VIB conference series

International Conference on Polyploidy

11-14 JUNE 2019 | GHENT, BE

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Preface

Yves Van de Peer

The study of polyploidy dates back more than 100 years to the work of biologists such as Hugo de Vries and G. Ledyard Stebbins Jr. It has since then been realized that polyploidy is widespread and commonplace in plants. Although polyploidy is much rarer in animals, there are also numerous cases of currently polyploid insects, fishes, amphibians and reptiles. For a long time, ancient polyploidy events, dating back millions of years, were much less well documented and it was not until the advent of genomic technologies that conclusive evidence of ancient whole genome duplications (WGD) events became available and we now have evidence for tens, or even hundreds, of ancient WGD events. Explanations of the short-term success of polyploids are usually centered on the effects of genomic changes and increased genetic variation, which are mediated by changes in gene expression and epigenetic remodeling. Increased genetic variation, together with the direct cytogenetic consequences of genome doubling, can potentially affect the morphology and physiology of newly formed polyploids and could lead to alterations of ecologically and environmentally suitable conditions. For instance, it has repeatedly been proposed that polyploids have increased environmental robustness than do diploids, potentially leading to evolutionary advantages during periods of environmental turmoil. Moreover, polyploidy has also sometimes been linked with higher diversification rates. Long(er)-term implications of WGD might be evolutionary innovation and increase in biological complexity by the biased retention of regulatory and developmental genes, which, given time, might diversify in function or cause rewiring of gene regulatory networks. Lastly, the impact of polyploidy on the adaptive potential of clonal systems seems to be greater than was initially appreciated. Adaptive effects of polyploidy in prokaryotes might, through increased mutational robustness, contribute to the direct survival of the individual. In many multi-cellular organisms, polyploidy does not generally affect whole organisms but arises, as part of normal development, in the somatic cells of otherwise diploid organisms. Unprogrammed polyploidy also occurs and, for instance, characterizes a substantial proportion of human tumours.

In short, polyploidy, or the duplication of entire genomes, has been observed in prokaryotic and eukaryotic organisms, and in somatic and germ cells. It is clear that the consequences of polyploidization are complex and variable, operate at different evolutionary time-scales and can differ greatly between systems and species. All this, and much more, will be discussed at this sixth International Conference on Polyploidy, where we welcome about 180 participants of more than 26 different countries. We wish you all a very inspiring and fun meeting!

Yves Van de Peer
VIB/GhentUniversity, BE

International Conference on Polyploidy

June 11th – 14th (Ghent, Belgium)

Tuesday June 11[(1)]

Registration and Poster set-up 15:30 -

Welcome and Introduction 16:00-16:10

Plenary session: Jonathan Wendel - Iowa State University, US **16:10-17:00**
The wondrous cycles of polyploidy in plants

Poster set-up & Welcome reception 17:00-19:00

Wednesday June 12

Session 1: Evolutionary and ecological significance of polyploidy **09:00-12:30**
Part 1: Short-term/micro-evolutionary

Douglas Soltis - University of Florida, Florida MNH, Gainesville, USA 09:00-09:30
Is there a single polyploidy paradigm?

Tia-Lynn Ashman - University of Pittsburgh, USA 09:30-10:00
Ecological and evolutionary advantages of polyploidy revealed by strawberries

Coffee break 10:00-10:30

Justin Conover - Iowa State University, US 10:30-10:45
Deleterious mutation accumulation during cotton allopolyploidization, speciation, and domestication

Polina Novikova - VIB-UGent Center for Plant Systems Biology, BE 10:45-11:00
Polyploidy and adaptation in Australian burrowing frogs Neobatrachus

Levi Yant - University of Nottingham, GB 11:00-11:15
Pervasive population genomic consequences of genome duplication in Arabidopsis arenosa

David Sankoff - University of Ottawa, CA 11:15-11:30
Distinguishing successive ancient polyploidy levels based on genome-internal syntenic alignments

Matthias Hartmann - Institute of Botany Czech Acedemy of Science, Pruhonice, CZ 11:30-11:45
When triploids take over: phylogeography of geographical parthenogenesis in Hieracium alpinum

Filip Kolář - Charles University, CZ 11:45-12:00
Adaptation and genotype-environment interaction in polyploid populations: lessons from primary contact zones

Lei Gong - Northeast Normal University, CN 12:00-12:15
Phylogenetic and population structural inference from genomic ancestry maintained in present-day common wheat Chinese landraces

Kentaro Shimizu - University of Zurich, CH 12:15-12:30
Are homeologs redundant? Resequencing of the allotetraploid Arabidopsis kamchatica using subgenome classification approaches

[(1)] Please find the complete abstract book on the USB flash drive in your conference bag.

Lunch 12:30-14:00

Session 2: Evolutionary and ecological significance of polyploidy **14:00-17:30**
Part 2: Long-term/macro-evolutionary

Martin Lysak - CEITEC, Masaryk University, CZ 14:00-14:30
Post-polyploid diploidization and karyotype evolution

Rie Shimizu-Inatsugi - University of Zürich, CH 14:30-15:00
From Genome to Ecology: niche separation in genus Cardamine through polyploidization

Coffee break 15:00-15:30

Michael Barker - University of Arizona, US 15:30-15:45
Impact of polyploidy on the genetic diversity of vascular plants

Gökce Aköz - Gregor Mendel Institute and Vienna Graduate School of Population Genetics, AT 15:45-16:00
The Aquilegia genome reveals a hybrid origin of core eudicots

Rob Denton - University of Minnesota Morris, US 16:00-16:15
The evolutionary history of nuclear genomes trapped in a polyploid salamander lineage

Kira Zadesenets - The Federal Research Center Institute of Cytology and Genetics, Siberian Branch of the Russian Academy of Sciences, RU 16:15-16:30
Genome reorganization after whole genome duplication in evolution of free-living flatworms of the genus Macrostomum

R. Shawn Abrahams - University of Missouri, US 16:30-16:45
Gene Duplication in the Evolution of the Glucosinolate Biosynthesis Pathway

Mathieu Rousseau-Gueutin - INRA, FR 16:45-17:00
Co-evolution of chloroplast and nuclear genomes in flowering plants and impact of allopolyploidy on fine-tuned cytonuclear interactions

Jonathan Spoelhof - University of Florida, US 17:00-17:15
Does reproductive assurance explain the incidence of polyploidy in plants and animals?

Victor Albert - University at Buffalo, Nanyang Technological University, US 17:15-17:30
Deep-time morphological stasis in the carnivorous plant genus Drosera despite different trajectories of genomic upheaval

Poster session & networking reception 17:30-20:00

Thursday June 13

Session 3: Genetics, epigenetics, and physiology of polyploidy **09:00-12:30**

Christian Parisod - University of Bern, CH 09:00-09:30
Integration of genomic and ecological changes in polyploids

Maurine Neiman - University of Iowa, US 09:30-10:00
Does polyploidy influence genomic accumulation of repetitive elements?

Coffee break 10:00-10:30

Mimmi Eriksson - University of Vienna, AT 10:30-10:45
Transposable elements and early stages of polyploid evolution. A perspective from sibling allopolyploid marsh orchids (Dactylorhiza).

Ning Li - Northeast Normal University, CN 10:45-11:00

DNA methylation repatterning accompanying hybridization, whole genome doubling and homoeolog exchange in nascent segmental rice allotetraploids

Clayton Visger - California State University Sacramento,, US 11:00-11:15
Integrating multiple RNA-seq normalizations to assess the impact of autopolyploidy and drought stress on gene expression in Tolmiea

Guanjing Hu - Iowa State University, US 11:15-11:30
Chromatin structure and evolution of duplicate gene expression

Armel Salmon - University of Rennes 1 - UMR CNRS ECOBIO, FR 11:30-11:45
Small-RNAs and PAH-stress tolerance in polyploid Spartina species (Poaceae)

Eric Jenczewski - Institut Jean Pierre Bourgin, INRA, FR 11:45-12:00
Evolution by gene loss: meiotic recombination genes could have their say

Anne-Marie Chèvre - INRA, FR 12:00-12:15
A reshuffling machinery for generating new diversity in a polyploid context: Model Brassica napus

Elvira Hoerandl - University of Goettingen, DE 12:15-12:30
The interplay of polyploidy, epigenetic patterns, niche shifts and apomixis in plants

Lunch 12:30-14:00

Session 3: Genetics, epigenetics, and physiology of polyploidy **14:00-15:00**

Simon Sandve - Norwegian University of Life Sciences, Aas, NO 14:00-14:30
Gene regulatory evolution following whole genome duplication

Rebecca Heald - University of California, USA 14:30-15:00
Investigating the link between genome size and cell size and physiology using amphibian systems

Coffee break & poster session (continued) 15:00-16:00

Boat tour & conference dinner (registration required) 16:15 -

Friday June 14

Session 4: 'Somatic' polyploidy **09:00-11:00**

Zuzana Storchova - Technical University Kaiserslautern, DE 09:00-09:30
Proteome changes upon whole genome doubling

Don Fox - Duke University Medical Center, US 09:30-10:00
Critical roles for somatic polyploidy in organ development and repair

Jinsong Liu - The University of Texas MD Anderson Cancer Center, US 10:00-10:15
Polyploidy and Origin of Human Tumors

Thomas Jungas - CBI CNRS UMR5547, FR 10:15-10:30
Characterization of polyploid neurons in the developing mouse neocortex

Coffee break 10:30-11:00

Session 5: Evolutionary and ecological significance of polyploidy **11:00-12:30**
Short-term/micro-evolutionary (continued)

Sílvia Castro - CFE, University of Coimbra, PT 11:00-11:15
What are the ecological mechanisms involved with the allopatric distribution of a diploid-tetraploid complex?

Anna Klepikova - The Institute for Information Transmission Problems RAS, RU 11:15-11:30
The fate of homeologous genes which control transition to flowering in recent allotetraploid Capsella bursa-pastoris

Ovidiu Paun - University of Vienna, AT 11:30-11:45
Genomic interactions and their eco-physiological implications for adaptation to soil chemistry in allopolyploid marsh orchids

Emma Jane Morgan - Charles University in Prague, CZ 11:45-12:00
Pre- & postzygotic barriers to across-ploidy gene flow in Arabidopsis arenosa: testing the role of spatial scale, genetic divergence & reinforcement

Warren Albertin - ISVV, FR 12:00-12:15
The yeast species B. bruxellensis: a diploid-allotriploid complex showing adaptation to anthropic environments

Reiko Akiyama - University of Zurich, CH 12:15-12:30
Fine-scale empirical data on ecology and transcriptomics reveal niche differentiation of an allopolyploid from diploid parents in Cardamine

Lunch 12:30-14:00

Session 6: Evolutionary experiments, computational models and statistical approaches to study polyploidy **14:00-17:30**

Itay Mayrose - Tel Aviv University, IL 14:00-14:30
Revisiting global biogeographic trends of polyploid plants

Emanuel Gaquerel - CNRS/University of Strasbourg, FR 14:30-15:00
Joining forces: allopolyploidy-mediated innovations in the specialized metabolic networks of Nicotiana species

Coffee break 15:00-15:30

Robin Burns - GMI, AT 15:30-15:45
Exploring interactions between the parental genomes in the young allotetraploid A.suecica

Thomas Abeel - Delft University of Technology, NL 15:45-16:00
Assembly graph cleaning through machine learning improves de novo long-read polyploid assembly

Paul Blischak - University of Arizona, US 16:00-16:15
Demographic inference in autopolyploid species

Kelley Leung - University of Groningen, NL 16:15-16:30
Polyploidy effects in the haplodiploid parasitoid Nasonia

Brittany Sutherland - University of Arizona, US 16:30-16:45
Supervised Learning Estimation of Duplicate Genomes (SLEDGe): A Machine Learning Approach to Characterizing Ancient Whole-Genome Duplications

Arthur Zwaenepoel - VIB-UGent Center for Plant Systems Biology, BE 16:45-17:00
Inference of ancient whole genome duplications and the evolution of the gene duplication and loss rate

Stan Oome - Genoome, NL 17:00-17:15
Taglotyping: Ploidy-independent TAG-SNP based haplotyping using only short reads

Tom Ruttink - ILVO, BE 17:15-17:30
Unraveling hybridization in the genus Phytophthora using phylogenomics and genome size estimation

Poster award and closing 17:30-18:00

Lighting the way with diagnostics

Since 2009, Sysmex Nederland and Sysmex Belgium are part of the international Sysmex organisation, with headquarters in Kobe, Japan. Under the former Goffin Meyvis name, the Benelux organisation has a long history as supplier of high-tech analytical systems for laboratory use. While the Sysmex product line is focussed mainly on haematology and urinalysis, several other reputable manufacturers are represented exclusively by us in the Benelux area. Our product lines meet high standards. Dedication and focus is achieved in a Business Unit structure. We value our partners in business and this is reflected in the long relationships we have with our suppliers. Professionalism is the keyword, technically and commercially. Sysmex is an ISO 9001:2008 certified company. Sysmex Nederland and Sysmex Belgium specialise in the distribution of High-Tech and Medium-Tech analytical equipment. With sales offices in the Netherlands and Belgium and a centrally located warehouse, the Benelux area is be efficiently served. Our markets are:

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Plenary session

The Wondrous Cycles of Polyploidy in Plants

Jonathan F. Wendel - Iowa State University, Ames, USA

One of the signal realizations of the genomics era is that all flowering plants are multiply polyploid. *Gossypium*, the cotton genus, exemplifies this, with both ancient polyploidy and more recent neoallopolyploids that originated following a biological reunion 1-2 MYA of divergent diploids from different hemispheres. This serendipitous merger between diploid genomes generated myriad genomic responses, including gene silencing, intergenomic gene conversion, novel cytonuclear interactions, and complex transcriptomic responses affecting co-expression patterns and variable cis- and trans-controls. Cyclical, recurring polyploidy occurring over time scales ranging from hundreds to millions of years sets in motion processes that lead to genome downsizing, genomic fractionation, and chromosomal diploidization. This polyploidy-induced dynamism is episodically and variably reiterated throughout the angiosperms. A major challenge is to connect these processes to the adaptive role of polyploidy and its importance to the generation of biodiversity and to agriculture.

Invited Speakers

Genome doubling: where do we go from here?

Invited speaker | Douglas Soltis, University of Florida, US

The past 30 years have seen enormous progress in our understanding of polyploidy (genome doubling), yielding many novel insights into the genetic, genomic, physiological, phenotypic and ecological consequences of this evolutionarily important process. Biologists have also added more naturally occurring models to the tool kit of available systems for study. Numerous advances in the study of polyploidy set the stage for the next generation of investigations, posing the question what next? Much remains to be done in the brave new world that awaits the intrepid researcher. With input from others, I propose several research avenues, including: Are there lineage dependent sets of rules to polyploidy? What is the status of gene balance and does this extend to gene networks? Do stressful environments increase the likelihood of polyploid establishment? What are the consequences of polyploidy for the soil microbiome? Do soil features (e.g., available nitrogen, phosphorus) contribute to the distribution of successful polyploids on a global basis? How widespread are chromosome substitution phenomena? What does genome doubling per se do? How do the genetic/genomic consequences of polyploidy extend to the phenotype? And can we extend from genomic to ecological consequences? Few polypoid systems have been investigated from genetics to physiology to morphology and ecology. A major community goal over the next 10 yr should be to fill these gaps from genes to ecology, providing a suite of well-studied model polyploids. Before meaningful synthesis is possible, more comprehensive data sets are needed–organismal systems that include comparable genetic, genomic, chromosomal, proteomic, morphological, physiological, and ecological data.

Ecological and evolutionary advantages of polyploidy revealed by strawberries

Invited speaker | Tia-Lynn Ashman, University of Pittsburgh, US

Polyploidy, or whole genome duplication often with hybridization, is common in eukaryotes and is thought to drive ecological and evolutionary success especially in plants. The mechanisms of polyploid success in ecologically relevant contexts, however, remain largely unknown. Here I discuss research on wild strawberry (*Fragaria*) that assessed functional trait divergence and plasticity as drivers of polyploid fitness advantage. First, we conducted an extensive test using worldwide genotype collection of six allopolyploid (ranging from 6x-10x) and five diploid (2x) taxa in three climatically different common gardens, and second, we created synthetic, autotetraploid (4x) plants derived from self-pollinated full sibs of two diploid taxa to examine the direct effects of genome doubling on functional traits and fitness components in the greenhouse. In the field experiment, we detected divergence in functional trait means but not plasticities between polyploids and diploids, moreover we found that across the heterogeneous garden environments polyploids exhibited fitness advantage, conferred by both trait means and adaptive trait plasticities. In the greenhouse, the direct effect of doubling recapitulated patterns of functional trait differences observed in wild polyploid *Fragaria* in the field. And although a reduction in clonal reproduction was observed in response to genome doubling, this effect was strongly genetic family dependent potentially providing variation upon which selection could act. Our findings elucidate essential ecological mechanisms underlying polyploid adaptation to heterogeneous environments, and also indicate that genome doubling alone can generate ecological divergence. Polyploids exhibit a 'jack-and-master' strategy for fitness advantage, and significant genetic variation in the immediate effects of whole genome doubling suggest that independent origins of polyploidy could allow for rapid short-term evolutionary adaptation and fuel genomic diversity of polyploids.

Post-polyploid diploidization and karyotype evolution

Invited speaker | Martin Lysak, Central European Institute of Technology (CEITEC), Masaryk University, CZ

Polyploidy (i.e., whole-genome duplications, WGD) is widespread across land plant phylogenies and particularly frequent in ferns and angiosperms. Such genome duplications spurred the evolution of key innovations associated with diversification in many angiosperm clades and lineages. However, the diversifications were not initiated by genome doubling per se. Rather, differentiation of the primary polyploid populations through a range of processes results in post-polyploid genome diploidization. Structural diploidization gradually reverts the polyploid genome to one functionally diploid-like through chromosomal rearrangements which frequently result in dysploid changes (i.e., reduction of chromosome number). Thus, the extant chromosome number variation in many plant groups, and especially monophyletic taxa with multiple base chromosome numbers (x), frequently results from clade-specific WGDs followed by diploidization. Dysploidies may lead to reproductive isolation among post-polyploid offspring and significantly contribute to speciation and cladogenetic events. The impacts of WGD events and post-polyploid diploidization on diversification and evolution of karyotype structures are particularly well described in a number of crucifer species and clades (the mustard family, Brassicaceae). This work was supported by the Czech Science Foundation (grant no. 19-06632S).

