

SWISSPLANT 2017



Symposium of the Plant Science Research Community in Switzerland

Scientific Program Committee

Sam Zeeman, Consuelo De Moraes, Willi Gruitsem,
Antía Rodríguez-Villalón, Clára Sánchez-Rodríguez,
and Alex Widmer
ETH Zurich

Organizer

Swiss Plant Science Web

swissplantscienceweb.ch – 26th edition



25–27 January 2017
Leukerbad (VS)

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SWISSPLANT 2017

**Symposium of the Swiss Plant Science Web
25–27 January 2017
Leukerbad (VS), Switzerland**

Venue

Thermalhotels Leukerbad, Leukerbad

Scientific Program Committee

Sam Zeeman, Consuelo De Moraes, Willi Gruissem,
Antía Rodríguez-Villalón, Clára Sánchez-Rodríguez, and Alex Widmer
ETH Zurich

Conference Organization

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SwissPlant 2017 – Welcome by the SPSW president

The Swiss Plant Science Web (SPSW) held its first scientific conference «SwissPlant» in 2011. These yearly conferences are now well established and highly popular in our community, and they have become a showcase of the current advances of plant science research in Switzerland. They bring together research from molecular biology to ecology, and thereby allow a broader view and better understanding of the complexity and diversity of plant life.

In addition, by choosing an attractive place for our conferences, we hope to foster informal discussions among peers. This year, like two years ago, it will take place in the beautiful resort of Leukerbad (canton of Valais/Wallis), where one of the best-known plant developmental biologists (J.W. von Goethe) spent a memorable night on November 9, 1779, 237 years ago.

I especially welcome all the new members of our SPSW network and hope that they feel at ease in our community. With their new ideas, they will help to shape the long-term continuation of the Swiss Plant Science Web. «SwissPlant 2017» is the ideal place to start discussions on new collaborations!

This year's scientific conference committee is from the ETH Zürich. I thank Sam Zeeman, Consuelo de Moraes, Willi Grissem, Antía Rodríguez, Clára Sanchez and Alex Widmer for setting up an exciting program. I am also very grateful to Sylvia Martínez, our SPSW coordinator, who took a big effort to organize all practical things and to make your stay here smooth and easy.

Enjoy the science – as well as the leisure in the spas and on the slopes of Leukerbad!

Thomas Boller,
University of Basel, SPSW president

The Swiss Plant Science Web is the umbrella organization for plant science research and education at universities in Switzerland.

Program

Wednesday, 25 January 2017

15:30	Swiss Plant Science Web strategic meeting for SPSW members
17:00	Welcome drink
17:25	Welcome by Thomas Boller , SPSW president Opening remarks by Samuel Zeeman , Scientific Program Committee
Session I, chair: Consuelo De Moraes	
17:30	Enrico Martinoia U Zurich page 24 My journey with malate – ups and downs with a good friend
18:00	Andrzej Kurenda U Lausanne page 23 Involvement of hydraulic signals in leaf-to-leaf wound signalling in <i>Arabidopsis</i>
18:20	Sam Zeeman ETH Zurich page 38 Protein targeting to starch: A new class of proteins that bring enzymes and substrates together
18:40	Markus Geisler U Fribourg page 15 ABCG36/PDR8 regulation by the LRR receptor-like kinase, ALK1
19:00	Dinner
Session II, chair: Alex Widmer	
20:20	Stefan Hörtensteiner U Zurich page 17 Biochemical and molecular analysis of dephytylation during chlorophyll breakdown
20:40	Felix Mauch U Fribourg page 25 Finally: A biochemical function for the PATHOGENESIS-RELATED PROTEIN 1
21:00	Antía Rodríguez-Villalón ETH Zurich page 28 Perturbing phosphoinositide homeostasis during vascular cell differentiation: a tale of two halves?
21:20	Samuel Wuest U Zurich page 37 Explaining the reproductive allocation strategy of <i>Arabidopsis</i>
21:50	Get-together at the hotel bar

Thursday, 26 January 2017

7:00	Breakfast begins
	Session III, chair: Willi Gruissem
8:00	<u>Anne Roulin</u> U Zurich page 29 Genome wide analysis of transposable elements in <i>Brachypodium distachyon</i> : insights from natural populations
8:20	<u>Christian Fankhauser</u> U Lausanne page 14 Shade promotes phototropism through phytochrome B-controlled auxin production
8:40	<u>Felix Kessler</u> U Neuchâtel page 22 VTE5 phytol kinase is essential for resistance to combined light and temperature stress in tomato
9:00	<u>Nicolas Bologna</u> ETH Zurich page 11 Dynamic nucleo-cytoplasmic shuttling of <i>Arabidopsis</i> AGO1 is required for RNA silencing
9:20	<u>Sebastian Streb</u> ETH Zurich page 33 Increasing the carbohydrate storage capacity of plants by engineering a glycogen-like polymer pool in the cytosol
9:40	Coffee break
	Session IV, chair: Sam Zeeman
10:10	<u>Ivan Baccelli</u> U Neuchâtel page 7 The priming molecule β -aminobutyric acid (BABA) is a natural product of plants: clues for a new stress hormone
10:30	<u>Katja Baerenfaller</u> ETH Zurich page 8 Modelling the dynamics of protein synthesis and degradation in flagellin 22-stimulated <i>Arabidopsis</i> cell cultures
10:50	<u>Matt Horton</u> U Zurich page 19 Identifying the genes important in adaptative variation requires an integrative approach
11:10	<u>Klaus Schlaeppli</u> Agroscope page 31 Deciphering functions of the plant root microbiome

11:30	<u>Matthias Thalmann</u> U Zurich page 34 Stress-induced changes of leaf carbohydrate metabolism are modulated by the circadian clock and ABA
11:50	Leisure time (lunch on your own, skiing, hiking, snowshoeing, thermal spa...)
	Session V, chair: Sebastian Streb
17:00	<u>Célia Baroux</u> U Zurich page 9 How shape constrains cell fate: a 3D look at ovule growth in <i>Arabidopsis</i>
17:20	<u>Gavin George</u> ETH Zurich page 16 EGES-2: A system for the accurate measurement of photosynthesis and respiration of herbaceous plants with digital control of environmental parameters
17:40	<u>Michael Hothorn</u> U Geneva page 20 Inositol pyrophosphate signaling molecules control plant phosphate homeostasis by binding to SPX sensor domains
18:00	<u>Sarah Robinson</u> U Bern page 27 Intrinsic cell wall properties uncouple growth from growth hormones
18:20	<u>Thomas Städler</u> ETH Zurich page 32 Genomic imprinting in the endosperm is systematically perturbed in abortive hybrid tomato seeds
18:40	Dinner
	Poster session (drinks will be served) pages 40–41

