

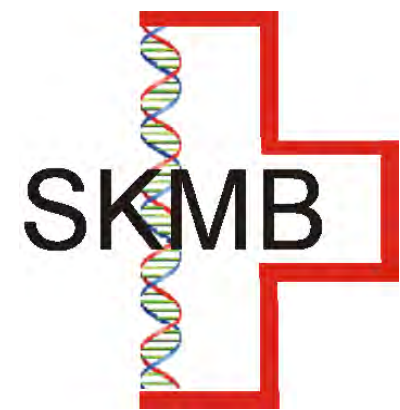


SWISSPLANT 2014

29–31 January 2014
Parkhotel du Sauvage, Meiringen

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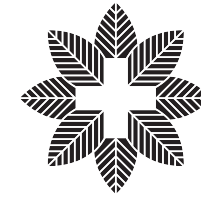
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Swiss Committee for Molecular Biology



The Swiss Plant Science Web gratefully acknowledges the financial support for the SWISSPLANT 2014 conference.



SWISSPLANT 2014

**Symposium of the Swiss Plant Science Web
29–31 January 2014, Meiringen, Switzerland**

Scientific Committee

Prof. Dr. Teresa Fitzpatrick
Prof. Dr. Michel Goldschmidt-Clermont
Prof. Dr. Luis Lopez-Molina
Dr. Yamama Naciri
Prof. Dr. Roman Ulm

Conference Organization

Swiss Plant Science Web
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- 36 Posters (in alphabetical order)
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Welcome

The Swiss Plant Science Web (SPSW) held its first scientific conference «SwissPlant 2011» in the Hotel du Sauvage in Meiringen. As many of you are aware, this meeting is a sequel of sorts of the «Swiss Plant Molecular and Cell Biology Conferences» that started in 1992 in Les Diablerets. Meanwhile it is well established and highly popular in the plant science research community.

The first «SwissPlant» conference was organized in 2011 by Barbara Hohn, Thomas Hohn and myself. This new format was scientifically very successful, because plant molecular and cell biology topics were expanded to include plant ecology, biodiversity and global change. From a social point of view the Hotel du Sauvage proved to be a very nice meeting place and easy to reach for participants from all regions in Switzerland.

By now, the «SwissPlant» conference has become a nice tradition and an excellent opportunity for senior researchers to showcase current advances in plant science research in Switzerland and to trigger new collaborations.

The scientific conference committee from the University of Geneva has compiled an attractive program. Our special thanks go to Teresa Fitzpatrick, Michel Goldschmidt-Clermont, Luis Lopez-Molina, Yamama Naciri, and Roman Ulm. I am glad that Sylvia Martínez, our new SPSW coordinator, has successfully tackled all organizational aspects. Furthermore, I am happy that with her experience and with the idealistic support of all members, the Swiss Plant Science Web will continue to thrive. Enjoy science and leisure in Meiringen

Thomas Boller
University of Basel, SPSW president

Program

Wednesday, 29 January

14:00	Opening remarks by the Scientific Comittee	
14:05	Opening remarks by Thomas Boller	
	Session I:	Biotic Interactions, Part I
14:10	Philippe Reymond	Insect eggs induce a systemic acquired resistance against pathogens in Arabidopsis
14:30	Edward E. Farmer	Signalling in the first two minutes of insect attack
14:50	Beat Keller	Molecular approaches to improve durable fungal disease resistance in wheat
15:10	Coffee break	
	Session II:	Biotic Interactions, Part II
15:40	Delphine Chinchilla	Pattern recognition receptors in plant immunity
16:00	Didier Reinhardt	Identification of regulatory components in arbuscular mycorrhizal symbiosis
16:20	Hannes Gamper	How miscible are natural arbuscular mycorrhizal fungal assemblages?
16:20	Robert Dudler	An endophytic rhizobial strain produces protein inhibitor syringolin A
17:00	Apéro	
	Session III:	Evolution
17:30	Sylvain Aubry	Deep evolutionary comparison of gene expression identifies parallel recruitment of trans-factors in independent origins of C4 photosynthesis
17:50	Kentaro Shimizu	Genome-wide quantification of homeolog expression ratio revealed non-stochastic gene regulation in synthetic allopolyploid Arabidopsis kamchatica
18:10	Christian Parisod	Genome evolution: causes and consequences of the dynamics of transposable elements
18:30	Cris Kuhlemeier	The genetic architecture of pollination syndromes
18:50	Dinner	
	Session IV:	Plant Development
20:40	Célia Baroux	Nuclear organization dynamics in plant developmental transitions: resolving causality
21:00	Christian Hardtke	Automated quantitative histology reveals vascular morphodynamics during Arabidopsis hypocotyl secondary growth
21:20	Daniele Roppolo	Plasma membrane domains and local cell wall modifications
21:40	Markus Geisler	TWISTED DWARF1 regulates cytoskeleton dynamics
22:00	Socializing at the hotel bar	

Thursday, 30 January

07:00	Breakfast	
	Session V:	Photosynthesis & Metabolism
08:00	Stefan Hörtensteiner	Analysis of catabolite modifying reactions during chlorophyll breakdown
08:20	Felix Kessler	ABC1K1, a chloroplast kinase, couples the response of photosynthesis to highlight with metabolism
08:40	Samuel Zeeman	New insights into starch synthesis through work in model plants, non-model plants and heterologous systems
09:00	Oliver Kötting	Plastidial NAD-dependent malate dehydrogenase is critical for night-time plastidial redox homeostasis and embryo development in Arabidopsis
09:20	Christian Körner	Why growth controls photosynthesis
09:40	Coffee Break	
	Session VI:	Plant Biodiversity
10:10	Yamama Naciri	Species boundaries and molecular markers
10:30	Chris Kettle	Conservation genetics in a changing world, Genes, Trees, and toy planes
10:50	Christoph Küffer	Island biogeography: moving beyond species numbers
11:10	Christophe Randin	What can we learn from the past and current geographic distribution of a rare and endemic alpine plant when making future projections under climate change?
11:30	Jake Alexander	How do changing competitive interactions shape a species' response to climate change?
11:50	Leisure time (Lunch on your own, skiing, swimming, hiking...)	
17:00	Poster Session (drinks will be offered)	
18:30	Dinner	
	Session VII:	Gene Expression & Signaling
20:20	Katja Bärenfaller	Different Arabidopsis leafcell types and the proteasome
20:40	Emilie Demarsy	Plasma Membrane H ⁺ -ATPase regulation is required for auxin gradient formation during phototropism
21:00	Yves Poirier	A rice cis-natural antisense RNA acts as a translational enhancer for its cognate mRNA and contributes to phosphate homeostasis and plant fitness
21:20	Michel Goldschmidt-Clermont	Nuclear control of chloroplast gene expression: why so complex?
21:40	Closing remarks	
21:50	Socializing at the hotel bar	

Friday, 31 January

Breakfast & departure

1	Philippe Reymond	Session I: Biotic Interactions, Part I
2	Edward Farmer	
3	Beat Keller	
4	Delphine Chinchilla	Session II: Biotic Interactions, Part II
5	Didier Reinhardt	
6	Hannes Gamper	
7	Robert Dudler	
8	Sylvain Aubry	Session III: Evolution
9	Kentaro Shimizu	
10	Christian Parisod	
11	Cris Kuhlemeier	
12	Célia Baroux	Session IV: Plant Development
13	Christian Hardtke	
14	Daniele Roppolo	
15	Markus Geisler	
16	Stefan Hörtensteiner	Session V: Photosynthesis & Metabolism
17	Felix Kessler	
18	Samuel Zeeman	
19	Oliver Kötting	
20	Christian Körner	
21	Yamama Naciri	Session VI: Plant Biodiversity
22	Chris Kettle	
23	Christoph Küffer	
24	Christophe Randin	
25	Jake Alexander	Session VII: Gene Expression & Signaling
26	Katja Bärenfaller	
27	Emilie Demarsy	
28	Yves Poirier	
29	M Goldschmidt-Clermont	



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Insect eggs induce a systemic acquired resistance against pathogens in *Arabidopsis*

Olivier Hilfiker, Raphaël Groux, **Philippe Reymond**

Department of Plant Molecular Biology, University of Lausanne

Although they constitute an inert stage of the insect's life, eggs trigger plant defenses that lead to egg dessication, drop-off, and mortality, or attraction of egg predators. However, the expression profile of *Arabidopsis* leaves after oviposition by *Pieris brassicae* is drastically distinct from the profile obtained after larval feeding. Strikingly, SA accumulates in response to oviposition by *P. brassicae*, both in local and systemic leaves. We discovered that insect eggs reduce bacterial infection in *Arabidopsis* through the activation of a systemic acquired resistance (SAR) response. This egg-induced SAR requires defense priming regulated by the newly discovered systemic signal pipecolic acid and depends on ALD1 and FMO1. This unique phenomenon might illustrate a strategy by eggs to prevent the detrimental effect of bacterial pathogens on feeding larvae.