From Genome to Ecology: niche separation in genus *Cardamine* through recurrent polyploidization

Invited speaker | Rie Shimizu-Inatsugi, University of Zürich, CH

Niche shift is an important factor for new polyploid species to avoid ecological conflict with progenitors as well as introgressive hybridization with them, which prevents establishment of the polyploid as a new species. We use genus *Cardamine*, a close relative of genus *Arabidopsis*, to study the niche separation between progenitors and allopolyploid and its molecular basis. By both of ecological census and transcriptomic analysis, we have shown that the most important factor to separate the niche between two diploids (*C. hirsuta* and *C. amara*) and their tetraploid (*C. flexuosa*) is soil moisture (also refer to the talk by Reiko Akiyama, on Day4, at 12:15). The two progenitors favor either of dry or wet habitat as specialists, while the tetraploid can be found in wider range of soil moisture as a generalist. The adaptive radiation as a consequence of recurrent polyploidizations in genus *Cardamine* could be attributed to the two contrasting types of specialists as progenitors.

Eco-genetic additivity of diploids in allopolyploid wild wheats

Invited speaker | Christian Parisod, Institute of Plant sciences, University of Bern, Switzerland, CH

Underpinnings of the distribution of diploids and allopolyploids in space and along ecological gradients are elusive. Allopolyploidy expectedly yields species with divergent ecological niches to escape competition from diploid progenitors, but departure from genetic and ecological additivity remains to be tested. Here, four diploid wild wheats that differentially combined into four allopolyploid species are used to assess the impact of historical and ecological constraints on species ranges. Genetic variation relating diploid progenitors to allopolyploids supports their genetic additivity. Comparative phylogeography and modelling of climatic niches further support ecological additivity of locally adapted diploid progenitors into allopolyploid species that expanded to become widespread. Diploids further occupy only a small fraction of their potential distribution, whereas allopolyploids largely fill suitable range with specific lineages. Genetic and ecological additivity thus promote the expansion of such polyploid species under environmental changes. The apparent paradox between such conservative evolution and the patent diversification of wild wheats under the influence of transposable elements will be discussed.

Does polyploidy influence genomic accumulation of repetitive elements?

Invited speaker | Maurine Neiman, University of Iowa, US

Powerful insights into the consequences and biological importance of polyploidy can come from comparing otherwise similar conspecifics that differ in ploidy level. We use this approach to characterize the distribution and abundance of repetitive genomic elements in diploid, triploid, and tetraploid *Potamopyrgus antipodarum*, a freshwater New Zealand snail. Repetitive genomic regions like transposable elements often make up a substantial fraction of eukaryotic genomes and contribute to a great deal of observed genomic variation. Despite their ubiquity, we still know very little about the evolutionary dynamics of repetitive genomic elements. Polyploidy provides an especially interesting framework in which address this important unanswered biological question because of the largely untested hypotheses suggesting that increased mutation rate and/or mutational shielding in polyploids relative to diploid counterparts should result in a relatively high rate of accumulation of repetitive elements in polyploid genomes. Our analyses reveal that in general, polyploidy is not associated with an increased burden of repetitive genomic elements in *P. antipodarum*. Nevertheless, we find that triploid and tetraploid *P. antipodarum* have 10-100x more tandemly repeated histone and ribosomal (rDNA) units than do diploid snails. Subsequent comparisons of the load of these repetitive elements across and within the three ploidy levels revealed no evidence for differences in accumulation of these linked histones and rDNAs between triploid and tetraploid *P. antipodarum*. This result suggests that asexual reproduction, which characterizes polyploid *P. antipodarum*, rather than polyploidy per se, is likely the main driver of the dramatic proliferation

Gene regulatory evolution following whole genome duplication

Invited speaker | Simen Sandve, Norwegian University of Life Sciences, Aas, NO

The explosive increase in gene redundancy that follows an autopolyploidization event is believed to release selective constraint, which facilitate functional divergence and evolution of novel molecular adaptations. However, the role of selective forces in shaping novel molecular phenotypes following whole genome duplications (WGDs) is not well understood. In this talk I discuss how we have applied comparative transcriptomics to address this knowledge gap from a gene regulatory perspective, using salmonid fish as a model system. A common ancestor of salmonid fish underwent a WGD event (Ss4R) 100-80 million years ago. Present-day salmonid genomes have retained 50-60% of these Ss4R duplicates, and co-expression analyses across tissues reveal widespread tissue regulatory divergence. Integrating additional species, including a non-salmonid outgroup, further show that tissue-regulatory divergence is highly asymmetric between duplicates. To further understand how selection shaped this divergence, we used a phylogenetic approach to model gene expression evolution following Ss4R in liver. Focusing on the subset of Ss4R duplicates retained as 2:1 copy number ratio between salmonids and outgroup species, we indeed found Ss4R to have fueled adaptive shifts of gene expression levels. However, the majority of Ss4R duplicates that displayed such adaptive shifts were found to be 'down tuned'. These genes were enriched for functions related to translation, splicing, and cellular energy homeostasis, and also displayed strong selective constraints at the sequence level. Taken together, our results support a strong genome wide bias of one duplicate evolving under stronger purifying selection pressure, while selective constraints are relaxed for the other copy. For Ss4R duplicates conserved across multiple salmonid species, this asymmetry is not linked to an increase in adaptive gains in expression level. Instead, our data point to gene dosage balance as a particularly strong factor in shaping duplicated salmonid genomes across 80 million years of evolution.

Investigating questions of size scaling and speciation using frogs with different genome sizes

Invited speaker | Rebecca Heald, University of California, US

Determining how size is controlled is a fundamental question in biology that is poorly understood at the organismal, cellular and subcellular levels. The *Xenopus* species, *X. laevis* (an allotetraploid with 36 chromosomes) and *X. tropicalis* (a diploid with 20 chromosomes) differ in size at all three of these levels. We take advantage of cytoplasmic extracts prepared from *Xenopus* eggs to reconstitute nuclear and spindle assembly in vitro and identify cytoplasmic factors that scale the size of subcellular structures. To study mechanisms of cell and organism size scaling, we utilize hybrids produced through fertilization of *X. laevis* eggs with *X. tropicalis* sperm, which are intermediate in size between the parental species. Interestingly, the reverse cross is inviable, as embryos produced when *X. tropicalis* eggs are fertilized by *X. laevis* sperm lose specific paternal chromosomes and die abruptly as blastulae. Together, our studies aim to reveal underlying principles of biological size control, as well as the molecular basis of variation that contributes to genomic instability and evolution.

Proteome changes upon whole genome doubling

Invited speaker | Zuzana Storchova, Technical University Kaiserslautern, DE

Whole genome doubling has occurred frequently during evolution. At the same time, genome wide sequencing documented whole genome doubling in more than 30 % of malignant tumors. Doubling of the genome was shown to increase genomic instability and accelerate evolvability. Additionally, it increases tolerance to chromosome segregation errors as well as to various stresses. In patients with malignant tumors, whole genome doubling is associated with increased metastasing capacity, drug resistance and poor prognosis. What molecular mechanisms underlie these striking phenotypic changes remains poorly understood. Several laboratories have analysed the transcriptome of yeast and mammalian cells after whole genome doubling. While these analyses identified some changes in gene expression, they could not explain the observed phenotypes. We used mass spectrometry of tetraploid budding yeast and human cells and compared them with the respective diploid parental cells. This analysis revealed unexpected proteome changes occurring upon whole genome doubling. The observed differences suggest marked proteome remodelling in cells that underwent whole genome doubling and might help to determine the mechanisms that drive their phenotypic changes.

Critical roles for somatic polyploidy in organ development and repair

Invited speaker | Don Fox, Duke University Medical Center, US

Specific tissues and cell types within diploid organisms contain whole genome duplications. Such cells are referred to as endopolyploid (hereafter: polyploid), and are formed by either truncated cell cycle or cell fusion mechanisms. Despite increasing evidence that polyploid cells contribute to organ development, organ injury repair, and cancer, it remains largely unclear why certain cells or tissues in otherwise diploid organisms are polyploid, as well as how polyploidy may alter fundamental cellular properties. To uncover how polyploidy impacts cell and tissue biology, we established new, accessible tissue models to identify the functional roles of polyploid cells in otherwise diploid organisms. My laboratory showed that construction of an organ- the Drosophila rectal papillae- depends on precise levels of both polyploidy and polyploid cell division. Further, we identified a tissue repair event in the Drosophila pylorus that restores tissue mass and function without a single division. Instead, this repair event involves increases in ploidy that precisely match the number of pre-injury genomes. This latter finding mirrors the use of polyploidy for tissue repair in injured tissues such as the mammalian liver and heart. More recently, we have uncovered a role for polyploidy in protecting tissue integrity under conditions of chronic tissue injury. We also uncovered multiple mechanisms by which polyploid cells side-step the potential negative outcomes associated with error-prone cell divisions. These mechanisms include segregation of broken acentric chromosomes and disassembly of conjoined chromosomes through a novel chromosome separation process. More recently, we have found evidence of cytoplasm sharing between polyploid cells formed by aberrant cell divisions, which we argue is a mechanism to negate the high frequency of genomic instability in polyploid cells. Collectively, our findings impact our understanding of the regulation of polyploid cell divisions in nature and disease.

Revisiting global biogeographic trends of polyploid plants

Invited speaker | Itay Mayrose, School of Plant Sciences and Food Security, Tel Aviv University, Tel Aviv, Israel , IL

Understanding the distributional patterns of polyploids is of key importance in understanding the biological relevance of polyploidy. For over a century, revealing such patterns has been a major goal of polyploidy research, motivating botanists to collect and assemble wide karyological data. Nevertheless, the lack of large comparative datasets has restricted such studies to local floras and to narrow taxonomical scopes, limiting our understanding of the underlying drivers of polyploid plant distribution. In this talk, I will describe the construction of a large phylogenetic database that provides the inferred ploidy level for each plant species having both cytological and sequence data. I will then describe how this database was combined with extensive spatial occurrence data to compute a map portraying the worldwide distribution of polyploid abundance. I will then discuss the inferred underlying drivers affecting the global distribution of polyploid plants and whether these are consistent with previous hypotheses.

Joining forces: allopolyploidy-mediated innovations in the specialized metabolic networks of *Nicotiana* species

Invited speaker | Emanuel Gaquerel, Institute of Plant Molecular Biology (CNRS / University of Strasbourg), , FR

Plants adapt to their environments by diversifying their phenotypes in various ways. This diversification is intimately linked and reflected in plants' fascinating capacity to evolve novel specialized metabolites. It is now clearly recognized that all modern flowering plant genomes derive from processes set in motion by a history of repeated and episodic auto- or allopolyploidy events and that these polyploidy events act as important drivers of phenotypic innovations. However, the contribution of polyploidy events to the diversification of plant specialized metabolism defenses against insects has only rarely been examined. My group investigates specialized metabolism innovations in the genus *Nicotiana*. During this presentation, I will present first data from our study on the relative contributions of the whole genome duplication at the base of the Solanaceae, of *Nicotiana*-specific-duplications and of transposable elements insertion in regulatory regions, in the assembly of the nicotine biosynthetic pathway, a key innovation within the *Nicotiana* genus. Interestingly, half of the *Nicotiana* species are allopolyploids of different ages. Using a multidisciplinary approach combining transcriptomics, metabolomics and chemical ecology methods, we are currently dissecting biochemical pathways leading to "transgressive" defense-related metabolic characters for two well-mapped allopolyploidy cases corresponding to sections Polydiclae and Repandae within the *Nicotiana* genus. Species from the section Repandae derive from a single ancient allopolyploidy event, for which *N. sylvestris* and *N. obtusifolia* are the closest paternal and maternal genomes, respectively. We discovered that Repandae species are unique among all *Nicotiana* species with respect to their capacity to synthesize long chain fatty acid-based N-acyl-nornicotine compounds in their trichomes where they act as superior toxins against insects. These metabolites are central in the arms race between these species and specialized herbivores. Hence, during the second part of my presentations, I will present results of our recent investigations on the contribution of allopolyploidization in the evolution of this novel defense trait.

Selected Speakers

Deleterious Mutation Accumulation During Cotton Allopolyploidization, Speciation, and Domestication

First author | Justin Conover, Iowa State University, US
Co-authors | Daojun Yuan, Iowa State University, US, Corrinne Grover, Iowa State University, US, Joshua Udall, Iowa State University, US, Jonathan Wendel, Iowa State University, US

Following genome duplication, theory predicts that genes face a reduced evolutionary selective pressure due to the genetic redundancy inherent in polyploid genomes. It's predicted that polyploids should harbor more deleterious recessive mutations, as the effect of these mutations can be masked by functional homoeologs and more quickly drift to fixation. While this theory has largely gone untested, the vast amounts of genome sequencing available now presents the opportunity to study the short- and long-term effects of deleterious mutation accumulation following polyploidization. Here, we use genomic resequencing of multiple individuals sampled from two progenitor diploid and seven polyploid Gossypium species (all derived from the same polyploidization event 1-2 million years ago) to understand how the number, genomic distribution, subgenomic distribution, and site frequency spectra of deleterious has changed between diploid and polyploid cotton. We look at how these dynamics change during post-polyploidy speciation to understand if there is an initial influx of deleterious mutations associated with the population bottleneck of polyploid formation; whether multiple polyploid species arising from the same polyploidization event show similar patterns of deleterious mutations accumulation; and whether the domestication of polyploid species has led to an overall increase of deleterious mutations.

Polyploidy and adaptation in Australian burrowing frogs *Neobatrachus*

First author | Polina Yu. Novikova, VIB-UGent Center for Plant Systems Biology, BE
Co-authors | Yves Van de Peer, VIB-UGent Center for Plant Systems Biology, BE

Polyploidy is rare in animals, and most polyploid animals reproduce asexually. Amphibians represent a dramatic vertebrate exception, with multiple independent sexually reproducing polyploid lineages. The Australian burrowing frog genus *Neobatrachus* is comprised of 6 diploid and 3 polyploid species and offers a powerful model animal polyploid system. We generated exome-capture sequence data from 87 individuals representing all 9 species of *Neobatrachus* to investigate population genomic effects of polyploidy on genus-wide demography. We document widespread gene flow between the tetraploids, asymmetric inter-ploidy gene flow directed from sympatric diploids to tetraploids, and current isolation of diploid species from each other. Changes in ecologically suitable areas corresponded to estimates of demographic histories and suggested that diploids may be suffering the early impacts of climate-induced habitat loss, while tetraploids appear to be avoiding this fate. At the same time, polyploids have to adapt their cellular machinery to ensure proper segregation of chromosomes during meiosis. We have assembled a draft genome of *N. pictus* and conducted a first selection scan between the *N. pictus* (2n) and the N. sudellae (4n). Our preliminary results show that selected genes in the tetraploids are enriched for microtubule motor activity function. This suggests modifications of the homologous pairing process during meiosis. Continuing this work we hope to provide the first description of adaptation mechanism(s) to autotetraploidy in animals. Overall, we demonstrate that *Neobatrachus* is an attractive model to study the effects of ploidy on evolution of adaptation in animals.

Pervasive population genomic consequences of genome duplication in *Arabidopsis arenosa*

First author	Levi Yant, University of Nottingham, GB
Co-authors	

Ploidy-variable species allow direct inference of the effects of chromosome copy number on fundamental evolutionary processes. While an abundance of theoretical work suggests polyploidy should leave distinct population genomic signatures, empirical data remains sparse. We sequenced$\sim$ 300 individuals from 39 populations of *Arabidopsis arenosa*, a naturally diploid-autotetraploid species. We find that the impacts of polyploidy on population genomic processes are subtle yet pervasive, such as reduced efficiency of purifying selection, differences in linked selection and rampant gene flow from diploids. Initial masking of deleterious mutations, faster rates of nucleotide substitution and interploidy introgression likely conspire to shape the evolutionary potential of polyploids.

Distinguishing successive ancient polyploidy levels based on genome-internal syntenic alignments

First author	Yue Zhang, University of Ottawa, CA
Presenting author	David Sankoff, University of Ottawa, CA
Co-authors	Chunfang Zheng, University of Ottawa, CA, David Sankoff, University of Ottawa, CA

A basic tool for studying the polyploidization history of a genome, especially in plants, is the distribution of duplicate gene similarities in syntenically aligned regions of a genome. Often there are two or more peaks, each representing a different polyploidization event. These distributions may be generated by means of a discrete time, non-homogeneous branching process, followed by a standard sequence divergence model. While the similarities data allows for inference of fractionation rates and other parameters they usually cannot pin down the ploidy level of each event. For a sequence of two events of unknown ploidy, either tetraploid or hexaploid, we base our analysis on high-similarity triples of genes – triangles. We calculate the probability of the four triangle types with origins in one or the other event, and impose a mutational model so that the distribution resembles the original data. Using a ML transition point in the similarities between the two events as an discriminator for the hypothesized origin of each similarity, we calculate the predicted number of triangles of each hypothesized type for each model combining hexaploidization and/or tetraploidization. This yields a profile of triangle type for each model, which can then be used to assess real genomic data.

When triploids take over: phylogeography of geographical parthenogenesis in *Hieracium alpinum*

First author: M Hartmann, Institute of Botany Czech Acedemy of Science, Pruhonice, CZ
Presenting author: Matthias Hartmann, Institute of Botany Czech Acedemy of Science, Pruhonice, CZ
Co-authors: J Chrtek, Institute of Botany Czech Acedemy of Science, Pruhonice, CZ, T Fér, Department of Botany, Charles University, Prague, CZ, P Choler, University Grenoble Alpes CNRS LECA, FR, P Taberlet, University Grenoble Alpes CNRS LECA, FR, P Mráz, Department of Botany, Charles University, Prague, CZ

The differential geographical distribution often observed between closely related sexuals and asexuals is termed geographical parthenogenesis (GP). The arcto-alpine *Hieracium alpinum* (Asteraceae) encompasses two geographically allopatric cytotypes. Diploid sexuals occur in the South-Eastern Carpathians. Triploids reproduce asexually via seeds (apomixis) and occupy the remaining and much larger part of the species range; a clear-cut example of GP. Assessments of the intraspecific phylogeny in a spatial-temporal context represents a powerful method to understand the evolutionary history of this diploid-triploid plant complex. We applied a combination of several molecular markers (more than 1.000 ortologous nuclear genes, AFLPs and three cpDNA intergenic spacers) to reveal the geographical distributions of genotypes assessed within 100 populations sampled across the species range. The molecular data shows that triploids arose multiple times independently from diploids outside the range of extant diploids, i.e. after auto-polyploidization the newly formed triploids replaced the diploids due to some advantages of the former. We detected a very strong phylogeographical signal, in which many haplotypes were confined to a specific region. However, one asexual haplotype was omnipresent, representing over 95% of the sampled plants in Scandinavia, Scotland, Greenland, and Iceland, over 40% of the plants sampled in the Alps and a few plants sampled in the Western Carpathians. The phylogeographic pattern, which was strongly correlated with cytogeography, helped us to understand (1) the evolution of this diploid-triploid complex, and particularly (2) the origin of triploids and apomixis.