Friday, 27 January 2017

7:00 Breakfast begins

Session VI, chair: Antía Rodríguez-Villalón

- 8:00 Beat Keller | U Zurich | page 21
Pathogen-derived proteins recognized by wheat NLR immune receptors
- 8:20 Daniel Croll | U Neuchâtel | page 13
What pathogen genomes tell us about rapid disease evolution in agro-ecosystems
- 8:40 Michel Goldschmidt-Clermont | U Geneva | page 18
Perception of UV-B by the photoreceptor UVR8 induces protection of the photosynthetic apparatus against excess light in *Chlamydomonas*
- 9:00 Elisabeth Truernit | ETH Zurich | page 35
OCTOPUS-LIKE 2, a novel player in *Arabidopsis* root and vascular development, reveals a role for OCTOPUS family genes in root metaphloem sieve tube differentiation
- 9:20 Joop Vermeer | U Zurich | page 36
Cell volume regulation during lateral root formation in *Arabidopsis*
- 9:40 Coffee break
- Session VII, chair: Elisabeth Truernit**
- 10:10 Betty Benrey | U Neuchâtel | page 10
Cascading effects of early-season herbivory on late-season herbivores and their parasitoids
- 10:30 Lorenzo Borghi | U Zurich | page 12
The strigolactone transporter PDR1 regulates plant biomass production on nutrient-poor soil
- 10:50 Julia Santiago | U Lausanne | page 30
Mechanistic insights into floral abscission and cell wall remodeling
- 11:10 Bruno Müller | U Zurich | page 26
Cytokinin distribution and transport to support plant morphogenesis
- 11:30 Tobias Züst | U Bern | page 39
Novel defenses as an escape from co-adapted herbivores: cardenolides in the Brassicaceae
- 11:50 Sam Zeeman, closing remarks, Scientific Program Committee

The priming molecule β -aminobutyric acid (BABA) is a natural product of plants: clues for a new stress hormone

Ivan Baccelli¹, Damien Thevenet², Andrea Balmer¹, Victoria Pastor¹, Armelle Vallat³, Reinhard Neier², Gaétan Glauser³, and Brigitte Mauch-Mani¹

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Plants can be sensitized to respond faster and/or stronger to stress situations by application of the non-protein amino acid β -aminobutyric acid (BABA), a well-known priming agent. BABA can increase resistance against a wide range of stresses, such as attacks by pathogens, nematodes, and arthropods, as well as abiotic stressors like heat, cold or salt. Plants treated with BABA can deploy more rapidly the signaling pathway most appropriate to counteract the given stress situation. For instance, against *Plectosphaerella cucumerina* infection, BABA priming leads to an ABA-dependent enhancement of the callose response at the sites of attempted penetration, and thus to increased resistance. With the present study we provide evidence that BABA, which has been considered a xenobiotic for more than 50 years, is actually produced by plants and its endogenous levels increase after stress. After developing a sensitive and selective detection method based on ultra-high pressure liquid chromatography tandem mass

spectrometry (UHPLC-MS/MS), we were able to separate BABA from its two isomers alpha (AABA) and gamma (GABA) that are present in plant tissues, and to quantify it. Subsequent analyses revealed that BABA is present in various plant species, including *Arabidopsis* and crops like maize and wheat. Importantly, BABA levels were found to increase following infection with necrotrophic, biotrophic and hemibiotrophic pathogens, as well as after salt stress and submergence. Interestingly, virulent, avirulent and non-pathogenic strains, or elicitors like flagellin, all induce BABA.

By considering the broad efficacy of this molecule on plants and the perception mechanism recently revealed in *Arabidopsis*, the endogenous finding of BABA and its variation patterns suggest that it may be a novel stress hormone.

> SPSW researcher portfolio:
<https://swissplantsciencweb.ch/nc/research/home/portfolio/mauch-mani>

Modelling the dynamics of protein synthesis and degradation in flagellin 22-stimulated *Arabidopsis* cell cultures

Phillip Ihmor¹, Wilhelm Gruissem¹, Mark Robinson² and **Katja Baerenfaller**¹

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² Institute of Molecular Life Sciences, University of Zurich, Switzerland

As proteins are the main effectors inside cells, their levels need to be tightly regulated. Protein levels can be regulated via changes in transcript levels and post-transcriptionally through changes in the rates of translation and protein degradation. To assess the relative contributions of transcription, translation and protein degradation towards protein level regulation in response to a biotic stress we have challenged cultured *Arabidopsis* cells with the bacterial pathogen-associated molecular pattern (PAMP) flagellin 22 or did a mock treatment.

Over a time course we have then acquired the high-throughput datasets: 1) RNA sequencing (RNA-Seq) for transcription, 2) ribosome sequencing (Ribo-Seq) for translation, 3) SILAC (stable isotope labelling with amino acids in cell culture) MS/MS (tandem mass spectrometry) for protein abundance and 4) pulse-chase SILAC MS/MS for protein turn-over. Of special interest are the pulse-chase SILAC data on protein turn-over, as to our knowledge such data have never been acquired in plants. Analysing the different datasets we observed that treatment with flagellin 22 induces a multi-level response on all regulatory levels. Combining the different datasets allows us now to determine the relative contributions of protein synthesis and degradation towards protein level regulation in response to a biotic stress.

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How shape constraint cell fate: a 3D look at ovule growth in *Arabidopsis*

Ethel Mendocilla Sato¹, Nuno Pires¹, Daphné Autran² and **Célia Baroux**¹

¹ Institute of Plant and Microbial Biology, University of Zurich, Switzerland

² Institute for Research and Development (IRD), Montpellier, France

The reproductive lineage of flowering plants is established late in development. The differentiation of male and female spore mother cells (SMC) marks the somatic-to-reproductive fate transition. In *Arabidopsis*, a single SMC differentiate in a stereotypical, subepidermal and centrally located position in the ovule primordium. Starting as a dome of 3–4 cells at the surface of the placenta, the ovule primordium achieves an elongated, digit-shape upon a limited number of cell division and elongation events. SMC differentiation is tightly linked to ovule growth but whether a causal relationship exists is unknown.

Does tissue patterning influences SMC fate acquisition? Reciprocally, does SMC fate acquisition influences ovule shape? Using high-resolution 3D imaging at different developmental time points, 3D image segmentation and a customized, quantitative analysis pipeline, we reconstructed cell division and anisotropic growth (patterning events) contemporary to SMC differentiation. Interestingly, mutant analysis suggested a functional link between patterning perturbation and SMC fate. The work in progress allows to propose a novel conceptual framework to understand the influence of organ shape on cell fate.

> SPSW researcher portfolio:
<https://swissplantscienceweb.ch/nc/research/home/portfolio/baroux>

Cascading effects of early-season herbivory on late-season herbivores and their parasitoids

Betty Benrey and Johnattan Hernandez-Cumplido

Institute of Biology, University of Neuchatel, Switzerland

Individuals of different species interact in local communities and can influence each other's fitness both directly or indirectly through intermediaries. This is particularly true for the interactions among plants, insect herbivores and the natural enemies of these herbivores. Using a combination of field and laboratory experiments, we examined how early season herbivory by beetles on lima bean plants affects plant performance and the abundance and performance of seed predators and their parasitoids, which occur on the plants at the end of the growing season. In addition, we determined the consequences of early herbivore-induced defenses for plant performance. We hypothesized that early-season induction will affect plant reproduction and alter the suitability of seeds for late season seed-eating beetles, and the vulnerability of these seed beetles to parasitoids.