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Edward Farmer

Signalling in the first two minutes of insect attack

Edward E Farmer

Department of Plant Molecular Biology, University of Lausanne

The leaf gets its resilience to attack by combining great tolerance to damage with the ability to synthesize molecules that target the digestive tract of herbivores. Central to leaf survival in the face of herbivory is jasmonate (JA), a defence prohormone that begins to accumulate within 30 s of wounding a leaf and that will lead to the activation of 1000 genes in the plant. It takes about 90 s to detect JA increases in distal leaves that share vascular connections to the wounded leaf. What signals lead to distal JA accumulation? We found that feeding *Spodoptera littoralis* larvae elicited electrical activity of a similar velocity (3 - 8 cm per min) to that expected for the long distance wound signal that leads to JA accumulation distal to wounds. Several genes that underlie the production of these signals were identified. It has also been necessary to identify the first enzyme of jasmonate synthesis that is operative distal to a wound. Only one of four 3-lipoxygenases in *Arabidopsis* fulfils this function allow us to define part of what appears to be a new signal pathway that functions the first 2 min of the leaf response to attack.



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Beat Keller

Molecular approaches to improve durable fungal disease resistance in wheat

Beat Keller, Simon Krattinger, Daniel Stirnweis, Severine Hurni, Tina Jordan, Susanne Brunner

Institute of Plant Biology, University of Zürich

New genomic tools have allowed us to develop approaches and strategies which are revolutionizing the way specific questions can be tackled in wheat biology. The availability of partial or complete genomic sequences, although mostly in a non-ordered form, allows a more efficient characterization of agronomically important genes for their use in classical or transgenic wheat breeding.

The study of natural diversity allows the identification of new functional disease resistance alleles as well as the characterization of the molecular basis of resistance gene function and specificity. Ultimately, this might also allow the design of synthetic genes with new specificities and, ideally, to develop a more durable type of resistance based on major genes. Such approaches will be discussed for the allelic series of Pm3 resistance genes against powdery mildew. Natural diversity can also reveal mechanisms of resistance gene evolution.

We are also exploring the transgenic use of Pm3 resistance alleles. A field trial has shown that these alleles confer improved resistance to powdery mildew when overexpressed. Moreover, resistance was improved when transgenic lines with different alleles (differing in the specificity of resistance) were mixed. Although such experiments are at a very early phase in relation to plant breeding, they show the potential of the approach. In particular, wild relatives of crop plants hold great allelic and gene diversity for resistance. It is often difficult to transfer genes from wild species into crops, as it can take decades to derive agronomically useful material from such introgressions.

In addition to the work on wheat as the host species, genomic tools on the pathogen side start to play an important role in molecular analysis of host-pathogen interactions, and will ultimately be useful for improving resistance breeding strategies. This approach will be described based on our work on powdery mildew genomics.



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Delphine Chinchilla

Pattern recognition receptors in plant immunity

Delphine Chinchilla, Ana Dominguez, Marta Kiss-Papp

Institute of Botany, University of Basel



Plants are exposed to a myriad of potentially pathogenic microbes but disease is rather the exception than the rule. The plant immune system confers the capacity to recognize and respond to microbes, notably via perception of microbe associated molecular patterns (MAMPs). MAMPs are characteristic of a whole class of microbes and are present in both foes and friends. They are often essential molecules for microbial life, and it is therefore difficult for microbes to escape perception by the plant’s immune system. Detection of MAMPs by plant cells induces vigorous defense responses including production of reactive oxygen species, rapid changes in gene expression and secretion of antimicrobial compounds.

MAMPs are sensed at the cell surface by receptors called pattern recognition receptors (PRRs). For example, bacterial flagellin is perceived by the flagellin receptor FLS2, and bacterial elongation factor EF-Tu is perceived by the receptor EFR. Both FLS2 and EFR are receptor kinases with an extracellular ligand-binding domain and an intracellular protein kinase domain.

Our understanding of PRR activation and signal transduction made an important step forward with the discovery that the flagellin receptor FLS2, upon activation, became physically associated with another receptor kinase, called BRI1 associated kinase 1 (BAK1). BAK1 had first been identified as a co-receptor for the brassinosteroid receptor BRI1 in Arabidopsis, but it proved to be equally required for immune signalling. Thus BAK1 seems to be shared by several signalling pathways controlling developmental as well as defense responses.

Our current work makes use of different biochemical, physiological and genetic approaches to understand how BAK1 interacts and activates PRRs, what is the repertoire of BAK1 activated PRRs, and whether BAK1 has other functions in plant immunity.

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Didier Reinhardt

Identification of regulatory components in arbuscular mycorrhizal symbiosis

Laure Bapaume, Mélanie Rich, Sabine Laukamm, Martine Schorderet, **Didier Reinhardt**

University of Fribourg, Department Biology

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. To identify regulatory components involved in AM development, we have conducted forward mutant screens in the transposon line W138 of petunia, with the main focus on mutants affected in the intraradical development of the AM fungus. A first component identified by this strategy is VAPYRIN, an ankyrin-related protein that consists of two protein:protein interaction domains. Although VAPYRIN itself does not carry a typical domain for association or insertion into membranes, it localizes to small subcellular structures in root cells, suggesting a localization to a specific subcellular compartment. In order to reveal the nature of these structures, various fluorescent subcellular markers are used for colocalization studies. Furthermore, Biochemical approaches are being employed to identify interacting proteins for VAPYRIN.

A second component, *ATYPICAL ARBUSCULE1 (ATA1)*, that was recently identified by transposon mutagenesis, is a transcription factor of the GRAS family. *ATA1* is required for the development of the intracelular arbuscules which mediate the nutrient transfer between the symbiotic partners. In *ata1* mutants, the arbuscules develop poorly and the mycorrhizal fungus cannot establish itself in the root. *ATA1* is expressed exclusively in roots, and is strongly induced during AM development, suggesting that it may represent a symbiosis-related master regulator that triggers the expression of downstream simbiosis-related genes required for the intracellular accommodation of the fungal symbiont and for the establishment of the symbiotic machinery. We are currently testing which genes are under the control of *ATA1*.



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Hannes Gamper

How miscible are natural arbuscular mycorrhizal fungal assemblages?

Hannes A Gamper

Environmental Systems Science, ETH Zurich

This is a question arising from recently heightened efforts to inoculate biotechnologically propagated arbuscular mycorrhizal (AM) fungi to crop plants as enhancers of mineral nutrient uptake from soil. The question is similarly pressing given there are more opportunities for accidental global dispersal of AM fungal propagules by modern human activities.

To determine the drivers and determinants of AM fungal strain establishment and survival at introduction sites with already naturally occurring residents, a manipulative field experiment was set at eight distant Swiss grassland sites among which we reciprocally transferred top soil samples. Half of those study sites were placed in the North and half in the South of the Alps to utilize the fact that there may be a biogeographic dispersal barrier. Two sites each on both sides of the Alps were further put on either slightly acidic or alkaline soil to include contrasting edaphic conditions as a possible environmental filter to fungal immigration. Incorporation of foreign soil samples into the local soils was supposed to mimicked massive AM fungal propagule dispersal and mixed taxon inoculation events. With eight experimental subplots at each of eight study sites, the experiment comprises 56 field plots with pairs of mixed natural AM fungal assemblages and eight control plots in which the local AM fungi were left to re-assemble after soil disturbance and removal of the original vegetation cover. To enable an equal sampling of the symbiotic AM fungal assemblages all the plots were planted to a genetically and developmentally uniform population of ribwort plantain (*Plantago lanceolata*) bioassay seedlings.