Adaptation and genotype-environment interaction in polyploid populations: lessons from primary contact zones

First author: Filip Kolář, Charles University, CZ
Co-authors: Guillaume Wos, Charles University, CZ, Magdalena Bohutínská, Charles University, CZ, Martin Čertner, Charles University, CZ

In theory, polyploidy has multifarious effects on selection and adaptation. Whole genome duplication (WGD) is a severe mutation directly affecting fitness and leading to later accumulation of genetic load. On the other hand, phenotypic effects of WGD per se, increased number of mutational targets and polysomic masking may bring benefits. Evolution experiments comparing the adaptation process in diploids and polyploids are, however, generally scarce and virtually non-existing in multicellular organisms. Studies of established populations, in contrast, are often hampered by unclear relationships between ploidies or allopolyploidy confounding the effects of WGD and hybridisation. We attempted to overcome these issues by focusing on primary contact zones - "natural laboratories" of polyploidy where diploid populations still co-exist with their genetically close autopolyploid derivatives. Ecological experiments in Knautia serpentinicola showed better performance of locally formed tetraploid cytotype compared to its diploid progenitor, particularly in nutrient-rich soils in the presence of a competitor. In contrast, ploidy had negligible phenotypic effect in *Arabidopsis arenosa* from primary contact zone spanning ∼2000 m elevational gradient in Western Carpathians. This was further corroborated by non-significant difference in fitness response towards stressful alpine environment, variable strength of genotype-environment interaction in transcriptomic data and even reduced number of adaptive candidate variants in genomes of polyploid populations. In summary, our results based on genetically close diploid-autopolyploid systems suggest that inter-ploidy differences may get inflated in systems where divergent ploidies are compared

Phylogenetic and population structural inference from genomic ancestry maintained in present-day common wheat Chinese landraces

First author | Lei Gong, Northeast Normal University, CN
Co-authors | Bao Liu, Northeast Normal University, CN

Hexaploid common wheat is one of the most important food crops worldwide. Common wheat domestication began in the Fertile Crescent of the Near East approximately 10,000 years ago and then spread west into Europe and eastward into East Asia and China. However, the possible spreading route into and within China is still unclear. In this study, we successfully extracted DNA from single ancient wheat seeds and sequenced the whole genome of seven ancient samples from Xiaohe and Gumugou cemeteries in Xinjiang, China. Genomic inference and morphological observation confirmed their identity as hexaploid common wheat grown in prehistoric China at least 3200 years before present (BP). Phylogenetic and admixture analyses with RNA-seq data of modern hexaploid wheat cultivars from both China and Western countries demonstrated a close kinship of the ancient wheat to extant common wheat landraces in southwestern China. The highly similar allelic frequencies in modern landraces of Qinghai-Tibetan plateau with the ancient wheat support the previously suggested southwestern spreading route into the highland China. A subsequent dispersal route from the Qinghai-Tibetan plateau margins to the Yangtze valley was proposed in this study. Furthermore, the common wheat populations grown in the Middle and Lower Yangtze valley wheat zones were also proposed to be established by population admixture with the wheat grown in the Upper Yangtze valley. Our study reports ancient common wheat sequences at a genome-wide scale, providing important information on the origin, dispersal, and genetic improvement under cultivation of present-day wheat landraces grown in China.

Are homeologs redundant? Resequencing of the allotetraploid *Arabidopsis kamchatica* using subgenome classification approaches

First author | Kentaro K. Shimizu, University of Zurich, CH
Co-authors | Tim Paape, University of Zurich, CH, Gwyneth Halstead-Nussloch, University of Zurich, CH, Jun Sese, Humanome Laboratory, JP

Little is known about genome-wide selection in homeologs in the early stages of polyploid speciation. Although homeologs are often considered redundant and most were lost in paleopolyploid lineages, it is not clear whether homeologs are lost neutraly or kept by negative selection against loss-of-function mutations in new polyploids. One major difficulty to study it is to dis-entangle homeologs in RNA-seq or genome-wide polymorphism analysis. We developed bioinformatic workflow for subgenome-classification approaches such as HomeoRoq and EAGLE-RC. We conducted resequencing analysis of distribution-wide 25 accessions of the model allotetraploid *Arabidopsis kamchatica*, which was derived from two diploid species, *A. halleri* and *A. lyrata*. We used Sanger sequencing to verify the accuracy of sorting to subgenomes (0.2% error, 3/1,375 SNPs). By comparing the frequency spectrum of replacement with synonymous mutations, we found genome-wide negative selection in *A. kamchatica*, although it was slightly weaker than in diploid parents. High-impact mutations causing loss of function were rarely fixed in the species. These results suggest that homeologs were not totally redundant in early polyploid evolution and that loss of homeologs was disadvantageous. Pairs of homeologs tends to show different signature of selection, suggesting that homeologs had different evolutionary trajectories. We also found that the proportion of adaptive substitution was positive in *A. kamchatica*, in contrast to most diploid species studied so far. This suggests that the existence of homeologs could enhance neofunctionalization. Future goals are to identify the general patterns of genome-wide selection in diverse polyploid species.

Impact of polyploidy on the genetic diversity of vascular plants

First author | Michael Barker, University of Arizona, US
Co-authors |

Polyploidization, or whole genome duplication (WGD), is hypothesized to significantly impact variation and selection. Genome doubling ise xpected to reduce the efficacy of natural selection as increased numbers of alleles mask substitutions more severely than in diploid populations. This may lead to an increase in genetic variation in polyploid populations. In contrast, polyploid speciation itself may often involve substantial bottlenecks that could reduce genetic diversity. These and other genetic changes associated with WGD fuel many hypotheses about the evolutionary impacts of polyploidy. Here, I present new analyses of substitution rates and genetic diversity of diploid and polyploid plants to explore these hypotheses. Using a dataset representing more than 54 families and 32 orders of vascular plants, we observed a similar level of genetic variation in polyploids and diploids. Polyploid species had significantly higher substitution rates than their diploid relatives, consistent with the masking hypothesis. We also evaluated the influence of paleopolyploidy on genetic variation in diploids. We found a significant negative correlation between the age of their most recent ancient WGD and the expected heterozygosity of each species. The time since an ancient WGD explained approximately 10% of the difference in genetic diversity among species. These results suggest that whole genome duplication casts a "long shadow" lasting millions of years, with profound, long-lasting effects on the evolution of vascular plant genomes and diversity.

The *Aquilegia* genome reveals a hybrid origin of core eudicots

First author | Gökçe Aköz, Gregor Mendel Institute and Vienna Graduate School of Population Genetics, AT
Co-authors | Magnus Nordborg, Gregor Mendel Institute, AT

Whole-genome duplications (WGD) have dominated the evolutionary history of plants. One consequence of WGD is a dramatic restructuring of the genome as it undergoes diploidization, a process under which deletions and rearrangements of various sizes scramble the genetic material, leading to a repacking of the genome and eventual return to diploidy. Here, we investigated the history of WGD in the columbine genus *Aquilegia*. The tetraploidy in this basal eudicot was previously reported to be independent of the ancient gamma hexaploidy found in all core eudicots. However, our novel analyses of synteny between the two well-preserved genomes reveal that the columbine genome shares diploidization driven rearrangements with the grape genome, suggesting that they have descended from a common tetraploid ancestor. Thus, the triplicate genome structure of core eudicots must be a product of a two-step process: first tetraploidy in the ancestry of all eudicots, then hexaploidy in the ancestry of core eudicots. We further argue that the latter involved allopolyploidization, and that core eudicots thus have a hybrid origin. These findings highlight the value of basal eudicots as an outgroup to the core eudicots and will hopefully encourage larger scale analyses to understand what hybridization has meant for core eudicots — a group which comprises more than 70% of all living flowering plants!

The evolutionary history of nuclear genomes trapped in a polyploid salamander lineage

First author	R.D. Denton, University of Minnesota Morris, US
Presenting author	R. D. Denton, University of Minnesota Morris, US
Co-authors	A.E. Morales, Ohio State University, US, H.L. Gibbs, Ohio State University, US

Quantifying genetic introgression between sexual species and polyploid lineages traditionally thought to be asexual is an important step in understanding what factors drive the longevity of putatively asexual groups. However, the presence of multiple distinct subgenomes within a single lineage provides a significant logistical challenge to evaluating the origin of genetic variation in polyploids. Here, we measure the extent and tempo of introgression over the evolutionary history of an allopolyploid lineage of all-female salamanders (genus *Ambystoma*) and two ancestral sexual species. We collected variation from more than a thousand ultraconserved element loci using a reference mapping method and inferred subgenome specific patterns of variation in the all-female lineage by gauging support for sets of historical models that reflected different patterns of introgression and divergence. Our analyses support a scenario in which the genomes sampled in unisexuals salamanders were present in the lineage ∼3.4 million years ago, followed by an extended period of divergence. Recent secondary introgression has occurred at different times between each sexual species and their representative genomes within the unisexuals during the last 500,000 years. Sustained introgression of sexual genomes into the polyploid unisexual lineage has been the defining characteristic of their reproductive mode, but this study provides the first evidence that unisexual genomes have also undergone long periods of divergence without introgression. The alternating periods of divergence and introgression between polyploid salamanders and their sexual relatives could reveal the scenarios in which the influx of novel genomic material is favored.

Genome reorganization after whole genome duplication in evolution of free-living flatworms of the genus *Macrostomum*

First author: Kira Zadesenets, The Federal Research Center Institute of Cytology and Genetics, Siberian Branch of the Russian Academy of Sciences, RU

Co-authors: Nikita Ershov, The Federal Research Center Institute of Cytology and Genetics, Siberian Branch of the Russian Academy of Sciences, RU, Ilyas Jetybayev, The Federal Research Center Institute of Cytology and Genetics, Siberian Branch of the Russian Academy of Sciences, RU, Eugene Berezikov, European Research Institute for the Biology of Ageing, University of Groningen, University Medical Center Groningen, NL, Lukas Schärer, Evolutionary Biology, Zoological Institute, University of Basel, CH, Nikolay Rubtsov, The Federal Research Center Institute of Cytology and Genetics, Siberian Branch of the Russian Academy of Sciences, RU

Macrostomum is a diverse genus of rhabditophoran flatworms with more than 200 species characterized by global diversity around the world. These species are widespread in fresh, brackish-water and saline basins. The *Macrostomum* species karyotyped to date can be divided into three groups depending on their chromosome number (species with 2n=6, 2n=8-10, and 2n=12 karyotypes). The most interesting finding is the group containing three *Macrostomum* species — namely *M. lignano* (2n=8), *M. janickei* (2n=10), and *M. mirumnovem* (2n=9) — that showed unstable karyotypes. The revealed instability is linked with numerical and structural rearrangements. Two of these species, *M. lignano* and *M. janickei*, have undergone a round of recent WGD in their evolution, while the latter, *M. mirumnovem* resulted from a recent allopolyploidization event. The karyotypes of both *M. lignano* and *M. janickei* contain large chromosome that was resulted from chromosome fusions of one ancestral chromosome set, and therefore encodes presumably one copy of ancestral genome. Moreover, bioinformatics analysis of the *M. lignano* genome assembly revealed the presence of three subgenomes coexisting and presumably co-evolving in the modern genome. NGS data for microdissected DNA probes specific to the *M. lignano* chromosomes detected the presence of chimeric contigs in the existing genome assemblies of *M. lignano* and allowed to differentiate chromosome-specific DNA sequences and explore the early stages of evolution of duplicated DNA in paralogons. These findings open up new possibilities for studying early stages of genome evolution after a recent WGD in animals. This study is supported by the Russian Science Foundation (RSF) under grant 19-14-00211.

Gene Duplication in the Evolution of the Glucosinolate Biosynthesis Pathway

First author: R. Shawn Abrahams, University of Missouri, US

Co-authors: Merijn Kerstens, Wageningen University, NL, Klaas Bouwmeester, Wageningen University, NL, J. Chris Pires, University of Missouri, US, Eric Schranz, Wageningen University, NL

Glucosinolates (Mustard oils) are a class of specialized defense metabolites and a key innovation of the order Brassicales. With approximately 140 known compounds, glucosinolates can exhibit functions beyond direct herbivory defense, including forms of signaling, nutrient transport, and anti-bacterial activity. Innovation in the biosynthesis pathway has shown to be mediated by a series of whole genome duplication events and small scale duplications. However, much of glucosinolate research has been done within an intra-species context without reference to phylogeny or through pairwise species comparisons. This leaves a limited understanding to extent of lineage diversity in the biosynthesis pathway. In this study, we used a phylogenomic synteny network approach, using over 35 plant genomes in the Brassicaceae & Cleomaceae to understand patterns of gene family expansion for the biosynthetic loci. We investigated variation following the Alpha duplication event at the base of the Brassicaceae, subsequent whole genome duplication events, and small scale duplication patterns. Our findings reconstruct dosage mediated patterns of locus duplication and uncover lineage specific shifts in genetic architecture that may underlie trait innovation and convergence.

Co-evolution of chloroplast and nuclear genomes in flowering plants and impact of allopolyploidy on fine-tuned cytonuclear interactions

First author: Mathieu Rousseau-Gueutin, INRA, FR
Co-authors: Julie Ferreira de Carvalho, INRA, FR, Jérémy Lucas, CEA, FR, Gwenaëlle Deniot, INRA, FR, Cyril Falentin, INRA, FR, Héloïse Archambeau, INRA, FR, Marie Gilet, INRA, FR, Fabrice Legeai, INRA, FR, Maryse Lode, INRA, FR, Jérôme Morice, INRA, FR, Gwenn Trotoux, INRA, FR, Emilie Montès, INRA, FR, Jean-Marc Aury, CEA, FR, Valérie Barbe, CEA, FR, Rod Snowdon, Justus Liebig University, DE, Zhesi He, University of York, GB, France Denoeud, CNRS, FR, Patrick Wincker, CEA, FR, Ian Bancroft, University of York, GB, Anne-Marie Chèvre, INRA, FR

Plant eukaryotic cells were formed more than 1 billion years ago through the engulfment of a cyanobacterium, which was then converted into the chloroplast. Since this endosymbiotic event that enabled plants to perform photosynthesis, most chloroplast genes were functionally transferred to the nucleus. To date, several chloroplast protein complexes, including photosystems, are encoded by both nuclear and plastid genes, leading to chloroplast and nuclear genome co-evolution. Allopolyploidy, resulting from the hybridization and genome doubling of two divergent species, may disrupt these fine-tuned cytonuclear interactions as allopolyploids present biparental nuclear chromosomes but only uniparental organelle inheritance. Using the *Brassica* system, we studied the effects of ancient and recent polyploidy events on 102 nuclear genes involved in cytonuclear complexes. In *Brassica* diploid species that have been subjected to a whole genome triplication event (22 million years), we observed that genes involved in such cyto-nuclear complexes are preferentially retained in duplicates, are nearly all transcribed and undergoing strong purifying selection. Furthermore, by performing comparative genomics and transcriptomics (DNA and RNA-Seq) between resynthesized and natural *B. napus* individuals with their diploid parents, we did not identify any homogenization of paternal copies and observed subgenome dominance regardless of the maternal progenitor. We also recently set up a crispr-cas9 experiment to decipher if maternal and paternal nuclear copies have equal abilities to form functional chloroplast protein complexes. These results provide new insights on the impact of hybrid and allopolyploid speciation on cytonuclear interactions.

Does reproductive assurance explain the incidence of polyploidy in plants and animals?

First author: Jonathan Spoelhof, University of Florida, US
Co-authors: Rachel Keeffe, University of Florida, US

The incidence of polyploidy varies greatly among eukaryotes. Most hypotheses that address this variation focus on broad differences between plants and animals, particularly developmental factors that hinder polyploidy in well-studied animal groups. This presentation examines a complementary hypothesis: facultative modes of reproductive assurance greatly increase the probability of polyploid establishment and diversification. Polyploid lineages in plants and animals often express reproductive assurance through asexual reproduction or self-fertilization, and models of polyploid establishment consistently predict that reproductive assurance will mitigate density-dependent mating disadvantages among incipient polyploid populations. However, plant and animal clades that diversified following ancient genome duplications generally retain sexuality and outcrossing mating systems, so facultative forms of reproductive assurance, which do not disrupt the ability to outcross, may facilitate both the establishment eventual diversification of polyploid lineages. To explore this hypothesis, we summarize the availability of eukaryotic ploidy, chromosome counts, and genome size data from publicly available databases and compare them with modes of reproductive assurance that are expressed in major clades. The prevalence of both polyploidy and facultative reproductive assurance are broadly congruent among major plant clades, but data from animal clades that express facultative reproductive assurance are relatively scarce. Data from fungi and microscopic eukaryotes are even rarer. Gathering ploidy data from these groups is essential to understand the incidence of polyploidy and its influence on eukaryotic diversity.

