





# From place to space -

# Tracing the spatial dimension of plant sciences

7<sup>th</sup> December 2022 ETH Zurich





Universität Zürich ETH Zürich Universität Basel Plant Science Center









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### ORGANIZATION

### **PhD Organising Committee**

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### Venue

ETH Zurich, Auditorium Maximum (HG F30) Rämistrasse 101, 8006 Zurich

### Symposium Website

ightarrow www.pscsymposium2022.ethz.ch

Admission is free of charge.

### PROGRAM

	BRUNO STUDER
09:15	ETH Zurich, CH
	Welcome and opening by PSC chair
	Lucia Piro & Trang Dang
	both Institute of Integrative Biology , ETH Zurich
	Session chairs
	363300 Chan 3
09:30	Prof. Dr. SARA SIMONINI
	Molecular Embryology, Department of Plant and Microbial Biology, University of Zurich, CH
	When it is the right time to divide: parental regulation of cell division during reproduction
10:00	Dr. MARKUS GEISLER
10100	Regulation of Auxin Transport, Plant and Microbial Sciences, University of Fribourg, CH
	A twist in the thale: an update on auxin-mediated cell elongation
10:30	Dr. STEFANIA GIACOMELLO
	Spatial Research, KTH Royal Institute of Technology, SE
	A journey in spatially resolved transcriptomics: from animal tissues to plants

### 11:00 BREAK AND POSTER SESSION

### Katja Stengele & Charlotte Joller

both Department of Environmental Sciences, University of Basel

Session chairs

11:30	Dr. CHRYSOULA PANTAZOPOULOU Plant-Environmental Signaling, Environmental Biology, University of Utrecht, NL	
	How can Arabidopsis perceive neighbors in space and time?	
12:00	Prof. Dr. PHILIPPE REYMOND Molecular Studies of Plant-Insect Interactions, Department of Plant Molecular Biology, University of Lausanne, CH	
	Intra- and interplant responses to insect egg deposition in Arabidopsis	
12:30	Dr. DESALEGN ETALO Laboratory of Phytopathology, Department of Plant Sciences, Wageningen University and Research, NL	

The tri-partite interaction between parasitic plants, host, and their microbiome

13:00 LUNCH AND POSTER SESSION

Alexandra Siffert & Iciar Giménez Department of Plant and Microbial Biology, University of Zurich & Institute of Agricultural Sciences, ETH Zurich

Session chairs

 
 14:30
 Prof. Dr. SABINE RUMPF Ecology, Department of Environmental Sciences, University of Basel, CH

 Effects of environmental change on arctic and alpine vegetation

 15:00
 Prof. Dr. ELIANA M JIMÉNEZ Tropical Rainforest Ecology, Universidad Nacional de Colombia-Sede Amazonia, CO Spatial and temporal variation of forest net primary productivity components

on contrasting soils in northwestern Amazon

15:30 BREAK AND POSTER SESSION

Alexandra Siffert & Iciar Giménez

Department of Plant and Microbial Biology, University of Zurich & Institute of Agricultural Sciences, ETH Zurich

Session chairs

#### **FLASH TALKS**

STEPHANIE RUAUD 16:00 Institute of Agricultural Sciences, University of Zurich Pyrenoid formation in hornworts: genomic hints for green algal-like pyrenoid scaffolding mechanisms 16:10 YULING YUE Department of Systematic and Evolutionary Botany, University of Zurich Shared transcriptional response in independent lineages of land plants during plant- cyanobacteria symbiosis shed light on its evolutionary origin 16:20 APHRODITE KANTSA Department of Environmental Systems Science, ETH Zurich Discriminability of floral colors explains global biogeographical patterns of pollinators **OLIVER REUTIMANN** 16:30 Institute of Integrative Biology, ETH Zurich Developing a sampling design for a genetic diversity monitoring program in Switzerland Alexandra Siffert & Iciar Giménez Department of Plant and Microbial Biology, University of Zurich & Institute of Agricultural Sciences, ETH Zurich

Session chairs

17:00 Prof. Dr. STEFANIA DE PASCALE Horticulture, Department of Agricultural Sciences, Università degli studi di Napoli Federico II, IT Bioregenerative systems to sustain human life for Long-Term Space Missions: the challenges of plant cultivation

#### 17:30 POSTER AWARDS and concluding remarks

# Invited speakers

# When it is the right time to divide: parental regulation of cell division during reproduction

### Sara Simonini



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During the process of fertilization of sexually reproducing organisms, maternal and paternal gametes, egg and sperm respectively, fuses together to give rise to the zygote. Differently from animal, in flowering plants the so-called double fertilization involves a second female gamete, the central cell, from which originates the endosperm, a triploid and ephemeral tissue that nurtures and sustains the growth of the embryo. The fusion of the paternal and maternal gametes generates a series of dramatic events, including the re-activation of the cell cycle that is, somehow, strongly inhibited before fertilization to avoid premature division. Genetic evidences show that both parents exert a tight control over cell cycle progression: the mother reins cell division in the seeds, whereas the father provokes the opposite. The lack of this control then has dramatic and conflicting effects as the development of seed-like structure from unfertilized ovules, or suicidal cell divisions as result of unbalanced DNA content after fertilization. The molecular mechanisms underlying these processes are yet to be fully understood. I will present some of our recent data about the characterization of such mechanisms, showing how maternal factors keep female gametes quiescent, and how paternally-derived signals trigger cell cycle progression specifically at fertilization.

# A twist in the thale: an update on auxin-mediated cell elongation

### **Markus Geisler**

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Plants own an amazing high degree of developmental plasticity by regulating cell growth and division in response to internal and external signals. This plasticity is controlled by local maxima and minima of the signalling molecule, auxin. These are generated by the cell-to-cell movement of auxin, a unique process not yet described in non-plant organisms or for other hormones. This so-called polar auxin transport is thought to be mainly provided by the action of auxin exporters of the ABCB and PIN families. Interestingly, *abcb* loss-of-function mutants reveal a strong developmental phenotype, including a helical, non-handed disorientation of epidermal layers.

In my talk, I will address the morphological and molecular background for this "twisted syndrome" by dissecting the individual roles of ABCB proteins. It appears that all auxintransporting ABCBs are regulated on the transport level by transient cis-trans isomerization of a conserved and diagnostic D/E-P motif. This catalytic activity is provided by PPlases, including the FKBP42, TWISTED DWARF1. Beside acting as PPlase, TWISTED DWARF1 functions also as a co-chaperone of HSP90 stabilizing ABCBs at the plasma membrane, indicating a dual role during ABCB regulation.

Our findings classify the TWISTED DWARF1-HSP90 module as a positive regulator of polar auxin transport providing plasticity to ABCB-controlled auxin transport and plant development.



# A journey in spatially resolved transcriptomics: from animal tissues to plants





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Spatial context is fundamental in understanding how tissues are formed and how single cells function and interact together. Most of the available spatial technologies are only available for mammalian tissues limiting the exploration of plant systems. In this talk, I will present our recent developments in Spatial Transcriptomics with a specific focus on plants and microbes.

# How can Arabidopsis perceive neighbors in space and time?

### Chrysoula Pantazopoulou



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Modern agriculture is characterized by the intensification of agricultural practices and cultivation of plants in dense stands. It is crucial for plants growing at high densities to perceive and respond to upcoming shade from a neighboring plant rapidly. The light quality in the canopy is determined by the Red:Far-Red light (R:FR) ratio, with high R:FR indicating sufficient light for photosynthesis and low R:FR indicating shade caused by proximate neighbors. We found previously that leaf tip touching between individuals in a dense vegetation of Arabidopsis is the earliest neighbor detection in shoots. Following touching, the leaves respond with an upward leaf movement (called hyponasty). Due to this hyponastic response the canopy architecture changes from a horizontal to a more vertical one. This vertical alignment of leaves generates FR light reflection, leading to a low R:FR signal inside the canopy. Under these conditions, the plant can detect the low R:FR signal in different parts of the leaves and respond with a further hyponasty and/or elongation of the leaf petiole. All these canopy alterations are part of the shade avoidance strategy of plants to consolidate light capture. Interestingly, touchinduced hyponasty involves a signal transduction pathway that is distinct from light-mediated hyponasty. This indicates that a canopy develops progressively though different signaling pathways towards the final shade avoidance phenotype.

# Intra- and interplant responses to insect egg deposition in Arabidopsis

### **Philippe Reymond**



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Insect eggs deposited on plant leaves are recognized and induce defenses that inhibit egg development or attract egg predators. Oviposition by the Large White butterfly *Pieris brassicae* leads to salicylic acid accumulation and local cell death in *Arabidopsis thaliana*. These responses are activated by a phospholipid elicitor perceived at the cell surface and share molecular similarities with generic innate immunity. Surprisingly, we discovered that oviposition inhibits growth of bacterial and fungal pathogens through the establishment of an intra- and interplant systemic acquired resistance (SAR). This finding suggests that eggs manipulate plant signaling by increasing resistance to pathogens, for the potential benefit of feeding larvae.

