Botany and Plant Biology, University of Geneva

In the chloroplasts of plants and algae the post-transcriptional steps of gene expression are remarkably complex. Polycistronic transcripts are processed and spliced, and in plants specific C residues are further edited to U. The stability of the resulting mRNAs is determined by RNA-binding proteins, and their translation is strongly regulated. A striking feature of these processes is that they are mediated by numerous nucleus-encoded proteins that act on single chloroplast transcripts or small subsets of transcripts. The number and specificity of the factors that act in post-transcriptional steps of chloroplast gene expression raise many questions about their functions. Some of them may be involved in anterograde signaling from the nucleus to the chloroplast in response to environmental cues. In Chlamydomonas, some of the nucleus-encoded factors may also play a role in the negative feedback regulation of translation that acts on chloroplast subunits of the photosynthetic complexes to ensure their stoichiometric assembly (Control by Epistasy of Synthesis or CES) (1). Finally, it has also been argued that some of the nucleus-encoded factors may just be there to "debug" the chloroplast genetic program (2), and that this could contribute to the complexity of gene expression. To investigate these questions, we have analyzed the function of nucleus-encoded factors that control the expression of specific chloroplast-encoded subunits of PSI and PSII at the level of splicing, mRNA stabilization and translation in Chlamydomonas.

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(2) Maier UG, Bozarth A, Funk HT, Zauner S, Rensing SA, Schmitz-Linneweber C, Börner T, Tillich M. (2008) BMC Biol. 6:36.

Michel Goldschmidt-Clermont

Notes

1	Emmanuel Boutet
2	Lucien Bovet
3	Robert Dudler
4	Doris Herrmann
5	Stefan Hörtensteiner
6	Nataliya Komarova
7	Andrea R Pluess
8	Sarah Robinson
9	Klaus Schlaeppi
10	Philipp M. Schlüter
11	Samuel Wuest
12	Samuel C. Zeeman

Poster Session

How to make use of the curation expertise of SIB Swiss Institute of Bioinformatics in collaborative research projects: the CASP family case study

Emmanuel Boutet[1], Daniele Roppolo[3], Brigitte Boeckmann[1], Lydie Bougueleret[1], Ioannis Xenarios[1,2], Sylvain Poux[1] and the Swiss-Prot group

[1]Swiss-Prot group, SIB Swiss Institute of Bioinformatics, CMU
[2]Vital-IT group, SIB Swiss Institute of Bioinformatics, University of Lausanne
[3]Department of Plant Molecular Biology, University of Lausanne

Manual biocuration of proteins is essential to provide high quality datasets that can be automatically parsed, and to allow the scientific community a rapid access to reliable information. The Swiss-Prot group of the SIB Swiss Institute of Bioinformatics as part of the UniProt consortium produces and maintains UniProtKB/Swiss-Prot, the manually curated section of the UniProt.

High quality information is added by experienced biocurators and manual curation process includes the protein sequence validation, verification of results from computational analyses, extraction and structuring of information from the literature.

Biocuration is however not limited to extraction of information from external sources and can provide essential value for research. We will present a case study showing how the expertise of our biocurators can be used in the framework of an experimental project.

On the basis of informal discussions that took place at the 2009 SWISSPLANT meeting, we established a scientific collaboration with Niko Geldner's research group at the University of Lausanne to better characterize the CASP proteins, a family that specifically localize to Casparian strips. This collaboration was the first step in deciphering this structure essential for the plant physiology and integrity.

In-depth in-silico analysis of this family required the identification and cleaning of additional homologous protein sequences. For the sake of this project, we focused our curation effort on CASP-related proteins and annotated 379 of them in a total of 59 organisms. All of these entries were integrated in the UniProtKB/Swiss-Prot database, and subsequently used for a phylogenetic analysis to classify members into subfamilies. This study will soon be published and shows that the CASP-related plant specific proteins are related to a well-studied protein family present in protists and opisthokonts. Site-directed mutagenesis of meaningful amino-acids was conducted for functional analysis. The CASP subset of this large family, characterized by two conserved extracellular loops involved in subcellular location, is required for the correct formation of Casparian strips and is present only in root-forming organisms.