We found strong support for these hypotheses. In the field, early-season herbivory negatively affected plant reproduction and seeds of these plants suffered lower infestation by seed-eating beetles, which in turn suffered less parasitism. Laboratory assays with field-collected seeds confirmed that the performance of beetles and parasitoids was lower on seeds from plants that had been subjected to early season herbivory. Further analyses revealed that seeds produced by control plants contain larger quantities of two important cyanogenic compounds and had a higher total protein content than seeds from plants subjected to herbivory. Our results provide insight into how direct and indirect interactions between and within different trophic levels affect the dynamics and structure of complex communities

> SPSW researcher portfolio:
<https://swissplantsciencweb.ch/nc/research/home/portfolio/benrey>

Dynamic nucleo-cytoplasmic shuttling of *Arabidopsis* AGO1 is required for RNA silencing

Nicolas G. Bologna, Raphael Iselin, Luciano A. Abriata, Nathan Pumplin, Alexis Sarazin, Pauline E. Jullien and Olivier Voinnet

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Small (s)RNAs, ~20-24-nucleotide (nt) in size, are major eukaryotic regulators of gene expression. While they may vary in terms of their biogenesis and size, all sRNAs are incorporated into RNA-induced silencing complexes (RISCs) to regulate target gene expression through transcriptional gene silencing (TGS) in the nucleus or post-transcriptional gene silencing (PTGS) in the cytoplasm. ARGONAUTE (AGO) proteins are the main component of RISCs. Canonical eukaryotic AGOs contain four main domains: a variable N-terminal domain and the more highly conserved PAZ, MID, and PIWI domains that fold into a bi-lobal structure displaying a central groove for sRNA binding. The *Arabidopsis* genome encodes 10 AGO proteins, defining three major phylogenetic clades: (i) AGO1, -5, and -10; (ii) AGO2, -3, and -7; and (iii) AGO4, -6, -8, and -9. Whereas AGO4, -6, and -9 associate mostly with 24-nt siRNA mediating TGS, AGO1, -2, -5, -7 and -10 bind 21–22-nt molecules acting in PTGS. To characterize *Arabidopsis* AGO proteins we systematically analyzed fluorescence reporter of these proteins and found a clear correlation between clades, function and subcellular localization.

Unexpectedly, however, we found that despite its main cytoplasmic localization, AGO1 exhibits a non-canonical nuclear localization signal (NLS) and a highly conserved canonical nuclear export signal (NES) whose activity is inhibited by drugs antagonizing the CRM1 pathway. Both signals, located in a non-structured part of the N-terminal domain, are functional in vivo and required for the normal PTGS functions of AGO1, suggesting that a crucial step in the biology of AGO1-bound sRNAs occurs in the nucleus. Directed mutagenesis and highly stringent nuclear purification coupled to high-throughput sequencing have now provided insights into the nuclear step of AGO1 biology, which will be discussed alongside a dynamic structure-based model possibly explaining how AGO1 might constantly shuttle between the nucleus and the cytoplasm.

> SPSW researcher portfolio:
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The strigolactone transporter PDR1 regulates plant biomass production on nutrient-poor soil

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⁴ Carnegie Institution for Science, Washington DC, USA

⁵ Goethe-Universität Frankfurt am Main, Germany

The phytohormone strigolactones (SLs), initially characterized as germination stimulant of parasitic weeds, are carotenoid derivatives that regulate plant architecture and induce hyphal branching of arbuscular mycorrhizal fungi (AMF). The ABCG protein PDR1 from *Petunia hybrida* is up-to-date the only reported SL cellular exporter. PDR1 is asymmetrically localized in root cortex cells, compatible with a shoot-ward transport of SL, and in hypodermal passage cells (HPC), entry gates to AMF. PDR1 is induced by low phosphate (Pi) and hormones such as auxins and SLs.

To investigate if PDR1 acts at the interface between soil nutrient uptake and plant growth, we studied the effects of de-regulating SL transport by over-expressing PDR1 (PDR1 OE). PDR1 OE plants produce more biomass on Pi-poor soils compared to wildtype, due to higher mycorrhization levels but also to dramatic changes in root cell identity, root architecture and cell division rates. By mean of X-ray computed tomography, a non-destructive method, we investigated *Petunia* below-ground development and found that PDR1 OE induces lateral roots and root hair elongation, thus integrating nutrient availability with plant development. SLs, in antagonism to auxins and ABA, also regulate the number of HPC along the main root. We show here that HPC are preferential entry gates to K⁺ and Mg²⁺ ions. Therefore, on K⁺ and Mg²⁺ poor soils plants mutant for SL biosynthesis or transport are disadvantaged compared to wildtype, on the contrary PDR1 OE better counteract such conditions.

At support of the higher SL transport capability, the SL biosynthesis pathway is induced in PDR1 OE plants, possibly as effect of feedback regulation. We propose therefore that PDR1 OE, because of its enhanced Pi and ion uptake, could be a useful tool to increase plant yields in nutrient-scarce soils.

> SPSW researcher portfolio:

<https://swissplantsciencweb.ch/nc/research/home/portfolio/martinoia>

What pathogen genomes tell us about rapid disease evolution in agro-ecosystems

Daniel Croll^{1,2}, Fanny Hartmann², Clémence Plissonneau², Andrea Sanchez-Vallet², Bruce A. McDonald², and Javier Palma-Guerrero²

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Most plants face attacks by pathogens. In agriculture, outbreaks of fungal diseases are frequent and pose a significant threat to sustainable food production. What enables pathogens to overcome host defenses and cause damage is poorly understood. A key evolutionary step for pathogens is to evolve effectors that specifically target and disable the plant immune system. We use experimental and population genomics tools to identify the genes underlying pathogenicity. Our main model is the fungus *Zymoseptoria tritici*, which causes one of the most important diseases on wheat. Based on large collections of sequenced pathogen genomes, we performed genome-wide association studies (GWAS) to identify the genes linked to the breakdown of host resistance. These genes encoded small secreted proteins that were highly expressed during plant infection.

Then, we assembled reference-quality genomes to analyze the chromosomal regions surrounding effector genes. We found that these regions were undergoing rapid chromosomal sequence evolution driven by repetitive elements. We found substantial gene deletion polymorphism segregating in pathogen populations and, hence, functional differences among pathogen strains. Genes located in highly dynamic chromosomal regions provide pathogen populations with evolutionary potential to rapidly adapt to environmental changes or new hosts.