Community sequencing of a 1.8 kb long nuclear ribosomal DNA and 443 bp long glutamine synthetase gene fragment revealed the presence of highly site-specific AM fungal assemblages, at the onset of the experiment, as well as, 1.5 years post manipulation. The arbuscular mycorrhizal fungal assemblages recorded in plots on slightly acidic soil were generally more taxon-rich and more receptive to immigration. Immigration success of AM fungal strains was largely independent of the soil pH conditions at the site of origin and raised the total phylotaxon richness at certain sites above that observed in the control plot with only the local AM fungal assemblage. This clearly showed that taxon numbers of natural AM fungal assemblages are limited by dispersal, but also evidenced high susceptibility of natural AM fungal assemblages to biological invasion and thus receptiveness for AM fungal inoculants. Determination of the ecophysiological, agronomic, and ecological implications requires follow-up projects, which should particularly focus on plant growth and nutrition, but also symbiont selection by plants.



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Robert Dudler

An endophytic rhizobial strain produces protein inhibitor syringolin A

Alexey Dudnik, Robert Dudler

Institute of Plant Biology, University of Zurich

A syringolin A synthetase-like gene cluster was identified in the α -proteobacterial strain *Rhizobium* sp. *AP16*, which has been isolated as an endophyte from poplar roots. The gene cluster exhibits some unusual features. We show that the strain produces syringolin A and related variants *in vitro* under the appropriate conditions. The results of a mutational analysis of the gene cluster's unusual features as well as of experiments aimed at the elucidation of the biological function of proteasome inhibitor production for an endophytic bacterium will be presented.



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Sylvain Aubry

Deep evolutionary comparison of gene expression identifies parallel recruitment of trans-factors in independent origins of C₄ photosynthesis

Sylvain Aubry, Steve Kelly, Britta MC Kümpers, Richard D Smith, Julian M Hibberd

Department of Plant Sciences. University of Cambridge, UK / Institute of Plant Biology, University of Zürich

With at least 60 independent origins spanning monocotyledons and dicotyledons, the C₄ photosynthetic pathway represents one of the most remarkable examples of convergent evolution. The recurrent evolution of this highly complex trait involving alterations to leaf anatomy, cell biology and biochemistry allows an increase in productivity by ~50% in tropical and subtropical areas. The extent to which separate lineages of C₄ plants use the same genetic networks to maintain C₄ photosynthesis is unknown. We developed a new informatics framework to enable deep evolutionary comparison of gene expression in species lacking reference genomes. We exploited this framework to compare gene expression in independent C₄ lineages (*Cleome gynandra* and *Zea mays*) whose last common ancestor diverged ~140 million years ago. We show that genes encoding proteins of the C₄ cycle are recruited into networks defined by photosynthesis-related genes. Despite the wide evolutionary separation and independent origins of the C₄ phenotype, we report that these species use homologous transcription factors to both induce C₄ photosynthesis and to maintain the cell specific gene expression required for the pathway to operate. We define a core molecular signature associated with leaf and photosynthetic maturation that is likely shared by all angiosperm species derived from the last common ancestor of the monocotyledons and dicotyledons. We show that deep evolutionary comparisons of gene expression can reveal novel insight into the molecular convergence of highly complex phenotypes and that parallel evolution of trans-factors underpins the repeated appearance of C₄ photosynthesis. Moreover, the transcription factors that are shared by independent C₄ lineages are key targets for engineering the C₄ pathway into C₃ crops such as rice.



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Kentaro Shimizu



Genome-wide quantification of homeolog expression ratio revealed non-stochastic gene regulation in synthetic allopolyploid *Arabidopsis kamchatica*

Kentaro K Shimizu¹, Rie Shimizu-Inatsugi¹, Jun Sese², Satoru Akama²

¹ Institute of Evolutionary Biology and Environmental Studies, University of Zurich

² Tokyo Institute of Technology, Japan

Genome duplication with hybridization, or allopolyploidization, occurs commonly in plants, and is considered to be a strong force for generating new species. However, genome-wide quantification of homeolog expression ratios was technically hindered because of the high homology between homeologous gene pairs. To quantify the homeolog expression ratio using RNA-seq obtained from polyploids, a new method named HomeoRoq was developed, in which the genomic origin of sequencing reads was estimated using mismatches between the read and each parental genome. To verify this method, we first assembled the two diploid parental genomes of *Arabidopsis halleri* and *A. lyrata* collected in Far East, then generated a synthetic allotetraploid, mimicking the natural allopolyploid *A. kamchatica*. The quantified ratios corresponded well to those obtained by Pyrosequencing. We found that the ratios of homeologs before and after cold stress treatment were highly correlated (r=0.870). This highlights the presence of non-stochastic polyploid gene regulation despite previous research identifying stochastic variation in gene expression. Moreover, our new statistical test incorporating overdispersion identified 226 homeologs (1.11% of 20,369 expressed homeologs) with significant ratio changes, many of which were related to stress responses. HomeoRoq would contribute to the study of the genes responsible for polyploid-specific environmental responses. We then compared the homeolog expression ratios in *A. kamchatica* with the ratios of their corresponding genes in the parental species. The result shows that many new expression patterns including silencing and averaging were emerged, while at most 11% of homeologs keep the expression levels in the parental species. The high ratio of the non-additive expressions may indicate the functional improvement for new environmental adaptation.

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Christian Parisod



Genome evolution: causes and consequences of the dynamics of transposable elements

Christian Parisod, Céline Geiser, Amélie Bardil, Benjamin Dauphin, Natacha Senerchia

Laboratory of Evolutionary Botany, Institute of Biology, University of Neuchâtel

Transposable elements (TEs, also called jumping genes) represent the most abundant and dynamic fraction of genomes in almost all eukaryotes. The evolutionary significance of TEs and the genome instability they cause remains hotly debated. In particular, we poorly understand to what extent TE dynamics drives and is driven by the evolutionary trajectory of host taxa. The proximal and ultimate causes of genome instability can be suitably tackled using polyploid plants as the origin of such species couples rapid genome reorganization with the evolution of novel traits of ecological significance. Polyploidy reveals genomic conflicts resulting in drastic restructuring and methylation changes in TE genome fractions, offering a deeper understanding of the proximate factors of TE dynamics. The merging of increasingly different TE loads in both natural and experimental hybrids in the Buckler Mustard (polyploids within a species) and wild wheats (polyploids between species) indeed results in the activation of increasing numbers of TEs and rising levels of genome reorganization. These empirical observations fit our ‘diverge, merge and diverge’ model, providing a coherent framework predicting genome instability from the divergence between progenitors of a given polyploid. The ultimate consequences of genome instability are addressed by surveying natural hybrid zones between polyploids. In wild wheats, divergent TE fractions show significant reorganization after experimental hybridization and reduced introgression in natural hybrid populations, suggesting that TE dynamics is central to the maintenance of species boundaries. In a natural hybrid zone between ecotypes of the Buckler Mustard, genomic incompatibilities and divergent selection by habitat factors support the association of genome-wide variation to fine-scale environmental heterogeneity, despite gene flow. Such a case of incipient ecological speciation offers an outstanding opportunity to assess how TE dynamics interacts with adaptive processes in sustaining the build up of reproductive isolation.

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Cris Kuhlemeier



The genetic architecture of pollination syndromes

Cris Kuhlemeier, Korinna Esfeld, Michel Moser, Hester Sheehan, Avichai Amrad, Holly Summers

Institute of Plant Sciences, University of Bern

The genus *Petunia* (Solanaceae) comprises species that are adapted to distinct pollinators, bees, nocturnal hawkmoths and hummingbirds. We use a combination of genetics, genomics and behavioral studies to identify the genes underlying evolutionary shifts in pollination syndromes. The genetic architecture of such shifts is surprisingly simple. Even the modification of single genes can strongly affect pollinator preference and thereby cause reproductive isolation. We propose that such potential speciation genes, rather than neutral markers, can resolve phylogenetic relationships during recent radiations.