# The tri-partite interaction between parasitic plants, host, and their microbiome

### **Desalegn Etalo**



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Plant parasitic weeds belonging to the Orobanchaceae family are the major threat for global food security. They are challenging to control because their life cycle is intimately intertwined with the host physiology. Furthermore, most of the damage on the host occurs while these parasites are at the subterranean life cycle stages. The interaction between the hosts and the parasitic weeds mainly takes place in the rhizosphere where lively microbial activity takes place. However, In the past decades, most studies on host-parasite interactions focused on genetics, biochemistry, and physiology, while the plant-associated microbiome was kept aside, neglecting its value as a source of unmeasured host genetic variation. In my talk, I will discuss the reciprocal interactions between Sorghum and the parasitic weed *Striga hermonthica*, at the microbiome level, by emphasizing on the impact the microbiome has on the fitness of both the parasite and the host.

# Effects of environmental change on arctic and alpine vegetation

### Sabine Rumpf

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Climate is currently warming at a rapid pace, causing species to shift their ranges to follow the conditions they are adapted to. In arctic and alpine ecosystems, climate is warming at an even higher pace than the global average. Species range shifts to higher latitudes and elevations are a globally observed consequence, and species richness and vegetation productivity are increasing at highest latitudes and elevations. Yet, the limited empirical evidence available so far suggests that species' warm range limits shift at least as fast as the cold limits at the global scale, resulting in contracting distributions of many species and, hence, increased extinction risks. Furthermore, both range limits seem to lag behind temperature trends, and the vast majority of publications report considerable amounts of variation between species-specific responses. These idiosyncratic responses imply asynchronous shifts and might result in reshuffled plant communities with novel biotic interactions. An improved understanding of the factors and processes determining the magnitude and velocity of species responses is pressing in a conservation context as arctic and alpine ecosystems harbour disproportionately high biodiversity, including rare and endangered species, and are in general poorly protected.

# Spatial and temporal variation of forest net primary productivity components on contrasting soils in northwestern Amazon

### Eliana M. Jiménez

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Climate is a strong determinant of tropical forest productivity; therefore, it is often assumed that Amazonian forest growing on the same local rainfall regime responds similarly to fluctuations in rainfall, independently of soil differences among them. We evaluated intra- and inter-annual variation of net primary productivity (NPP) components, and forest dynamics during 2004-2012 yr in five forests on clay, clay-loam, sandy-clay-loam, sandy-loam and loamysand soils, and the same local rainfall regime in northwestern Amazonia (Colombia). The questions were as follows: (1) Do NPP components and forest dynamics respond synchronously to temporal rainfall fluctuations? (2) Are the responses between above and belowground components and forest dynamics similar for different forest stands? A slight and complex synchronicity among different NPP components in their response to temporal rainfall fluctuations were found; few plots showed that aboveground biomass (AGB) and stem growth were susceptible to rainfall fluctuations, while belowground components (fine roots) showed correlation with one-month lagged rainfall. Furthermore, despite that northwestern Amazonia is considered relatively aseasonal, litterfall showed high seasonality in the loam-soil forest group, as well as the fine-root mass, particularly during the 2005 drought. Litterfall correlation with rainfall of sandy-loam terra-firme forest was time lagged as well as fine-root mass of the loamy-sand forest. The correlation between mortality and rainfall was weak, except for the loamy-sand forest (white-sand forest, 77%). High mortality rates occurred in the non-flooded forests for the censuses that included the dry years (2004-2005, 2005-2006). Interestingly, litterfall, AGB increment, and recruitment showed high correlation among forests, particularly within the loam-soil forest group. Nonetheless, leaf area index (LAI) measured in the most contrasting forests (clay and loamy-sand soil) was poorly correlated with rainfall, but highly correlated among them, which could be indicating a phenotypic response to the incident radiation in these sites; also, LAI did not reflect the differences in NPP components and their response to rainfall. Overall, the different temporal behavior of NPP components among forests in relation to rainfall fluctuations suggests the important role that soil exerts on the responses of plant species in each site, besides their effect on forest dynamics and community composition.

# Bioregenerative systems to sustain human life for Long-Term Space Missions: the challenges of plant cultivation

### Stefania de Pascale



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Human exploration beyond Low Earth Orbit (LEO) will require technologies regenerating resources like air and water, and producing fresh food while recycling consumables and waste. Bioregenerative Life Support Systems (BLSSs) are artificial ecosystems in which appropriately selected organisms, including bacteria, algae and plants, are assembled in consecutive steps of recycling, to reconvert the crew waste into oxygen, potable water and edible biomass. Plants are considered the most promising biological regenerators to accomplish these functions, thanks to their complementary relationship with humans, however, cultivation in Space requires the knowledge of their response to Space factors (e.g. altered gravity and ionizing radiation) and specific cultivation conditions (e.g. controlled environment, hydroponic systems). The presentation will summarize the research activity carried out at the Department of Agricultural Sciences of the University of Naples Federico II on plant-based BLSSs.

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# Poster abstracts

in alphabetic order

# Fast chlorophyll fluorescence as a tool to evaluate plant vitality in natural ecosystems

Reinhard Bachofen

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Over the past decades fast chlorophyll fluorescence, the polyphasic kinetics observed during the first second after illumination, has developed to a technique to evaluate plant vitality, mainly in experiments under controlled laboratory conditions. Lack or excess of water, light intensity, missing nutrients, presence of pesticides or pest organisms and more, all effect plant vitality and influence fluorescence kinetics.

We show with 4 examples obtained in field experiments significant changes in the kinetics of fast chlorophyll fluorescence:

Euphorbia infected by Uromyces, grass and shrubs in a natural nitrogen gradient, Sphagnum peatmoss at different water level and the lichen Xanthoria in dry-wet cycle.

Signal changes correlate with changes in specific environmental factors, allowing estimating the broad spectrum of plant vitality within small-scale populations in the field.

Additional poster authors with groups of Albanian students from the universities of Tirana and Pristina

## P2 Spatial distribution patterns of transcription in Arabidopsis nuclei

### Célia Baroux

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Transcription can be rapidly reprogrammed upon environmental cues, sometimes within minutes. This conveys novel cellular properties which enable physiological adaption of the organism. Whether such reprogramming, detected by transcriptome profiling, also involves changes in the nuclear distribution of transcription is not known. Probing for the spatial organisation of transcription sites in the nucleus is challenging but become accessible thanks to super-resolution imaging and image processing solutions (Dumur et al, 2019). Here, we employed 3D STED microscopy to localize active RNA Pol II. We developped a customized, image analysis workflow to resolve the spatial distribution of RNA Pol II clusters. Light is known to incude rapid transcriptional changes along with chromatin reorganisation in the young seedling (reviewed in Bourbousse et al, 2020). We found a light-specific pattern of transcriptional activation engaging prominently clusters at the nuclear periphery. Our findings argue for a non-random, functional distribution of the transcriptional compartment responding to light.

Additional poster authors Ricardo Randall, Department of Plant and Microbial Biology, University of Zurich Devin Routh, Service and Support for Science IT (S3IT), University of Zurich

# Unravelling molecular mechanisms underlying the superior performance of grass stomata

#### Trang Dang

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In higher plants, stomata are found on most aerial parts. Thanks to the guard-cell-pair structure, stomatal pores are adjustable in response to exogenous and endogenous signals, thereby controlling the gas exchange between the plants and the ambient environment. By balancing the trade-off between photosynthetic  $CO_2$  assimilation and transpirational water loss, stomata determine plant water use efficiency, which is among the essential traits of crops to adapt to the changing environment.

Based on the morphology, two types of stomata exist in angiosperms including the dumbbellshaped stomata, found only in grass (Poaceae) family, and the kidney-shaped ones, found in most other land plants. Interestingly, the dumbbell-shaped stomata show more efficient responses to rapidly fluctuating environments comparing to the kidney-shaped counterparts<sup>1</sup>. The superior performance of grass stomata is partly thanks to the addition of two subsidiary cells surrounding the pair of guard cells, allowing rapid shuffling of osmolytes between the two cell types, resulting in rapid stomatal movement. Understanding the molecular mechanisms underlying this osmolyte exchange will contribute significantly to the knowledge of plant responding to changing environments, thereby, providing novel approach to improve plant water use efficiency.

Very recent studies revealed that starch-derived metabolites in Arabidopsis guard cell causes stomatal movement in response to fluctuate light<sup>2</sup>. However, these studies have not yet been demonstrated in the unique dumbbell-shaped stomata of Poaceae. The sugar transportation pathway between guard cells and subsidiary cells is still under-investigated. Hence, my project aims to understand the starch metabolic and sugar and transportation pathways within the dumbbell-shaped stomata complex, thereby, elucidating their roles in the stomatal responses to abiotic factors.

<sup>1</sup>Lawson, T. and Vialet-Chabrand, S. (2019) Speedy stomata, photosynthesis and plant water use efficiency. New Phytol. 221, 93–98 <sup>2</sup>Flütsch, S. et al. (2020) Guard cell starch degradation yields glucose for rapid stomatal opening in Arabidopsis. Plant Cell 32, 2325–2344

Additional poster authors Diana Santelia, Department of Environmental Science, ETH Zurich

# Understanding polycomb repressive complex 2 (PRC2) recruitment during plant embryogenesis

### <u>Yixuan Fu</u>

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The Polycomb group (PcG) proteins, and particularly the Polycomb Repressive Complex 2 (PRC2), are key developmental regulators employed by both plants and animals. The most studied role for PRC2 is the deposition of trimethylation of histone H3 at lysine 27 (H3K27me3), which in turn represses gene expression. However, the mechanism of how PRC2 is recruited to its target genes in plants is not well understood. We aim to identify PRC2 recruiters and characterize their mode of action, with a specific focus on seed development and embryogenesis of Arabidopsis.