To retrieve CASP-related proteins:
http://www.uniprot.org/uniprot/?query=family:%22Casparian+strip+membrane+proteins+%28CASP%29+family%22

2 Lucien Bovet

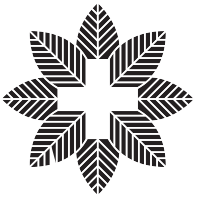
Gene expression changes during tobacco curing

Lucien Bovet, Florian Martin, Markus Klein

Philip Morris International R&D, Philip Morris Products S.A.

As traditional agronomical practices, tobacco leaves are harvested in the field and then undergo two successive curing steps, full leaf maturation and drying. There are three major curing processes, namely air-, flue- and sun-curing. These are usually associated with the three main tobacco types, burley, bright and oriental tobaccos, respectively. Before harvesting and/or during the early curing phase, green leaves are senescing. This results in a significant alteration of the leaf constituents via enzymatic activities. Acquiring knowledge about gene regulation and its effects on leaf chemistry during early curing is of particular interest in order to target specific traits linked to both the types of tobacco and the curing process.

To monitor changes in gene expression during the tobacco lifecycle we have used an Affymetrix Tobacco Exon Array that covers a large set of *N. tabacum* functional genes identified from the Tobacco Genome Initiative (TGI), thereby representing a feasible alternative to a full whole genome gene array. This exon array was used in this study to identify genes that are differentially expressed during early curing in the three major tobacco types. Large sets of senescence-regulated genes encoding i.e. proteases (SAG, cysteine proteases), chlorophyll catabolic enzymes (chlorophyllases), Rubisco and starch degradation enzymes were identified. Time-course experiments have shown the evolution of the transcripts during the first phase of the curing, providing useful information about the major chemical changes that occur before the last drying phase of the curing process.



3 **Robert Dudler**

ISOCHORISMATE SYNTHASE 2 is required for stomatal immunity in Arabidopsis

Myriam Schaufelberger, Leonie Luginbühl, Barbara Schellenberg, and **Robert Dudler**

Institute of Plant Biology, University of Zurich

In Arabidopsis, *ISOCHORISMATE SYNTHASE 1* (*ICS1*) is responsible for the biosynthesis of the bulk of salicylic acid (SA) required for systemic acquired resistance (SAR) and *PR* gene activation, which are abrogated in homozygous ics1 mutant plants. Such plants contain only about 10% of the SA observed in the wild type1. Some of this residual SA is synthesized by the product of the *ICS2* paralog, which, in contrast to *ICS1* is not induced by UV light or pathogens[1,2]. No phenotype has been reported for homozygous *ics2* mutant plants.

To our surprise and in contrast to a report in the literature[3], we have found that stomatal immunity in Arabidopsis, which is dependent on SA, is functional in homozygous ics1 plants, but not in *ics2* mutants. Stomatal immunity in *ics2* mutants can be restored by transformation of an *ICS1* gene placed under the control of the guard cell-specific MYB60 promoter. Plants carrying an *ICS2::GUS* reporter gene show that *ICS2* is expressed in guard cells.

We conclude from these experiments that for stomatal immunity in Arabidopsis, guard cells exhibit a cell-autonomous requirement for SA biosynthesis which is met by the activity of the *ICS2* gene product.

References:

[1] Garcion et al. (2008). Characterization and biological function of the ISOCHORISMATE SYNTHASE2 gene of Arabidopsis. Plant Physiol 147, 1279-1287.

[2]Wildermuth et al. (2001). Isochorismate synthase is required to synthesize salicylic acid for plant defence. Nature 414, 562-565.

[3]Melotto et al. (2006). Plant stomata function in innate immunity against bacterial invasion. Cell 126, 969-980.

Indo-Swiss Collaboration in Biotechnology (ISCB)

Doris Herrmann, Sonia Aubry

ISCB, Cooperation & Development Center, EPFL Lausanne

4 Doris Herrmann

ISCB is a well established bilateral research and development programme, jointly funded and steered by the Department of Biotechnology (DBT), Ministry of Science and Technology, Government of India and the Swiss Agency for Development and Cooperation (SDC), Federal Department of Foreign Affairs, Government of Switzerland.