Deep-time morphological stasis in the carnivorous plant genus *Drosera* despite different trajectories of genomic upheaval

First author	Tanya Renner, Penn State University, US
Presenting author	Victor Albert, University at Buffalo, Nanyang Technological University, US
Co-authors	Tanya Renner, Penn State University, US, David Sankoff, University of Ottawa, CA, Chunfang Zheng, University of Ottawa, CA, Qiao Wen Tan, Nanyang Technological University, SG, Kenji Fukushima, University of Wurzburg, DE, Chloe Drummond, Penn State University, US, Tianying Lan, University at Buffalo, US, Stephan Schuster, Nanyang Technological University, SG, Luis Herrera-Estrella, Texas Tech University, US, Enrique Ibarra-Laclette, Institute of Ecology INECOL, MX, Martin Mata-*Rosa*s, INECOL, MX, Sitaram Rajaraman, Nanyang Technological University, SG, Todd Michael, JCVI, US, Victor Albert, University at Buffalo, Nanyang Technological University, US, Jarkko Salojärvi, Nanyang Technological University, US

Recent studies of carnivorous plant genomes have focused on genome-scale issues in the evolution of carnivory, including the influence of polyploidy. The tiny 100 Mb genome of the bladderwort (*Utricularia*) is at least 4:1 relative to grapevine, whereas the large 1.6 Gb genome of the Australian pitcher plant (*Cephalotus*) has not duplicated past the ancient gamma triplication shared with *Vitis*. We used Dovetail Chicago and HiRise Hi-C data to perform chromosomal scaffolding of PacBio draft assemblies for two species from a third independently carnivorous plant lineage, the genus *Drosera*. Published phylogenies place *Drosera regia* sister to the remaining ∼250 species radiation, within which *D. capensis* occupies a derived position. Although *D. regia* is divergent from other *Drosera* in its woody rhizome, undivided styles, and operculate pollen, it shares the same stereotyped flypaper-trap morphology common to all species. Syntenic analyses reveal no post-gamma polyploidies prior to the species split ∼90 million years ago (Mya), based on Ks values for orthologous syntelogs and a calibration of ∼120 Mya for the gamma event. After speciation, a triplication occurred specifically in *D. regia* ∼50 Mya, then an independent *D. capensis*-specific genome duplication ∼30 Mya. The *D. regia* triplication is considerably more fractionated than the *D. capensis* duplication, and *D. regia* retains far fewer conserved syntenic blocks descending from gamma. As such, despite the species' substantial deep-time phenotypic stasis, their gene space has evolved along considerably different gene birth/death trajectories, albeit with the accrual of similar overall functional enrichments, including related to carnivorous enzymology.

Transposable elements and early stages of polyploid evolution. A perspective from sibling allopolyploid marsh orchids (*Dactylorhiza*).

First author	M. C. Eriksson, University of Vienna, AT
Presenting author	Mimmi C. Eriksson, University of Vienna, AT
Co-authors	T Wolfe, University of Vienna, AT, M. K. Brandrud, University of Vienna, AT, J McCann, No affiliation, AT, E Temsch, University of Vienna, AT, M Hedrén, Lund University, SE, O Paun, University of Vienna, AT

The formation of an allopolyploid triggers a genomic shock and a plethora of cascading responses, including epigenetic relaxation and activation of transposable elements (TEs). In this context, TE activity is initially stochastic with regards to TE type, the activated allele and the insert positions, but its phenotypic effects (if any) are immediately subject to natural selection. We study here the role and fate of TEs in a series of six postglacial, sibling allotetraploid marsh orchids (*Dactylorhiza*) that are ecologically distinct. The unidirectional hybridization associated with their origin combined two diploid genomes of different sizes, likely associated with different demographic histories in their history. The two parental species have diverged six million years ago and feature today broadly distinct transcriptomes and TE content. The TE landscape and genome size for the six investigated allopolyploids are largely additive and exhibit only little differences among each other, indicating constraints in TE regulation. We focus in greater detail on two of the sibling allopolyploids, an older *D. majalis* (formed around 2,000 generations ago), and *D. traunsteineri* (ca 1,000 generations old). In stark contrast to the former, the latter could be considered an extremophile confined to low-competition habitats, characterized by depleted soils of both macro and micro-nutrients. Our molecular analyses indicate more extensive compensatory misregulations and TE activity in the younger, poor-soil specialist *D. traunsteineri* than in *D. majalis*. Altogether, our results suggest that genomic conflicts and misregulations continue to sort out over thousands of generations after initial allopolyploidizations.

DNA methylation repatterning accompanying hybridization, whole genome doubling and homoeolog exchange in nascent segmental rice allotetraploids

First author	Ning Li, Northeast Normal University, CN
Co-authors	Chunming Xu, Northeast Normal University, CN, Ai Zhang, Northeast Normal University, CN, Lei Gong, Northeast Normal University, CN, Bao Liu, Northeast Normal University, CN

Allopolyploidization, which entails interspecific hybridization and whole genome duplication (WGD), is associated with emergent genetic and epigenetic instabilities that are thought to contribute to adaptation and evolution. One frequent genomic consequence of nascent allopolyploidization is the homoeologous exchange (HE), which arises from compromised meiotic fidelity and generates genetically and phenotypically variable progenies. In this study, we used a genetically tractable synthetic rice segmental allotetraploid system to interrogate genome-wide DNA methylation and gene expression responses and outcomes to the separate and combined effects of hybridization, WGD and HEs. Progenies of the tetraploid rice were genomically diverse due to genome-wide HEs that affected all chromosomes, yet they exhibited overall methylome stability. Nonetheless, regional variation of cytosine methylation states was widespread in the tetraploids. Transcriptome profiling revealed genome-wide alteration of gene expression, which at least in part associated with changes in DNA methylation. Intriguingly, changes of DNA methylation and gene expression could be decoupled from hybridity and sustained and amplified by HEs. Our results suggest that HEs, a prominent genetic consequence of nascent allopolyploidy, can exacerbate, diversify and perpetuate the effects of allopolyploidization on epigenetic and gene expression variation, and hence may contribute to allopolyploid evolution.

Integrating multiple RNA-seq normalizations to assess the impact of autopolyploidy and drought stress on gene expression in *Tolmiea*

First author	CJ Visger, California State University, Sacramento, US
Presenting author	Clayton J. Visger, California State University Sacramento,, US
Co-authors	PS Soltis, Florida Museum of Natural History, US, DE Soltis, University of Florida, US

We investigated the transcriptomic basis of the ecophysiolgical divergence in drought response between diploid and autopolyploid *Tolmiea*. Using polyethylene glycol treated hydroponic cultures, we subjected *T. diplomenziesii* and *T. menziesii* to negative osmotic potential, inducing extreme drought stress. We then compared gene expression over time in response to our treatment and determined the gene functions most likely to contribute to the physiological differences between *T. diplomenziesii* and *T. menziesii*. Using recently developed methods, we accounted for variation in transcriptome size and cell size/density, enabling our comparisons of gene expression to take place in the context of change per cell, per biomass, and per transcriptome. We found that in response to drought, tetraploid *Tolmiea* exhibits an extreme degree of transcriptome size plasticity, both between individuals and within individual drought responses. Additionally, we found that between the diploid and tetraploid, 9.2% of all loci investigated were differentially responsive to drought. Based on the integration of a functional enrichment analysis and prior physiological investigations, our results suggest that the tetraploids may reduce their photosynthetic machinery in response to drought.

Chromatin structure and evolution of duplicate gene expression

First author	Guanjing Hu, Iowa State University, US
Co-authors	Pei-Yau Lung, Florida State University, US, Xin Sui, Florida State University, US, Daniel L. Vera, Florida State University, US, Corrinne E. Grover, Iowa State University, US, Justin Conover, Iowa State University, US, Sentil Balaji BeeJay, Florida State University, US, Jinfeng Zhang, Florida State University, US, Hank W. Bass, Florida State University, US, Jonathan F. Wendel, Iowa State University, US

Polyploidy is one of the most prominent mechanisms in plant speciation and adaptation. Notwithstanding many remarkable advances in our understanding of polyploid evolution, the mechanistic understandings of duplicated gene regulation remain largely mysterious. Recent studies of chromatin structure suggested that the dynamics of chromatin accessibility plays a pivotal role in orchestrating gene regulation. To explore this phenomenon, we characterized genome-wide nucleosome organization and chromatin accessibility in allopolyploid (AD genome) cotton, *G. hirsutum*, relative to its two diploid parents and their synthetic F1 hybrid, using a technique known as differential nuclease sensitivity mapping (DNS). The degree of promoter chromatin accessibility, as measured by DNS scores, was positively correlated with gene expression levels and also expression differences between homoeologs, providing insight into the well-known phenomenon of homoeolog expression bias in allopolyploids. A general reduction of promoter accessibility was found in the synthetic F1 hybrid compared to its diploid progenitors. More interestingly, this reduction was asymmetric between subgenomes, leading to an unbalanced pattern of homoeolog expression bias towards the more highly expressed D- than A- homoeologs. In the allotetraploid, both subgenomes contained nearly twice as many hypersensitive footprints as their counterparts in the diploids and F1, suggesting that genome doubling and subsequent sequence evolution have introduced more potential cis-regulatory elements into the duplicated gene promoters. Taken together, our study provides a detailed view of the genome-wide cis-regulatory landscapes and how these are altered by genome merger and doubling.

Small-RNAs and PAH-stress tolerance in polyploid *Spartina* species (Poaceae)

First author: A Salmon, University of Rennes 1 - UMR CNRS ECOBIO, FR
Presenting author: Armel Salmon, University of Rennes 1 - UMR CNRS ECOBIO, FR
Co-authors: A Cavé-Radet, University of Rennes 1 - UMR CNRS ECOBIO, FR, D Giraud, University of Rennes 1 - UMR CNRS ECOBIO, FR, J Boutte, University of Rennes 1 - UMR CNRS ECOBIO, FR, O Lima, University of Rennes 1 - UMR CNRS ECOBIO, FR, A El-Amrani, University of Rennes 1 - UMR CNRS ECOBIO, FR, ML Ainouche, University of Rennes 1 - UMR CNRS ECOBIO, FR

Investigating epigenetic mechanisms in non-model species confronted to challenging environments is an important question in evolutionary ecology, but requires the development of adequate resources and methodologies. We present here the approach we developed in salt-marsh species *Spartina* (Poaceae) which are notorious for recurrent hybridization and genome duplication events that resulted in highly successful invasive species. *Spartina* species play an important ecological role in the sedimentary dynamics of coastal saltmarshes as ecosystem engineers, and some exhibit particular tolerance and resilience to chemical pollution (e.g. heavy metals, polycyclic aromatic hydrocarbons PAHs) which make them excellent candidates for phytoremediation. The most complex genome (namely the invasive allo-dodecaploid *Spartina anglica*) arose recently in Europe c.a. 150 years ago, by genome duplication of the homoploid hybrid *S.* × *townsendii* resulting from an interspecific cross between the introduced *S. alterniflora* (2n=6x=62) and the European native *S. maritima* (2n=6x=60). Allopolyploidy was accompanied by significant DNA methylation alteration following hybridization, mostly in regions flanking transposable elements, and gene expression evolution. We explored the genomes and transcriptomes of these species, and developed bioinformatic approaches and tools for detecting the different putative orthologous copies originating from the parents (duplicated homoeologs) in *S. anglica*. We also evaluated the repetitive compartment, which allowed us to identify small RNAs (siRNAs and miRNAs) involved in transcriptomic and epigenomic responses to PAH-induced stress in parental species, the hybrid and the highly tolerant allopolyploid.

Evolution by gene loss: meiotic recombination genes could have their say

First author: Adrián Gonzalo, Institut Jean Pierre Bourgin, INRA, FR
Presenting author: Eric Jenczewski, Institut Jean Pierre Bourgin, INRA, FR
Co-authors: Marie-Odile Lucas, INRA Rennes, FR, Catherine Charpentier, Institut Jean Pierre Bourgin, INRA, FR, Greta Sandmann, Institut Jean Pierre Bourgin, INRA, FR, Andrew LLoyd, : Institute of Biological, Environmental, and Rural Sciences, Aberystwyth University, GB

Many eukaryotes descend from polyploid ancestors that experienced massive duplicate gene loss. This genomic erosion is particularly strong for duplicated (meiotic) recombination genes that return to a single copy more rapidly than genome average following polyploidy. To better understand the evolutionary forces underlying duplicate loss, we analysed how varying copy numbers of MSH4, an essential meiotic recombination gene, influences crossover formation in allotetraploid *Brassica napus*. We first demonstrate that MSH4 systematically returns to single copy following numerous independent polyploidy events, a pattern that is probably not by chance. We then show that faithful chromosome segregation and crossover frequencies between homologous chromosomes are unchanged with MSH4 duplicate loss; by contrast, crossovers between homoeologous chromosomes (which result in genomic rearrangements) decrease with reductions in MSH4 copy number. These results lead to considering a simple and general route for meiotic adaptations in allopolyploids: i.e. through the loss of a key meiotic recombination gene. Our study opens new directions to better appreciate the evolutionary role of gene loss in the context of polyploidy, where this phenomenon is pervasive.

A reshuffling machinery for generating new diversity in a polyploid context: Model *Brassica napus*

First author	A PELE, INRA, FR
Presenting author	Anne-Marie CHEVRE, INRA, FR
Co-authors	F BOIDEAU, INRA, FR, S PAILLARD, INRA, FR, M TABUREL, INRA, FR, A LAPERCHE, Agrocampus Ouest, FR, G TROTOUX, INRA, FR, C FALENTIN, INRA, FR, M-M Gilet, INRA, FR, F LEGEAI, INRA, FR, J MORICE, INRA, FR, M ROUSSEAU-GUEUTIN, INRA, FR

Recombination is main mechanism for genetic reshuffling and it has been shown that increase of ploidy leads to an increase of crossovers (COs). However, it remains strictly controlled with one obligate CO per pair of homologous chromosomes and rarely more than three. Additionally, these COs are non-randomly distributed, as centromeric and pericentromeric regions are deprived of COs. We recently showed that is possible to break down this tight meiotic control. In an AAC allotriploid hybrid obtained by crosses between the allotetraploid crop oilseed rape (*Brassica napus*, AACC, 2n=38) and one of its progenitors (B. rapa, 2n=20), the frequency of COs is increased by more than 3-fold with modification of the recombination landscape especially in pericentromeric regions compared to AA diploid hybrids. Preliminary results indicate a similar recombination pattern in ACC allotriploids produced by crossing *B. napus* with its other diploid progenitor (*B. napus*, CC, 2n=18). We decided to take advantage of these modified recombination rules in *Brassica* allotriploids to deeply increase the genetic diversity of *B. napus* that has been severely eroded by human selection. By crossing a *B. napus* variety with individuals from B. rapa (10 pop) and *B. napus* (9 pop) core collections, we produced 1,178 AAC and 357 CCA F1 hybrids, and their following generations by backcrosses. As 1 to 5% of the F1 progenies were expected to be allotetraploids such as *B. napus*, we screened more than 32,000 plants to identify these tetraploid plants. We then intercrossed these hybrids and genotyped 1,600 plants using a 15K SNP array, revealing that novel genetic diversity from diploid progenitors were introgressed all along the chromosomes in *B. napus*.

The interplay of polyploidy, epigenetic patterns, niche shifts and apomixis in plants

First author	CCF Schinkel, University of Goettingen, DE
Presenting author	Elvira Hörandl, University of Goettingen, DE
Co-authors	E Syngelaki, University of Goettingen, DE, S Klatt, University of Goettingen, DE, S Dullinger, University of Vienna, AT

Apomixis, the asexual reproduction via seed, occurs almost exclusively in polyploid plants. Apomictic plants tend to occur in colder climates and at higher altitudes/latitudes, and occupy much larger distribution areas than their sexual relative (geographical parthenogenesis). However, genetic and epigenetic mechanisms behind these phenomena are still enigmatic. Here we present our studies on epigenetic patterns and geographical parthenogenesis in diploid and tetraploid populations of the alpine plant *Ranunculus kuepferi* based on methylation-sensitive AFLPs (MSAPs), environmental data and experimental work. MSAP patterns differed significantly between cytotypes, but also between modes of reproduction. In combined groups, striking differences in MSAP patterns occurred between mixed and obligate asexual mode of reproduction. Correlations to climatic variables suggest that MSAP patterns correlate to niche shifts to colder climates and higher altitudes of tetraploids. Experiments in climate growth chambers confirm that cold treatments both influence directly mode of reproduction and methylation patterns. We develop an evolutionary scenario for geographical parthenogenesis: during postglacial migration into colder areas, diploid *R. kuepferi* conducted shifts to apomixis and underwent polyploidization; the newly formed tetraploid cytotype changed its epigenetic profile in adaptation to colder climates. Apomixis stabilizes epigenetic patterns by avoiding meiotic resetting, hence creating a self-perpetuating system that established apomictic reproduction. The combination of apomixis and niche shifts helped tetraploids to colonize faster the higher parts of the Alps.

Polyploidy and Origin of Human Tumors

First author	Jinsong Liu, The University of Texas MD Anderson Cancer Center, US
Co-authors	

Polyploid giant cancer cells (PGCCs) have long been observed in cancer and were thought originally to be nondividing. Surprisingly, the formation of a blastomere by cleavage division is the first step in embryogenesis, after the formation of the zygote also shows abundant polyploidy. The evidence from our laboratories and others demonstrated that the stress-induced PGCCs can divide by endoreplication (endocycle and endomitosis), which lead to dedifferentiation of somatic cells and acquisition of embryonic stemness. Therefore, formation of PGCCs in somatic cells may represent a previously overlooked endogenous embryonic program that can be activated to dedifferentiate somatic cells into stem cells of various potencies for tumor initiation. Based on these data, here I propose that human tumors originate from a stem cell at a specific developmental hierarchy, which can be achieved by dualistic origin: dedifferentiation of the zygote (sexual) via the blastomere-mediated cleavage division during normal development, or transformation from damaged or aged mature somatic cells via a blastomere-like embryonic program (asexual) via formation of polyploidy of somatic cells. Initiation of the tumor begins with a stem cell that has uncoupled the differentiation from the proliferation program which results in stem cell maturation arrest. Thus, the birth of a tumor can be viewed as a triad that originates from a stem cell via dedifferentiation through a blastomere or blastomere-like program, which then differentiates along along Waddington's landscape, and arrests at a developmental hierarchy. The significance of polyploid blastomere-like cancer stem cells in cancer therapy will be discussed.