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Célia Baroux

Nuclear organization dynamics in plant developmental transitions: resolving causality

Wenjing She¹, Daniel Grimanelli², Kinga Rutowicz³, Marek WJ Whitehead¹, Marcin Puzio³, Maciej Kotliński³, Andrzej Jerzmanowski³, **Célia Baroux¹**

1 Institute of Plant Biology, University of Zürich (CH)
 2 IRD Montpellier (F)
 3 IBB, University of Warsaw (PO)

Plants retain a remarkable cellular plasticity after embryogenesis, in contrast to most animals. Whereas it is recognized that meristem activity, positional information and asymmetric divisions, together with the expression of key transcription factors contribute to plant cell lineage establishment, cytogenetic research from the past decades revealed an unsuspected dimension to the process: Nuclear organization is surprisingly dynamic during developmental transitions fueling a novel concept that large-scale chromatin reprogramming underlies cellular plasticity in plants. Yet, the challenge is now to decipher the causality and functional relevance of nuclear organization dynamics. As a case study, I will report on our findings that chromatin reprogramming during the somatic-to-reproductive transition in *Arabidopsis* contributes to establish gametophytic competence.



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Christian Hardtke

Automated quantitative histology reveals vascular morphodynamics during *Arabidopsis* hypocotyl secondary growth

Martial Sankar¹, Kaisa Nieminen¹, Laura Ragni¹, Ioannis Xenarios², **Christian S Hardtke¹**

1 Department of Plant Molecular Biology, University of Lausanne
 2 Swiss Institute of Bioinformatics

Among various advantages, their small size makes model organisms preferred subjects of investigation. Yet, even in model systems detailed analysis of numerous developmental processes at cellular level is severely hampered by their scale. For instance, expansion of *Arabidopsis* hypocotyls through secondary growth creates a radial pattern of highly specialized tissues that comprises several thousand cells starting from a few dozen. This dynamic process is difficult to follow because of its scale and because it can only be investigated invasively, precluding comprehensive understanding of the cell proliferation, differentiation and patterning events involved. To overcome such limitation, we established an automated quantitative histology approach. We acquired time series of hypocotyl cross-sections from tiled high-resolution images and extracted their information content using custom high-throughput image processing and segmentation. Coupled with automated cell type recognition through machine learning, we could establish a cellular resolution atlas that reveals vascular morphodynamics during secondary growth.



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Daniele Roppolo

Plasma membrane domains and local cell wall modifications

Daniele Roppolo

Institute of Plant Sciences, University of Bern, SNSF Ambizione fellow

How cells spatially control modifications of their extracellular space is a fundamental question in cell biology. Non-uniform secretion of extracellular materials can result in the establishment of polar domains, while fine tuned secretion produces local modifications of a minor portion of the cell surface. In plants, the presence of a uniform primary cell wall in all cells does not prevent secondary modifications to occur as full part of a cell differentiation process or in response to environmental conditions. Such cell wall modifications alter the physical properties of the apoplast and as a consequence its physiological behavior. We are approaching the question of how local cell wall modification is achieved in plants by looking at proteins that define membrane subdomains potentially associated with export of the cell-wall modifying machinery. We are focusing on the CASPL protein family, whose founder members are the Casparian strip membrane domain proteins (CASPs). CASPs are expressed in root endodermis, form a plasma membrane domain and localize secreted peroxidases that mediate lignin deposition. CASPLs are present in all land plants and in algae, and are related to animal MARVEL proteins. In *A. thaliana*, there are 5 CASPs and 33 CASPLs. CASPLs are expressed in a tissue-specific manner: in root, different CASPLs are expressed in the endodermis at different development stages, in the epidermis, in the lateral root cap, in the pericycle, in the vasculature. Above-ground CASPLs are found in the stomata lineage, in anther walls, in the floral organ abscission zone. In most of these tissues local or polar cell wall modifications have been described; we are then investigating the association between CASPL localization and such cell wall modifications. By comparing cell-type specific transcriptomes and proteomes, we aim at identifying CASPL molecular partners involved in 1) their localization and 2) the recruitment of the cell-wall modifying machinery.

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Markus Geisler

TWISTED DWARF1 regulates cytoskeleton dynamics

Markus Geisler

University of Fribourg, Department of Biology

The immunophilin-like FKBP42, TWISTED DWARF1 (TWD1), controls the polar transport of the plant hormone, auxin, by regulating the activity and plasma membrane presence of several members of the ABCB/PGP family of auxin transporters.

In order to understand the molecular basis of disoriented growth found for *twd1* plants both on the cellular and tissue level, we employed a co-immunoprecipitation-MS/MS approach in order to identify novel TWD1 interacting proteins that might be responsible for the “twisted syndrome”. We identified several actin isoforms that were themselves partially known to control root length and waving. Employing single-cell systems we found that *twd1* reveals previously overseen developmental defects and that actin single and double-mutant combinations phenocopy defects found in *twd1*. Quantification of auxin distribution and transport suggests that these defects are most likely caused by altered auxin transport capacities. Strikingly, actin reporter gene and VAEM analyses revealed that these defects correlate with altered actin bundling and dynamics in *twd1*, which is inline with the concept that TWD1 interferes with the stability of the actin cytoskeleton.

In summary, our findings contribute to our understanding of the role of the actin cytoskeleton in actin transport and underline the versatile role of TWD1 as auxin transport regulator.

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Stefan Hörtensteiner

Analysis of catabolite modifying reactions during chlorophyll breakdown

Bastien Christ, Mareike Hollenstein, **Stefan Hörtensteiner**

Institute of Plant Biology, University of Zurich

Chlorophyll degradation is a key feature of different developmental processes of plants such as fruit ripening and leaf senescence. The end products of degradation are colorless linear tetrapyrroles that are stored in the vacuoles of senescing cells. These products, termed “phylobilins”, are derived from chlorophyll by a multi-step pathway that involves an oxygenolytic opening of the porphyrin ring by PAO as the key step. Therefore, this pathway, which is commonly present in all higher plants, is termed “PAO/phylobilin” pathway.

Comparison of the chemical structures of phylobilins from different plant species uncovered that modification at different side positions of the common tetrapyrrolic backbone occur in a species-specific manner. In *Arabidopsis thaliana*, a total of four reactions are involved in the formation of more than 10 structurally different phylobilins: carboxymethylester hydrolysis, oxidative deformylation, ethyl side chain hydroxylation and subsequent glucosylation.

We aimed at identifying the enzymes that catalyze these phylobilin modifications. Genes of two of the involved enzymes, i.e. a methylesterase (MES16) and a cytochrome P450 monooxygenase (CYP89A9), could be identified because of their close co-expression with other known chlorophyll catabolic genes. The ethyl side chain hydroxylation is catalyzed by a Rieske-type monooxygenase, which is highly homologous to PAO. Different subcellular compartments are involved in phylobilin modification as concluded from the localization and substrate specificity of the three proteins. Thus, hydroxylation takes place at the chloroplast envelope and occurs before oxidative deformylation by CYP89A9 at the endoplasmic reticulum and carboxymethyl ester hydrolysis by MES16 in the cytosol.

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ABC1K1, a chloroplast kinase, couples the response of photosynthesis to highlight with metabolism

Jacopo Martinis¹, Gaétan Glauser², **Felix Kessler¹**

1 Laboratory of Plant Physiology, University of Neuchâtel
2 CAC Chemical Analytical Center, University of Neuchâtel

Plants are exposed to ever changing light conditions. High light intensity forces the photosynthetic machinery in chloroplasts to adapt and dissipate excess energy. This implicates non-photochemical quenching (NPQ) via the xanthophyll cycle. Alpha-tocopherol is produced to protect the thylakoid membrane against lipid oxidation. Here we demonstrate that ABC1K1, a member of the ABC1-like kinase family in chloroplasts, is identical with PGR6 (Proton Gradient Regulation 6). The *abc1k1/pgr6* mutant is strongly compromised in non-photochemical quenching and subject to photoinhibition at photosystem II after short high light exposure but recovers after longer exposure. Non-targeted metabolomics show that the photosynthetic defect under high light stress is accompanied by a reduction in carotenoid and tocopherol concentrations when compared to the wild type. These results indicate that the ABC1K1 kinase acts to upregulate key metabolic pathways to protect the photosynthetic machinery under high light stress.