We identified several interactors of MEDEA (a subunit of PRC2) by IP/MS and validated the interaction by yeast-two hybrid experiments. Many of these interactors fall into two families of protein. The first class is the HISTONE DEACETYLASE, with mutants showing defected phenotype during seed development. The second class of MEDEA interactors are members of the PHD finger domain-containing protein which binds H3K4me3 (an activation mark). We therefore hypothesize that HISTONE DEACETYLASEs and PHD proteins, through removal of histone acetylation (an activation mark) and recognition of H3K4me3-marked genes, create a favourable environment for PRC2 functioning and recruitment.

To test our hypothesis, we plan to create tagged lines and knock-out lines (including higherorder mutants) for genes of interest, use immunoprecipitation to validate the protein interactions, and analyse histone modifications by ChIP-Seq and CUT&TAG during seed development.

Additional poster authors Sara Simonini, Institute of Plant and Microbial Biology (IPMB), University of Zurich

### Ρ5

# Uncovering divergent drought responses of two forage grass species

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In European grasslands, tall fescue (Festuca arundinacea Schreb.) and perennial ryegrass (Lolium perenne L.) are extensively cultivated for fodder production. Perennial ryegrass is vulnerable to water deficit, and drought causes significant reductions in biomass yield. Contrastingly, tall fescue exhibits tolerance to water deficit and maintains biomass yield under drought conditions. However, the mechanisms underlying drought tolerance in tall fescue remain unclear. This study revealed several traits of tall fescue that enable it to outperform perennial ryegrass under water deficit. For example, image analysis revealed tall fescue had a higher stomata density in comparison to perennial ryegrass. Whilst other studies have shown a high stomata density leads to increased water loss and decreased survival under water deficit, we found that tall fescue responded earlier to water deficit by closing its stomata to reduce water loss and improve survival under drought. Furthermore, tall fescue maintained its leaf elongation at a significantly lower soil moisture content than perennial ryegrass (tall fescue 35%, perennial ryegrass 69%, p = 0.013). These results suggest that in addition to the known root characteristics, tall fescue has several leaf physiological and morphological features which contribute to improved plant performance under water deficit. Our results demonstrate how two species sharing an ecosystem space have evolved different responses to water deficit. This knowledge can help breeders develop drought tolerant forages, ensuring the survival and productivity of European grasslands.

Additional poster authors

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# Evaluating two targeted metagenomics protocols to analyze root-associated fungal communities in ferns and lycophytes

### Thais Guillen-Otero

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Detailed studies of the fungi associated to lycophytes and ferns provide important insights on the early evolution of land plants. However, most investigations so far have assessed fern-fungus interactions based uniquely on visual root inspection. In the present research, we establish and evaluate two metagenomic protocols to analyze the fungal communities associated to fern and lycophyte roots. The first approach targets the ITS rRNA region to screen the general fungal community, whereas the second one uses the 18S rRNA as a marker targeting Glomeromycota fungi. To test these protocols, we collected and processed roots from 12 phylogenetically distant fern and lycophyte species. The protocols yielded a total of 477213 and 1658998 quality reads corresponding to 4909 and 2172 ASVs, respectively. Dominant fungal lineages were Glomerales, Pleosporales and Helotiales. We found remarked compositional differences between the ITS and 18S datasets. Nevertheless, the latter detected the greatest diversity in Glomeromycota. NMDS ordination suggested an important role of the geographical region in determining samples similarities. The ITS-based protocol is a consistent and replicable tool to analyze the general fungal communities associated with fern and lycophyte roots. The 18S protocol is more appropriate for studies focused on the detailed screening of arbuscular mycorrhizal fungi.

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Additional poster authors

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Peter Szoevenyi and Michael Kessler, Department of Systematic and Evolutionary Botany, University of Zurich

# The cell space - Identification and analysis of candidate genes regulating pollen number in *A. thaliana*

### Naoto-Benjamin Hamaya

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In angio- and gymnosperms, pollen grains contain the male gametes. Their number is an important factor for reproductive success. Compared with extensive studies on the cellular differentiation in pollen development, little is known about the molecular basis of this quantitative trait. Recently, we reported the first gene controlling male gamete number, *REDUCED POLLEN NUMBER1 (RDP1)*, from the predominantly selfing species *Arabidopsis thaliana*. This gene encodes a homolog of yeast Mrt4, an assembly factor of the ribosomal large subunit. Null mutants of *RDP1 (rdp1-3)* showed a reduction of 53% in pollen number while their viability was not affected.

In order to understand the underlying gene network, we aimed to identify genes that act together with *RDP1*. Here we present candidate genes for the regulation of pollen grain number. We performed a differential gene expression analysis of tissue-specific RNA-Seq data using *rdp1-3* and wild-type microspore mother cells extracted by laser-assisted microdissection. From this analysis, we selected nine candidate genes and created two to three mutants for each of these genes using the CRISPR/Cas9 system. We found significantly different pollen grain numbers in all mutants of four candidate genes compared to the wild type (on average approx. 10% difference). Two candidate genes are putative positive regulators of pollen grain number, while the other two are putative negative regulators. With RT-qPCR, we further confirmed that these four genes lay downstream of *RDP1* and that there is no feedback regulation among the genes including *RDP1*.

Next, we will assess the pollen number of complementation lines to rule out possible off-target effects from the CRISPR/Cas9 system. Our results constitute the first steps towards understanding the genetic network that regulates male gamete number, which will provide insights into this developmental process and might help to explain the variation of male gamete number found in different species in nature.

Hiroyuki Kakui \*, Department of Evolutionary Biology and Environmental Studies, University of Zurich and Graduate School of Agriculture, Kyoto University

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## P8 Evolutionary constraints at low-latitude range edges under climate change

#### Jessica Heblack

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Warm edges of species distribution have been shown to be under particular threat by climate warming as range retractions are commonly observed. It is unclear why many populations cannot adapt to and long-term persist under warmer, dryer, or hot-drier conditions. Theory suggests that changes in the selection regime and genetic limitations can play an important role in setting range limits: Selection regimes may contribute if environmental gradients become too steep or are multivariate. Genetic limitations may include low genetic variation for environmental tolerances, or genetic correlations antagonistic to the direction of selection. In a greenhouse experiment, manipulating temperature and watering, we investigated how genotypic variation for growth and performance changed from no stress to univariate to combined stress, and the presence of trade-offs among stress tolerances. We raised full-sib plants of 120 families of a genetically diverse central population of Arabidopsis lyrata under average southern edge conditions, as well as under heat or drought as can sometimes occur, or combined heat and drought. By tracking growth, development, and allocation strategies we produced genetic variance-covariance matrices (G-matrix) within and across experimental environments. We will present results that shed light on the role of stress on evolutionary potential in multi-trait space, the presence of trade-offs in coping under multiple stressors, and their effect on adaptation at warm range limits.

Additional poster authors

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### P9 Phosphatases in regulating LRX1-mediated cell wall integrity sensing

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Plant cells are surrounded by cell walls that provide shapes to different types of cells and physically limit cell expansion. For cell growth to take place, the controlled loosening of cell walls is a prerequisite. Thus, control mechanisms that closely survey the different steps of cell wall remodeling are necessary, implying that signals from the apoplast to the cell wall and vice versa ascertain the exchange of information. LRR-extensins (LRXs) of Arabidopsis are regulators of cell wall development. LRXs are high-affinity binding sites for RALF (RAPID ALKALINIZATION FACTOR) peptide hormones that are known to be involved in cell growth regulations by contracting cell wall acidification. The transmembrane protein kinase FERONIA (FER) functions in cell wall integrity sensing and interacts with RALFs and LRXs. Therefore, we propose an LRX-RALF-FER module that regulates cell wall development. In Arabidopsis, LRX1 and LRX2 are predominantly expressed in root hairs and mutations in these genes cause defects in root hair development. Our group uses Arabidopsis root hairs as a model system to study the function of LRX/FER/RALF in cell wall integrity sensing.

In order to decipher the regulatory mechanism of LRX1-mediated cell wall integrity sensing, we screened for repressor of *Irx1* (*rol*) mutants that reconstitute the root hair development in the root hair defective mutant background *Irx1*. One of the characterized mutants, *rol23*, has a mutation in a gene encoding for a type 2C protein phosphatase. Protein phosphatases have been shown to regulate signaling pathways by controlling kinase turnover. This study aims to understand how the induction and retention of LRX1-mediated cell wall integrity sensing signaling are regulated by type 2C protein phosphatase.

Additional poster authors Garbor Kadler, Shibu Gupta Amandine Guérin, Caroline Levasseur and Christoph Ringli, Institute of Plant and Microbial Biology, University of Zurich



### Imputed plant trait data and the bias within

<u>Julia S. Joswig</u> Department of Geography, University of Zürich

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Plant traits are functional facets of biodiversity which manifest plant responses to environmental pressures. Data on plant traits can be sampled in situ or remotely sensed. However, the usage of different trait data streams is often limited by their coverage or mismatch.