Characterization of polyploid neurons in the developing mouse neocortex

First author	Thomas Jungas, CBI CNRS UMR5547, FR
Presenting author	Alice Davy, CBI CNRS UMR5547, FR
Co-authors	Alice Davy, CBI CNRS UMR5547, FR

The textbook view of mammalian brain is that it is composed only of diploid cells. Yet, a few studies have evoked the presence of aneuploid/polyploid neurons in the adult brain. In humans, it has been described that polyploid neurons are enriched at preclinical stages of Alzheimer Disease and are preferentially targeted by cell death as the disease progresses. These studies have revealed that a small fraction of polyploid neurons can also be detected in control tissues, suggesting that neuronal polyploidy is a physiological process. In the rat, polyploid neurons have been detected in Layer V of the neocortex. To characterize this population of physiological polyploid neurons, we are using the mouse neocortex as a model system. We performed cytometry and FISH analyses at various embryonic, perinatal and postnatal stages to determine the window of production of these neurons. Our data show that the population of polyploid neurons appears at the earliest stages of corticogenesis, that it rapidly increases while neurogenesis proceeds, and finally dramatically drops just after birth concomitantly with the end of neurogenesis. At the adult stage this polyploid population remains detectable but rare. We are currently refining our description of polyploid neurons, coupling DNA content analysis with neuronal identity markers and cell sorting. We are currently addressing their mode of production, testing two hypotheses: 1) neural progenitors' cytokinesis failure and 2) endoreplication. In conclusion, we hypothesize that – as was demonstrated in plants or in the few polyploid mammalian tissues – polyploidy could confer modified or specific functions to neurons and we will test this possibility in the future.

What are the ecological mechanisms involved with the allopatric distribution of a diploid-tetraploid complex?

First author	S Castro, CFE, Centre for Functional Ecology and Department of Life Sciences, University of Coimbra, PT
Presenting author	Silvia Castro, CFE, University of Coimbra, PT
Co-authors	M Castro, CFE, Centre for Functional Ecology and Department of Life Sciences, University of Coimbra, PT, A Figueiredo, Departamento de Geografia, Faculdade de Letras, Universidade de Coimbra, PT, H Castro, CFE, Centre for Functional Ecology and Department of Life Sciences, University of Coimbra, PT, C Siopa, CFE, Centre for Functional Ecology and Department of Life Sciences, University of Coimbra, PT, C Dias, CFE, Centre for Functional Ecology and Department of Life Sciences, University of Coimbra, PT, M Serrano, Laboratorio de Botánica, Facultade de Farmacia, Universidade de Santiago, ES, BC Husband, Department of Integrative Biology, University of Guelph, CA, J Loureiro, CFE, Centre for Functional Ecology and Department of Life Sciences, University of Coimbra, PT

Polyploidy is a widespread phenomenon in the evolutionary history of flowering plants. Since it can result in immediate reproductive isolation from its diploid progenitor, polyploidy has been proposed as a mechanism of rapid sympatric speciation. Under random mating, the establishment of new polyploid cytotypes is limited by positive frequency-dependent selection due to inter-cytotype mating, which may be overcome by ecological/reproductive circumstances that enhance within-cytotype mating. One such mechanism promoting polyploid persistence is ecological differentiation. Previous research has found ploidy-mediated effects on morphology, breeding system and ecological tolerances; however, evidence is limited to a few well-studied species and, the evolutionary consequences of these differences remains largely unexplored. Here, our goal was to assess the contribution of polyploidy to ecological divergence using the mixed ploidy (diploid and tetraploid) species, *Jasione maritima*. First, we explored the distribution of cytotypes throughout the species range and the role of environmental variables in shaping their distribution. Second, we tested for niche differentiation using reciprocal transplants and measured differential competitive ability under controlled conditions using diploids, synthetized tetraploids and established tetraploids. Here we present the results obtained to-date on the immediate effects of genome duplication in this polyploid complex and discuss the ecological processes that drive plant speciation via polyploidization.

The fate of homeologous genes which control transition to flowering in recent allotetraploid *Capsella bursa-pastoris*

First author	AV Klepikova, The Institute for Information Transmission Problems RAS, RU
Presenting author	Anna V. Klepikova, The Institute for Information Transmission Problems RAS, RU
Co-authors	AS Kasianov, The Institute for Information Transmission Problems RAS, RU, MD Logacheva, The Institute for Information Transmission Problems RAS, RU, AA Penin, The Institute for Information Transmission Problems RAS, RU

Polyploidization following by sub- or neofunctionalization along with loss of duplicated genes is one of the mainstream mechanisms of plant genome evolution. A cosmopolitan weed *Capsella bursa-pastoris* (shepherd's purse) is a recently emerged allotetraploid which allows to explore the early events after polyploidization. Using transcriptome map comprising ten organs of European *Capsella* we found 44% of homeologous pairs with significant differences in expression between homeologs. We did not observe so-called "genomic dominance" – a prevalent expression of one of the subgenomes: for a half of differentially expressed homeologous pairs gene from subgenome A was expressed at higher level, and for the second half expression of gene from subgenome B dominated. For further investigation of subgenomes interaction in dynamics we have collected a time series of Middle East *C. bursa-pastoris* shoot apical meristems and leaves during transition to flowering. The differential expression analysis and clustering of homeologs showed 14-19% of homeologous pairs with significant difference in expression. The expression dominance was symmetrical between subgenomes A and B. In gene regulatory network controlling transition to flowering the number of higher expressed homeologs A and B was similar and the prevalently expressed homeologs were not organized in gene cascades. These facts suggest the absence of coordinated expression dominance of subgenomes at the early stages of polyploid evolution.

The research was funded by the Russian Foundation for Basic Research (project no. 18-34-00682) and Ministry of Science and Higher Education RF (0053-2019-0005).

Genomic interactions and their eco-physiological implications for adaptation to soil chemistry in allopolyploid marsh orchids

First author	Thomas Wolfe, University of Vienna, AT
Presenting author	Ovidiu Paun, University of Vienna, AT
Co-authors	Marie K. Brandrud, University of Vienna, AT, Mimmi Eriksson, University of Vienna, AT, Wenjia Gu, Queen Mary University of London, GB, Gert Bachmann, University of Vienna, AT, Andrew Leitch, Queen Mary University of London, GB, Ovidiu Paun, University of Vienna, AT

Early-generation allopolyploids need to quickly accommodate divergent genomes into one nucleus by adjusting organization and function, with ecological consequences. The relative contribution of environmental and intrinsic drivers of polyploid evolution is still not fully clarified, and recurrently formed allopolyploids offer excellent study systems. Here, we show that in *Dactylorhiza* (Orchidaceae) recurrent allopolyploid evolution shapes distinct eco-physiological features in sibling allotetraploids, which match their respective environments. Our system of choice is a pair of widespread, sibling allopolyploids, estimated through ABC inference to be less than 1,000 and, respectively, less than 2,000 generations old. *Dactylorhiza traunsteineri* occupies fens featuring extremely depleted soils in macro- (N-NO3, P, K) and micronutrients. In contrast, its sibling, *D. majalis*, prefers meadows with mesic soils. By using integrative eco-physiological and transcriptomic studies we reveal large-scale, webbed differences between the two polyploids at the level of nutrient transport, leaf elemental chemistry, light harvesting and use, stomata movements, together with photoprotection, which permit occupation of the distinct niches. At the transcriptomic level, we show that recurrent allopolyploid evolution is shaped by partly constrained regulatory interactions, but that stabilization of initial cis-trans compensatory divergence between the diploid parental genomes is still incomplete thousands of generations after allopolyploidization. In the short evolutionary timescale relevant for this system, trans-acting factors appear as key drivers of individual phenotypic traits between independently-originated, sibling allotetraploids.

Pre- & postzygotic barriers to across-ploidy gene flow in *Arabidopsis arenosa*: testing the role of spatial scale, genetic divergence & reinforcement

First author: E J Morgan, Charles University in Prague, CZ
Presenting author: Emma Jane Morgan, Charles University in Prague, CZ
Co-authors: M Lučanová, Charles University in Prague, CZ, M Čertner, Charles University in Prague, CZ, K Kubíková, Charles University in Prague, CZ, F Kolář, University of Innsbruck, AT

Polyploidy, the result of whole genome duplication, is considered a strong reproductive barrier which can lead to population divergence and, eventually, speciation. However, it has been argued that new species should only arise if neo-polyploids can spread to new geographical areas or are able to establish a new ecological niche to avoid competing with parental lineages. Although polyploidy is generally considered a strong reproductive barrier, recent advances have shown that ploidy barrier is often permeable and interploidy gene flow regularly occurs. The diploid-autopolyploid complex of *Arabidopsis arenosa* provides a suitable system for addressing triggers and evolutionary consequences of interploidy gene flow. Several stable ploidy contact zones have been identified across Europe, with a gradient of genetic divergence among diploids and their tetraploid derivatives. We explore the distribution of ploidies across these contact zones, at various spatial scales, as well as the strength of reproductive isolation between them, and the potential for admixture. To investigate the ecological and evolutionary consequences of natural ploidy contact we use a combination of field sampling, ecological niche modelling, flow cytometry and microscopy. We aim to understand the potential for gene flow and the role of triploid block/bridge in this system, namely endosperm development and seed, seedling and adult triploid plant fitness and fertility. Preliminary results indicate a strong yet permeable triploid block barrier, with no evidence of reinforcement.

The yeast species *B. bruxellensis*: a diploid-allotriploid complex showing adaptation to anthropic environments

First author: Warren Albertin, ISVV, FR
Co-authors: Marta Avramova, ISVV, FR, Alice Cibrario, ISVV, FR, Julie Maupeu, Microflora, FR, Amélie Vallet-Courbin, Microflora, FR, Chris Curtin, AWRI, AU, Paul Grbin, University of Adelaide, AU, Anthony Borneman, AWRI, AU, Cristian Varela, AWRI, AU, Joseph Schacherer, University of Strasbourg, FR, Marguerite Dols-Lafargue, ISVV, FR, Isabelle Masneuf-Pomarede, ISVV, FR

The yeast *Brettanomyces bruxellensis* is associated with various fermentation processes including wine, beer, tequila, etc. In some, it is considered as a spoiler (wine) while it can have a positive contribution in others (beer). Previous work showed the existence of diploid (2N) and allotriploid (3N) individuals, yet the significance of the 3N remained elusive. In this work, we collected ¿1400 isolates from 29 countries and 9 substrates. The strains were genotyped using mic*Rosa*tellite markers. 47% of the isolates were diploids, clustered in 2 subpopulations. The remaining 53% strains were putative allotriploids distributed in 4 distinct groups, suggesting the occurrence of independent allotriploidisation events. The genetic diversity of the species was strongly related to substrate origin, with a wine 2N group, a kombucha 2N group, two distinct wine 3N clusters, a tequila/bioethanol 3N group and finally a beer 3N group. The subpopulations were also discriminated by specific traits: 145 isolates were phenotyped in four media containing increasing content in sulfur dioxide, a preservative frequently used in winemaking to limit spoilage. Most subpopulations were sensitive to SO2 treatments, except the two wine 3N groups that showed sulfites tolerance/resistance. Finally, we performed competition assays between a sensitive (2N) strain and a tolerant (3N) one, and we showed that the 2N is fitter in absence of sulfites treatment, while the 3N outcompetes the 2N in presence of sulfites. This work highlights unusual high significance of allotriploids among yeast species and suggests specific adaptation of *B. bruxellensis* to human-related environments.

Fine-scale empirical data on ecology and transcriptomics reveal niche differentiation of an allopolyploid from diploid parents in *Cardamine*

First author: Reiko Akiyama, University of Zurich, CH

Co-authors: Jianqiang Sun, National Agriculture and Food Research Organization, JP, Masaomi Hatakeyama, University of Zurich; Functional Genomics Center Zurich; Swiss Institute of Bioinformatics, CH, Heidi Lischer, University of Zurich; Swiss Institute of Bioinformatics, CH, Roman Briskine, University of Zurich; Functional Genomics Center Zurich, CH, Angela Hay, Max Planck Institute for Plant Breeding Research, DE, Xiangchao Gan, Max Planck Institute for Plant Breeding Research, DE, Miltos Tsiantis, Max Planck Institute for Plant Breeding Research, DE, Hiroshi Kudoh, Kyoto University, JP, Masahiro Kanaoka, Nagoya University, JP, Jun Sese, Humanome Lab, Inc.; AIST; AIST-Tokyo Tech RWBC-OIL, JP, Kentaro Shimizu, University of Zurich; Yokohama City University, CH, Rie Shimizu-Inatsugi, University of Zurich, CH

It has been hypothesized that allopolyploids occupy a niche with intermediate, broader or fluctuating environmental conditions compared with parental diploids. Yet, it remains unclear whether empirical data support this hypothesis, and whether specialization of expression patterns of the homeologs relates to habitat environments. We studied the ecology and transcriptomics of a wild allopolyploid *Cardamine flexuosa* and its diploid parents *C. hirsuta* and *C. amara* in their native range in Switzerland. We analyzed habitat environment data at a fine geographical scale during two seasons to capture temporal fluctuation. The diploid parents favored opposite environmental extremes in terms of soil moisture, soil carbon-to-nitrogen ratios, and light availability. The habitat of the allopolyploid *C. flexuosa* was broader compared with those of its parental species, and overlapped with those of the parents, but not at its extremes. In *C. flexuosa*, the genes related to water availability were over-represented among those that both the expression level and the expression ratio of homeolog pairs varied among habitat environments. Both ecological and transcriptomic data in this study indicated water availability to be the key environmental factor for niche differentiation, and provide empirical evidence for niche differentiation between an allopolyploid and its diploid parents at a fine scale.

Exploring interactions between the parental genomes in the young allotetraploid *A. suecica*

First author: Robin Burns, GMI, AT

Co-authors: Polina Novikova, VIB, BE, Magnus Nordborg, GMI, AT

The young allopolyploid *A. suecica* is a hybrid between *A. arenosa* and the model plant *A. thaliana* that originated in the wake of the last glacial maximum (circa 16kya). The deep divergence of the parent species, natural self-fertilization of the hybrid and the ability to generate synthetic lines, make *A. suecica* an attractive model for research into the biology of polyploidy, particularly pertaining to recent changes in the genome. Here, we present a chromosome-level assembly of natural *A. suecica*, together with DNA, RNA, ChiP and HiC sequencing of the hybrid, parental and synthetic lines. We explore critical questions about how the function and structure of a genome can change in response to a polyploidy event, and how such a response is shaped by evolution. We explore functional changes by analyzing signatures of genome dominance, changes in gene regulation, and the impact of transposable element (TE) insertions on gene expression. We also investigate how the two different but closely related parental genomes of *A. suecica* are organized in a single nucleus and whether homeologous regions interact on a 3D level. Combined with structural analyses on the organization of centromeric and rDNA repeats in the allotetraploid, we provide the first global overview on how the *A. suecica* genome became established in a short period of time, despite the many challenges associated with polyploidy. Furthermore, through the analysis of herbarium samples, we identify closely related *A. arenosa* from the candidate refugia of *A. suecica*, supporting and completing previous inferences on the demographic history of the species

Assembly graph cleaning through machine learning improves de novo long-read polyploid assembly

First author | Ramin Shirali Hossein Zade, Delft University of Technology, NL
Co-authors | Thomas Abeel, Delft University of Technology, NL

Determining accurate genotypes is important for associating phenotypes to genotypes. The DNA sequence of plant crops can be used to link agronomic traits to genome regions which can help to develop improved crop varieties for agriculture purposes. De novo genome assembly is a critical step to determine the complete genotype. The main challenges of de novo genome assembly, particularly for plants, are repetitive DNA sequences and polyploidy within their genome. The introduction of third generation sequencing and long reads has promised to resolve repeat-related problems. While there have been notable improvements, reads originating from these repeats are still introducing assembly mistakes because they introduce false overlaps in the assembly graph. This work focuses on detecting repeat induced overlaps and improving performance of existing de novo assembly methods. Removing repeat induced overlaps leads to a cleaner graph in de novo assembly process and produces higher quality assemblies. For this purpose, a machine learning classifier is trained to detect repeat induced overlaps and remove them from the set of overlaps during the assembly process. The method was evaluated on genomes from two species: *S. cerevisiae* and *S. tuberosum*. We show favorable results in assembly quality compared to baseline assembly metrics. Particularly for the more complex *S. tuberosum* genome, the impact is more pronounce.

Demographic inference in autopolyploid species

First author | Paul Blischak, University of Arizona, US
Co-authors | Mike Barker, University of Arizona, US, Ryan Gutenkunst, University of Arizona, US

Understanding the historical forces that have shaped the distribution of genetic variation in contemporary species is a central goal of evolutionary genetics. For diploid species, there exists a wealth of theoretical and empirical work illustrating the interplay of evolutionary factors affecting genome-wide patterns of genetic diversity. Models and tools for inferring past demographic events from resequencing data are also helping to make these inferences accessible to an increasing array of organisms. However, much of this prior work does not consider species experiencing whole genome duplication (WGD) events and the complexities that may follow, including potentially large changes in effective population size or changes in mating systems that result in inbreeding. Here we explore extensions of the Wright-Fisher diffusion applied to autopolyploids to study patterns of genetic variation using the sample frequency spectrum (SFS). In the simplest case, polyploidy leads to a doubling of the number of chromosomes, effectively scaling the rate at which genetic drift removes variation from the population. Using simulations, we model different demographic scenarios for autopolyploids to better understand how drift, inbreeding, and selection affect genome-wide patterns of polymorphism, as well as how we might infer these patterns from the observed SFS. Taken together, our work provides an important extension of diploid models in population genetics that will help to improve evolutionary inference in autopolyploids.

Polyploidy effects in the haplodiploid parasitoid *Nasonia*

First author	K Leung, University of Groningen, NL
Presenting author	Kelley Leung, University of Groningen, NL
Co-authors	L. van de Zande, University of Groningen, NL, L.W. Beukeboom, University of Groningen, NL

In Hymenoptera (wasps, ants, bees, and sawflies), haploid males develop from unfertilized eggs and diploid females from fertilized eggs. Hymenopteran polyploidy is considered highly deleterious, because diploid males are often sterile or inviable in species with the complementary sex determination mode. However, there is a large gap in polyploid knowledge for species where diploid males are fertile, such as *Nasonia*. A Whiting polyploid *Nasonia* line (WPL) has been maintained in the laboratory for ¿70 years. We recently generated a new polyploid line by RNAi knockdown of a the sex determination gene transformer. There are major differences between the old and new lines. The WPL polyploids exhibit high male mate competition ability, low female fecundity, and cell reduction. The polyploids of the transformer knockdown line have low male mate competition ability, high female fecundity and no cell reduction. These results demonstrate variation in polyploid phenotypes that depend on polyploidisation pathway, and possibly involve co-adapted gene complexes. They also represent the possibility of numerous evolutionary trajectories, with some more likely to be deleterious dead ends and others more conducive to downstream polyploid advantage. Expanding on this, additional polyploid lines can be generated by targeting other genes, transformer-2 and womanizer. These have different knockdown phenotypes from transformer, although polyploid effects are currently unknown. Cumulatively, these resources reflect great potential for developing *Nasonia* into a model for studying animal polyploid evolution.