The ISCB programme was initiated in 1974. In the first 25 years a wide range of biotechnology projects were supported. In 1999, the new ISCB programme was started. The main goal is to contribute towards food security in the Indian context through innovative life sciences and biotechnology approaches, supporting sustainable and climate resilient agriculture. Innovative biotechnological products and processes addressing small and marginal farmers' needs and demands are developed and validated.

The key achievements of the first decade of the new ISCB are:

- Successful development of prototype biofertilizer, leading up to 40% yield increase in wheat under marginal conditions
- Establishment of transgenic chickpea plants resistant against insect pests (pod borer or aphid)
- Development of a biopesticide at pilot scale against the insect pest pod borer
- Partnerships (non exclusive licensing agreements) with Indian companies and public institutes for final steps of product development for biofertilizer, chickpeas and biopesticide
- > 40 years equivalents of scientific exchanges and build-up of molecular laboratories
- > 350 articles and scientific publications as well as > 400 posters and presentations at conferences

A new phase (2013-2016) was launched recently. In the new phase, ISCB will follow-up projects which are in the poduct development stage until they reach the farmers. In addition, ISCB will fund several new networks, consisting of technical and socio-economics projects within the scope and guidelines of ISCB.

A call for these new networks will be launched in spring 2013.

Website: http://iscb.epfl.ch

5 Stefan Hörtensteiner

Arabidopsis thaliana cytochrome P450 CYP89A9 is involved in the formation of novel major chlorophyll catabolites during leaf senescence

Bastien Christ,[1] Iris Süssenbacher,[2] Nicole Bichsel,[1] Aurelie Egert,[1] Thomas Müller,[2] Bernhard Kräutler,[2] **Stefan Hörtensteiner[1]**

[1]Institute of Plant Biology, University of Zurich
[2]Institute of Organic Chemistry and Centre of Molecular Biosciences, University of Innsbruck, Austria

In *Arabidopsis thaliana* several nonfluorescent chlorophyll catabolites (NCCs) have been described as 'final' products of chlorophyll breakdown. NCCs are linear tetrapyrroles derived from chlorophyll by oxygenoytic chlorin macrocycle opening. NCCs are formed from respective fluorescent chlorophyll catabolite (FCC) precursors by a non enzymatic isomerization inside the vacuole of senescing cells. Here we identified a group of at least four dioxobilane-type NCCs (DNCCs) as novel major chlorophyll breakdown products in Arabidopsis. The constitution of the most abundant DNCC, *At*-DNCC-1, was resolved by one and two-dimensional nuclear magnetic resonance spectroscopy. We further identified cytochrome P450 monooxygenase CYP89A9 as being responsible for DNCC accumulation in wild type Arabidopsis; *cyp89a9* mutants that are deficient in CYP89A9 function were devoid of DNCCs, but accumulated proportionally higher amounts of NCCs. CYP89A9 localized to the endoplasmic reticulum implying that FCCs occurring in the cytosol might be their natural substrates. Using recombinant CYP89A9, we could confirm FCC specificity and show that dioxobilane-type FCCs (DFCCs) are the products of the CYP89A9 reaction. We conclude that Arabidopsis CYP89A9 is involved in the formation of novel dioxobilane-type catabolites of chlorophyll. The enzyme specifically forms DFCCs, which after import into the vacuole isomerize to respective DNCCs. In wild type Arabidopsis, DNCCs are the major degradation products of chlorophyll, representing more than 90% of the chlorophyll present in green leaves.

6 Nataliya Komarova

Sorting of membrane proteins between plasma membrane and tonoplast

Nataliya Komarova, Stefan Meier, Anna Meier, Patrizia Ascone, Marianne Suter Grotemeyer, and Doris Rentsch

Institute of Plant Sciences, University of Bern

The tonoplast (TP) and the plasma membrane (PM) are two endpoints of membrane traffic along the plant secretory pathway. Correct targeting of membrane proteins is essential for the generation and maintenance of membrane identity and integrity and proper cell function, cell division, and growth. Though the general trafficking mechanisms seem conserved in eukaryotes, the endomembrane system of higher plants displays distinct organizational features that may entail adaptive specializations in membrane trafficking. As limited information is available on structural determinants required for targeting of plant membrane proteins, we used the five differentially localized members of the PTR/NRT1 subfamily II from Arabidopsis to identify signals required for their specific targeting to the PM and TP, respectively. We found that an N-terminal peptide of TP-localized AtPTRs was necessary and sufficient for targeting PM-localized PTRs and Arabidopsis sucrose transporter SUC2 to the TP, whereas the predicted large central loop region of the PM-localized AtPTR1 and 5 was required for targeting to the PM. Furthermore, we investigated intracellular trafficking of PTRs using dominant negative mutants of Rab GTPases to further elucidate the exact traffic routes.