For example, in situ sampled data is sparse and of heterogeneous origin: we lack systematic efforts at trait diversity monitoring. This sparsity (in trait space completeness, geographical distribution and resolution) limits the potential for analyses.

In this study, we provide insights for how to make use of plant trait data. In the case of in situ data, we assess an imputation or "gap-filling" approach which is often used to fill missing values in trait data and thus enable multivariate analysis: the Bayesian Hierarchical Probabilistic Matrix Factorization (BHPMF). We investigate whether BHPMF imputation leads to biases in trait space, and identify aspects influencing bias.

Our study extends the criteria for the evaluation of gap-filling beyond classical approaches, providing insight into statistical data structure and allowing better-informed use of imputed trait data, with improved practice for imputation. For the future, citizen science and remote sensing are promising data streams to fill trait data gaps.

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# Wheat responses to drought stress studied by Asian varieties as under-explored genetic and genomic resources

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Wheat is one of the most important food crops in the world. Due to the changing climate, wheat-growing regions are affected by extreme weather conditions such as drought stress, which will increasingly harm wheat production. To mitigate yield losses due to global climate change, one strategy is to breed wheat varieties with better adaptation to scarce water resources. Our project aims to identify wheat genotypes with a better drought adaptation compared to modern varieties and to understand the underlying genetic basis.

We work with an Asian nested association mapping (NAM) population, which has been developed for explorative trait mapping in 24 diverse and highly under-explored Asian genotypes, including many landraces adapted to diverse precipitation patterns. During the development of the NAM population, all genotypes were crossed with the same parental Asian modern variety, namely a Japanese modern variety Norin 61, to increase the genetic diversity compared to a bi-parental mapping population, while keeping a known population structure.

Screening of the parental genotypes in a large-scale field experiment at the International Maize and Wheat Improvement Center (CIMMYT) in Mexico revealed varying responses to drought stress. While modern varieties tended to be highly susceptible to drought stress, landraces showed lower relative yield losses. To understand the genetic basis of this variation, we phenotyped a subset of the NAM population in a comparable experiment at CIMMYT and genotyped it using the GRAS-Di technology. Besides yield, additional traits were measured to investigate the physiological causes of drought adaptation: phenology, plant height, canopy temperature, and spectral indices. We conducted preliminary Quantitative Trait Loci (QTL) mapping on individuals that derived from the crossing of two parental genotypes with Norin 61. This revealed significant QTLs for different traits under drought as well as under irrigated conditions, which makes the planned combined analysis of all phenotyped lines very promising.

The detected QTLs, as well as the identified drought-tolerant wheat lines, should become useful in breeding varieties with increased drought tolerance and in helping the scientific community to better understand the mechanisms underlying drought response.

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# Discriminability of floral colors explains global biogeographical patterns of pollinators

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Flash talk

Flowers are known to emit multisensory cues (visual, chemical, tactile, and acoustic) to communicate with other organisms. Given that pollinators, like most animals, can make decisions based on what they see, floral color is considered a key stimulus for biocommunication that has tightly evolved alongside the sophisticated visual systems of pollinators. Here we ask how important the pollinators' ability to discriminate floral colors is for expanding their geographical ranges. To address this guestion, we tested the hypothesis that pollinators' capacity in telling different colors apart predicts their biogeographical status. Specifically, we compiled an extensive, global dataset of floral reflectance spectra from natural communities, covering a wide range of ecosystems, and we compared the performance of pollinators in correctly discriminating the existing flower colors. For this, we used visual models of color discrimination developed from combining advances in the physiology of vision and behavioral assays for specific species of bees, moths, and flies. Our preliminary results confirm that pollinators' biogeographical status is related to their capacity to effectively discriminate different flower colors. Some of the most successful and widespread bees, which often become invasive, like the honeybee and buff-tailed bumblebee, show remarkably high capacities to accurately discriminate flower colors in almost all environments examined. Exceptions to this include highly isolated areas, like New Zealand, in which the Apidae family has never occurred naturally. Our findings also have interesting implications for the long-standing debate on the origin of stingless bees, given that the performance of American species appears to be better in Asian ecosystems. Finally, non-bee pollinators, like the hummingbird hawkmoth, show markedly lower accuracy in color discrimination than bees, probably reflecting the fact that these animals are not as strong selective forces of global significance as bees. We conclude that the chromatic structure of flower communities, when examined on a macroecological scale, can narrate the biogeographical stories of pollinators, highlighting the pivotal importance of visual biocommunication for shaping biotic environments.

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### P13

# Identification of QTL conferring resistance to stem rust in Italian ryegrass

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Stem rust, caused by the ascomycete *Puccinia graminis* Pers. f. sp. graminicola, is one of the most frequent fungal diseases in Italian ryegrass (*Lolium multiflorum* Lam.). The main damage from its infection occurs late in the season, right before ripening of the seeds, and has a negative impact on forage quality and seed yield. A detailed understanding of the inheritance of plant resistance against stem rust is needed to enable efficient breeding strategies.

In this study, a nested association mapping (NAM) population, which combines the advantage of linkage analysis and association mapping, was established. The NAM population consists of 728 F2 plants. These were derived by crossing an individual genotype of the cultivar Rabiosa (common founder) with 23 diverse founders, followed by open pollination among their respective F1 progeny. Genotyping data were produced by using genotyping-by-sequencing, resulting in more than 70,000 SNP markers. For the common founder Rabiosa, a high quality genome assembly was recently published. The NAM population was phenotyped for stem rust resistance at seed harvest in three different environments with two replicates each. Stem rust infection was scored on a 1 to 9 scale, where 1 indicates the absence of symptoms (resistant) and 9 indicates strong infection (susceptible). A genome-wide association analysis identified two major QTL for stem rust resistance on chromosomes 6 and 7. Blocks of linkage disequilibrium, which are significantly associated to stem rust resistance, were used to narrow down the region of potential causal genes linked to the corresponding peak. Eight possible candidate genes were identified on chromosome 6 and ten on chromosome 7. Together with the knowledge of known stem rust resistance genes, we identified receptor-like serine/threonine-protein kinases on both chromosomes as putative resistance genes against stem rust in L. multiflorum. Although further investigations are needed to find the causal gene, markers to efficiently introgress stem rust resistance can directly be provided to the breeders.

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### Parallel reduction of sporophyte complexity in the moss family Funariaceae – How genome evolution is shaping sporophyte architecture

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Whether evolutionary trajectories leading to a particular function or morphology are predictable is a fundamental question of evolutionary biology. Yet, there is little consensus on this issue and experimental evidence is contentious. The phenomenon of convergent evolution, the repeated evolution of traits in independent lineages, provides ideal replicates to test for constraints on the trajectory of the evolutionary processes. We investigate the genetic bases of parallel morphological evolution in a closely related group of moss species, the family Funariaceae. Fitting the classical example of parallel evolution, a reduced sporophyte phenotype has evolved multiple times independently in the funarioid mosses.

To understand the molecular mechanisms underlying their divergent sporophyte morphologies we use the two species system *Funaria hygrometrica – Physcomitrium patens*. We carried out comparative transcriptomic analysis to identify genes with divergent expression dynamics throughout sporophyte development in the two species. In addition, we generated a chromosome-scale assembly of the *F. hygrometrica* genome to facilitate research on the role of structural variations of the genome in evolution of sporophyte morphology.

Our data suggests that divergent sporophyte morphologies are mainly achieved by heterochronic expression of conserved developmental regulators. However, we also found a significant enrichment of species-specific genes among genes preferentially expressed during sporophyte development. Despite a shared history of ancient whole genome duplications and high collinearity, the genomes of *F. hygrometrica* and *P. patens* differ significantly in size and repetitive element content and composition, which raises the question to which extent genome structure is contributing to the differing sporophyte morphologies.

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### P15

### Is the spatial distribution of native Metarhizium populations in Swiss grassland soils driven by abiotic soil factors?

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Entomopathogenic fungal strains of the genus Metarhizium are widely used as biocontrol agents (BCAs). However, the factors that drive establishment and abundance of applied BCA strains in the soil are still not fully understood. Examine factors, which correlate with the presence of native Metarhizium populations in the soil, may unravel key environmental drivers for successful BCA establishment.

Presence and abundance of native Metarhizium spp. was evaluated at 72 Swiss grassland sites, representing different elevations and management intensities. All sites have previously been char-acterized in the frame of the former EU-project BIOINVENT. Data included soil physicochemical parameters, weather conditions, vegetation composition, and soil bacterial and fungal amplicon sequences.

Overall, we detected 9,269 fungal (ITS) and 23,482 bacterial (16S) sequence variants (SVs). Tax-onomic classification allowed assignment of 10 fungal SVs to the genus Metarhizium. Four of them were included in clade 1, which contains all currently commercialized BCA isolates. The four clade 1 Metarhizium SVs were assigned to *M. robertsii* (SV3), *M. majus/M. guizhouense* (SV82), and *M. brunneum* (SV15 and SV22). Metarhizium clade 1 cumulated sequence abundance correlated well with (R =0.82, p < 2.2e-16) clade 1 ITS-qPCR quantification.