Supervised Learning Estimation of Duplicate Genomes (SLEDGe): A Machine Learning Approach to Characterizing Ancient Whole-Genome Duplications

First author	B Sutherland, University of Arizona, US
Presenting author	Brittany Sutherland, University of Arizona, US
Co-authors	G Tiley, Duke University, US, M Barker, University of Arizona, US

Whole-genome duplication (WGD) is often a cyclic process, with taxa returning to diploidy following WGD. In these diploidized taxa, inferring polyploid ancestry can be difficult. The most common method for inferring ancient WGDs, Ks plots, visualizes synonymous substitution rates across duplicate genes in the genome. A characteristic peak can be found in these distributions, resulting from a burst of gene duplications occurring with WGD. Although conceptually simple, interpretation of these Ks plots can be inexact. Peak height and prominence can be affected by multiple factors and is highly variable. Peaks are often inferred by eye and may be prone to observer bias. Quantitative methods that fit normal distributions to Ks plots frequently identify multiple significant peaks and make it difficult to assess which result from WGD. Despite difficulties in analysis and interpretation, Ks plots may reveal a wealth of information about WGDs, including number and age of WGDs and whether WGD resulted from auto- or allopolyploidy. Here, we present a machine learning approach for the inference and characterization of WGDs. Using simulated gene age distributions reflecting presence or absence of WGD, varying ages of duplication, and different types of duplication, we trained a set of machine learning models for WGD inference. With these models, we have achieved 100% accuracy against an empirical dataset of taxa previously analyzed by syntenic methods, and 94% congruence with classifications made with existing methods for a larger (∼1400 specimen) dataset representing a wide sampling of green plants. Overall, our new machine learning approach provides a robust, repeatable, and objective method to infer ancient WGDs in genomic data.

Inference of ancient whole genome duplications and the evolution of the gene duplication and loss rate

First author	Arthur Zwaenepoel, VIB-UGent Center for Plant Systems Biology, BE
Co-authors	Yves Van de Peer, VIB-UGent Center for Plant Systems Biology

Gene tree - species tree reconciliation methods have been employed for studying ancient whole genome duplication (WGD) events across the eukaryotic tree of life. Most approaches have relied on using maximum likelihood trees and the maximum parsimony reconciliation thereof to count duplication events on specific branches of interest in a reference species tree. Such approaches do not account for uncertainty in the gene tree and reconciliation, or do so only heuristically. The effects of these simplifications on the inference of ancient WGDs are unclear. In particular the effects of variation in gene duplication and loss rates across the species tree have not been considered. Here, we developed a full probabilistic approach for phylogenomic reconciliation based WGD inference, which allows to assess the statistical support for WGD hypotheses from alignments of multi-copy gene families while accounting for both gene tree and reconciliation uncertainty using a method based on the principle of amalgamated likelihood estimation. The model and methods are implemented in a maximum likelihood and Bayesian setting and account for variation of duplication and loss rate across the species tree, using methods inspired by phylogenetic divergence time estimation. We applied our newly developed framework to ancient WGDs in land plants and investigate the effects of duplication and loss rate variation on reconciliation and gene count based assessment of these earlier proposed WGDs.

Taglotyping: Ploidy-independent TAG-SNP based haplotyping using only short reads

First author	Stan Oome, Genoome, NL
Co-authors	Sean Stijven, Datastories, BE

Handling potato genomic data can be quite difficult, as this crop is a highly heterozygous autotetraploid. To face this challenge, we have developed a methodology that requires nothing more than a single haploid reference genome (DM4.03 will suffice for potato) and an Illumina short read collection of a few dozen genotypes (diploid, tetraploid etc. can be mixed in an analysis). Our algorithm will sort each locus to its haplotypes and allele dosage level for every genotype provided, and produces a list of all haplotype specific SNPs that can be directly used for marker development. The output of this algorithm can also be used to place unmapped scaffolds in their correct location, determine exact recombination sites, and approximate pedigrees. We will also be testing if the Taglotypes can serve as a backbone in polyploid phased genome assembly. We see no reason to assume that our approach will not work in any other species.

Unraveling hybridization in the genus *Phytophthora* using phylogenomics and genome size estimation

First author	Kris Van Poucke, ILVO, BE
Presenting author	Tom Ruttink, ILVO, BE
Co-authors	Annelies Haegeman, ILVO, BE, Thomas Goedefroit, ILVO, BE, Fran Focquet, ILVO, BE, Leen Leus, ILVO, BE, Marillia Horta Jung, Mendel University, CZ, Corina Junker, Julius Kühn Institut, DE, Miguel Angel Redondo, SLU, SE, Claude Husson, INRA, FR, Anne Chandelier, CRAW, BE, Slavtcho Slavov, ABI, BG, Arthur De Cock, CBS, NL, Peter Bonants, WUR, NL, Sabine Werres, Julius Kühn Institut, DE, Jonas Oliva Palau, SLU, SE, Benoit Marcais, INRA, FR, Thomas Jung, Mendel University, CZ, Jan Stenlid, SLU, SE, Tom Ruttink, ILVO, BE, Kurt Heungens, ILVO, BE

Hybrid species have previously been identified in at least five of the 12 phylogenetic clades of *Phytophthora*, a genus that comprises many economically and ecologically important plant pathogens. Hybridization potentially can lead to an increased host range and an enhanced virulence and as a consequence can pose a serious threat to agriculture as well as natural ecosystems. Early and correct identification of new hybrids is therefore required but is hampered by the limitations of traditional molecular methods. Identification of hybridization events is also important in the study of phylogenies as positioning of recent hybrids in a phylogenetic tree can be dubious and lead to suboptimal topologies. To improve the identification of hybrids we have combined Genotyping-by-Sequencing (GBS) and genome size estimation on a genus-wide collection of 671 *Phytophthora* genotypes. Analysis based on locus- and allele counts and especially on the combination of species-specific loci and genome size allowed us to confirm and characterize 9 previously described hybrid species and to discover 16 new diploid and polyploid hybrids. Our method was also valuable for species identification, at an unprecedented resolution, and allowed to correct mis-assigned species in the reference collections. We used both a concatenation- and a coalescent-based phylogenomic method to construct a reliable phylogeny using the GBS data of the non-hybrid *Phytophthora* isolates. Hybrid species were subsequently linked to their progenitors in this phylogenetic tree via bi- or trifurcate links. Our study paves the way for relatively low cost but high resolution identification of hybrids and their phylogenetic relations in members of many other phyla.

Poster Presentations

(1) SARSim: A user-friendly simulation tool for sequence reads and SNP array genotyping data in polyploid species

First author	Y Liao, Wageningen University and Research, CN
Co-authors	T Giorgio, Wageningen University and Research, IT

With the development of next-generation sequencing, genetic studies in polyploids have become achievable. Making use of SNP arrays, studies have been done on polyploid species, for example, potato, sweet potato, and chrysanthemum. However, sequencing based genotyping methods may be more flexible. Also, different types of pedigreed or non-pedigreed populations could be used for genetic analysis. In order to develop and evaluate methods and analysis tools, simulation studies are essential. Here we present SARSim, a user-friendly interface which makes use of PedigreeSim (Voorrips and Maliepaard 2012) to generate bi-allelic marker dosage scores. Based on these simulated dosage scores, SNP allele intensities from arrays, sequence read count and read depth from sequence reads could be simulated for different ploidy levels (even in crosses involving different ploidy levels), and different types of chromosomal pairing. This tool was evaluated using real data and genetic situations from potato, alstroemeria, and hexaploid chrysanthemum in SNP array and sequence reads separately.

(2) Near-infrared spectroscopy as a non-destructive technique to detect cytotypes

First author	D C Albach, Carl von Ossietzky-University, DE
Presenting author	D Albach, Carl von Ossietzky-University, DE
Co-authors	

Near-infrared spectroscopy (NIR) is a technique commonly used for non-destructive differentiation of vegetation types in remote sensing or in fodder- and grain quality as a low cost, rapid and yet highly precise and repeatable technique. Spectral reflectance is a proxy for various phytochemical compounds such as phenolics or proteins. NIR sometimes has been used in species identification but not explicitly in the differentiation of cytotypes. However, based on our initial analyses it is possible to differentiate conspecific cytotypes under greenhouse conditions. Here, I explore further the use of NIR to differentiate between cytotypes of various plant species in the wild, in the greenhouse under different stresses and in herbarium specimens with examples from herbaceous (Veronica) and succulent dicots (*Salicornia*), ferns (*Polypodium*) and grasses (*Festuca*). I will present results using NIR to rapidly identify cytotypes in the field by screening populations of hundreds of individuals in the German saltmarsh. Another application is the identification of ploidy information from herbarium specimens, for which flow cytometry is not feasible. Issues to be discussed are the careful calibration, potential species-specific problems such as certain surface features and nutritional and water status of the plants.

(3) Changes in allopolyploid gene expression patterns and phenotypes: Evidence from the yarrow species (*Achillea*, Asteraceae)

First author | Duo Chen, College of life Sciences, Beijing Normal University, CN
Co-authors | Yan-Ping Guo, College of life Sciences, Beijing Normal University, CN

Due to the merging of two parental genomes, allopolyploid species often exhibit dramatic changes at genomic, transcriptomic and phenotypic levels. Allopolyploid species commonly arise from multiple origins. Questions arise: Do different lineages formed recurrently exhibit similar changes in gene expression? May those changes be linked to relavant phenotypic variation? We did transcriptom analyses on two allotetraploid yarrows, *Achillea alpina* and *A. wilsoniana* which origined independently from *A. acuminata* and *A. asiatica*. These species are distinct in leaf morphology: *A. acuminata* has serrate while *A. asiatica* carries 3-pinnatisect leaves, and the allotetraploids show intermediate leaf forms: pinnatilobed (typical of *A. alpina*) to bipinnatisect (typical of A. wilsoniana). We sequenced the transcriptomes of apical shoots and leaves of the tetraploid and diploid parental species. The result showed that both tetraploid species coexpressed more genes with *A. asiatica* which is the maternal progenitor. Regarding the tetraploid gene expression levels, *A. acuminata* homeologs often displayed expression-level dominace although more genes showed homeolog expression bias to *A. asiatica*. We further checked expression patterns of several leaf dissection regulatory genes by qRT-PCR. For most genes tested, the total expression levels in the tetraploids did not show transgression patterns, which may explain the intermediate leaf shapes of the allotetraploids. In sum, the transcriptome changes resulted from hybridization and polyploidization appear to be conserved between the allotetraploid yarrows that share the parental species but derived independently from different progenitor populations.

(4) Cytogeographic pattern of *Urtica dioica* in Europe relates to morphology and habitat preferences

First author | Ludmila Rejlová, Institute of Botany, Czech Academy of Sciences, Průhonice, Czech Republic, CZ
Co-authors | Jindřich Chrtek, Institute of Botany, Czech Academy of Sciences, Průhonice, Czech Republic, CZ, Pavel Trávníček, Institute of Botany, Czech Academy of Sciences, Průhonice, Czech Republic, CZ, Zuzana Chumová, Institute of Botany, Czech Academy of Sciences, Průhonice, Czech Republic, CZ, Magdalena Lučanová, Institute of Botany, Czech Academy of Sciences, Průhonice, Czech Republic, CZ, Petr Vít, Faculty of Environmental Sciences, Czech University of Life Sciences Prague, Prague, Czech Republic, CZ, Tomáš Urfus, Department of Botany, Faculty of Science, Charles University, Prague, Czech Republic,

Although European flora belongs among the best explored, there are still several plant groups which have only been marginally studied. One of the striking examples is the polyploid complex of *U. dioica*, with multiple diploid taxa often found in remote and partly relict geographic ranges, in contrast to 4x individuals with an unknown evolutionary history that occur in a variety of synanthropic habitats. We have evaluated the cytogeographic pattern, morphological variation in relation to ecology and Bioclim data. The tetraploid cytotype strongly prevails (87%) over diploid cytotype (13%; most of currently accepted subspecies are included) in an extensive dataset of 7012 samples from 1317 populations collected across Europe and southwestern Asia. For the first time we detected the rare 3x and 5x ploidy levels in both mature plants and seeds. The ploidy of the endosperm suggests gene flow between the two dominant ploidy levels. The diploid subspecies of *U. dioica* do not differ from each other in the genome size (Cx), while Cx-values of other closely related species significantly differed from those in *U. dioica*. Exactly recorded locations were used to get a grasp of the ecological preferences of major ploidy levels. We applied simple correlation, Bioclim modelling and affinity to human-affected habitats. European populations of diploid individuals prefer lower altitudes, have a closer ecological niche and occur in less human-affected habitats in comparison to the ubiquitous 4x cytotype. Distance-based morphometrics perform well at distinguishing diploid taxa. Morphological variation will be interpreted in the context of results of molecular analyses (in progress). Key words: polyploidy, morphometrics, Bioclim data

(5) Effect of ploidy level on the content of phenolic compounds from leaf, root and stem of Taiwan jewel orchid, a valuable medicinal plant

First author: Hsiao-Hang Chung, Department of Horticulture, National Ilan University, TW
Presenting author: Jen-Tsung Chen, Department of Life Sciences, National University of Kaohsiung, TW
Co-authors: Shu-Kai Shi, Department of Life Sciences, National University of Kaohsiung, TW, Bin Huang, Department of Biomedical Science and Environmental Biology, Kaohsiung Medical University, TW, Jen-Tsung Chen, Department of Life Sciences, National University of Kaohsiung, TW

Taiwan jewel orchid (*Anoectochilus formosanus Hayata*), a native terrestrial orchid and aslo a traditional medicinal plant in Taiwan, has a wide range of pharmaceutical effects on human health such as pain relief, liver protection, anti-inflammatory, treatments of diabetes, fever and nephritis, prevent oxidative stress, cancer and cardiovascular diseases. The previous report showed a significant enhancement on biomass, total flavonoid and gastrodin contents in the tetraploids when compared with the diploids of Taiwan jewel orchid. In this present report, we further evaluate the effect of chromosome doubling on the total content of phenolic compounds which are generally considered as antioxidant with health benefits. The results showed that there was a significant higher total content of phenolic compounds in leaf as well as stem in the tetraploids. Altogether, it is proposed that ploidy level could affect the level of secondary metabolites such as flavonoids, gastrodin as well as phenolic compounds in different organs of Taiwan jewel orchid.

(6) *Triticale* (× *Triticosecale* Wittmack) - a human-made polyploid, which needs to be improved

First author: M. Kwiatek, Poznań University of Life Sciences, PL
Co-authors:

Triticale (× *Triticosecale* Wittmack) is a man-made artificial polyploid which was created by fusing wheat (*Triticum aestivum* L.) and rye (*Secale cereale* L.) genomes in order to consolidate the quality properties of wheat with the specific traits of rye, such as tolerance to biotic and abiotic stresses. The narrow genetic variation of this crop is a result the lack of evolution process and a small number of patental forms used for production of modern cultivars. In this talk I will present the current state of knowledge and directions of future studies of hexaploid triticale concerning chromosome manipulations. I will try to specify the main goals for creating chromosome aberrations in this artificially generated crop, which are referred to as introgression of genes that are responsible for quality traits, biotic stresses resistance, and heterosis. I will discuss the breeding methods, supported by cytomolecular analyses, which are based on development of chromosome aberrations induced by meiotic restitution, chromosome elimination, chromosome fragmentation or random fusion of chromatin fragments into chromosome structures. This kind of chromosome manipulations can be generated through induced cross-hybridizations, which are alternatives to genome editing technologies, associated with the production of genetically modified organisms (GMOs). I will also show the newest modifications and improvements in triticale breeding strategies, which involve recent achievements in cytogenetics and genomics. At the end I will discuss whether the new methods, such as gametocidal factor system or induced homoeologous recombination, can be exploited to accelerate the breeding considering particular end use properties of triticale.

(7) Ph2 gene phenotype scoring in wheat-rye hybrids with terminal deletions of 3D chromosome

First author: R Svačina, Institute of Experimental Botany AS CR, CZ
Co-authors: J Bartoš, Institute of Experimental Botany AS CR, CZ, P Sourdille, INRA Génétique, Diversité, Ecophysiologie des Céréales, FR, H Serra, INRA Génétique, Diversité, Ecophysiologie des Céréales, FR, M Malurová, Institute of Experimental Botany AS CR, CZ, M Karafiátová, Institute of Experimental Botany AS CR, CZ, TR Endo, Faculty of Agriculture, Ryukoku University, JP, J Doležel, Institute of Experimental Botany AS CR, CZ

Wheat (*Triticum aestivum* L.) developed through hybridization of three related species, resulting in coexistence of three highly similar sub-genomes in its nuclei. The development of diploid-like chromosome pairing during meiosis is necessary to allow formation of viable gametes. In wheat, this system is being enforced genetically by means of Ph genes, mainly Ph1 and Ph2. In absence, both Ph1 and Ph2 lead to increased level of homoeologous chromosome associations in metaphase I in hybrids, while the former has higher effect over the latter. Analysis of a mutant ph2a has narrowed down the position of Ph2 gene to a distal 80 Mb of a short arm of chromosome 3D. However, the size of such deletion hampers the identification of candidates and therefore it would be useful to reduce the deletion size. The goal of our project is to reduce the Ph2 gene region through deletion mapping, so that analysis of candidate genes can be performed. We have established a new set of deletion lines for a short arm of chromosome 3D. We utilized the 2C gametocidal chromosome from A. cylindrica to develop the deletion lines after monosomic introduction into wheat cv. 'Chinese Spring'. Through this tool, we are able to induce non-lethal terminal deletions to chromosomes which can be transferred into progeny. The novel deletion lines were characterized using molecular markers and crossed with rye for subsequent observation of ph2 mutant phenotype on a haploid background in meiocytes isolated from young anthers. Recently, 27 various deletion lines in the ph2a deletion area were crossed with rye and analyzed. Through this study, we managed to narrow down the region of Ph2 gene to an area varying from 63–67 Mb to 77–79 Mb, containing 86–133 genes.