> SPSW researcher portfolio:

<https://swissplantsciencweb.ch/nc/research/home/portfolio/croll>

EGES-2: A system for the accurate measurement of photosynthesis and respiration of herbaceous plants with digital control of environmental parameters

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² IFEVA, Facultad de Agronomía, Universidad de Buenos Aires and CONICET, Buenos Aires, Argentina

Phototropism is an asymmetric growth response enabling plants to optimally position their organs. In flowering plants, the phototropin (phot) blue light receptors are essential to detect light gradients. In etiolated seedlings the phototropic response is enhanced by the red/far-red (R/FR) sensing phytochromes (phy) with a predominant function of phyA. In this study, we analyzed the influence of the phytochromes on phototropism in green (de-etiolated) *Arabidopsis* seedlings. Our experiments in the laboratory and outdoors revealed that in open environments (high R/FR) phyB inhibits phototropism. In contrast, under foliar shade where access to direct sunlight becomes important the phototropic response was strong. phyB modulates phototropism depending on the R/FR ratio by controlling the activity of three bHLH transcription factors of the PHYTOCHROME INTERACTING FACTORS (PIFs) family. Promotion of phototropism depends on PIF-mediated induction of several members of YUCCA family leading to auxin production in the cotyledons.

Our study identifies PIFs and YUCCAs as novel molecular players promoting phototropism in photoautotrophic but not etiolated seedlings. Moreover, we reveal fundamental differences in the phytochrome-phototropism crosstalk in etiolated versus green seedlings. We propose that in natural conditions where the light environment is not homogeneous the uncovered phytochrome-phototropin co-action is important for plants to optimize their growth strategy and hence photosynthetic light capture.

> SPSW researcher portfolio:
<https://swissplantscienceweb.ch/nc/research/home/portfolio/fankhauser>

ABCG36/PDR8 regulation by the LRR receptor-like kinase, ALK1

Markus Geisler

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The PDR-type ABCB transporter, ABCG36/PDR8/PEN3, functions in the export of multiple substrates, including the active auxin, IBA, and indole glucosinolates. An outward facing, lateral root location and altered pathogen resistance of the *abcg36* mutant suggest that ABCG36 might secrete phytochemicals into the rhizosphere critical for plant-microbe interaction. How this lateral subdomain is achieved and ABCG36 activity is controlled is at present unclear.

Several proteomics studies indicate that ABCG36 is phosphorylated upon elicitor treatments. Employing a co-immunoprecipitation approach we identified several receptor-like kinases as ABCG36 interactor. The LRR kinase, ALK1, was found to negatively regulate IBA export of ABCG36 upon heterologous co-expression in yeast and tobacco. In line with this role for ALK1, we found altered polar transport of IBA and IBA levels in *alk1* and *abcg36* alleles. Interestingly, root ABCG36 expression and location is drastically distorted upon *F. oxysporum* infection, which can be mimicked by IBA treatments, indicating that ABCG36 expression might be controlled by auxin. Our findings identify ALK1 as a regulator of ABCG36 transport activity by means of protein phosphorylation and contribute to our understanding of the role of auxins in plant-pathogen interaction.

> SPSW researcher portfolio:
<https://swissplantscienceweb.ch/nc/research/home/portfolio/geisler>

EGES-2: A system for the accurate measurement of photosynthesis and respiration of herbaceous plants with digital control of environmental parameters

Gavin M. George and Samuel C. Zeeman

Institute of Agricultural Sciences, ETH Zurich, Switzerland

Performing accurate measurement of photosynthetic and respiration rates are vital to a large proportion of plant-based studies. While several commercial systems exist to perform such measurements we found none of them ideal for small herbaceous plants such as *Arabidopsis*. We, therefore, designed, a multi-chamber, computer controlled, infrared-gas analyzer coupled system for the continuous measurement of whole plant rosettes. This system was called ETH Gas Exchange System-1 (EGES-1). We have since upgraded this system to accommodate a wider variety of species while providing precise control over environmental parameters. Critically, we have i) increased the flow rates through each of the eight chambers, ii) introduced a computer-controlled feedback loop for the precise introduction of CO₂, and iii) added an additional feedback loop for the introduction of humidity. The advantages of this new system (EGES-2) is illustrated in the context of a variety of physiological experiments.

> SPSW researcher portfolio:

<https://swissplantsciencweb.ch/nc/research/home/portfolio/zeeman>

Biochemical and molecular analysis of dephytylation during chlorophyll breakdown

Luzia Guyer, Damian Menghini, Mareike Hauenstein and Stefan Hörtensteiner

University of Zurich, Switzerland

Chlorophyll degradation is a key feature of different developmental processes of plants, such as fruit ripening and leaf senescence. One of the key features of breakdown is the removal of the hydrophobic phytol chain from chlorophyll. On the one hand, this is a prerequisite to enable vacuolar storage of the final degradation products, on the other hand, chlorophyll-derived phytol has been considered to be important for tocopherol biosynthesis. Two enzymes are known to catalyze the dephytylation reaction: chlorophyllase (CLH) and pheophytinase (PPH). While CLH hydrolyses phytol- or methylesters from many different porphyrins, PPH is specific for pheophytin, i.e. Mg-free chlorophyll. Using a recombinant system, we explored the biochemical properties of the *Arabidopsis thaliana* PPH in detail.

Furthermore, we analyzed the role of both CLH and PPH in plants. Despite that CLH had been considered for over a century to hydrolyze chlorophyll in planta our analyses demonstrates that (1) chloroplast-located PPH, but not extraplastidial CLH is required for chlorophyll breakdown during leaf senescence in *A. thaliana* and tomato; (2) mis-expression of CLH inside the chloroplast causes strong phototoxicity, indicating that chlorophyll together with CLH could be a two-component defense system of plants against herbivores; (3) surprisingly, neither CLH or PPH play a major role in chlorophyll dephytylation or tocopherol biosynthesis during fruit ripening and seed maturation. This calls for the involvement of additional, so far unidentified phytol hydrolases in chlorophyll degradation and tocopherol biosynthesis in seeds and fruits.

> SPSW researcher portfolio:

<https://swissplantsciencweb.ch/nc/research/home/portfolio/hoertensteiner>

Perception of UV-B by the photoreceptor UVR8 induces protection of the photosynthetic apparatus against excess light in *Chlamydomonas*

Michel Goldschmidt-Clermont

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> SPSW researcher portfolio:

<https://swissplantscienceweb.ch/nc/research/home/portfolio/goldschmidt-clermont>

Identifying the genes important in adaptive variation requires an integrative approach

Matt Horton

Department of Plant and Microbial Biology
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One of the main challenges in ecology and evolution is to understand the genetic basis of adaptation. The standard approaches for identifying adaptive loci include genome-wide tests of selection (selection scans), the genetic analysis of ecologically relevant traits, and evaluating the association between genetic data and environmental variables. Here, we show how these approaches can be combined to pinpoint promising candidate selection genes. We note, however, that despite the strength of these approaches – many of the genes that are important in ecology and evolution will not be identified by these approaches.