Felix Kessler

Literature:
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New insights into starch synthesis through work in model plants, non-model plants and heterologous systems

Samuel C Zeeman

Department of Biology, ETH Zurich

Starch is a vital plant product for society. Learning more about its metabolism gives us options for crop improvement by altering starch structure, properties and yields. Starch is primarily composed primarily of the branched glucan, amylopectin, which has an architecture that allows the formation of insoluble, semi-crystalline granules.

Starch synthesis requires multiple isoforms of the biosynthetic enzymes (starch synthases and starch branching enzymes) that initiate granules and elaborate amylopectin. The crystallisation process is also facilitated by a specialised sub-class of the debranching enzymes, which are thought to selectively remove misplaced branch points. Despite this knowledge, starch biosynthesis has not yet been recreated in-vitro or in a heterologous system, suggesting that other, as-yet undiscovered protein factors may also be involved.

Much progress has been made by studying starch metabolism in the model plant *Arabidopsis thaliana*, where it is a primary product of photosynthesis, stored temporarily in chloroplasts during the day. Functional genomic and biochemical studies have advanced our understanding of the roles of known starch-metabolising enzymes and facilitated the discovery of new ones. However, non model plants can also provide novel insight. The myrmecophytic plant (ant-plant), *Cecropia peltata*, makes starch in its leaves like other plants, but also produces glycogen-rich food bodies for mutualistic ants that dwell within its hollow stems. We used next-generation sequencing and shotgun proteomics reveals how the biosynthetic apparatus is reconfigured to achieve this.

We are now using this knowledge to help steer our genetic studies in *Arabidopsis* and the expression of combinations of enzymes in heterologous systems, with the aim of defining a minimal set of enzymes/proteins required for starch production.



Samuel Zeeman

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Plastidial NAD-dependent malate dehydrogenase is critical for night-time plastidial redox homeostasis and embryo development in *Arabidopsis*

Seraina Beeler, Tina Schreier, Martha Stadler, **Oliver Kötting**

Institute of Agricultural Sciences, ETH Zürich

In illuminated chloroplasts, one mechanism involved in redox poise is the malate valve: Excess electrons from photosynthetic electron transport in the form of NADPH are used for the reduction of oxaloacetate to malate, thus regenerating the electron acceptor NADP. Malate is readily transported to the cytosol (in exchange for oxaloacetate) where it can serve various metabolic functions. The key enzyme of the malate valve is NADP-dependent malate dehydrogenase (MDH) – a strictly redox-regulated, light-activated enzyme that is inactive in the dark. However, not much is known about redox homeostasis in the dark, when glycolytic substrate-level phosphorylation of starch breakdown products leads to the coupled formation of ATP and NADH, which are used for biosynthetic processes at varying demands – indicating the need for redox poisoning mechanisms also in the dark.

Here, we show that the second plastidial MDH, which is strictly dependent on the coenzyme NAD (pdNAD-MDH), is crucial for night-time plastid redox homeostasis in *Arabidopsis*. A *pdnad-mdh* null mutation is embryo-lethal. Plants with reduced pdNAD-MDH levels by means of artificial microRNA are viable, but exhibit strong pleiotropic effects, for instance dwarfism, reductions in chlorophyll levels, and disordered chloroplast ultrastructure. Daytime metabolite levels seem unchanged except for reduced starch accumulation, which reflects the reduced photosynthetic rate. In the night, however, levels of malate and total glutathione, the major cellular thiol-disulfide buffer, are increased by about 100% compared with the wild type indicating that pdNAD-MDH is involved in redox poise in the dark. Moreover, deficiency of pdNAD-MDH cannot be functionally complemented by NADP-MDH illustrating distinct roles for NAD- and NADP-linked redox homeostasis.



Oliver Kötting

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Why growth controls photosynthesis

Christian Körner

Institute of Botany, University of Basel

In this presentation I will explain why the concept of photosynthesis driving growth does only apply to very special situations and is commonly irrelevant under natural life conditions. I will recall the classical rules of element stoichiometry and the known action of various stresses and limitations on tissue formation. These insights are not really new, but they neither reached textbook authors nor the modeling community, but they are acknowledged among crop physiologists for years. If growth and carbon cycle models arrive at plausible results, this is for the wrong reason, not reflecting the underlying mechanisms.

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Christian Körner

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Species boundaries and molecular markers

Yamama Naciri, Daniel Jeanmonod

Plant Systematics and Biodiversity Laboratory, University of Geneva and Conservatoire et Jardin botaniques de Genève

The use of few DNA sequences to assign specimens to species or to analyse species relationships implicitly suggests that species boundaries are easily captured using one or several markers. The situation is, however, much more complex. The study of plants offers many examples of molecules giving divergent signals that can, moreover, disagree with the accepted taxonomy of the organisms concerned. This can even be the case when species are well-defined morphologically and ecologically. Different factors explain such discrepancies, among them the inherent stochasticity of lineage sorting, chloroplast capture and gene flow. Several examples will be given from recent studies on *Gentiana*, *Zelkova* and *Silene* that question the traditional views of species concepts.



Yamama Naciri

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Conservation genetics in a changing world, genes, trees, and toy planes

Chris J Kettle

Ecosystem Management, Department of Environmental Science, ETH Zurich



Tropical deforestation and fragmentation continue to be the major drivers of biodiversity loss and compromise ecosystem services and rural livelihoods in many developing regions. Developing policies which ensure the resilience of tropical forests within complex agro-forest landscapes is thus one of the major challenges of the 21 Century. Ensuring that the foundation tree species can survive and provide the structural component of these landscapes is a basic requirement. I will present an overview of our research which informs conservation and restoration of forest trees and their genetic resources in a changing world.

Chris Kettle

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Island biogeography: moving beyond species numbers

Christoph Kueffer

Institute of Integrative Biology, Department of Environmental Systems Science, ETH Zurich



The island biogeography theory of McArthur and Wilson (1967) is one of the most influential theories in ecology. It is a powerful way to explain and predict the number of species present in an isolated land area as a consequence of immigration and extinction. However, the theory does not account for evolutionary processes, nor does it address patterns of phylogenetic or functional diversity. Thanks to the rapidly growing availability of phylogenetic and ecological data from oceanic islands, it becomes increasingly possible to address such dimensions of island biodiversity.

In this talk I will give an overview of research on the phylogenetic and functional diversity of island floras at local to global scales that we do in my research group and in collaboration with partners from around the world. We have recently compiled a global database of floristic data from c. 120 island archipelagos that allows us to investigate what determines phylogenetic diversity and drives adaptive radiations on islands. In the Seychelles (Western Indian Ocean) and Canary Islands (Macaronesia) we combine such information on speciation patterns with comprehensive data on functional traits collected in the field.

I will conclude by introducing a number of recent initiatives aimed at networking plant diversity research on islands including a global research network, an international conference in summer 2014 in Hawaii, and the launch of a new society and academic journal. Oceanic islands have long been used as model systems for research in biogeography, ecology, and evolution. With the increasing availability of data on phylogenies and functional plant diversity from many islands worldwide, islands have the potential to once again move to the forefront of ecological and evolutionary research.

Christoph Kueffer

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What can we learn from the past and current geographic distribution of a rare and endemic alpine plant when making future projections under climate change?

Christophe Randin¹, Theofania Patsiou^{1,2,3}, Spyros Theodoridis^{2,3}, Elena Conti²

1 Plant Ecology Unit, Botany, Department of Environmental Sciences, University of Basel
2 Institute of Systematic Botany, University of Zurich
3 Zurich-Basel Plant Science Center, Zurich



Ongoing rapid climate change is predicted to cause local extinction of plant species in mountain regions. However, some plant species have been able to persist during Quaternary climate oscillations without shifting their range. Here, we tested two candidate mechanisms of persistence by comparing the macrorefugia and microrefugia hypotheses. We used the rare arcto-tertiary endemic alpine plant *Saxifraga florulenta* as a model organism and combined ensembles of species distribution models (SDMs) with a high-resolution paleoclimatic and topographic dataset to reconstruct its potential current and past distribution since the last glacial maximum. To test the macrorefugia hypothesis, we first verified whether the species could have persisted in or shifted to geographic areas defined by its realized niche (macrorefugia). We then identified potential microrefugia based on the climatic and topographic properties of the landscape and applied refined scenarios of microrefugia dynamics and functions over time. Last, we quantified the number of known occurrences that could be explained by either the macrorefugia or microrefugia model.