7 Andrea R Pluess

Microevolution along environmental gradients in *Fagus sylvatica*

Andrea R. Pluess

D-USYS, Ecosystem Management, ETH

Species with large climatic envelopes might be less susceptible to global change due to a multitude of adapted genotypes along environmental gradients. In a first step and in collaboration with P. Weber (WSL) we study if European beech (*Fagus sylvatica*) at dry and mesic sites is genetically differentiated to understand the potential for long-term resistance of forests in the predicted drier summers of Central Europe. With an AFLP genome scan approach, we investigated neutral and potentially adaptive genetic variation in three regions containing a dry and a mesic site each ($n_{ind.}$ = 241, $n_{markers}$ = 517). We linked this dataset with dendrochronological growth measures and local moisture availabilities based on precipitation and soil characteristics. Tree height and median basal growth increments were reduced and genetic diversity decreased slightly at dry sites. Overall genetic differentiation was low (F_{st} = 0.028) and Bayesian cluster analysis grouped all populations together suggesting high (historical) gene flow. The Bayesian outlier analyses indicated 13 markers with three markers differing between all dry and mesic sites and the others between the contrasting sites within individual regions. A total of 41 markers, including seven outlier loci, changed their frequency with local moisture availability, but marker presence/absence was not related to dendrochronological characteristics. Given the indications for microevolutionary processes operating within short geographic distances, in a next step I will study variation in genes of abiotic-stress response and phenology within the framework of ADAPT (C. Heiri et al., WSL) and in collaboration with G.G. Vendramin (CNR, Florence), S. Oddou-Muratorio and H. Lalagüe (INRA, Avignon). Thereby we will investigate adaptation to a multitude of environmental gradients using 80 European beech provenances sampled across Switzerland.

Based on the first study, the general genetic similarity among the six sites suggests that 'preadaptive' genes can easily spread across the landscape. Yet, due to the long live span of trees, fostering saplings originating from dry sites and grown within mesic sites might increase resistance of beech forests during the anticipated longer dry periods.

8 Sarah Robinson

Probing the mechanical changes underlying Hypocotyl elongation

Sarah Robinson[1], Michal Huflejt[1], Gabriella Mosca[1], Alain Weber[1], Pierre Barbier de Reuille[1], Anne-Lise Routier-Kierzkowska[1], Dimitrios Felekis[3], Tobias Preuten[2], Christian Fankhauser[2], Bradley Nelson[3], Richard Smith[1], Cris Kuhlemeier[1]

[1]Institute of Plant Sciences, University of Bern
[2]Center for Integrative Genomics, University of Lausanne
[3]Institute of Robotics and Intelligent Systems, ETH Zurich

A fundamental question in development is how can an organ acquire a specific overall shape by adjusting the rate and direction of growth in a local manner? Much progress has been made in understanding how gene expression patterns are established during development. However, much less is known about how these patterns are related to the physical growth of biological shapes. What mechanical changes are driving growth? Where do they occur? And do they feed back onto the genes that generated them?

To answer these questions we explore the mechanical properties of the Arabidopsis hypocotyl which has a simple shape, growing almost entirely along one axis, and with few cell divisions. Using a newly developed technology called Cellular Force Microscopy (CFM), we have directly measured the mechanical properties of cells in the hypocotyl. We have miniaturized the system, and combined it with confocal microscopy, allowing us to compare cfm measurements molecular markers. We have already used the system to compare cfm measurements with with osmotic treatments. Osmotic treatments, enable us to estimate the pressure of the cell by finding the plasmolysis point of the cell. Osmotic treatments also enable us to calculate the elasticity of the cell in all 3 dimensions by measuring the deformation of the cell wall using the 3D visualization and analysis software MorphoGraphX.