We observed higher abundances of the two SVs belonging to *M. brunneum* in intensively man-aged grassland and a very low presence of the *M. majus/M. guizhouense* SV in mountain regions. We further explored the difference in 14 abiotic factors among sites, where specific Metarhizium SVs were present or absent. Each SV revealed associations with distinct factor compositions. Available soil phosphorous content, pH and temperature correlated with several SVs while, e.g., the soil carbon to nitrogen ratio was correlated to a single SV. Furthermore, we modeled the in-fluence of abiotic factors on the bacterial-, fungal- and plant communities. When taking the geo-graphical, soil and weather parameters into account, Metarhizium presence did not significantly correlate with community structures of plants, bacteria and fungi.

Results of the study indicate that different species or isolates of Metarhizium clade 1 occupy diverse grassland habitats defined by characteristic environmental factors. These findings may contribute to the development of new and more habitat related approaches for biological control.

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### P16

# BRANCHING ENZYME 1 aids plastidial intron splicing through interaction with Maturase K

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Branching enzymes introduce α-1,6-linked branches into starch, the plastidial storage carbohydrate of plants. Impairments in starch metabolism typically moderately affect plant growth but generally lead to viable plants. However, in case of BE1, one of the three branching enzymes in Arabidopsis, homozygous mutants arrest at the heart stage of embryogenesis. Using co-immunoprecipitation in Arabidopsis and tobacco, we found that BE1 interacts with Maturase K (MatK), an essential splicing factor encoded in the plastid genome. Embryo-specific expression of BE1 complements embryo lethality, but the rescued plants produce chlorotic true leaves with aberrant chloroplasts and are seedling lethal. Chemically-induced silencing of BE1 at seedling stage results in pale newly emerging leaves, which show splicing defects in several MatK-associated introns. Preliminary RNA-immunoprecipitation data further suggest that the BE1 complex binds plastid intron RNA, which will be further explored using Chip analysis and RNA sequencing. Together, these data indicate that BE1 has a vital function in chloroplast development rather than in starch metabolism. We propose that BE1 has diverged from canonical branching enzymes to facilitate plastid intron splicing through interaction with MatK.

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# P17 Deposition of histone variant H3.3 in plants

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Organization of eukaryotic DNA into chromatin has many implications in the regulation of all cellular processes that use DNA as a substrate, such as transcription, replication, recombination and DNA damage repair. Nucleosomes are the fundamental unit of chromatin and are composed of an octamer of histones H2A, H2B, H3 and H4. Histone H3 has two main variants, H3.1 and H3.3, and their differential incorporation affects nucleosome stability and high order chromatin organization. Both variants are essential for animal and plant development as loss-of-function mutants are not viable or sterile. H3.1 is enriched in heterochromatic regions, while H3.3 is associated with transcriptionally active regions in the genome. In addition, H3.3 is also enriched at telomeres, regulatory regions and rDNA repeats. For the appropriate locus-specific incorporation into chromatin, histone variants require specific histone deposition machineries. In animals, H3.3 deposition is carried out by the Alpha Thalassemia-mental Retardation X-linked (ATRX) and the Histone Regulator A (HIRA) complexes. ATRX is a highly conserved chromatin remodeler of the SWI/SNF family and was shown to interact with the Death domain-associated protein (DAXX) in animals that brings specificity to H3.3 over H3.1.

In plants, H3.3 incorporation and its impact on gene expression are less well understood. The plant orthologue of ATRX has been shown to bind histones, and *atrx*<sup>-/-</sup> knock-out plants display reduced H3.3 occupancy. No homologue of DAXX has been identified in plants and it remains to be determined whether plant ATRX associates with other histone binding proteins to incorporate H3.3. In my project, I identify the interaction partners of ATRX in *Arabidopsis thaliana* by using immunoprecipitation followed by mass spectrometry (IP-MS) and found that plant ATRX is part of a large chromatin remodeling complex. I show a novel interaction between ATRX and PICKLE (PKL), a chromatin remodeling factor of the CHD3/CHD4 subfamily. Altogether, the characterization of the ATRX-PKL complex will bring insights on how the histone variant H3.3 is incorporated in plants and how its genome-wide distribution pattern defines different chromatin states and regulates transcription.

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# Repeated adaptation in wild carnations: the role of shared genetic variation and novel mutations in convergent selection signatures in the genome

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Understanding the genomic origins of adaptation is a major challenge of contemporary evolutionary biology. Studying selection pressures driving repeated adaptation across a single species can provide insights into the constraints required for adaptation in nature. Despite frequent reports of genomic convergence, we know little about the mechanisms determining these processes. Specifically, we lack comparative assessments of the relative contribution of shared versus novel genomic variation during rapid convergent adaptation between closely related species, i.e. in the 'gray zone of speciation'. We will leverage the repeated adaptation of two Dianthus species to substrate drivers such as ionic stress and drought, to address the role of spatially fluctuating selection in convergent adaptation to harsh environments. Through genome scanning approach we will identify genes involved in fundamental processes such as ion transport, stress-signaling, and mineral nutrition. Furthermore, studying convergent evolution in newly established model species of Dianthus will enable valuable comparisons outside the leading plant model family Brassicaceae. In summary, using an integrative approach of statistical genomics, eco-evo experiments and functional approaches, we aim to identify the shared genomic basis governing convergence and adaptation to challenging soils within the Caryophyllaceae family.

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# P19 Efficient monitoring of plant genetic diversity in multispecies meadows

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Plant biodiversity can influence ecosystem functioning in grasslands (e.g., yield stability, resilience towards pathogens and environmental stress). Much of what is known about the biodiversity-ecosystem functioning interactions in grassland is at the level of species richness, i.e., the between-species diversity. However, plant genetic diversity (PGD), i.e., the within-species diversity, also plays a role in these interactions. We recently developed a PGD assessment method that is based on multispecies amplicon sequencing. This method greatly reduces analysis costs, potentially enabling large-scale applications.

In this study, we aim to establish this method to monitor PGD changes under experimental field conditions. We set up a field experiment in Langenthal (BE, Switzerland), where 60 meadow plots were oversown. The experimental treatments are combinations of the following variables: time of overseeding (October 2021 or March 2022), seed mixture (Schweizer 431U and 440U), seeding intensity (20 or 40 kg/ha), and management (mowing or grazing). The plots were set up in randomized order in triplicates, including controls without overseeding. Multispecies plant samples were taken before overseeding and at three different times over the course of the growing season of 2022. Those samples will be used for multispecies amplicon sequencing, aiming to assess sequence differentiation at the community- and species-levels among the treatments.

The outcome of this work will provide new insights on the efficacy of overseeding and its effect on grassland PGD. This will also help in establishing a cost-effective pipeline for large-scale, multispecies PGD monitoring in grasslands.

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# How do different plant species recruit their microbiome?

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The communication between plants and microbes can result in beneficial, pathogenic, or neutral interactions. Previous research looked at belowground plant-microbe interactions mainly from the microbial perspective; however, we aim at understanding how plants shape their microbial community. For this, we examine a set of phylogenetically diverse plant species for their exudation profiles (nutrients or chemical signals from root tissues) in various environments. We aim to understand which metabolites that are exuded from the plant result in association with different microbes. Preliminary results have shown that root exudate profiles are dynamic within and across species and are influences by their environment. This trend is seen in root exudates of plants grown in both hydroponic and soil systems. With the knowledge of the metabolites play key roles in the plant-microbe interaction interface. This can help inform us of the structure and function specific plant and microbe interactions improve plant health and yield for more sustainable agricultural practices.

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# Inferences on the plant-Nostoc symbiosis

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The plants' microbiome holds many valuable features. For many decades, research was directed towards nodule-forming bacteria and mycorrhizal fungi which provide nitrogen and phosphorus, respectively. With the advances in sequencing techniques, microbiome studies reveal a highly diverse composition of microbes. However, cyanobacteria are often overlooked because they are codescendants of the photosynthetic precursor of the chloroplast and are thus harder to distinguish. Diazotrophic cyanobacteria of the genus Nostoc show great potential to be used as biofertilizers and to study the impact of the cyanobacterial symbiosis of the first land plants.

While the microbial side of the plant-cyanobacteria beneficial symbiosis is well established, the molecular details on the plant host still need to be unraveled. In this work, we identified the genes involved in initiating, establishing, and maintaining symbiosis in the well-established model system consisting of the hornwort *Anthoceros agrestis* and the cyanobiont *Nostoc punctiforme*. In addition, the hornwort orthologs of the genes of the common symbiosis pathway, which play a role in nodule forming and arbuscular mycorrhiza symbioses, have been examined to test whether a conserved module exists. To enable inferences on phytohormones attracting Nostoc and to verify specific plant genes, a *Nostoc punctiforme* mutant was created. Hormogonia are the motile filaments of Nostoc that are critical to infect a plant. We created fusions of GFP to three genes expressed during the differentiation of hormogonia. This allows for an easy quantitively assessment of hormogonia induction.

