

ZMBP Summer Academy 2022

Speaker Abstracts

Emergence and propagation of epigenetic patterns during somaclonal reproduction

J. Isaac Rodríguez¹, Anjar Wibowo², Ilja Bezrukov², Detlef Weigel², CLAUDE BECKER¹

¹ LMU Biocenter, Faculty of Biology, Ludwig Maximilians University Munich, 82152 Martinsried

² Department of Molecular Biology, Max Planck Institute for Developmental Biology, 72076 Tübingen

Stress and other environmental cues lead to changes at the epigenetic level. While some of these induced changes positively affect the plant's response to a given stress and therefore are of potential interest in increasing stress tolerance, they are usually unstable and get reset during sexual reproduction. Many plants, including several major crops, reproduce or are propagated asexually. We have explored how asexual reproduction influences epigenome conservation and dynamics. In my talk, I will highlight how somaclonal propagation in *Arabidopsis thaliana* enables the reprogramming of the DNA methylation landscape and how these altered methylated states can persist across ensuing sexual reproduction events. We used different genetic backgrounds and various regeneration methods to investigate how these features influence the genetic and epigenetic mutation rates and patterns. Our findings provide insights into the influence of epigenetic variation on somaclonal phenotypic variation in plants.

Leveraging evolutionary diversity to discover new autophagy mechanisms in plants and humans

Yasin Dagdas, @PlantoPhagy, yasin.dagdas@gmi.oeaw.ac.at
Gregor Mendel Institute (GMI), Austrian Academy of Sciences,
Vienna BioCenter (VBC), Vienna, Austria

Selective autophagy is a highly conserved quality control mechanism that remodels cytoplasmic contents during cellular reprogramming. It is induced by a wide spectrum of biotic and abiotic stress responses to keep the cytoplasm in tune with the changing environment. Selective nature of the process is ensured by selective autophagy receptors (SARs) that bridge the cargo with the core autophagy machinery to mediate selective cargo degradation. SARs recruit the autophagy machinery through interacting with ATG8, a ubiquitin-like protein conjugated to the phagophore. Despite recent advances in metazoans, the catalogue of SARs, therefore our understanding of autophagy mediated cellular quality control, is incomplete in plants. Here, we leverage a state-of-the-art proteomics approach to identify novel autophagy players conserved across land plants. Unlike conventional proteomics approaches, we devised a simple, high-throughput approach to identify new cargo receptors and validated them using in vivo and in vitro approaches. I will present our latest findings on the mode of action of one of these cargo receptors that turned out to be also conserved in humans. Altogether our findings highlight the power of leveraging evolutionary diversity to discover new autophagy pathways across the eukaryotes.

Immune signalling of intracellular plant immune receptors

Farid El Kasmi¹

¹ZMBP. University of Tübingen, Tübingen, Germany

Perception of potential microbial phyto-pathogens happens first at the plasma membrane by cell-surface pattern-recognition receptors (PRRs) that initiate a wide range of immune responses. Adapted pathogens, however, have evolved effector proteins to dampen this PRR-triggered immunity. In resistant plants these effectors are recognized by intracellular nucleotide-binding leucine-rich repeat receptors (sensor NLRs) and NLR activation results in the re-initiation of PRR-triggered immune outputs and often in the programmed like cell death of the infected cell. Many sensor NLRs and also some PRRs require the presence of a conserved immune signalling convergence hub formed by the plant-specific lipase like proteins EDS1, PAD4 and SAG101 and a small NLR subfamily – the RNLs – for proper immune activation. Recently we showed that RNLs localize and function at the plasma membrane in a phospholipid-dependent manner. This is in accordance with the findings that RNLs and other cell death inducing NLRs form oligomeric ‘resistosomes’ at the plasma membrane, which function as cation channels. I will summarize our contribution to the current model of NLR function and also give an overview of the recent breakthrough findings and still open questions of the field of plant NLR biology.

Site and Mode of Action Identification and Characterization of Early Herbicidal Leads

PHILIPP JOHNEN¹

¹ BASF SE, Speyerer Straße 2, D-67117 Limburgerhof, Germany

Understanding the site and mode of action of a herbicide is key for its efficient development, the evaluation of its toxicological risk, efficient weed control and resistance management. Therefore, a robust procedure for the identification thereof is key for the efficient development of new crop protection agents.

After lead identification in the green house, we routinely use a combination of physionomics, metabolomics, chemo-proteomics and biochemical assays to exclude unwanted and identify new sites of action (SoAs) of herbicidal compounds. This setup led to the identification of acyl-acyl carrier protein (ACP) thioesterases (FATs) as the SoA of cinmethylin, a pre-emergence herbicide for the control of cool season grasses in cereals (1). Analysis of the downstream effects caused by cinmethylin treatment indicated that the class of FATs is indeed the predominant SoA in planta. Knowledge of FATs as the target of cinmethylin led to the identification of a hidden FAT inhibitor cluster among commercially available herbicides and showed the unique characteristic of cinmethylin in terms of wheat selectivity (2, 3). Furthermore, identification of a diverse set of FAT inhibitors provides a new tool set to study the role FATs in plant growth and development.