Measurements on hypocotyls during early development showed a 4 fold increase in the apparent stiffness of the tissue during the first week of growth. Osmotic treatments, however, showed a gradient of pressure and wall elasticity in the opposite direction, with young tissues being much more resistant to osmotic stress. We are still investigating the influence of osmotic regulation on this result. Using finite element models of our experimental system we try to determine the actual stiffness of the cell wall and the contribution of the different layers in the tissue.

In the longer term this setup will be used to monitor directly the effect of gene regulation on the mechanical properties at cellular resolution and to apply forces to look for changes in gene expression.

9 **Klaus Schlaeppi**

Composition and evolution of root-associated bacterial communities of *Arabidopsis thaliana* and relative species

Klaus Schlaeppi, Nina Dombrowski, Davide Bulgarelli, Matthias Rott, Emiel Ver Loren Van Themaat And Paul Schulze-Lefert

Department of Plant Microbe Interactions, Max Planck Institute for Plant Breeding Research, Cologne

Plants touch with their roots one of the richest microbial ecosystems on earth and engage at this contact zone with soil in intimate associations with a multitude of mainly commensalistic and mutualistic microbes. The composition of root-associated bacterial communities (RABCs) is markedly different from the surrounding soil microbiota indicative of specific colonization events. Little is known about assembly principles of RABCs and microbial services they provide to the plant. In replicate experiments we investigated RABC structures of the model plant *Arabidopsis thaliana* in comparison with the phylogenetically related species *Arabidopsis halleri*, *Arabidopsis lyrata* and *Cardamine hirsuta* both at natural sites and under controlled conditions in the greenhouse. RABC composition was determined by pyrosequencing of amplicons, which were derived from the taxonomically informative 16S rRNA gene. In all tested conditions RABCs are dominated by Proteobacteria, Bacteroidetes, and Actinobacteria and composition varies most as a function of the environmental condition, i.e. independent natural sites or replicate batches of soil in the greenhouse experiments. The tested plant species largely shared the same root microbiota within a given condition suggesting that the tested species recruit a RABC core that satisfies the common host needs dependent on the environmental condition. In all experiments we also identified taxonomically defined RABC subunits that were characteristic for the host species. Such species dependent RABC subunits possibly reflect host-specific requirements in a given environmental situation. For example, all bacteria assigned to the family of Thermomonosporaceae accumulated to reduced levels at *C. hirsuta* roots compared to the roots of Arabidopsis species. Interestingly, within the phylogenetic framework of host plants, we identified most such species distinctive subunits for the phylogenetically oldest *C. hirsuta* indicating that the composition of RABCs is linked to the evolutionary divergence time of the host species.

Canidate genes for specific pollinator attraction as revealed by transcriptome and proteome data

Khalid E.M. Sedeek, Weihong Qi, Monica A. Schauer, Alok K. Gupta, Lucy Poveda, Shuqing Xu, Zhong-Jian Liu, Ueli Grossniklaus, Florian P. Schiestl, **Philipp M. Schlüter**

Institute of Systematic Botany, University of Zurich

Sexually deceptive orchids of the genus *Ophrys* mimic the mating signals of their pollinator females to attract males as pollinators. This mode of pollination is highly specific and leads to strong reproductive isolation between species, such as the closely related species *O. exaltata*, *O. sphegodes* and *O. garganica*. In order to identify candidate genes responsible for pollinator attraction and reproductive isolation between these species, floral reference transcriptomes and proteomes were generated using a combination of next-generation sequencing and shotgun proteomics. Floral traits such as odour, colour and morphology are necessary for successful pollinator attraction, and candidate genes for anthocyanin (colour) and hydrocarbon (odour) biosynthesis were identified. Moreover, transcription factors putatively involved in the regulation of flower odour, colour and morphology were annotated, including Myb, MADS and TCP factors.

Genetic bases and ecological relevance of correlative controls during plant reproduction

Samuel Wuest, Ueli Grossniklaus and Bernhard Schmid

Institute of Evolutionary Biology and Environmental Studies, University of Zurich

11 Samuel Wuest

The study of heritable variation in ecologically-relevant plant traits (e.g. flowering time, drought tolerance) can result in a better understanding of evolutionary processes, and lead to increased agricultural output. Recent technological advances in molecular biology and genetics have opened new possibilities to dissect genetic networks underlying developmental traits. However, to understand the evolutionary forces that shape these networks, an ecological context is necessary.