This study greatly contributes to unraveling the symbiotic mechanism of the cyanobacteria hosts and it complements what is known about plant interactions with nodule-forming bacteria and arbuscular mycorrhizal fungi. In addition, given the multiple origins of cyanobacteria symbiosis in the plant tree of life, future work can build upon this hornwort study to examine other plant lineages, thereby testing if there is a unified molecular mechanism behind plant-cyanobacteria symbiosis. Furthermore, the essential genes and metabolites identified through this research will lay the foundation for future efforts to assess and design crop-cyanobacteria symbiosis.

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# How to decrease phosphate (P) losses from acid sulfate soils in the lower Mekong delta while maintaining optimum crop yields?

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Acid sulphate soils (ASS), when oxidized develop high acidity, high levels of mobile toxic elements and very low P availability due to the high levels of Al and Fe oxides. In the Vietnamese Mekong Delta region, around 1.2 million ha are covered by ASS, including salt-affected area, accounting for 40% region's total land area and nearly 10% and 25% of world and Asia ASS areas, respectively. Promising crops currently grown on upland ASS are pineapple and yam because of their economic value and adaptability to these soils. It is hypothesized that high application rates of P fertilizers are applied to attain optimal crop yields, causing not only low P use efficiency (PUE) but also high risk of water pollution due to P losses from soils. However, there is little available information on P dynamics and input-output budgets and P nutrient management for yam and pineapple grown in ASS.

We aim to understand P dynamics in ASS and develop tailored soil management options that will improve PUE in yams and pineapple and decrease P losses to water. To reach these goals, a systematic approach is employed: Firstly, we are studying the forms and availability of P and their controlling processes across ASS profiles by conducting soil profile description, soil horizons sampling, general soil characteristics analyses, measuring P availability with anion exchange resin method, and broad and in-depth P speciation with Hedley extractions, P K-edge X-ray adsorption near edge structure spectroscopy (XANES, for inorganic P), and 31P Nuclear magnetic resonance (for organic P). Secondly, we will assess how soil management options can improve P use efficiency by studying their effects on i) P availability and dynamics in incubation and in pot experiments and on ii) soil P availability, crop yields and PUE in field experiments. Finally, to identify the options that help reduce P losses to water, we will evaluate the P losses at plot level via runoff and via bypass flow using brilliant blue in the field experiments and then simulate P losses and transfer in water at a larger scale employing SWAP model coupled with the Mike 11 HD/Ecolab model.

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# Homoeologous gene expression analysis of wheat cultivars and synthetic hexaploid wheats as an example of cold response

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Wheat is an important allohexaploid crop that is established through interspecific hybridization and whole genome duplication. Polyploidization is a common phenomenon in many plants and animals. Although it is considered as an important driving force of evolution, how polyploidization contributes to the evolution and environmental adaptation is still under discussion. In this study, we investigated the relationship between homoeologous gene expression and cold response.

We conducted RNA-seq analysis using nine accessions: three hexaploid wheat cultivars, one tetraploid wheat cultivar (Langdon), two lines of *Aegilops tauschii* Coss., one diploid wild wheat relative, and two lines of synthetic hexaploid wheat (Syn) obtained by crossing between tetraploid wheat and *Ae. tauschii*. The obtained RNA-seq reads were mapped on A, B, and D genomes of the wheat reference sequence (CS RefSeq v1.1, IWGSC 2018), respectively, using STAR (Dobin et al. 2013). Homoeolog triads, the set of homoeologous genes on A-, B-, and D-subgenome, were identified by homology search based on their CDS sequences. Then, the origin of the reads was defined using EAGLE-RC (Kuo et al. 2020) to evaluate the expression level of each homoeolog in the homoeolog triads.

First, we compared the change of gene expression pattern by one-week cold (7°C) treatment in each line. Patterns and numbers of the differentially expressed genes (DEGs), including cold-responsive genes (e.g. COR15), were different in each line. The diversity of DEGs among cultivars was sometimes greater than the diversity between cultivars and synthetic hexaploids. Interestingly, the cold-responsive genes were more up-regulated under 7°C treatment in a Japanese cultivar than in a Swiss cultivar. This may refer to the adaptation to the growing environment of each line, reflecting more sensitivity of Japanese cultivar due to milder Japanese winter climate than Switzerland.

Next, the expression ratio of homeologs from AB and D genomes (A+B:D) were calculated. Most homoeolog triads showed the same response before and after cold treatment, but about 1% of homoeolog triads showed a significant difference in response to cold between subgenomes. This suggests that there is no drastic change, so-called "genome shock", at least for gene expression regulation.

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# Deciphering the chemical communication in diverse plant and insect communities

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Extensive work in chemical ecology has demonstrated the critical role of chemical traits in modulating biotic interactions. In the case of plants, for example, by either deterring or attracting particular insect species, such as pollinators and herbivores. Most studies in chemical ecology have been conducted on few species at a time. Biological communities, in contrast, are characterized by a diversity of species and interactions. However, the lack of practical and conceptual frameworks has hindered the development of scalable approaches capable of addressing community-level questions. For instance, what is the role of plant secondary metabolites in structuring species-rich trophic networks?

To address this central question, my PhD project proposes to characterise the plant chemical traits related to floral advertisement (volatile organic compounds emitted by flowers and flower color) and floral rewards (nectar and pollen chemistry) represented by a diverse plant community in the eastern Swiss Alps. Concomitantly, we will reconstruct the interaction network between plant species and flower-visitor insects, and observe how interactions by different insects relate to plant chemical traits. Lastly, by integrating an approach based on information theory, we will construct theoretical models to identify which evolutionary processes may produce a structure of community-level chemical information consistent with the one observed empirically.

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# Developing a sampling design for a genetic diversity monitoring program in Switzerland

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Many efforts have been made to integrate genetic diversity, one of the three main pillars of biodiversity, into monitoring projects and conservation efforts. The development and improvement of genetic diversity indicators is an intensively discussed topic. The type of genetic indicators that can be inferred from genetic monitoring is strongly dependent on the underlying sampling design. Here, we developed a universally applicable proportionally stratified random sampling strategy optimized for population genetic inferences. We tested and validated this protocol in a pilot study for a monitoring of genetic diversity in Switzerland. Our aim was to sample 30 populations from each of five species with 10 individuals each. Based on a 5x5 kilometer grid with 3,700 cells, we randomly selected 40 cells with recently documented occurrence data of the species, taking into account the species' distribution across the five major biogeographic regions of Switzerland, which we treated as sampling strata. The sampling design thus accounts for the unequal sizes of the five biogeographic regions while having a random sampling design within these regions. To avoid sampling bias within the 5x5 km cells, the order of locations at which to attempt sampling was again randomized based on available occurrence data. We show that samples obtained by a proportionally stratified random sampling design cover the entire environmental climate-space (temperature and precipitation) of the corresponding species in Switzerland. The developed sampling design provides the foundation to infer population genetic indicators such as e.g. neutral and adaptive genetic diversity, inbreeding, connectivity or adaptive potential.

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# Pyrenoid formation in hornworts: genomic hints for green algal-like pyrenoid scaffolding mechanisms

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Flash talk

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Biophysical carbon concentrating mechanisms (CCMs) operating at the single-cell level have evolved independently in various lineages of eukaryotic algae and a single land plant lineage, the hornworts. An essential component for an efficient eukaryotic CCM is a pyrenoid, a specialized compartment inside the chloroplast that mainly comprises the CO<sub>2</sub>-fixing enzyme RuBisCO. Knowledge on pyrenoid assembly could help to induce its formation in pyrenoid-free plants, potentially increasing photosynthetic efficiency. Information on pyrenoid biology is primarily available for the unicellular green alga, Chlamydomonas reinhardtii, suggesting that pyrenoid assembly is achieved by scaffolding the RuBisCO with the aid of intrinsically disordered proteins (EPYC1 in Chlamydomonas). We investigate the assembly mechanism of pyrenoids using hornworts as a model system. In hornworts, pyrenoids have repeatedly been gained and lost during the last 50 million years, which is in line with the hypothesis that their assembly is controlled by a few master steps. We provide genomic evidence for the presence of a linker protein in a pyrenoid-bearing hornwort and its absence in a species without pyrenoids. Furthermore, we use gene expression evidence to show that the linker is localized in cells where pyrenoid assembly is observed. Finally, we provide computational evidence that the linker protein of hornworts may be more amenable to form pyrenoid-like structures with the RuBisCO of vascular plants than its Chlamydomonas analog. To identify CCM genes in hornworts we use a combination of tools including (1) protein co-IP of pyrenoid components, (2) localization of candidates homologous to CCM genes of Chlamydomonas, and (3) CO<sub>2</sub> assimilation measurements in pyrenoid-bearing and pyrenoid-free species. We provide evidence that the scaffolding candidate and the RuBisCO co-localize using fluorescent reporter lines. We show that the LCIB hornwort homolog is less intimately linked to the pyrenoid than in Chlamydomonas and localized to the outer thylakoid membranes and/or chloroplast envelop. Surprisingly we observed that CO<sub>2</sub> assimilation efficiency does not differ between pyrenoid-bearing and absent hornworts suggesting that CCM may be functional without pyrenoids. Our results provide insight into the pyrenoid-based CCM of hornworts implying molecular mechanisms potentially like those in Chlamydomonas, while others may be unique.