A consensus of two or three SDM techniques predicted extinctions between 14-10, 3-4 ka and 1 ka BP, which did not support the macrorefugia model. In contrast, we showed that *S. florulenta* could have contracted into microrefugia during periods of extinction predicted by the SDMs and later re-colonized suitable areas via diffusion. A limited dispersal scenarios consistently explained a large number of the current occurrences (61-96%). Additionally, we showed that microrefugia could have facilitated species range expansions or shifts of *S. florulenta*. Finally, we found that the most recent and the most stable microrefugia were the ones closest to species occurrences.

Hence, we propose a novel paradigm to explain plant persistence by highlighting the importance of supporting functions of microrefugia when forecasting the fate of plant species under climate change.

Christophe Randin

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How do changing competitive interactions shape a species' response to climate change?

Jake Alexander

ETH Zürich, Institute of Integrative Biology

Climate affects species distributions through direct effects on demographic rates, and through indirect effects mediated by biotic interactions, such as competition. While it is assumed that these indirect effects are important, we know very little about the extent to which they could influence predictions of species distributions when climate changes. This is partly because of uncertainty about the species composition of future communities. This problem is largely circumvented along steep elevational gradients in mountains, because the species that will enter communities as climate warms tomorrow will most likely be those that are found only a few hundred meters lower down today.

We simulated different scenarios for the competitive environment that a species could encounter following climate change, in which species and/or their surrounding community either do or don't migrate to higher elevations following climate warming. We did this by transplanting focal alpine plants and monoliths containing intact plant communities along an elevational gradient. Already after one year, the growth and survival of the focal plants was much poorer in competition with a community from low elevation than one from high elevation when growing under a warmer climate; however, community identity did not affect performance under a cool climate. These results suggest that the ability of plants to persist in the face of climate warming will depend strongly on the rate of immigration of competitors from warmer areas. If this occurs more slowly than the pace of climate change, species might persist outside of areas where they would be predicted to occur based on their current observed climatic envelope.



Jake Alexander

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Different *Arabidopsis* leafcell types and the proteasome

Julia Svozil, Matthias Hirsch-Hoffmann, Robert Dudler, Wilhelm Gruissem, **Katja Baerenfaller**

ETH Zurich, University of Zurich

As proteins are the main effectors inside cells, their levels need to be tightly regulated. This is partly achieved by specific protein degradation via the Ubiquitin-26S proteasome system (UPS). In plants an exceptionally high number of proteins are involved in UPS-mediated protein degradation and it is known to regulate all important cellular processes. We investigated the response to the inhibition of the proteasome at the protein level treating leaves with the specific inhibitor Syringolin A (SylA) in a daytime specific manner. The patterns of protein level changes indicate that the accumulating proteins cause proteotoxic stress that triggers various responses. To distinguish between direct and indirect targets of the UPS we also enriched and identified ubiquitylated proteins after inhibition of the proteasome. The comparison of the ubiquitylated proteins with those changing in abundance after SylA-mediated inhibition of the proteasome confirmed the complexity of the response and revealed that some proteins are regulated both at transcriptional and post-transcriptional level.

As the contribution of different cell types is lost in the analysis of an entire organ, we developed the MeSeLeCT method to mechanically separate the leaf cell types epidermis, mesophyll and vasculature. In these different leaf cell types we determined the proteins that were exclusively identified in one of the cell types, that accumulated after inhibition of the proteasome or that were direct targets of the proteasome. With this, we gained insights into cell type specific processes regulated by the proteasome that are important for the functioning of the leaf.



Katja Baerenfaller

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Plasma Membrane H⁺-ATPase regulation is required for auxin gradient formation during phototropism

Emilie Demarsy^{1,4}, Tim Hohm^{2,3}, Clément Quan¹, Laure Allenbach Petrolati¹, Sven Bergmann^{2,3}, Christian Fankhauser¹

1 Center for Integrative Genomics, University of Lausanne
 2 Department of Medical Genetics, University of Lausanne
 3 Swiss Institute for Bioinformatics, Lausanne
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Plants have the ability to perceive light direction and adapt their growth accordingly, which optimizes photosynthetic yield. Although this process called phototropism has been observed for decades, the molecular mechanisms underlying it are still not completely elucidated. It is known that phototropins are the major photoreceptors that control this response. Downstream of the phototropin signalling takes place a lateral redistribution of the plant growth hormone auxin, leading to asymmetric growth. Nevertheless, it remains elusive which exact mechanisms are responsible for gradient formation. Recently we identified the plasma membrane localized H⁺-ATPases as interactors of phototropin1 and its substrate PKS4 (Phytochrome Kinase Substrate⁴).

Using a combination of genetic, pharmacological, physiological and biochemical approaches we could show that H⁺-ATPases regulation is required during phototropism and this regulation is phototropin dependent. Furthermore, a computational modelling approach suggested that differential cell wall pH regulation is a crucial factor for establishing auxin gradient. We recently validated this model prediction using transgenic *Arabidopsis* seedlings expressing a fluorescent reporter of auxin distribution.



Emilie Demarsy

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A rice *cis*-natural antisense RNA acts as a translational enhancer for its cognate mRNA and contributes to phosphate homeostasis and plant fitness

Mehdi Jabnoune1, David Secco1, Cécile Lecampion2, Christophe Robaglia2, **Yves Poirier1**

1 Department of Plant Molecular Biology, University of Lausanne,
2 Laboratory of Plant Genetics and Biophysics, Aix Marseille University

Cis-natural antisense transcripts (*cis*-NATs) are widespread in plants and often associated with down-regulation of their associated sense genes. We found that a *cis*-NAT positively regulates the level of a protein critical for phosphate homeostasis in rice. *OsPHO1;2*, a gene involved in phosphate loading into the xylem, and its associated *cis*-NAT^{PHO1;2} are both controlled from promoters active in the vascular cylinder of roots and leaves. While the *OsPHO1;2* promoter is unresponsive to the plant phosphate status, the *cis*-NAT^{PHO1;2} promoter is strongly up-regulated under phosphate deficiency. Expression of both *cis*-NAT^{PHO1;2} and the *OsPHO1;2* protein increased in phosphate-deficient plants, while *OsPHO1;2* mRNA level remained stable. Down-regulation of *cis*-NAT^{PHO1;2} expression by RNA interference resulted in a decrease in *OsPHO1;2* protein, impaired the transfer of phosphate from root to shoot and decreased seed yield. Constitutive over-expression of NATPHO1;2 in trans led to a strong increase of *OsPHO1;2* protein, even under phosphate-sufficient conditions. Under all conditions, no changes occurred in the level of expression, sequence, or nuclear export of *OsPHO1;2* mRNA. However, expression of *cis*-NAT^{PHO1;2} was associated with a shift of both *OsPHO1;2* and *cis*-NAT^{PHO1;2} towards the polysomes. These findings reveal an unexpected role for *cis*-NATPHO1;2 in promoting *OsPHO1;2* translation and affecting phosphate homeostasis and plant fitness.



Yves Poirier

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Nuclear control of chloroplast gene expression: why so complex?

Linnka Legendre, Karen Loizeau, Damien Douchi, **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 cleaved, processed at the 5’ and 3’ ends and spliced. 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 proteins that act in post-transcriptional steps of chloroplast gene expression raise many questions about their functions. Some may be involved in anterograde signaling from the nucleus to the chloroplast in response to environmental cues. In *Chlamydomonas*, 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 in *Chlamydomonas* the function of nucleus-encoded proteins that control the expression of specific chloroplast-encoded subunits of PSI and PSII at the level of splicing, mRNA stabilization and translation.

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M Goldschmidt-Clermont

1	Emmanuel Boutet
2	Etienne Bucher
3	Gina Cannarozzi
4	Claudia Cosio
5	Urs Feller
6	Manfred Heinlein
7	Roman Kellenberger
8	Doris Rentsch
9	Diana Santelia
10	Wenjing She
11	Sebastian Streb
12	Zerihun Tadele
13	Stefan Torriani

Posters



1	Emmanuel Boutet
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UniProtKB manual and automatic biocuration processes

Emmanuel Boutet 1, Lydie Bougueleret 1, Ioannis Xenarios 1,2 and the Swiss-Prot group

1 Swiss-Prot group, Swiss Institute of Bioinformatics, CMU, Geneva
2 Vital-IT Group, Quartier Sorge, Bâtiment Génopode, Lausanne

UniProt Knowledgebase (UniProtKB) is an expertly curated database consisting of two sections: UniProtKB/Swiss-Prot, a reviewed section containing manually-annotated protein sequences enriched with curator-evaluated computational analysis and functional information extracted from literature, and UniProtKB/TrEMBL, an unreviewed section with automatically annotated records.