Indeed, continuing improvements in sequencing technology have revealed that higher organisms are colonized by diverse microbial communities that contribute to host fitness. The number of genes within the microbiome are on the order (or outnumber) those of the host, which leads naturally to the question if the microbiome is responsible for metabolic functions not encoded by the host genome. We discuss recent attempts to identify the microbial species and genes that can improve host fitness.

> SPSW researcher portfolio:

<https://swissplantscienceweb.ch/nc/research/home/portfolio/horton>

Inositol pyrophosphate signaling molecules control plant phosphate homeostasis by binding to SPX sensor domains

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Phosphorus is a macronutrient taken up by cells as inorganic phosphate (Pi). How cells sense cellular Pi levels is poorly characterized. I will report that that SPX domains, which are found in eukaryotic phosphate transporters, signaling proteins, and inorganic polyphosphate polymerases, provide a basic binding surface for inositol polyphosphate signaling molecules (InsPs), the concentrations of which change in response to Pi availability. Substitutions of critical binding surface residues impair InsP binding in vitro, inorganic polyphosphate synthesis in yeast, and Pi transport in *Arabidopsis*. In plants, InsPs trigger the association of SPX proteins with transcription factors to regulate Pi starvation responses. Our findings suggest that InsPs communicate cytosolic Pi levels to SPX domains and enable them to interact with a multitude of proteins to regulate Pi uptake, transport, and storage.

> SPSW researcher portfolio:

<https://swissplantsciencweb.ch/nc/research/home/portfolio/hothorn>

Pathogen-derived proteins recognized by wheat NLR immune receptors

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Major resistance (R) genes in wheat are of great importance in many wheat breeding programmes. However, R-gene based resistance can be short-lived under agricultural conditions due to pathogen adaptation. Therefore, there is a need to develop more sustainable uses of R genes in plant breeding. In our research we explore possible ways how to make R-gene based resistance more durable. Our experimental work is mostly done in the wheat-powdery mildew pathosystem as a model for the interaction of wheat with an obligate biotrophic pathogen. R genes frequently encode nucleotide-binding, leucine-rich repeat immune receptors (NLRs). We are functionally analyzing their interactions with the cognate avirulence factors in order to determine the molecular basis of specificity.

Transient expression experiments are performed in the heterologous plant species *N. benthamiana* to functionally study the molecular determinants of specificity of interactions in host as well as pathogen genes. Based on such experiments we have identified improved NLRs that recognize additional haplotypes of fungal Avr genes in transient expression assays. On the pathogen side, we are characterizing avirulence gene products in the pathogen that are recognized by NLR proteins. The goal of this work is to (i) improve our molecular understanding of the molecular interactions of host and pathogen molecules and (ii) in the development of pathogen-informed strategies to achieve broader recognition spectra of NLR proteins. Ultimately, resistance breeding based on the molecular understanding of the interactions should result in a more sustainable use of the precious R gene resources.

> SPSW researcher portfolio:

<https://swissplantsciencweb.ch/nc/research/home/portfolio/keller>

VTE5 phytol kinase is essential for resistance to combined light and temperature stress in tomato

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In global warming scenarios, plants need to cope with the combined threat of increasing temperatures and high light (HL) intensity. Here, we use a lipidomics approach in the tomato model system to identify lipophile antioxidants that may contribute to protection against this combined stress. Among several hundred compounds, the two most strongly upregulated compounds were α -tocopherol and plastoquinone/-ol. Both have known qualities as lipid antioxidants and contribute to protection of photosystem II from photodamage under environmental stress.

To perturb α -tocopherol levels we used a tomato *vte5* knock down line. VTE5 encodes phytol kinase that acts in the biosynthetic pathway of tocopherols and the corresponding mutant has reduced levels of α -tocopherol. When exposed to combined high temperature and high light stress *vte5* suffered strong photoinhibition and -bleaching while either stress alone did not produce a visible phenotype. As *vte5* had wild type levels of plastoquinone under combined stress, the phenotype was attributed to the lack of α -tocopherol. In summary, the data indicate that tocopherol is essential for protection against combined high light and high temperature stress.

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Involvement of hydraulic signals in leaf-to-leaf wound signalling in *Arabidopsis*

Andrzej Kurenda and Edward E. Farmer

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Plant specific, wound-induced electrical signals called variation potentials or slow wave potentials (VPs or SWPs) are key phenomena enabling leaf-to-leaf wound signalling to activate defense gene expression. The putative mechanism involves the induction of a hydraulic pressure wave in the wounded organ, axial transmission of the wave throughout the plant through xylem vessels, conversion of axial pressure to radial mechanical signals in distal organs, and finally mechanostimulus-induced plasma membrane depolarisation in xylem associated cells.

Despite the seemingly simplicity of the proposed mechanism it has not yet been experimentally confirmed mainly because of lack of methods of direct measurements of hydraulic pressure in xylem and phloem. Since wound-induced pressure waves propagating through the vasculature cause deformation of the whole leaf detectable as an increase of leaf thickness, it is possible to indirectly evaluate pressure wave dynamics using non-invasive turgor pressure or force sensors. Our measurements with external turgor pressure sensors and micro force sensing probes confirmed the existence of wound-induced hydraulic signal correlated highly with duration of electrical signals and activation of *JAZ10*, a marker of jasmonate pathway activity.

> SPSW researcher portfolio:

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My journey with malate – ups and downs with a good friend

Enrico Martinoia

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One of my main interest during my scientific career has been the characterization of vacuolar malate transport and the identification of corresponding transporters. Malate plays a multitude of different roles in plants and is an intermediate in many metabolic pathways. It is also an important osmolyte, a temporary store for CO₂ and reduction equivalents in C₄ and CAM plants, and is involved in aluminum detoxification. Nevertheless, the cytosolic malate concentration has to be kept constant. The vacuole plays a central role in this regulation, as it buffers an excess of malate or releases malate if plant metabolism requires it.

I will talk about what lead us to investigate vacuolar malate transport and about our latest results on vacuolar malate transporters and channels.

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Finally: A biochemical function for the PATHOGENESIS–RELATED PROTEIN 1

Jordi Gamir and Felix Mauch

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The PATHOGENESIS–RELATED PROTEIN 1 (PR-1) was discovered more than fifty years ago as the most prominent of several pathogenesis-related proteins that accumulated in the apoplastic space of virus-challenged tobacco plants. In support of an immune related function, PR-1 proteins from different plant species were later shown to possess *in vitro* antimicrobial activity. However, in contrast to other groups of PR-proteins, the biochemical activity and mode of action of PR-1 proteins has remained elusive. We have made a new effort to solve this longstanding question. We show here a) that PR-proteins of tobacco and tomato can rescue the sterol export defect of a yeast mutant suggesting that PR-1 binds sterols in the secretory pathway to mediate their export, b) that PR-1 proteins have an *in vitro* sterol binding activity and c) that the sterol binding capacity is the cause of the antimicrobial activity as addition of sterols titrates the inhibitory activity.