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# Temperature-induced plasticity and reciprocal selection in a plant-pollinator-herbivore system

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The geographic mosaic of coevolution predicts variation in abiotic and biotic factors to change coevolutionary dynamics between organisms. Although reciprocal selection is the first step in coevolution and essential for its understanding, it is poorly understood how environmental variation influences reciprocal selection between organisms. This is especially relevant under the current global warming. Global warming increases temperatures in many habitats, possibly disrupting plant-insect interactions because plant and insect physiology are strongly influenced by temperature. To understand how plants and insects (co)adapt to global warming, we need to study how temperature influences their interactions, the underlying phenotypic traits, and the resulting reciprocal selection. In this study, we investigated how temperature changes reciprocal selection between the field mustard (Brassica rapa) and its pollinating herbivore, the small cabbage white (Pieris rapae), using ambient (average 23°C) and hot (average 27°C with weekly 30°C day) greenhouse environments. We show that temperature induces extensive plasticity in plant and butterfly phenotype. Heat-treated plants produced more but smaller flowers compared to non-treated plants. Flowers of heat-treated plants produced less scent and less nectar. Butterflies had faster flower visitation speed in the hot compared to the ambient environment. Such temperature-induced plasticity changed the mutualistic and antagonistic interaction strength of the plant-pollinating-herbivore interaction. In the hot environment, butterflies visited more flowers, but plant fitness increased less per butterfly visit. In contrast, butterfly fitness was correlated with flower visitation and butterflies laid more eggs in the hot environment. Plants better tolerated herbivory in the ambient compared to the hot environment. Combining trait and fitness measurements, we show that temperature changes reciprocal selection between plants and butterflies, including flower traits such as flower scent and nectar, and butterfly foraging traits such as tongue length and foraging speed. Therefore, temperatureinduced plasticity in the interaction between a plant and its pollinating herbivore likely reshapes their coevolutionary trajectory.

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# Are soil development and changing plant competition important drivers of Arctic Greening?

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The Arctic has been warming faster than the global average during the last decades, resulting in the temporal and spatial expansions of vegetation, termed "Arctic Greening". This process arises from higher temperatures, prolonged season length and greater plant productivity. At the same time, an increasing number of non-native plants is being recorded in Arctic ecosystems. On Svalbard, 98 alien species have already been identified, especially in disturbed and nutrient enriched soils near settlements. Previous research has focused on climatic factors as the main cause of Arctic Greening. However, an accelerated rate of soil development could also play a key role in shifting vegetation patterns. Thus, variation in the responses of plants to a warmer Arctic may be controlled not only by temperature, but also by an interplay of soil development, associated soil microbial community, human disturbance and novel plant introductions, as well as the ability of native plants to adapt genetically to these changes. With time, we might therefore see dramatic shifts in the composition of Arctic plant communities, with range-expanding and non-native species outcompeting highly specialized tundra species.

We hypothesize that the modification of environmental conditions, soil properties and biological processes will play a key role in plant community composition, the spread of plant species and their functional variability. To disentangle these different components, we conducted vegetation surveys and trait measurements on different geologies on Svalbard this summer. Due to its high variability in surface geology, topography and soil nutrient status in a relatively small spatial scale, Svalbard offers ideal conditions to examine how the interplay between geology, species interactions and soil development might influence Arctic Greening. Furthermore, we will conduct a fully factorial experiment at ETH Zurich next spring, to dissect the effects of plant competition between non-native, range-expanding and native tundra species and the role of different soil characteristics and microbial communities in this context. Here, we present a first insight into this interdisciplinary project, which began in early 2022 to help improve our understanding of how sensitive Arctic ecosystems might change in the future.

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# Transmission-enhancing effects of a plant virus depend on symbionts of insect vector

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Vector-borne pathogens frequently alter traits of their primary hosts and vectors in ways that enhance their own transmission. Such effects may be modulated by the presence of other microbial symbionts, whose fitness interests may or may not diverge from those of the pathogen, but few studies to date have examined such interactions. In this study, we explored how the effects of a plant virus on interactions between host plants and aphid vectors are influenced by the presence of different aphid endosymbionts, which have previously been shown to influence such interactions via effects on aphid behavior and reproduction. We documented the effects of five different aphid endosymbionts on traits of pea aphids (Acyrthosiphon pisum) relevant for the transmission of pea enation mosaic virus, including effects of the pathogen on plant profile, aphid's attraction to and dispersal from infected plants, and aphid's performance on healthy and virus-infected hosts. Analysis of metabolites using gas chromatography-mass spectrometry revealed that virus infection altered plant phenotype by downregulating general hormonal defenses against aphids and by enhancing nutritional profile of fava beans. Subsequent experiments revealed that these plant changes influenced only aphids associated with certain species and strains of endosymbiotic bacteria, Regiella insecticola or Hamiltonella defensa. Aphids harboring these endobacteria had foraging behaviors conducive to pathogen acquisition and consecutive dispersal in dual-choice assays. Reciprocally to behaviors facilitating the pathogen spread, virus infection of host plants reversed phenotypic and reproductive costs of aphid symbiosis with these specific endosymbionts in performance assays. Additional analyses of metabolites revealed that aphids harboring these endobacteria had higher levels of critical amino acids, sugars, and neuromodulator molecules when feeding on virus-infected plants, which indicate some of the mechanisms involved on the combined effects of virus endosymbionts on aphid traits. Taken together, these findings illustrate that virus manipulative effects on host plants and aphid vectors are indeed modulated by aphid's endosymbiotic bacteria. Furthermore, the specificity of these interactions has intriguing implications for aphid bacteria and vector-virus mutualisms, as well as potentially important implications for plant epidemiology.

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# Neighbor GWAS: incorporating neighbor genotypic identity in genome-wide association study of field-grown *Arabidopsis thaliana*

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Trait expression of an individual plant often depends on neighboring plants besides the plant's own genotype in field environments. Yet, this fact has not been considered in genome-wide association studies (GWAS) of field-grown plants. Based on the Ising model of ferromagnetism, we have recently proposed a new method that incorporates neighbor genotypic identity into GWAS, named "Neighbor GWAS" (Sato et al. 2021 Heredity). We are applying the neighbor GWAS to insect herbivory on field-grown accessions of *Arabidopsis thaliana*. So far, we have found that neighbor GWAS explained a significantly larger fraction of herbivory variation than standard GWAS. By implementing the neighbor GWAS as genomic prediction, we also uncovered key genotype pairs that mitigated herbivory by mixed planting. To dissect the genomic basis of the neighbor effects, we are also applying the neighbor GWAS (eGWAS) detected significant expression quantitative trait loci (eQTLs) with respect to the neighbor effects. Overall, our study provides a new GWAS approach to analyze plant-plant interactions as a complex trait under spatially structured environments.

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# Sampling historical DNA to assess the impact of habitat loss on genetic diversity in a peatland-specialist

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Genetic diversity is the basis for adaptation to changing environmental conditions such as climate change. Human-induced habitat loss and landscape fragmentation are important drivers of population size reduction which can lead to genetic diversity loss. As a peat bog specialist, the cottongrass Eriophorum vaginatum has undergone major habitat loss in Switzerland during the past two centuries as a consequence of wetland drainage and peat extraction. Habitat loss has mainly occurred in the central plateau, a densely populated and relatively warm biogeographic region of Switzerland. In the pilot study for a monitoring of genetic diversity in Switzerland, E. vaginatum is monitored retrospectively using samples from natural history collections to understand the potential loss or change of genetic diversity during the last century. We developed a sampling scheme to select available E. vaginatum vouchers and corresponding contemporary samples for whole-genome re-sequencing (WGS) to assess whether past habitat loss has mediated the extinction or isolation of genotypes that are potentially adaptive under future climate change. From >700 vouchers available in seven different Swiss herbaria, we selected 226 specimens from across Switzerland considering age, spatial distribution, and habitat change. We tested and optimized DNA extraction protocols and performed WGS to identify methodological challenges and possible limitations of the data. With this project, we aim to measure genetic diversity change in E. vaginatum and to better understand consequences of habitat loss on the potential of species to adapt to climate change.

Additional poster authors

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# Impacts of human excreta-derived fertilizers for carrot and spinach growth: comparing four different human excretaderived fertilizers to no-fertilizer, NPK and green compost

## Sarah van den Broek

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The use of human excreta-derived fertilizers (HEDFs) contributes to sustainable agriculture by reusing valuable nutrients for plant growth that would otherwise be disposed of as waste. However, there are many risks associated with HEDFs, as they may contain antibiotics, pathogens and/or antibiotic resistance genes (ARGs).

To explore these potential negative side-effects in more detail, we conducted a 12-week greenhouse trial to measure the response of carrot (*Daucus carota*) and spinach (*Spinacia oleracea*) to four different HEDFs (two urine-based and two feces-based) grown in two contrasting soils. The HEDFs consisted of stored urine (South Africa), nitrified urine (produced from the stored urine), co-compost (South Africa), and vermicompost (Rwanda). As controls we used a no-fertilizer treatment, mineral nitrogen-phosphorous-potassium fertilizer (Switzerland) and green compost fertilizer (South Africa). We used two soil types from agricultural fields in Kenya: a sandy soil (80% sand) and a clay soil (75% clay).