(8) Functional consequences of transposable elements on duplicated gene transcription and retention in polyploid *Brassica* species

First author: J Ferreira de Carvalho, IGEPP - INRA, FR
Co-authors: J Boutte[1], L Maillet[1], T Chaussepied[1], S Letort, IRISA - INRIA, FR, J Lucas[1], G Deniot[1], C Falentin[1], M Gilet[1], F Legeai[1], M Lodé[1], J Morice[1], G Trotoux[1], A-M Chèvre[1], M Rousseau-Gueutin[1]; [1]IGEPP - INRA, FR

Allopolyploidy, which results from interspecific hybridization and genome doubling, entails the merger of more or less divergent parental species. In the long-term evolution of an allopolyploid, subgenome dominance is usually observed. The dominant genome is less fractionated, more expressed and exhibit overall higher fitness than the less expressed subgenome. The causes of genome dominance are still not completely unravelled but recent studies hint towards the implication of Transposable Elements (TEs) and epigenetic regulations in controlling gene expression and retention. Specifically, we investigate the subsequent questions: (1) Following allopolyploidy, are the genes retained in duplicates more impacted by TE proximity than genes in single copy, leading to highly variable levels of expression? (2) Are specific gene functions more prone to these epigenetic regulations? (3) Are these regulations immediately disrupted in the young resynthesized allotetraploid *B. napus*? In this study, we used *Brassica* to assess the long-term impact of TE dynamics on fractionation and gene transcription in duplicated genes. First, TEs were annotated using the REPET pipeline on the recently assembled B. rapa ssp. trilocularis genome. We then compared TE proximity and their impact on gene expression (RNA-Seq) in the three fractionated compartments of the paleohexaploid B. rapa. Additionally, dynamics of gene transcription in relation with TE proximity was also investigated in the B. rapa subgenome following interspecific hybridization with *B. napus* (F1), genome doubling (S0) and in polyploids. These results shed light on the repatterning of subgenome dominance following several rounds of polyploidy in *Brassica*.

(9) Modifications of recombination rules by changing the ploidy level: *Brassica* model

First author: Franz Boideau, INRA, FR
Co-authors: Cyril Falentin, INRA, FR, Virginie Huteau, INRA, FR, Gwenn Trotoux, INRA, FR, Maryse Taburel, INRA, FR, Frédérique Eber, INRA, FR, Marie Gilet, INRA, FR, Loeiz Maillet, INRA, FR, Jérôme Morice, INRA, FR, Julien Boutte, INRA, , Olivier Coriton, INRA, FR, Mathieu Rousseau-Gueutin, INRA, FR, Anne-Marie Chèvre, INRA, FR

Meiotic recombination is the main mechanism to generate new genetic diversity at each generation by reshuffling alleles inherited from the parents. However, crossovers (CO) are highly regulated both in frequency and in distribution, with rarely more than three COs per chromosome that are almost exclusively present in subtelomeric regions. In few polyploid plants, such as in *Brassica* or cotton, it has been shown that polyploidy can modify these rules as polyploids present a higher homologous recombination rate compared to their diploid progenitors. However, it is yet to deciphered the impact of the polyploidy level on the distribution of COs along the chromosomes. To that purpose, we produced two *Brassica* F1 hybrids having two different levels of ploidy: one allotetraploid (AACC) and one allotriploid (AAC). These hybrids result from crosses between oilseed rape (*Brassica napus*, AACC, 2n=38) and a synthetic oilseed rape line or one of its progenitors (B. rapa, AA, 2n=20), respectively. As the two hybrids share the same A sub-genome, we could compare the frequency and distribution of COs between A chromosomes, via the creation of genetic maps. These maps were obtained from thousands of SNP markers that we physically anchored on *Brassica* reference genomes in order to evaluate the COs distribution. Despite their lower ploidy level, our preliminary results indicate that allotriploids present a higher recombination rate than allotetraploids. The impact of polyploidy on COs distribution is undergoing. Altogether, this study will provide new ways to rapidly enhance the narrow genetic diversity of a major polyploid crop.

(10) Deciphering species-level phylogenetic relationships in the evolutionary complex genus *Rosa* using an amplicon-sequencing approach

First author: K. Debray, IRHS, Agrocampus-Ouest, INRA, Université d'Angers, SFR 4207 QuaSaV, Beaucouzé, France, FR
Co-authors: J. Marie-Magdelaine[1], J. Clotault[1], T. Thouroude[1], G. Michel[1], A. Bérard[2], M. C. Le Paslier[2], F. Foucher[1], IRHS, Agrocampus-Ouest, INRA, Université d'Angers, SFR 4207 QuaSaV, Beaucouzé, France, FR, V. Malécot, [1]IRHS, Agrocampus-Ouest, INRA, Université d'Angers, SFR 4207 QuaSaV, Beaucouzé, France, FR, [2]INRA, US 1279 EPGV, Université Paris-Saclay, Evry, France, FR

The genus *Rosa* comprises around 150 species well distributed throughout the Northern Hemisphere. Many rose species are polyploid ranging from 3x to 10x. Hybridization and polyploidization are evolutionary processes that played a key role in the spread of the rose species, but were never studied at the genus level. In the past years, several attempts to reconstruct a phylogeny for *Rosa* have been undertaken mostly using plastid sequences despite the fact that their maternal inheritance only reflects part of the evolutionary processes. Recently, several *Rosa* genomes have been released and give an unrecorded access to sequence variations in the genus *Rosa*. By taking advantage of this tremendously increase in genomic data, our objective is to develop a robust phylogenetic hypothesis able to reflect the complex evolutionary patterns that shaped the genus *Rosa*. We first mined all available genomic data from rose species to build a phylogenomic set of amplifiable orthologous single-copy nuclear tags containing sufficient phylogenetic signal at deep, medium, and shallow levels of the phylogenetic tree. Then, we chose an amplicon-sequencing approach to target these tags on a broader sample of ∼120 rose species enabling us to get allele sequences for each individual. After building a backbone species tree consisting of putative diploid progenitors, we tried to graft alleles obtained from hybrids and polyploids. The resulting network best represents evolutionary history of the genus *Rosa* than bifurcating trees. With this knowledge, we hope to study traits evolution and better characterize this gene-pool for breeding, while less than 10 species have contributed to create the thousands presently cultivated roses.

(11) Endless hybridization of couches (*Elymus*, Poaceae): an empirical evidence

First author: Romana Bartošová, Department of Botany, Charles University, Benátská 2, CZ-128 01, Prague, Czech Republic; Institute of Botany, Academy of Sciences of the Czech Republic, Prùhonice 1, CZ-252 43, Czech Republic, CZ

Co-authors: Tomáš Urfus, Department of Botany, Charles University, Benátská 2, CZ-128 01, Prague, Czech Republic, CZ, Václav Mahelka, Institute of Botany, Academy of Sciences of the Czech Republic, Prùhonice 1, CZ-252 43, Czech Republic, CZ, František Krahulec, Institute of Botany, Academy of Sciences of the Czech Republic, Prùhonice 1, CZ-252 43, Czech Republic, CZ

The study is focused on the extent of hybridization and polyploidization in three *Elymus* species (E. hispidus, E. repens and E. caninus) from agronomically important tribe Triticeae. We measured DNA-ploidy level and absolute genome size of 1081 plants from 302 natural populations from Central Europe. While hexaploidy (2n = 6x) prevails in *Elymus hispidus* and *E. repens, E. caninus* is exclusively restricted to tetraploid level (2n = 4x). Chromosome counts of a representative set of 21 plants were used as a calibration for flow cytometric data. Distance-based morphometrics (RDA and PCA) and correlation analysis were done for 367 *Elymus* plants. Introgressive hybridization between *Elymus hispidus* and *E. repens*, unidirectionally shifted towards *E. hispidus*, was indicated by a continual pattern of genome size of hexaploid individuals. We did not find any evidence for heteroploid hybridization involving tetraploid *E. caninus*. However, we detected minority cytotypes within *E. caninus* (hexaploid) as well as *E. repens*-*E. hispidus* group (heptaploid and nonaploid), suggesting the formation of unreduced gametes within several hybridizing populations. Morphometrics mirrored the continual homoploid pattern of absolute genome size (including the unidirectional shift) and a significant correlation of absolute genome size data with morphology was confirmed. Moreover, morphometrics detected additional characters for delimitation of the involved *Elymus* taxa. The extent of revealed introgressive hybridization is noteworthy, given the crossability of *E. hispidus* with *Triticum aestivum* (bread wheat). If this happens naturally, our data significantly contributed to a potential risk of gene-flow from a crop to a troublesome weed.

(12) Evolutionary dynamics of transposable elements and satellite DNA in polyploid *Spartina* species

First author: D. Giraud, UMR CNRS 6553 ECOBIO, University of Rennes 1, FR

Co-authors: A. Cavé-Radet, UMR CNRS 6553 ECOBIO, University of Rennes 1, FR, O. Lima, UMR CNRS 6553 ECOBIO, University of Rennes 1, FR, V. Huteau, INRA IGEPP Molecular Cytogenetics Platform, FR, O. Coriton, INRA IGEPP Molecular Cytogenetics Platform, FR, A. El-Amrani, UMR CNRS 6553 ECOBIO, University of Rennes 1, FR, M. Aïnouche, UMR CNRS 6553 ECOBIO, University of Rennes 1, FR, A. Salmon, UMR CNRS 6553 ECOBIO, University of Rennes 1, FR

Many studies on polyploid species report modifications of epigenetic control following polyploidization inducing a reactivation of transcriptional activity or a transposition burst of transposable elements. This new activity can result in gene expression changes and global genome reorganization. In this context, the aim of our study is to explore the dynamics of repeated sequences in *Spartina* species. This clade (Poaceae, Chloridoideae) is known for its well-documented events of interspecific hybridization and recurrent polyploidy which resulted in species with different ploidy levels (ranging from 4x to 12x). Recently, crosses between two hexaploid species led to the formation of two F1 hybrids and one allododecaploid species. The diversity of repeats was assessed from shotgun genomic sequencing of tetraploid (*S. versicolor*, *S. bakeri* and *S. spartinae*; 2n=4x=40) and hexaploid (*S. maritima*, *S. alterniflora*; 2n=6x=60-62) species, and after the annotation of clustered reads. Results show that genome size variation observed between species with same ploidy level can be explained by a greater amount of TEs in one of the tetraploid species (*S. spartinae*) and a burst of satellite DNA in one hexaploid species (*S. alterniflora*). Several transcriptionally active TE lineages were identified in all studied species, mostly belonging to the Ty1/copia group. The regulation of these elements was confirmed by the identification of homologous small-interfering RNAs.

(13) Origin and evolution of *Camelina* diploids and allopolyploids was accompanied by chromothripsis-like chromosomal rearrangements

First author: Terezie Mandakova, CEITEC, Masaryk University, CZ
Co-authors: Milan Pouch, CEITEC, Masaryk University, CZ, Jordan R. Brock, Washington University in St. Louis, US, Ihsan A. Al-Shehbaz, Missouri Botanical Garden, US, Martin A. Lysak, CEITEC, Masaryk University, CZ

False flax or gold of pleasure (*Camelina sativa*) is an increasingly popular oilseed crop closely related to *Arabidopsis* and canola. Despite the available genome sequence and other genomic resources, the origin and putative parental species of the allohexaploid *C. sativa* (2n = 6x = 40) were shrouded in mystery. Nearly nothing is known about the origin, genome evolution and phylogenetic relationships of the remaining neglected or almost unknown diploid and polyploid *Camelina* species (2n = 12, 16, 26 and 40). By employing state-of-the-art tools we established fine-scale comparative cytogenomic maps for six diploid, tetraploid and hexaploid *Camelina* species and elucidated their phylogenetic relationships. We were able to identify the parental genomes of the three known allopolyploid species (*C. microcarpa, C. rumelica* and *C. sativa*) and reconstruct the sequence of hybridization events. Besides common chromosomal rearrangements, such as translocations and inversions, chromothripsis-like events generated shattered chromosomes in *Camelina* diploids and polyploids. These complex chromosomal alterations, similar to those associated with several human disorders, have not been reported in a non-model plant species as yet. This study was supported by a research grant from the Czech Science Foundation (grant no. 17-13029S).

(14) Parallel adaptation to serpentine challenge in tetraploid *Arabidopsis arenosa*

First author: Veronika Konečná, Department of Botany, Faculty of Science, Charles University, CZ
Co-authors: Anita Bollmann, Department of Cell and Developmental Biology, John Innes Centre, Norwich Research Park, GB, Levi Yant, School of Life Sciences, University of Nottingham, GB, Filip Kolář, Department of Botany, University of Innsbruck, AT

Adaptation represents key evolutionary mechanism, which allows species and populations to succeed in a variable environment. Some of the outstanding adaptations in plants include adaptations to the hostile and toxic serpentine soils. This chemically extreme substrate provides multiple challenges to plant life such as extremely low Ca:Mg ratio and elevated levels of toxic metals. Therefore, serpentine barrens provide a powerful model for studying multi-challenge adaptations. Moreover, the island-like distribution of serpentines can lead to parallel evolution at the level of both genome and phenotype. Tetraploid *Arabidopsis arenosa* is a promising model for studying substrate adaptive evolution in serpentines as it colonized multiple serpentine sites in Central Europe and wide set of genomic and genetic tools developed for the closely related *A. thaliana* is available. By genotyping (whole genome resequencing) and phenotyping (ionome composition) five pairs of serpentine and close non-serpentine populations, we ask if *A. arenosa* serpentine populations are the result of parallel evolution and what is the genetic basis of serpentine adaptation. First results have shown multiple colonization events of serpentine barrens as well as several candidate genes for selection shared among multiple serpentine populations suggesting genomic parallelism. Furthermore, the candidate genes have relevant adaptive functions to serpentine stress such as in dehydration tolerance and ion homeostasis traits. Finally, an ongoing reciprocal transplant experiment suggests overall strong local adaptation to serpentine soils but with a different manifestation of the fitness response across different serpentine – non-serpentine population pairs.

(15) An examination of fitness traits in isogenic diploid, autotetraploid, and allotetraploid *Arabidopsis*

First author | Jonathan Spoelhof, University of Florida, US
Co-authors | Douglas Soltis, University of Florida, US, Pamela Soltis, University of Florida, US

Many factors influence the outcome of whole genome duplication. Hybridization, progenitor genotype, and the mechanism of genome doubling can all have significant impacts on the characteristics and fate of a new polyploid lineage. Extensive germplasm resources in *Arabidopsis* thaliana and *A. suecica* provide an opportunity to isolate these factors and determine their influence on key fitness traits. Germination, survival to flowering, and flowering time were recorded for two separate isogenic series of *Arabidopsis* lines grown in a common-garden experiment. One series comprises four isogenic pairs of diploid and synthetic autotetraploid *A. thaliana*. The other series comprises two semi-isogenic pairs of synthetic autopolyploid *A. thaliana* and *A. suecica* (both crossed to the same *A. arenosa* accession). The effects of ploidy, genotype, and hybridization on fitness traits are presented.

(16) Model adequacy for likelihood models of chromosome-number evolution

First author | A Rice, Tel-Aviv University, IL
Co-authors | I Mayrose, Tel-Aviv University, IL

Chromosome number is an important feature of the eukaryote genome. Describing its evolution within a group of interest is of prime importance when trying to detect polyploidy and dysploidy events. Indeed, inferring taxa as diploids or polyploids is a fundamental step for various downstream comparative analyses. ChromEvol is a probabilistic inference tool that given a phylogeny and respective chromosome counts, infers ancestral chromosome numbers and detects probable shifts in chromosome numbers along each branch of the phylogeny, allowing the categorization of tip taxa into diploids or polyploids. A basic step in this process compares between possible models of chromosome-number change. However, fitting a model does not necessarily mean that the model truly describes the evolutionary pattern of the underlying data. This vulnerability may lead to incorrect conclusions when the assumptions of the model are not met. Using a model adequacy framework as part of the inference process will allow researchers to compare the absolute fit of alternative models and not only their relative one. To date, model adequacy approaches are established for sequence evolution and continuous valued organismal traits. However, these tools cannot be applied to chromosome-number evolution due to the unique nature of this trait. We have thus developed a model adequacy methodology that can be specifically applied for the analysis of chromosome-number evolution. Using this methodology, allows to pinpoint phylogenies whose underlying evolutionary patterns deviate substantially from current modelling assumptions (e.g., due to hybridizations). We will further discuss the circumstances in which such deviations impact the inference of polyploidy events.

(17) Facultative apomictic orchids *Z. mackayi* favor sexual reproduction in a diploid-tetraploid hybrid zone in the Southeastern Brazilian highlands

First author	YA Moura, State University of Campinas, BR
Presenting author	S Koehler, State University of Campinas, BR
Co-authors	

Polyploidy and apomixis are two important and associated processes in plants. Knowledge on genetic diversity patterns is important to understand the role of these processes on plant speciation. Diploid and tetraploid cytotypes of the orchid *Zygopetalum mackayi* have a parapatric distribution in Southeastern Brazilian highlands, meeting in a contact zone where triploids are found. Facultative apomixis occurs in triploids and tetraploids, which produce sexual and apomictic embryos in the same seed. Our goal was to assess the role of facultative apomixis and intermediate cytotype in reproductive isolation between cytotypes of *Z. mackayi*. We described patterns of population genetic diversity and structure based on eight microsatellites of six populations in the contact zone (only sexual diploids, only facultative apomictic tetraploids and mixed-cytotype populations). We recovered similar and high levels of genetic diversity in all populations sampled which suggests sexual reproduction is dominant. Cytotypes emerged in three distinct genetic clusters. Our results suggest a secondary origin of the contact between diploids and tetraploids and the presence of a triploid block resulting in strong reproductive isolation between cytotypes. Apomixis is likely a consequence of polyploidization rather than an advantageous reproductive strategy in plants of *Z. mackayi*.