1. Campe R, Hollenbach E, Kammerer L, Hendriks J, Höffken HW, Kraus H, et al. A new herbicidal site of action: Cinmethylin binds to acyl-ACP thioesterase and inhibits plant fatty acid biosynthesis. *Pestic Biochem Physiol.* 2018;148:116-25.
2. Johnen P, Zimmermann S, Betz M, Hendriks J, Zimmermann A, Marnet M, et al. Inhibition of acyl-ACP thioesterase as site of action of the commercial herbicides cumyluron, oxaziclomefone, bromobutide, methylmymron and tebutam. *Pest Manag Sci.* 2022.
3. Brabham C, Johnen P, Hendriks J, M. B, Zimmermann A, Gollihue J, et al. Herbicide symptomology and the mechanism of action of methiozolin. *Weed Science.* 2020(16):18–30.

Dynamic signalling systems in plant shoot growth

Yanfei Ma^{1,#} & Jan U. Lohmann¹

¹Department of Stem Cell Biology, Centre for Organismal Studies, Heidelberg University; D-69120 Heidelberg, Germany

Current address: School of Ecology and Environment, Northwestern Polytechnical University; 710129, P.R.China

Abstract

Unlike animals, plants specified by postembryonic growth need to initiate organs continually, which is driven by robust stem cell systems. In the shoot, however, stem cell robustness faces fluctuation from a dynamic signalling of auxin, a potent phytohormone for differentiation. Here, we find that stem cells are not susceptible to auxin triggered differentiation in presence of WUCHEL, a core transcription factor for stem cell identity. WUS repressively binds to most auxin signalling components and response genes in such way as to restrict signalling output. However, a basal level of signalling is still required for stem cell activity. Finally, we demonstrate that to gate auxin output, WUS acts via regulating histone acetylation at target loci. We illuminate a developmental dynamic of how to keep stem cells close to differentiating cells undifferentiated involves a single master regulator mediated transcription network on cell differentiation programme.

Pathway evolution in plant secondary metabolism – pyrrolizidine alkaloids as model system

Dietrich Ober

Chemical Ecology and Molecular Evolution
Botanical Institute and Kiel Botanic Gardens
Kiel University, Kiel, Germany

Pyrrolizidine alkaloids (PAs) are a class of secondary compounds that are produced by plants as chemical defense against herbivores. They are part of fascinating interactions between the plant and specialized insects that developed counter adaptations to these plant toxins. Within the angiosperms, several plant lineages are described to produce PAs. Studies on the first pathway specific enzyme, i.e., homospermidine synthase, have shown that the pathway evolved several times independently during angiosperm evolution. Therefore, this system is a promising tool to study the evolution of pathways in plant secondary metabolism by comparative approaches. Recent results will be presented that on the strategies of gene identification, on models of gene evolution, and on some unexpected observations resulting from protein characterization.

How proteostasis shapes the plant response to environmental stimuli

Paul Gouguet¹, Gautier Langin¹, Jia Xuan Leong¹, Manuel Gonzalez Fuente¹, Shanshuo Zhu¹, Margot Raffeiner¹, SUAYB ÜSTÜN^{1,2}

¹University of Tübingen, Center for Plant Molecular Biology (ZMBP), 72076 Tübingen, Germany

²Faculty of Biology & Biotechnology, Ruhr-University Bochum, 44780 Bochum, Germany

Protein homeostasis is epitomized by a tight equilibrium of protein biosynthesis and degradation; the 'life and death' of proteins. Approximately one-third of newly synthesized proteins are degraded. As such, regulated protein turnover is required to maintain cellular integrity and survival. Autophagy and the ubiquitin-proteasome system (UPS) are the two principal intracellular degradation pathways in eukaryotes. Both degradation pathways orchestrate many cellular processes during plant development and upon environmental stimuli. As such, both pathways play a major role during plant-microbe interactions. We have recently identified that autophagy and the proteasome system are exploited by bacterial pathogens to reprogram host cellular pathways. By studying this intimate interplay, we can utilize plant pathogenic bacteria as tools to understand host cellular degradation machineries and to decipher novel components and functions. In my presentation, I will not only cover our recent work on the role of autophagy and the proteasome in plant-microbe interactions but will report on our attempts to identify new autophagy regulators and new functions of known UPS components. I will highlight different examples and discuss our recent advances.