The majority of UniProtKB records is based on automatic translation of coding sequences (CDS) provided by submitters at the time of initial deposition to the nucleotide sequence databases (INSDC). At this point, all the information included in the original submission, even the erroneous one, is transferred to the record. Selected entries are then manually annotated. Besides integrating, interpreting and standardizing data from literature and numerous resources, curators are also checking, and often correcting, gene model predictions. For plants, this task is limited to *Arabidopsis thaliana* and *Oryza sativa subsp. japonica*.

The development of manually curated annotation rules has also allowed to automatically enhance the information content associated with uncharacterized protein sequences with a high degree of accuracy. In this process, sets of aligned manually curated entries act as templates for rule creation that can then be applied to uncharacterized entries. This automated rule-based annotation systems allows annotation propagation in a controlled way to large sets of records in UniProtKB/TrEMBL. When templates are updated, the corresponding rules are also updated and so are the UniProtKB/TrEMBL records.

In response to the rapid increase of submitted sequences derived from massive genome sequencing, UniProtKB has developed the concept of complete proteome, defined as the entire set of proteins expressed by a specific organism. This provides to users the most relevant and best annotated protein sequences when searching instead of drowning in reports of redundant sequences. To ensure format standardization of sequences, UniProtKB had to implement pipelines for import of protein sequences from INSDC, Ensembl, EnsemblGenomes and RefSeq as sources for the underlying complete genomes. In UniProt release 2013_12, UniProtKB contains 2’220 complete proteomes of cellular organisms, of which 27 belong to the kingdom of green plants (Viridiplantae).

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Etienne Bucher

Epigenetic control of tissue and cell type specific gene expression

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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 specifically how TEs affect tissue specific gene expression.



In order to identify novel epigenetic factors, involved in this process we setup a mutant screen based on an epigenetically controlled and tissue specific reporter gene in Arabidopsis. Interestingly, we found mutants that were able to release transcription of the same transgene in various different tissues (young leaves, veins, leaf margin, flowers...). The causal mutations for some of these mutants have now been identified. Surprisingly, each mutant is involved in different silencing pathways acting in specific cells. This supports the notion that epigenetics is involved in cell type specific gene expression.

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Gina Cannarozzi

Genome and Transcriptome sequencing of Eragrostis tef

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Eragrostis tef is an allotetraploid cereal of central importance to the food security of the horn of Africa. The primary goal of the Tef Improvement Project is to provide new drought- and lodging- resistant cultivars to subsistence farmers in Ethiopia. Genomic sequencing of tef provides support for molecular breeding, enables us to find genes and networks responsible for important traits and allows comparison of the evolution of tef to that of other cereals. Currently, we have 50-fold coverage of the genome resulting in a genome size of 670 Megabases. Sequencing of a normalized library of the transcriptome revealed around 38,000 transcripts.



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Claudia Cosio

Effects of mercury and UV radiation on the aquatic plant *Elodea nuttallii*

Claudia Cosio

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In aquatic ecosystems, anthropogenic contamination with Hg is a topic of great concern, and threatens all levels of the ecosystems. *Elodea nuttallii* is a rooted submerged macrophyte tolerating a wide range of conditions and has been described as metal bioaccumulator¹. UV radiation has increased since the 1960, and is predicted to further increase in many regions².

We aimed to investigate the influence of enhanced UV radiation on the response of *E. nuttallii* to Hg. We analysed Hg accumulation, and fitness biomarkers. UV radiation decreased Hg uptake in shoots. Pigment content showed a decreasing trend in plants exposed to UV, high Hg concentration, and an additive effect of combined Hg and UV treatment. A similar pattern was observed on lipid peroxidation: no effect for Hg alone, but slightly decreased lipid peroxidation with UV. Addition of Hg to UV treatment significantly decreased lipid peroxidation in high Hg treatment. For oxidative stress enzymes, we observed an opposite effect of combined treatment: peroxidase activity was significantly decreased by UV and Hg treatments alone, whereas combined treatments abolished this effect. SOD activity was enhanced in UV and the high Hg treatments, but all other conditions did not affect SOD.

To summarize, the combination of Hg and UV stresses were additive for pigment content and lipid peroxidation, but antagonist for oxidative stress enzymes. Accumulation of Hg was in addition reduced by UV treatment, indicating that in natural ecosystems enhanced UV radiation could affect Hg fate in natural ecosystems, notably Hg distribution in biota.

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Urs Feller

Dehydrins: diagnostic tools for plants under abiotic stresses?

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Dehydrins (LEA group 2 proteins) are characterized by very special properties: they contain highly conserved domains (at least one K-segment and may also have one or several Y- and/or S-segments), are stable up to 95 °C, accumulate during late embryogenesis and change their expression pattern under various abiotic stresses (e.g. drought, cold or salt stress). They respond to adverse environment, including both stress and recovery phases, within a reasonable time period (days rather than minutes). Since distinct dehydrin (DHN) types drastically increase under abiotic stress in a typical manner and are quite stable *in vitro*, they may serve as diagnostic tools. Antibodies against the three well conserved domains allow the detection on immunoblots and evaluation and characterization of DHNs on protein level. A series of dehydrins containing K- and Y-segments are found to accumulate under drought. On the other hand, a different pattern is observed in plants subjected to cold or salt stress. The stress-provoked dehydrin profile disappears within a few days in the leaves of recovered from drought-plants. Interesting properties are observed on the m-RNA level. Distinct DHN transcripts accumulate under stress (some of them as a consequence of alternative splicing and/or alternative start codons). Recently an impressive series of natural DHN antisense transcripts (NATs) were identified. It is not yet clear whether and if so how they participate in the regulation of specific dehydrin prevalence. Dehydrin NATs were found to be encoded by the same locus (*cis*-NATs) or originate from different genomic loci (*trans*-NATs). Generally it is accepted that NATs may influence transcription, splicing of pre-mRNA, translation and mRNA stability. In summary, tools mentioned (the set of primers and the specific DHN antibodies) may be helpful in applied research to identify responses of plants to abiotic stresses.



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Manfred Heinlein



TMV movement in the context of plant defense responses

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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. Other parts of our work concentrate on cellular and long-distance signaling responses elicited by the virus. 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 and leads for the development of antiviral strategies in crops.

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Roman Kellenberger



Causes and consequences of epigenetic variation in plant interactions with pollinators and herbivores

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There is increasing evidence that epigenetic processes can stably alter phenotypic traits without introducing changes in DNA sequence, sometimes over multiple generations. In plants, interactions with enemies such as herbivores can trigger heritable epigenetic changes, and traits important for pollination such as floral symmetry can be regulated in an epigenetic fashion. However, the role of epigenetic mechanisms in co-evolution of different plant-insect relationships is still not fully understood.

This study aims to shed light on the epigenetic crosstalk between the model crop plant species *Brassica rapa* and its different insect visitors: 1) Do biotic stresses such as herbivory lead to changes in DNA methylation in *B. rapa*? 2) Are these epigenetic changes heritable? 3) Do these epigenetic changes alter the floral phenotype of *B. rapa*? 4) Does an altered floral phenotype affect plant-pollinator interactions? 5) What are the downstream genetic targets of the induced changes in DNA methylation?

Preliminary evidence suggests that herbivore damage in *B. rapa* can cause significant changes to DNA methylation patterns in leaves. Also, demethylation leads to changes in the expression in a number of floral volatile compounds that may affect plant-insect interactions. Future work will use a GWAS approach to test for associations between individual methylation sites across the genome that change as a result of herbivory, and correlations between these sites with floral signals and pollinator attractiveness.