In summary, we provide genetic and biochemical evidence for the capacity of PR-1 proteins to bind sterols and demonstrate that the sterol-binding activity is the mode of action of the antimicrobial activity. In line we the proposed function, PR-1 proteins are especially efficient inhibitors of sterol-auxotroph oomycete pathogens.

> SPSW researcher portfolio:

<https://swissplantscienceweb.ch/nc/research/home/portfolio/mauch>

Cytokinin distribution and transport to support plant morphogenesis

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Cytokinins direct essential cell-to-cell signalling systems to initiate and maintain differential cell functions during gametophyte development, embryogenesis and postembryonic shoot and root development. Guided by the live green fluorescent protein (GFP) imaging of the synthetic reporter, *TCSn::GFP* (Two Component signalling Sensor), we illuminate precise cell populations activating conserved phosphorelay signalling circuitry despite much broader intracellular cytokinin signalling competence and potential. I will report that *Arabidopsis* PURINE PERMEASE 14 (PUP14) plays a pivotal role in constraining the cytokinin signalling response throughout development by ligand sequestration. The expression patterns of *PUP14* are inversely correlated with the cytokinin signalling readout, indicating an antagonistic relation. Indeed, the conditional *PUP14* knock-down by an artificial microRNA (*amiRPUP14*) causes ectopic cytokinin signalling accompanied by aberrant morphogenesis in embryos, roots and the shoot apical meristem. PUP14 protein localises to the plasma membrane and imports

bioactive cytokinins as shown by analysing transport in mesophyll protoplasts, seedlings and microsomes. These activities of PUP14 reduce the apoplastic cytokinin pools and extracellular cytokinin perception by cytokinin sensors. The relevance of apoplastic cytokinins in initiating intracellular signalling is corroborated by our findings that signalling output is attenuated by secreted cytokinin-degrading enzymes, but not by intracellular variants. These results uncover the existence of a previously unknown and dedicated transport system that patterns the cytokinin signalling landscape of vascular plants.

> SPSW researcher portfolio:
<https://swissplantsciencweb.ch/nc/research/home/portfolio/mueller>

Intrinsic cell wall properties uncouple growth from growth hormones

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Complex shapes are produced from groups of cells by the process of morphogenesis. While, in animal tissues cellular rearrangement can play a role, in plants this is not the case. We can, therefore, investigate the development of complex form, by focusing our study on the regulation of cell expansion. For growth to occur, the cell wall must yield to the stress induced by turgor pressure, and irreversibly expand (Lockhart, 1965). Much of our knowledge of the cell wall and its mechanical properties comes from the vast body of work using extensometers, however, they typically provide low resolution information. By developing an automated confocal micro-extensometer (ACME) we were able to measure the mechanical properties of live *Arabidopsis* hypocotyls with cellular resolution, and in the direction of growth.

In doing so, we demonstrated that seedlings treated with the growth hormone GA had altered cell wall properties that correlated with increased growth. By eliminating complex geometry or mechanical feedback, we showed that the spatial restriction of growth in response to GA, is an intrinsic property of the cells and is independent of microtubule orientation. We were thus able to address the key question of how organ growth is regulated in a physiologically relevant system and provide insight into the complex process of morphogenesis.

> SPSW researcher portfolio:
<https://swissplantsciencweb.ch/nc/research/home/portfolio/kuhlemeier>

Perturbing phosphoinositide homeostasis during vascular cell differentiation: a tale of two halves?

Bojan Gujas, Elizabeth Kastanaki, Tiago Dias-Cruz, and **Antía Rodríguez-Villalón**

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In multicellular organisms, differentiation programs involve drastic changes in cell morphology and signal responsiveness to generate new structures and to acquire specialized functional features. An extreme case of this process is the differentiation of *Arabidopsis thaliana* (*Arabidopsis*) vascular cells into conductive units. To become transport elements, phloem and xylem cells undergo two different morphogenetic developmental programs which involve the reinforcement of their cell wall and partial or total cell clearance. Contrary to phloem cells, where vacuoles reduce their size and eventually disintegrate, xylem elements differentiate upon formation and subsequent rupture of a central vacuole. Yet, the molecular mechanisms regulating both processes remain poorly understood. We have previously reported that an increase in phosphatidylinositol 4,5-bis-phosphate (PIP2) levels negatively affects the correct progression of protophloem differentiation in *Arabidopsis* roots.

On the contrary, a genetic increase of PIP2 abundance causes premature protoxylem differentiation due to accelerated vacuolar fusion events. Genetic and pharmacological disturbance of PIP2 levels rewires intercellular trafficking towards the vacuole. Latter has been associated with the activity of phosphatidylinositol species phosphorylated at the 3' position, suggesting a novel regulatory pathway involved in cell differentiation. Altogether, our results demonstrated that a tight plasma membrane PIP2 homeostasis is strictly required to fine tune cell trafficking and vacuolar biogenesis which in turn, regulate vascular formation.

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<https://swissplantscienceweb.ch/nc/research/home/portfolio/rodriguez>

Genome wide analysis of transposable elements in *Brachypodium distachyon*: insights from natural populations

Anne Roulin

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Transposable elements (TEs) are mobile DNA sequences that have the capacity to replicate and move from one location to another in the genome. While TEs have long been seen as causing agents of genomic instability, it is now well established that they can produce diverse functional changes, from the disruption of coding sequences to the fine-tuning of gene expression. Therefore, they constitute a driving force of evolution. Plants are ideal systems to study TEs because they usually harbor a higher load of TEs than animals. However, with the reliance of most research on plants of economical interest, the functional impact and the potential adaptive role of TEs in natural systems have largely been overlooked. To better understand how TEs can affect natural systems, we used whole-genome sequencing data of 53 natural accessions of the grass *Brachypodium distachyon* originating from Turkey and Spain.

We investigated the recent transpositional activity and found a substantial number of TE variants (TE elements inserted at different loci) across the 53 genomes. Some of these variants are located in or nearby genes and an analysis of selection revealed that a subset of them also display signs of selection. Our results indicate that TEs may play an important role in the evolution of natural populations of *B. distachyon* and open new avenues of research focusing on their functional impact in natural systems.

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<https://swissplantscienceweb.ch/nc/research/home/portfolio/roulin>

Mechanistic insights into floral abscission and cell wall remodeling

Sebastian Augustin, Andra-Octavia Roman,
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Plants constantly renew during their life cycle and thus require to shed senescent and damaged organs. Floral abscission is controlled by the leucine-rich repeat receptor kinase (LRR-RK) HAESA, the peptide hormone IDA and the SERK co-receptors. The crystal structure of the ternary complex reveal the key residues involved in IDA sensing and receptor activation. I will present our in vivo validation of this signaling complex and the development of efficient IDA antagonist. In the future, we would like to connect IDA sensing to cell wall signaling and remodelling.