Our results showed that the sandy soil had significantly higher yield than the clay soil for both spinach and carrot. Spinach yield was highest in the no-fertilizer control (both soils) and lowest in stored urine (sandy soil) and co-compost (clay soil). Carrot yield was highest in vermicompost (sandy soil) and green compost (clay soil) and lowest in stored urine (both soils). Green compost had the highest survival rate of all treatments and plants. Stored urine and nitrified urine were among the worst treatments for both plants in both soils, going as low as 30% survival rate.

In conclusion, our study showed that soil type is more important for carrot and spinach yield than fertilizer type, but more studies are needed to confirm this in other soil types. While none of the HEDFs we investigated had a consistent yield increase, stored urine and nitrified urine decreased yield in almost all contexts compared to no-fertilizer controls. Further characterization of the experimental treatments and plants could explain the yield response. Namely, we will investigate soil physicochemical properties, soil microbial community composition, ARG diversity and abundance, enzyme activities, antibiotic soil pollution and plant antibiotic uptake, plant nutrient composition and plant RGB values to screen for changes in chlorophyll composition.

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# Intraspecific trait variation of beech seedlings (*Fagus sylvatica*) in a common garden experiment

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Beech (*Fagus sylvatica*) is common in Europe and commercially important. However, climate change, including droughts and heatwaves, is likely to change the distribution of beech and lead to local population declines. For beech to persist, it relies on intraspecific phenotypic and genetic diversity. We conducted a common garden experiment in with 200 beech seedlings from 16 European beech populations with known population genetic structure. Once placed in the common environment, we aimed to determine intraspecific trait variation that remained and its association with genetic variation. To determine phenotypic variation, we assessed tree growth rates, mortality, leaf damage and leaf optical properties using leaf spectroscopy throughout the growing season. We found strong signals of beech tree origin on the plant phenotype for plant height, stem diameter and leaf damage. Other traits, such as leaf spectra, were also highly variable between trees but lacked an origin-specific signal. Thus, traits varied in how much they are genetically determined. Our study provides a window into the mechanisms underlying the link between intraspecific genetic diversity and trait variation.

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# A tale of the YABBY family of transcription factors: YABBY function in hornwort sporophyte development

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YABBY transcription factors are a class of plant specific transcription factors found in every seed plant to date. As YABBY genes were neither found in fern genomes sequenced so far or in the genomes of model moss and liverwort species, they were thought to be specific to seed plants. However, YABBY genes were recently shown to be present in a lycophyte species, in hornworts and some streptophyte algal species. Previously, we found a single YABBY gene to be exclusively expressed in the sporophyte of the newly established model hornwort Anthoceros agrestis. YABBY function has only been investigated in flowering plants, where YABBY genes were shown to be involved in controlling aspects of lateral organ development. Since hornwort sporophytes don't posess any lateral organs, YABBY function in hornworts is unknown. Deciphering the role of YABBY during hornwort sporophyte development, in comparison to the well known roles of YABBY in Arabidopsis thaliana, could contribute to our understanding of land plant sporophyte body plan evolution. Therefore we set out to investigate the role of YABBY TF during the development of A. agrestis sporophytes. Using a newly established hornwort transformation method, we produced YABBY promoter-reporter lines to track expression of YABBY within the sporophyte. Using the same transformation method, we then attempted to produce AaYABBY CRISPR knock out lines and describe resulting phenotypes. Promoter reporter lines reveal YABBY expression to be restricted and highly specific to the archesporial tissue, suggesting its involvement in the development of sporogenous tissue. A putative YABBY knock out plant shows a strong sporophyte phenotype in agreement with a proposed role of YABBY in controlling cell division patterns within the archesporial tissue. In the future, additional YABBY KO lines need to be created, the sporophyte phenotype has to be investigated and described in detail, and YABBY rescue lines need to confirm the phenotype as caused by missing YABBY function.

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# The relative contribution of genetic and epigenetic variability to adaptation

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Trans generational epigenetic inheritance is a much-researched topic, yet its contribution to adaptation in an evolutionary context remains debated. Our project is aimed at getting a better understanding of the relative contributions of epigenetic and genetic variability to adaptation. To disentangle the genetic and epigenetic contribution to adaptation, we plan to generate starting populations of Funaria hygrometrica, that are only genetically or epigenetically variable and have these populations go through experimental evolution. To induce hypo methylation, we are going to utilize the methyl transferase blocker Zebularine, whereas the genetic mutations are going to be induced through UV-radiation. To balance the respective variabilities to reflect the natural state, preliminary experiments will need to be carried out to determine the dose effect relation of the two treatments. The selection pressure for the experimental evolution is going to be exerted by adding salt to the growth media inducing stress for the plants. Every generation will be created by mutagenesis or epimutagenesis followed by growth under the selective agent. During experimental evolution, the growth of the individual populations is going to be recorded as a proxy for adaptation. After the experimental evolution experiment, evolved populations will be allowed to grow without stress for two generations before sequencing, to ensure that the observed adaptation is inheritable across generations and not simply due to phenotypic plasticity. We plan to use a combination of whole genome bisulfite sequencing and whole genome resequencing, to record the changes in both the genome and epigenome. By combining the sequencing and phenotypic data we strive to elucidate the potential of genetic and epigenetic variability for adaptation.

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# Attract-and-infest strategy to biologically control adult and larval stages of *Popillia japonica*

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The Japanese beetle (*Popillia japonica*) is an invasive insect pest, threatening agricultural production in Europe with its extremely polyphagous lifestyle and fast invasion pattern. Damage of adult *P. japonica* is very conspicuous, and beetles may easily be detected with monitoring traps. However, larval stage is well concealed by its subterranean life-style and damage in grassland habitats is visible only after grubs reach high population levels. This difference in habitat of larval and adult life stages leads to a significant time-lag between the detection of beetles and the application of control measures to suppress build-up of larval populations. The implementation of attract-and-infest traps aims to close this gap by using adult beetles as vectors to disseminate a fungal biocontrol agent in both adult and larval habitats and population. We use lab and field experiments to test whether (1) infested adults horizontally transmit the fungal inoculum to other adults and (2) vertically carry the inoculum to larvae and their habitat. The attract-and-infest strategy may be an important tool to break the invasion pattern of Japanese beetles in Europe.

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# Genome-wide association study highlights escape from aphids by delayed growth in *Arabidopsis thaliana*

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Plant phenological and architectural traits modulate the likelihood of herbivore settlement, but their key genes remain largely unknown. Here, we conducted a genome-wide association study (GWAS) of aphid abundance in a field population of *Arabidopsis thaliana*. Near a significant peak of GWAS, we found growth-related genes, such as *ROOT HAIR DEFECTIVE3*, and candidate genes with unknown functions. Out of these unknown genes, a mutant of the putative ribosomal gene (AT3G13882) exhibited far slower growth and later flowering phenotype than a wild type under laboratory condition. The turnip aphid *Lipaphis erysimi* failed to settle a colony on the ribosomal gene mutant, probably due to the rosette size of this mutant being 61% smaller than the wild type. These findings suggest that the side effects of growth-related genes on herbivore abundance may be more important than currently recognized.

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# Shared transcriptional response in independent lineages of land plants during plant- cyanobacteria symbiosis shed light on its evolutionary origin

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While associations between plants and arbuscular mycorrhizal fungi as well as noduleforming bacteria are actively studied, the plant-cyanobacteria symbioses are poorly investigated. Initiation of the plant-cyanobacteria symbiotic interaction starts with the plants' secretion of chemoattractant and a specific hormogonium-inducing factor (HIF) to attract cyanobacteria. While the HIF is essential to establish the symbiotic interaction, its exact chemical identity remains to be determined.

Cyanobacteria-plant symbiotic interactions have independently evolved in isolated lineages of land plants, such as the hornworts, and liverworts. Here we employ the tractable system of the hornwort, Anthoceros agrestis, the liverwort, Blasia pusilla, and the cyanobiont, Nostoc punctiforme to investigate how the plant host attracts the cyanobacterial partner and establishes the symbiosis. Using two model systems with independently evolved symbiotic interactions provides a unique opportunity to identify shared molecular mechanisms. Plant-cyanobacteria interaction can be only initiated when plants are starved of combined nitrogen. Therefore, we examined gene expression of a hornwort (Anthoceros agrestis) and a liverwort (Blasia pusilla) during nitrogen-starvation. Comparative transcriptomic analyses show that nitrogen-starvation induces the upregulation of genes involved with flavonoid and carbohydrate synthesis in both bryophyte species. Importantly, these genes are not upregulated upon nutrient starvation in symbiont free plants or plants capable of forming symbiosis with AM mycorrhiza fungi and/or nodule forming bacteria. Flavonoids are efficient inducers of heterocyst-related genes in cyanobacteria and are known to be important in attracting nodule- forming bacteria. Lipid biosynthesis and transfer from the host plant to the symbionts are essential for mutualism with mycorrhizal fungi.

Our results suggest that bioactive molecules involved in initiating the plant-mycorrhiza and nodule-forming bacteria symbioses are produced under nitrogen starvation and are potentially involved as HIF in the hornwort/liverwort-cyanobacteria symbiosis. We propose that molecular signals involved in the hornwort/liverwort-cyanobacteria symbiosis may have evolved from a general starvation response by linking it with the activation of new pathways enabling the initiation of the plant-cyanobacteria interaction.

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