In my current project, I am studying how genetic variation that causes variation in ecologically-relevant traits affects the performance of individuals at different levels of competition. The focal traits are control mechanisms exerted by reproductive structures over maternal growth. Such mechanisms have been observed in several plant species -amongst them important crops- and are termed “correlative controls”. Hereby, the production of offspring induces senescence and/or growth arrest in maternal tissues, such that the life span of the parental plant is extended if reproduction is delayed. Correlative controls could well represent cases of offspring begging in plants, whereby the developing embryo affects maternal resource allocation patterns. However, neither the molecular bases of such mechanisms nor their ecological relevance are known. I will present my ongoing work that applies molecular genetics and genomics to examine the mechanistic bases of correlative controls, and experimental ecology to explain how evolutionary forces could have shaped these developmental pathways.

Glycogen and starch synthesis in different parts of the same plant: use of next-generation sequencing and proteomics to unedrstand the metabolic reprogramming in the non-model specias *Cecropia peltata*

Sylvain Bischof, Martin Umhang, Simona Eicke, Sebastian Streb, **Samuel C. Zeeman**

Institute of Agricultural Sciences, EZH Zurich

Starch is the most widespread storage carbohydrate in plants. Its biosynthetic pathway evolved from an ancestral capacity to make glycogen. The production of massive, semi-crystalline starch granules is more complex than that of small, soluble glycogen particles, but the factors determining whether glycogen or starch is formed are not fully understood. In 1971, Rickson reported in Science that the tropical tree *Cecropia peltata* is able to synthesize soluble glycogen in specialized myrmeophytic structures (Müllerian Bodies) in addition to starch in the leaves. Müllerian bodies are collected and eaten by ants that have a mutualistic relationship with the plant, dwelling in its hollow stems and protecting it from herbivores. It is proposed that glycogen is a better source of nutrition for the ants than starch would be. Here, we investigated how the plant is able to synthesise either polymer. Electron micrographs, sugar and glucan measurements show that soluble sugars and glycogen accumulated to very high levels in Müllerian Bodies, but not in leaves. Structural characterization revealed that the glycogen has greater numbers of short oligosaccharide branches than the leaf starch. An integrated RNA-seq and quantitative shotgun proteomic approach revealed how the expression and abundances of key biosynthetic enzyme isoforms (starch synthases, branching enzymes and debranching enzymes) are altered in the Müllerian Bodies. These data can explain the assembly of glycogen instead of starch. As starch constitutes the major carbohydrate for food and for industrial applications worldwide, these data are valuable as they can be used as an archetype for potential biotechnological reprogramming of storage carbohydrate synthesis. Furthermore, as *C. peltata* is an important invasive plant and has potential medicinal uses, our transcriptome and proteome datasets adds a valuable resource for further molecular studies.