> SPSW researcher portfolio:

<https://swissplantscienceweb.ch/nc/research/home/portfolio/santiago>

Deciphering functions of the plant root microbiome

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Diverse assemblages of microbes colonize plant roots and collectively function as a microbiome. Earlier work has described the composition of root microbiomes of numerous plant species, while how the root microbiome contributes to plant growth and health is less well understood. We have isolated ~200 bacteria isolates and they corresponded to ~25% of the abundant root microbiome members. Further, we developed an axenic microcosm system to conduct simplified microbiota inoculation experiments with plants. We compare effects on plant growth that were induced by individual bacteria or combinations of microbiome members.

First microbiota inoculation experiments indicated that a more diverse root community could alleviate the plant growth compromising activities of its individual members. A reductionist experimental approach that offers countless opportunities for future systematic and functional examinations of the plant root microbiome will be presented.

> SPSW researcher portfolio:

<https://swissplantscienceweb.ch/nc/research/home/portfolio/vanderheijden>

Genomic imprinting in the endosperm is systematically perturbed in abortive hybrid tomato seeds

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Hybrid seed failure represents an important postzygotic barrier to interbreeding among species of wild tomatoes (*Solanum* section *Lycopersicon*) and other flowering plants. We studied genome-wide changes associated with hybrid seed abortion in the closely related *Solanum peruvianum* and *S. chilense*, where hybrid crosses yield high proportions of inviable seeds due to endosperm failure and arrested embryo development. Based on differences of seed size in reciprocal hybrid crosses and developmental evidence implicating endosperm failure, we hypothesized that perturbed genomic imprinting is involved in this strong postzygotic barrier. Consequently, we surveyed the transcriptomes of developing endosperms from intra- and interspecific crosses using tissues isolated by laser-assisted microdissection.

We implemented a novel approach to estimate parent-of-origin-specific expression using both homozygous and heterozygous nucleotide differences between parental individuals and identified candidate imprinted genes. Importantly, we uncovered systematic shifts of 'normal' (intraspecific) maternal:paternal transcript proportions in hybrid endosperms; the average maternal proportion of gene expression increased in both crossing directions but was stronger with *S. peruvianum* in the maternal role. These genome-wide shifts almost entirely eliminated paternally expressed imprinted genes in *S. peruvianum* hybrid endosperm but also affected maternally expressed imprinted genes and all other assessed genes. These profound, systematic changes in parental expression proportions suggest that core processes of transcriptional regulation are functionally compromised in hybrid endosperm and contribute to hybrid seed failure.

> SPSW researcher portfolio:
<https://swissplantscienceweb.ch/nc/research/home/portfolio/staedler>

Increasing the carbohydrate storage capacity of plants by engineering a glycogen-like polymer pool in the cytosol

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Institute for Agricultural Sciences, Plant Biochemistry, ETH Zurich, Switzerland

Global demand for higher crop yields and more efficient utilization of agricultural products will grow over the next decades. Here, we present a new concept for boosting carbohydrate content of plants, by channeling photosynthetically fixed carbon into a newly engineered glucose polymer pool. We transiently expressed starch/glycogen synthases from either *Saccharomyces cerevisiae* or *Cyanidioschyzon merolae*, together with starch branching enzyme from *C. merolae*, in the cytosol of *Nicotiana benthamiana* leaves. This effectively built a UDP-glucose-dependent glycogen biosynthesis pathway. Glycogen synthesis was observed using Transmission Electron Microscopy and analysis of the glucose polymer structure. Within three days of enzyme expression, glycogen content was 5 to 10 times higher than control starch levels.

Further, production of starch and sucrose, the carbohydrate end products of photosynthesis, were reduced in abundance. We conclude that most of the new fixed carbohydrates were routed and trapped into the new glycogen sink. Our approach allows carbohydrates to be efficiently stored in a new subcellular compartment, thus increasing the value of vegetative crop tissues for biofuel production or animal feed. The method also opens new potential for increasing the sink strength of heterotrophic tissues.

> SPSW researcher portfolio:
<https://swissplantscienceweb.ch/nc/research/home/portfolio/streb>

Stress-induced changes of leaf carbohydrate metabolism are modulated by the circadian clock and ABA

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Starch is a versatile polymer of glucose that serves widely different functions depending on the cell type it is located or the environmental conditions the plant is exposed to. We have previously shown that starch in the leaves is rapidly remobilised during osmotic stress. Stress-induced starch degradation depends on two key enzymes, α -amylase 3 (AMY3) and β -amylase 1 (BAM1), which are induced in an ABA-dependent manner. The concerted action of AMY3 and BAM1 releases sugars which serve as osmoprotectants. Mutants lacking those two enzymes fail to remobilise starch and accumulate less osmolytes, affecting water uptake and root growth under osmotic stress.

The induction of AMY3 and BAM1 is mediated by the AREB/ABF-SnrK2 kinase pathway. This mechanism appears to be conserved amongst dicotyledon species, as ABA responsive elements are found in the promoters of BAM1 orthologs.

Interestingly, the induction of starch degradation in response to osmotic stress appears to be dependent on the time of stress onset. Starch metabolism is most affected during midday while it is almost insensitive to stress imposed during late afternoon. Here, we show that this difference is mediated by the circadian clock. Spell (TOC1), a key component of the circadian evening loop, appears to negatively regulate stress-induced starch degradation in the evening.

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<https://swissplantsciencweb.ch/nc/research/home/portfolio/santelia>

OCTOPUS-LIKE 2, a novel player in *Arabidopsis* root and vascular development, reveals a role for OCTOPUS family genes in root metaphloem sieve tube differentiation

M. Aguila Ruiz Sola, Mario Coiro, Simona Crivelli, Samuel C. Zeeman, Signe Schmidt Kjølnner Hansen, **Elisabeth Truernit**

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Phloem is essential for the long distance transport of carbohydrates and signalling molecules in plants. Protophloem is the first phloem cell type that differentiates in developing organs, while metaphloem is considered to be the part of the phloem that is functional for longer time periods. OCTOPUS (OPS) was previously identified as a master regulator of protophloem differentiation in *Arabidopsis*. Here, we investigated the function of *OCTOPUS-LIKE 2 (OPL2)*, a gene homologous to OPS. OPS and OPL2 display overlapping expression patterns in the developing vasculature, and several lines of evidence suggest a high degree of functional overlap. A mutation in OPL2 reveals redundant functions of OPS and OPL2 in developmental processes in which OPS was known to play a role, notably cotyledon vascular patterning and protophloem development.

Most interestingly, we also discovered a role for OPS and OPL2 in metaphloem sieve tube differentiation, which had not been described thus far. Taken together, our results provide further evidence for a key role of OPS(-like) genes in vascular patterning, root growth and phloem development.