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Regulators of peptide transport and metabolism in *Arabidopsis thaliana*

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Doris Rentsch

Transport of small peptides of two to three amino acids is suggested to be important for translocation of organic nitrogen within the plant, for plant nutrition and development. The plasma membrane-localized peptide transporter PTR1 of *Arabidopsis thaliana* mediates transport of di- and tripeptides with low selectivity and high affinity (1, 2). Consistent with AtPTR1 transport properties and the expression in roots, *atptr1* T-DNA insertion lines displayed reduced growth compared to wild type plants on medium with dipeptides as sole nitrogen source, while overexpressing lines (35S::AtPTR5) produced more biomass (3). To identify regulators of peptide transport or metabolism two different approaches were used. The current state of this work will be presented. So far, (i) five mutants with reduced growth on dipeptides as nitrogen source were identified by screening an EMS mutagenized M2 population. While three of them are allelic to *AtPTR1*, for the two remaining mutants next-generation mapping (NGM) identified the mutation-harboring region on different chromosomes. (ii) Comparison of 38 different *Arabidopsis* accessions identified accession Est-0 and Tanz-1 as most efficient in utilizing dipeptides as nitrogen source. Advanced intercross-recombinant inbred lines (Est-1xCol-0) (4) are currently being used for QTL mapping.

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Diana Santelia



Mechanism of starch degradation in *Arabidopsis* guard cells

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Stomata are small adjustable pores situated in plant epidermis. They are surrounded by a pair of guard cells which control their opening in response to many environmental stimuli. The central role of stomatal pores is to regulate water loss and CO₂ exchange into and out the leaves, allowing photosynthesis while conserving water to avoid dehydration. A complex signalling and metabolic network regulates guard cell function. While the role of ABA, potassium and chloride is well established, the metabolic network linking malate and other organic acids to sugars and/or starch remains unclear. Here, we show starch degradation in *Arabidopsis* guard cells occurs very rapidly during the first 3h hours of light, through the specific action of β-amylase 1 (BAM1), which releases maltose from the nonreducing ends of glucan chains. We also provide evidence that in guard cells starch degradation, but not synthesis, is dependent on blue light signaling and is necessary for proper stomatal opening under blue light.

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Wenjing She

An efficient method for quantitative, single-cell analysis of chromatin modification and nuclear architecture in whole-mount ovules in *Arabidopsis*

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In flowering plants, the somatic-to-reproductive cell fate transition is marked by the specification of spore mother cells (SMCs) in floral organs of the adult plant. The female SMC (megaspore mother cell, MMC) differentiates in the ovule primordium and undergoes meiosis. The selected haploid megaspore then undergoes mitosis to form the multicellular female gametophyte, which will give rise to the gametes, the egg cell and central cell, together with accessory cells. The limited accessibility of the MMC, meiocyte and female gametophyte inside the ovule is technically challenging for cytological and cytogenetic analyses at single cell level. Particularly, direct or indirect immunodetection of cellular or nuclear epitopes is impaired by poor penetration of the reagents inside the plant cell and single-cell imaging is demised by the lack of optical clarity in whole-mount tissues.

Thus, we developed an efficient method to analyze the nuclear organization and chromatin modification at high resolution of single cell in whole-mount embedded *Arabidopsis* ovules. It is based on dissection and embedding of fixed ovules in a thin layer of acrylamide gel on a microscopic slide. The embedded ovules are subjected to chemical and enzymatic treatments aiming at improving tissue clarity and permeability to the immunostaining reagents. Those treatments preserve cellular and chromatin organization, DNA and protein epitopes. The samples can be used for different downstream cytological analyses, including chromatin immunostaining, Fluorescence In-Situ Hybridization (FISH) and DNA staining for heterochromatin analysis. Confocal laser scanning microscopy (CLSM) imaging, with high resolution, followed by 3D reconstruction allows for quantitative measurements at single-cell resolution.



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Sebastian Streb



Establishing a universal method to predict protein complexes and generate protein interaction networks

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The era of whole genome sequencing has yielded unparalleled amounts of detailed genetic information. However, what genetic information cannot tell us is how these tens of thousands of proteins interact and work together in the cell. Many biological processes e.g. DNA replication, protein synthesis/degradation, primary and secondary metabolism require stable long term multimeric protein complexes. Unfortunately, our knowledge about their assembly is incomplete and a pastiche collection from several different species, often based on predictions. All attempts to define protein interactome maps required high investment of human resources and money.

Here, we show that with three basic native separation methods coupled to a proteomics approach, we can predict the subunit composition based on co-behavior of the proteins. Currently, we are able to define potential association in protein complexes for more than 2000 proteins in *Arabidopsis thaliana* and *Saccharomyces cerevisiae*. Surprisingly, the majority of the proteins assemble in multimeric long term complexes and the *in vivo* status of a protein in a monomeric form is rather exceptional.

The next goal is to develop a standard methodology which can be applied to any organism for which the genome is known with affordable cost for “standard” Laboratory to study protein complex composition and changes in their biological samples of interest.

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Zerihun Tadele



Boosting productivity of the orphan crop tef by breeding for semi-dwarfism and lodging tolerance

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Genetic improvement of native crops is a new and promising strategy to combat hunger in the developing world. Tef is the major staple food crop for about 50 million people in Ethiopia. As an indigenous cereal, it is well adapted to diverse climatic and soil conditions; however, its productivity is extremely low mainly due to susceptibility to lodging. Tef has a tall and weak stem which is liable to lodging or falling over which is aggravated by wind, rain or application of nitrogen fertilizer. To circumvent this problem, we developed the first semi-dwarf lodging-tolerant tef line, called *kegne*, from an EMS mutagenized population. Compared to the parental line, *kegne* is shorter in plant height and lodging-tolerant. The response of *kegne* to microtubule depolymerization and stabilizing drugs as well as subsequent gene sequencing and segregation analysis suggests that a defect in the α -tubulin gene is functionally and genetically tightly linked to the *kegne* phenotype. A single base pair mutation in the α -tubulin gene replaces the polar and uncharged amino acid threonine with the hydrophobic amino acid isoleucine. Introgression of *kegne* into locally adapted and popular tef cultivars in Ethiopia increased the lodging tolerance in the tef germplasm and will improve the productivity of this valuable crop. Our work highlights the advantages and opportunities associated with indigenous crop. Major yield gains achieved will efficiently reach subsistence farmers through well-established seed multiplication and distribution channels.

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Stefano Torriani



Transcriptome of the wheat pathogen Zymoseptoria tritici during a complete infection cycle

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Zymoseptoria tritici (aka *Mycosphaerella graminicola*) is a hemi-biotrophic fungus belonging to the Dothideomycetes, the largest class of ascomycetes that includes many plant pathogens. Like other hemibiotrophic pathogens *Z. tritici* uses different strategies for obtaining nutrition during its life cycle. For the first 10 days post inoculation (dpi) the pathogen survives as a biotroph without causing visible symptoms. The necrotrophic phase lasts until the affected plant cells have died. Depending on the strain-cultivar interaction, complete plant cell death occurs from 18 to 20 days after penetration. *Z. tritici* concludes its life cycle by surviving as a saprotroph on dead leaves for several months. Thus *Z. tritici* presents a powerful system to study host-pathogen interactions during different stages of disease development. Illumina sequencing technology was used to analyze changes in transcription during the complete infection cycle of *Z. tritici* on wheat across three biological replicates. Total RNA was extracted from inoculated plants at seven time points (3-, 7-, 11-, 13-, 14-, 21- and 56- dpi). The expression profile of 10,251 genes was analyzed. About 14% and 34% of the genes showed statistically significant differences in expression from the biotrophic to necrotrophic and from the necrotrophic to saprotrophic stages of infection, respectively. We identified five novel putative effector genes. Putative effector genes were preferentially transcribed at 11 dpi during the transition between biotrophy and necrotrophy. We also investigated the expression of gene clusters involved in the biosynthesis of non-proteinaceous metabolites. Two clusters of genes (PKS4 and PKS5-related genes) showed expression patterns similar to the putative effectors. A putative effector, PKS5 and a hemicellulase under diversifying selection were further characterized, using Agrobacterium-mediated transformation to determine their role in pathogenicity. From the functional analyses of the knockout strains we showed that eliminating the putative effector delayed the onset of necrosis by 48h. This study shows how next generation sequencing techniques can be used to screen the transcriptome during plant-pathogen interactions to identify novel virulence factors for further functional analyses.

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