Notes

Attendance List

Last Name	First Name	Affiliation	E-mail
Amelia	Amiguet	SPSW Staff	aamigue2@unil.ch
Aurélien	Bailly	University of Zurich	abailly@botinst.uzh.ch
Pierre	Barbier de Reuille	University of Bern	barbier@ips.unibe.ch
Thomas	Boller	University of Basel	thomas.boller@unibas.ch
Emmanuel	Boutet	Swiss-Prot SIB	emmanuel.boutet@isb-sib.ch
Lucien	Bovet	Philip Morris Intl	lucien.bovet@pmi.com
Etienne	Bucher	University of Basel	e.bucher@unibas.ch
Adeline	Chauvin	University of Lausanne	Adeline.Chauvin@unige.ch
Elena	Conti	University of Zurich	ContiElena@access.uzh.ch
Claudia	Cosio	University of Geneva	Claudia.Cosio@unige.ch
Mikael	Courbot	Syngenta	mikael.courbot@syngenta.com
Manuela	Dahinden	SPSW Staff	mdahinden@ethz.ch
Robert	Dudler	University of Zurich	rdudler@botinst.uzh.ch
Christian	Fankhauser	University of Lausanne	christian.fankhauser@unil.ch
Edward	Farmer	University of Lausanne	edward.farmer@unil.ch
Urs	Feller	University of Bern	urs.feller@ips.unibe.ch
Dan F. B.	Flynn	University of Zurich	dan.flynn@ieu.uzh.ch
Markus	Geisler	University of Fribourg	markus.geisler@unifr.ch
Niko	Geldner	University of Lausanne	Niko.Geldner@unil.ch
Michel	Goldschmidt-Clermont	University of Geneva	michel.goldschmidt-clermont@unige.ch
Pierre	Goloubinoff	University of Lausanne	pierre.goloubinoff@unil.ch
Sandrine	Gouingené	SPSW Staff	sandrine.gouinguene@unine.ch
Christian S.	Hardtke	University of Lausanne	christian.hardtke@unil.ch
Manfred	Heinlein	University of Basel	manfred.heinlein@unibas.ch
Doris	Herrmann	ISCB EPFL	doris.herrmann@epfl.ch
Barbara	Hohn	FMI Basel	Barbara.Hohn@fmi.ch
Thomas	Hohn	University of Basel	hohn@fmi.ch
Stefan	Hörtensteiner	University of Zurich	shorten@botinst.uzh.ch
Beat	Keller	University of Zurich	bkeller@botinst.uzh.ch
Felix	Kessler	University of Neuchâtel	felix.kessler@unine.ch
Nataliya	Komarova	University of Bern	komarova@ips.unibe.ch
Christian	Lexer	University of Fribourg	christian.lexer@unifr.ch
Verena	Liedschulte	Philip Morris Intl	Verena.Liedschulte@pmi.com
Luis	Lopez-Molina	University of Geneva	luis.lopezmolina@unige.ch
Enrico	Martinoia	University of Zurich	enrico.martinoia@botinst.uzh.ch
Felix	Mauch	University of Fribourg	felix.mauch@unifr.ch
Frederick	Meins	FMI Basel	meins@fmi.ch
Laurent	Mène-Saffrané	University of Fribourg	laurent.mene-saffrane@unifr.ch
Jean-Pierre	Métraux	University of Fribourg	jean-pierre.metraux@unifr.ch
Bruno	Müller	University of Zurich	bruno.mueller@uzh.ch
Heinz	Müller-Schärer	University of Fribourg	heinz.mueller@unifr.ch
Christiane	Nawrath	University of Lausanne	christiane.nawrath@unil.ch
Jean-Marc	Neuhaus	University of Neuchâtel	Jean-Marc.Neuhaus@unine.ch
John	Pannell	University of Lausanne	john.pannell@unil.ch
Melanie	Paschke	SPSW Staff	melanie.paschke@usys.ethz.ch
Andrea R.	Plüss	ETH Zurich	andrea.pluess@env.ethz.ch

Mikhail	Pooggin	University of Basel	mikhail.pooggin@unibas.ch
Carole	Rapo	SPSW Staff	carole.rapo@usys.ethz.ch
Didier	Reinhardt	University of Fribourg	didier.reinhardt@unifr.ch
Philippe	Reymond	University of Lausanne	Philippe.Reymond@unil.ch
Sarah	Robinson	University of Bern	sarah.robinson@ips.unibe.ch
Diana	Santelia	University of Zurich	dsantelia@botinst.uzh.ch
Silvia	Schelbert	University of Zurich	sschel@botinst.uzh.ch
Klaus	Schlaeppi	MPI Cologne	schlaeppi@mpipz.mpg.de
Philipp M.	Schlüter	University of Zurich	philipp.schlueter@systbot.uzh.ch
Anja	Schmidt	University of Zurich	aschmidt@botinst.uzh.ch
Kentaro K.	Shimizu	University of Zurich	shimizu@botinst.uzh.ch
Bruno	Studer	ETH Zurich	bruno.studer@usys.ethz.ch
Ted	Turlings	University of Neuchâtel	ted.turlings@unine.ch
Roman	Ulm	University of Geneva	roman.ulm@unige.ch
Irina	Vaseva	University of Bern	irina.vaseva@ips.unibe.ch
Franck	Vazquez	University of Basel	franck.vazquez@unibas.ch
Luca	Wacker	SPSW Staff	luca.wacker@unibas.ch
Samuel	Wuest	University of Zurich	samuel.wuest@ieu.uzh.ch
Samuel C.	Zeeman	ETH Zurich	szeeman@ethz.ch

Notes