> SPSW researcher portfolio:
<https://swissplantsciencweb.ch/nc/research/home/portfolio/truernit>

Cell volume regulation during lateral root formation in *Arabidopsis*

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Intercellular communication is central for the development of an organism. It is mediated by biochemical gradients as well as physical forces that collectively regulate differentiation and development. Lateral roots are initiated deep within the primary root from dividing pericycle cells, necessitating growth through overlying endodermal, cortical and epidermal cell layers. During its development, the lateral root heavily depends on spatial accommodating responses of these overlaying cell layers. We have shown that pericycle cells need to swell in order to undergo formative divisions, whereas the overlying endodermis undergoes a dramatic volume loss during lateral root formation. Through manipulating SHY2-mediated auxin signaling in the endodermis, we were able to completely block cell proliferation in the pericycle. It appears that the pericycle perceives the non-accommodating endodermis as an increased resistance to its expansion growth. The pericycle-endodermis interaction provides a unique opportunity to elucidate the molecular and cellular mechanisms underlying spatial

accommodation in plant development. We are using a multidisciplinary approach to elucidate their role during lateral root formation to start elucidating the mechanistic framework underlying spatial accommodation. Recently, we have used a transcriptomics approach to identify SHY2-dependent targets that mediate spatial accommodating responses. This has revealed that several transporters, ion channels, regulators of the cytoskeleton, cell wall and membrane trafficking were differently expressed. I will present our latest results in our quest towards understanding spatial accommodating responses.

> SPSW researcher portfolio:

<https://swissplantsciencweb.ch/nc/research/home/portfolio/vermeer>

Explaining the reproductive allocation strategy of *Arabidopsis*

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Since maternal resources are limited, it is generally assumed that trade offs exist between reproduction through seeds and other functions (e.g. vegetative growth or defense). Consequentially, plants will have to choose between alternative investment strategies. The mechanisms that govern such choices are still poorly understood.

I study the molecular and genetic determinants of allocation strategies in plants, especially how variation in monocarpic senescence leads to differences in reproductive investment and yield. In particular, I am interested in feedback controls of seeds over maternal senescence and growth. Focusing on *Arabidopsis* as a model, I will report observations that support the “seed dominance hypothesis”, i.e. that apically dominant flowering shoots and developing seeds have equivalent molecular and physiological effects on vegetative tissues. Furthermore, I will discuss results from a mutant screen for lines that exhibit seed number variation. Genetic mapping of the causal mutations suggests that variation in different genes and metabolic pathways can lead to similar increases in reproductive investment.

> SPSW researcher portfolio:

<https://swissplantsciencweb.ch/nc/research/home/portfolio/wuest>

Protein targeting to starch (PTST): A new class of proteins that bring enzymes and substrates together

Samuel C. Zeeman

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Starch is a vital plant product. It is the major nutritive component of our staple crops and an important feedstock for industry. Starch takes the form of insoluble, semi-crystalline granules composed of two glucose polymers: branched amylopectin and linear amylose. Amylopectin is the major component and responsible for the semi-crystalline nature of starch. It is made by a set of enzymes (starch synthases, branching enzymes and debranching enzymes). Amylose is made within the amylopectin matrix by a single enzyme – Granule-Bound Starch Synthase (GBSS) – that becomes trapped as amylopectin crystallises about it. There is much that we still do not understand starch biosynthesis, such as how the enzymes activities are coordinated and how starch granules are first initiated.

We recently discovered that a new class of proteins is required to localize some of the Starch Synthase activities. We called these proteins PTST (for Protein Targeting to Starch). *Arabidopsis* has three PTSTs. PTST1 binds GBSS and delivers it to the starch granule surface, whereupon it dissociates and leaves GBSS to synthesise amylose. In the absence of PTST1, GBSS fails to localize to the granule and the starch is composed solely of amylopectin. In contrast, PTST2 and PTST3 bind to another starch synthase, SS4, which is implicated in starch granule initiation. Defects in SS4 or in PTST2 or PTST3 significantly alter the number, size and shape of starch granules without necessarily affecting the structure or ratio of the constituent polymers. We believe that these PTSTs bind low-abundance precursors from a pool of malto-oligosaccharides and deliver them to SS4 for elaboration into starch granule initials. We hope that these discoveries may, in the long run, facilitate the improvement of our starch crops.

> SPSW researcher portfolio:
<https://swissplantscienceweb.ch/nc/research/home/portfolio/zeeman>

Novel defenses as an escape from co-adapted herbivores: cardenolides in the Brassicaceae

Tobias Züst

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Plants express a diverse array of defensive chemicals to fight off enemies. Complex chemical mixtures may interact with more target sites in herbivores, or may harbor more potential for facilitation among compounds to increase toxicity. Despite this potential benefit of chemical diversity, most plants are highly phylogenetically constrained in their chemotype. For example, plants of the Brassicaceae produce diverse mixtures of defensive compounds, but the vast majority belongs to a single compound class, the glucosinolates. Such phylogenetic conservatism in chemical defense may facilitate specialization of herbivores, as a gain of tolerance to one compound will increase the likelihood of tolerance to related compounds as well.

As a strategy to lose co-adapted herbivores, plants may occasionally gain the ability to produce completely novel defense classes in addition to the phylogenetically basal defense. Plants of the genus *Erysimum* (Brassicaceae) gained the ability to synthesize toxic cardenolides in addition to glucosinolates, making them resistant to several specialist herbivores of the Brassicaceae. Here I show how the genus underwent chemical diversification in both the basal and novel defense in a recent radiation, providing no evidence for redundancy of either defense. In addition, I found environmental regulation of the two defense types to operate largely independently, allowing for specific responses to diverse challenges by the plant. Diversification within and among compound classes therefore appears to serve different purposes in a plant's evolutionary defense response. In conclusion, events of gain-of-function and co-evolutionary escalation provide unique opportunities for the study of the selective forces that drive phytochemical diversification.

> SPSW researcher portfolio:
<https://swissplantscienceweb.ch/nc/research/home/portfolio/zuest>

Identifying regulators of di- and tripeptide utilization in *Arabidopsis*

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Plant protein annotation in UniProtKB/Swiss-Prot and sustainability of high quality expert curation

Damien Lieberherr¹, Emmanuel Boutet¹, Michel Schneider¹, Michael Tognolli¹, Sylvain Poux¹, Michele Magrane³, Cecilia Arighi⁴, Zhyong Lu⁵, Ioannis Xenarios^{1,2} and the Swiss-Prot group

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Organization and evolution of the b-amylase multigene family in land plants (embryophytes)

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Cytomechanical measurements reveal differential effects of cell wall components on pollen tube growth and integrity

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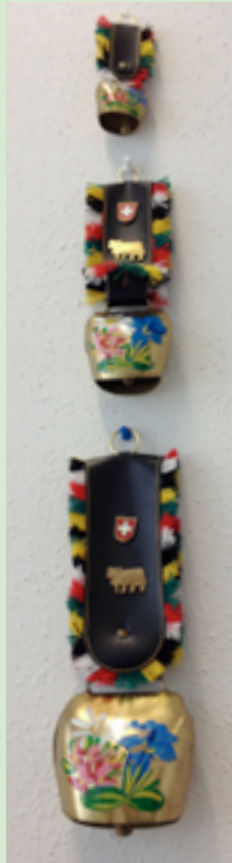
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