(18) Polyploidization and genome size variability in alpine plants of the American Cordilleras

First author	Jan Ptáček, Department of Botany, Faculty of Science, Charles University, Benátská 2, 12800, Prague, CZ
Co-authors	Tomáš Urfus, Department of Botany, Faculty of Science, Charles University, Benátská 2, 12800, Prague, CZ, Carolina I. Calviño, Department of Botany, INIBIOMA (CONICET – Universidad del Comahue), Quintral 1250, 8400 Bariloche, Rio Negro, AR, Katya Romoleroux, Pontificia Universidad Católica del Ecuador, Av. 12 de Octubre 1076 y Ramón Roca, 17012184, Quito, EC, Romina Vidal-Russell, Department of Botany, INIBIOMA (CONICET – Universidad del Comahue), Quintral 1250, 8400 Bariloche, Rio Negro, AR, Petr Sklenář, Department of Botany, Faculty of Science, Charles University, Benátská 2, 12800, Prague, CZ

Flow cytometry is a powerful tool for biosystematics and microevolutionary studies of plants. Hitherto published polyploidization and genome size data of European and North American floras reveal frequent occurrence of polyploids in the north temperate region. Virtually nothing is known about the occurrence of polyploids in the equatorial and temperate alpine regions of the Andes. The continuous range of Cordilleras provides unique settings for broad-scale evolutionary studies. We examined the absolute genome size variation and DNA ploidy level using flow cytometry of 1998 individuals and 936 alpine species of the temperate (Argentina) and (sub)tropical Andes (Bolivia, Ecuador) compared to the Rocky Mts. (USA). The highest rate of mixed ploidy species was detected in the Rocky Mts. (5.2%) followed by the temperate Andes of Argentina (2.4%), whereas the tropical zone contains only 1.6% of mixed ploidy taxa. On the other hand, the absolute genome size of alpine eudicots is lowest in the northern temperate zone (mean 4.58 $\pm$ 0.21 pg) and highest in the southern temperate zone (mean 6.86 $\pm$ 0.29 pg). We detected mixed ploidy species in 24 genera and particularly genus Caltha (Ranunculaceae) was identified as a highly appropriate model system for more detailed investigation of alpine polyploidy evolution. To sum up polyploidy doesn't seem to be preferably restricted to alpine zone of the Northern Hemisphere and its major role in plant evolution of the Andes has been so far substantially underestimated. Keywords: Alpine flora, Andes, flow cytometry, latitudinal gradients, polyploids, Rocky Mts.

(19) Restricted recombination leads to expansion and divergence of satellite DNA in allopolyploid dogroses

First author: Jana Lunerova, Intstitute of Biophysics of the Czech Academy of Sciences v.v.i., CS-61265 Brno, Czech Republic, CZ

Co-authors: Veit Herklotz, Melanie Laudien, Marco Groth, Christiane M. Ritz, Department of Botany, Senckenberg Museum of Natural History Görlitz, Am Museum 1, D-02826 Görlitz, Germany, DE, Ales Kovarik, Intstitute of Biophysics of the Czech Academy of Sciences v.v.i., CS-61265 Brno, Czech Republic, CZ

Despite the odd-ploidy (2n = 5x = 35) level dogroses (*Rosa* sect. *Caninae*) are capable for sexual reproduction due to their unique meiosis. During the canina meiosis two sets of chromosomes form bivalents and are transmitted by male and female gametes, whereas the remaining chromosomes form univalents and are exclusively transmitted by the egg cells. The evolution of chromosomes is expected to be driven by the selection pressure on recombining chromosomes (bivalents). To gain insight into a differential chromosome evolution we sequenced repeatoms of four pentaploid, two tetraploid and several diploid species. Fluorescence in situ hybridization was carried out using satellite and ribosomal DNA probes. We isolated and characterised a pericentromeric satellite repeat called CANR4. It has been found in all members of the genus *Rosa* including the basal subgenera *Hulthemia* and *Hesperhodos* (cca 200 mil. yrs). All pentaploid dogroses harboured high genome proportions (1.7–3.2%) and locus numbers (16–20 sites /35 chromosomes) of CANR4. In contrast, diploid (2n=2x=14) and tetraploid (2n=4x=28) roses displayed a variation in genome proportions (0.03–4.3%) and number of loci (5–16 sites per mitotic cell). All polyploids, and both early diverging diploid subgenera amplified two major CANR4 variants, while most diploids including candidate dogrose progenitors contained only a single variant. In dogrose meiosis, univalent chromosomes were enriched in CANR4 repeats compared to bivalents. SNPs and cluster analysis revealed higher intragenomic homogeneity of the satellite in dogrose. We hypothesize that satellite DNA expansion may contribute to divergence of univalent chromosomes in *Rosa* species with non-symmetrical meiosis.

(20) Chromosome-specific oligo painting elucidates large variation in *Eumusa* genome

First author: Denisa Šimoníková, Institute of Experimental Botany, Centre of the Region Haná for Biotechnological and Agricultural Research, Šlechtitelů 31, Olomouc, Czech Republic, CZ

Co-authors: Jaroslav Doležel, Institute of Experimental Botany, Centre of the Region Haná for Biotechnological and Agricultural Research, Šlechtitelů 31, Olom, CZ, Eva Hřibová, Institute of Experimental Botany, Centre of the Region Haná for Biotechnological and Agricultural Research, Šlechtitelů 31, Olom, CZ

The family Musaceae is classified into three genera: *Musa*, *Musella* and *Ensete*. The largest of them, *Musa*, comprises about 70 species, whereas closely related *Musella* (2n=18) and *Ensete* (2n=18) are represented by only a few species. Based on basic chromosome number and plant morphology, the genus *Musa* has been divided into four sections: *Eumusa* (x=11), *Rhodochlamys* (x=11), *Australimusa* (x=10) and *Callimusa* (x=9, 10). This work is focused on the largest section *Eumusa*, which showed surprisingly large variation in genome constitution in various subspecies of diploid *M. acuminata* (A genome), *M. balbisiana* (B genome), *M. schizocarpa* (S genome) and their triploid hybrid clones (AAA, AAB and ABB genomes). A set of 18 accessions representing *Eumusa* section was used for chromosome-specific oligo painting. A reference genome sequence of *M. acuminata* doubled haploid clone 'Pahang' (2n=22) facilitated the identification of chromosome arm-specific oligomers that were designed using Chorus program and synthetized by Arbor Biosciences (Ann Arbor, MI, USA). The oligos were labeled using reverse transcription by biotin- or digoxigenin-tagged reverse primer and used as probes for fluorescence in situ hybridization (Han et al., Genetics 200:771, 2015). This powerful approach allowed identification of all chromosomes within a karyotype and discovering chromosomal rearrangements, which accompanied the evolution of the family Musaceae. Acknowledgement: This work has been supported by the Grant Agency of the Czech Republic (award No. 19-20303S). The computing was supported by the National Grid Infrastructure MetaCentrum (grant No. LM2010005 under the program Projects of Large Infrastructure for Research, Development, and Innovations).

(21) A reference sequence for the highly polyploid and interspecific genome of sugarcane

First author	Olivier Garsmeur, CIRAD, FR
Presenting author	Angélique DHONT, CIRAD, FR
Co-authors	Gaetan Droc, CIRAD, FR, Bernard Potier, SASRI, ZA, Karen Aitken, CSIRO, AU, Marie-Anne Van Sluys, Universidade de Sao Paulo, BR, Blake Simmons, JBEI, US, Edwin van der Vossen, Keygene, NL, Robert Henry, QAAFI, AU, Jeremy Schmutz, JGI, US, Angéelique DHONT, CIRAD, FR

Sugarcane is the major crop for sugar and bioenergy production. Sugarcane cultivars (*Saccharum* spp.) have probably the most complex genome among crops, being highly polyploidy aneuploidy, highly heterozygous, and of interspecific origin (2n $\sim$ 12x $\sim$ 120, $\sim$10Gb). This complexity poses major challenges for producing a reference sequence. We exploited colinearity with sorghum to produce a BAC- based monoploid genome sequence of sugarcane. A minimum tiling path of 4660 sugarcane BAC that best covers the gene-rich part of the sorghum genome was selected based on whole-genome profiling, sequenced, and assembled in a 382 Mb single tiling path of a high-quality sequence. A total of 25,316 protein-coding gene models are predicted, 17% of which display no colinearity with their sorghum orthologs. We showed that the two species, *S. officinarum* and *S. spontaneum*, involved in modern cultivars differ by their transposable element content explaining their distinct genome size. A SNP-based genetic map was built and revealed a few large chromosomal rearrangements between *S. officinarum* and *S. spontaneum*, explaining their distinct basic chromosome numbers while also suggesting that polyploidization arose in both lineages after their divergence. This BAC-based sugarcane reference sequence represents an essential resource to explore hom(oe)ologous allelic variation and perform genetic and genomic studies in cultivars and sugarcane germplasm.

(22) Distinct 5S rDNA families mark bivalent and univalent chromosomes in pentaploid dogroses (*Rosa*, section Caninae)

First author	Radka Vozárová, Czech Academy of Sciences, Institute of Biophysics, CZ
Co-authors	Jana Lunerová, Czech Academy of Sciences, Institute of Biophysics, CZ, Veit Herklotz, Senckenberg Museum für Naturkunde Görlitz, DE, Yuri Tynkevich, Chernivtsi National University, Chernivtsi, UA, Roman Volkov, Chernivtsi National University, Chernivtsi, UA, Christiane Ritz, Senckenberg Museum für Naturkunde Görlitz, DE, Aleš Kovařík, Czech Academy of Sciences, Institute of Biophysics, CZ

The most of all *Rosa* species within the section Caninae are pentaploids (2n=5x=35), known as dogroses with an asymmetrical meiosis. During meiosis only two sets of chromosomes form bivalents while other sets are transmitted as univalents through the macrogamete. Here we isolated two specific sequences (5S rDNA A and 5S rDNA B) derived from the 5S rDNA intergenic spacers (IGS). Both variants show no or little sequence similarity. We analysed anthers meiosis in four 5x dogrose species originating from the Caninae and Rubigineae sections by 5S and 18S rDNA FISH. In *Rosa canina* (subsection Caninae) the type B variant was localised on both bivalents and univalents while the A variant was present on univalent chromosomes only. An opposite pattern was observed in *R. inodora* and *R. rubiginosa* (both subsection Rubigineae) in which the A variant was located on bivalents while the B and A variants occured on univalents. Type B variant colocalised with the 18S rDNA (NOR) in both groups. *Rosa dumalis*, a putative interspecific F1 hybrid between *R. canina* and *R. inodora* carried the type A 5S rDNA variant on bivalents; the univalents carried both A and B types. This suggests that at least some parental bivalent and univalent chromosomes may pair in the *R. dumalis* hybrid. Using genomic resources we quantified that the genome proportion of 5S rDNA ranged 0.02-0.06% corresponding to c.600-7,000 copies/1C. It appeared that members of the *Rosa* section (formerly Cinnamomeae) carry type A 5S rDNA variant only. In contrast most members of section Synstylae amplified both A and B variants at different ratios. We conclude that chromosomes may function as bivalents and univalents depending on species origin and direction of cross.

(23) DNA replication timing program in barley (Hordeum vulgare)

First author: J Cizkova, Institute of Experimental Botany, CZ
Co-authors: A Nemeckova, Institute of Experimental Botany, CZ, J Vrana, Institute of Experimental Botany, CZ, J Dolezel, Institute of Experimental Botany, CZ, E Hribova, Institute of Experimental Botany, CZ

Nuclear genome is replicated during the S phase of cell cycle under strict rules, which ensure accuracy and completeness of this important process. Replication timing programs have been described in prokaryotes, yeasts and many animal species. In plants, DNA replication was analysed in detail only in species with relatively small genomes, such as maize and *Arabidopsis*. To provide more insights, we studied the replication machinery in barley, a representative of plants with large genome (1C $\sim$ 5100 Mb) and high proportion of DNA repeats ($\sim$ 84%). In order to study DNA replication in time and space we combined flow sorting of EdU-labelled nuclei, 3D acrylamide FISH and Repli-Seq. Nuclei at different stages of the cell cycle (G1, G2, early, middle and late S-phase) were isolated and used for 3D FISH with probes specific to different DNA repeats and rRNA genes. We observed that replication process of different DNA repeats varied in time, e.g., centromeric retrotransposon Cereba was replicated during all stages of S phase, while sub-telomeric satellite repeat psc119 was replicated during the early and middle S phase. Difference in replication timing was observed also for rRNA genes, while 5S rRNA genes were replicated during early S phase, majority of 45S rRNA genes were replicated in late S phase, probably reflecting the presence of large amounts of pseudogenic 45S rDNA units. Genome-wide replication timing program in barley was described based on Repli-Seq, which identified genomic regions that replicate predominantly during early, middle and late S-phase. Our results provide the first picture of the complexity of DNA replication in barley, reflecting different types of DNA sequences and their role in genome organization.

(24) Segmental allotetraploidization generates extensive homoeologous expression rewiring and phenotypic diversity at the population level in rice

First author: Yue Sun, Northeast Normal University, CN
Co-authors: Ying Wu, Northeast Normal University, CN, Chunwu Yang, Northeast Normal University, CN, Shuai Sun, Northeast Normal University, CN, Xiuyun Lin, Jilin Academy of Agriculture, CN, Lixia Liu, Northeast Normal University, CN, Chunming Xu, Northeast Normal University, CN, Jonathan F. Wendel, Iowa State University, US, Lei Gong, Northeast Normal University, CN, Bao Liu, Northeast Normal University, CN

Allopolyploidization, that is, the doubling of two or more divergent genomes in a common nucleus/cytoplasm, is known to instantly alter genomewide transcriptome dynamics, a phenomenon referred to as "transcriptomic shock." However, the immediate effects of transcriptomic alteration in generating phenotypic diversity at the population level remain underinvestigated. Here, we employed the MassARRAY-based Sequenom platform to assess and compare orthologous, allelic and homoeologous gene expression status in two tissues (leaf and root) of a set of randomly chosen individuals from populations of parental rice subspecies (indica and japonica), in vitro "hybrids" (parental mixes), reciprocal F1 hybrids and reciprocal tetraploids at the 5th-selfed generation (S5). We show that hybridization and whole genome duplication (WGD) have opposing effects on allelic and homoeologous expression in the F1 hybrids and tetraploids, respectively. Whereas hybridization exerts strong attenuating effects on allelic expression differences in diploid hybrids, WGD augments the intrinsic parental differences and generates extensive and variable homoeolog content which triggers diversification in expression patterning among the tetraploid plants. Coupled with the vast phenotypic diversity observed among the tetraploid individuals, our results provide experimental evidence in support of the notion that allopolyploidy catalyses rapid phenotypic diversification in higher plants. Our data further suggest that largely stochastic homoeolog content reshuffling rather than alteration in total expression level may be an important feature of evolution in young segmental allopolyploids, which underlies rapid expression diversity at the population level.

(25) Hybrid speciation via genome merger in *Brassica*

First author	Elvis Katche, Justus Liebig University, DE
Presenting author	Annaliese Mason, Justus Liebig University, DE
Co-authors	Roman Gaebelein, Justus Liebig University, DE, Jacqueline Batley, The University of Western Australia, AU, Annaliese Mason, Justus Liebig University, DE

Hybridisation, where two species come together to form a new species with genetic material from both parents, is now known to be an important process in speciation and genome evolution. Hybridisation is generally conceptualized as occurring between species which are both diploid, and which contribute either half a set of chromosomes (homoploid hybrid) or a full set of chromosomes (allopolyploid hybrid) to a new hybrid plant. However, hybridisation can also occur between allopolyploid species. In particular, it has previously been hypothesized that some extant genomes may result from hybridisation between allotetraploid species which share one of two genomes in common, e.g. AABB $\times$ AACC $\rightarrow$ AABC, followed by recombination and restructuring of the two divergent haploid genomes (B and C in this example) to produce a novel genome. We aimed to experimentally test this evolutionary hypothesis in the *Brassica* genus, which contains three allotetraploid species which each contain two of three genomes (AABB, AACC and BBCC). Hybridisation between *B. juncea* (AABB) and *B. carinata* (BBCC) followed by self-pollination for six generations resulted in novel, stable karyotypes being formed by recombination between the closely related *Brassica* A and C genomes. Resulting hybrids were fully fertile and showed regular meiosis. Our results demonstrate that hybridisation between allopolyploids which share one of two genomes in common has the potential to produce new, recombined genomes, potentially confounding phylogenetic analyses of species origins.

(26) Confirmation that genes retained from ancient polyploidy events differ from tandem duplicable genes and singletons across a wide range of features

First author	Zoe Vance, Trinity College Dublin, IE
Co-authors	Aoife Mc Lysaght, Trinity College Dublin, IE

Gene duplication is known to be an important process in the evolution of genomes and creation of new genetic material. Duplicates retained following polyploidy events form a set which is largely non-overlapping with genes duplicated through tandem duplication. Many differences have been noted between these gene sets in function, basic gene features such as genomic length and more complex properties such as regulation of gene expression. However, existing work on this topic lacks cohesion regarding what features distinguish genes which are retained from ancestral polyploidy (ohnolog) from genes duplicable by tandem duplication; current studies in the literature are inconsistent in study species and definitions of the gene sets to be compared. Here we show, by comparing 16 features in consistently defined gene sets in human, that there is a general trend of higher constraint in ohnologs, and of lower constraint in tandem duplicated genes, relative to singletons. Ohnologs are generally more complex, slower evolving and more heavily regulated with tandem duplicates showing the opposite pattern. Additionally, we confirm previously reported trends for enriched functions in both gene sets; ohnologs are enriched for developmental, nervous system and regulatory functions while tandem duplicates show enrichment for functions associated with external triggers such as immune and sensory functions. As these comparisons are performed within a single species, using a consistent definition for the categories compared, they serve as a useful starting point to answer more complex questions about the nature of gene duplication, such as which differences can be interpreted to give insight into mechanisms of gene duplication and retention.

(27) Homeologous pairing is necessary for fertility and later establishment of hybrids formed between two allotetraploid species sharing a common genome

First author | Elvis Katche, Justus-Liebig University Giessen, DE
Co-authors | Annaliese Mason, Justus-Liebig University Giessen, DE

Interspecific hybridization is a major driver of speciation and hence evolution. Two major modes of speciation by hybridization have been described: homoploid hybrid speciation, which involves hybridization without a change in chromosome number (e.g. AA + BB → AB) and allopolyploid speciation, where hybridization is accompanied by a doubling of chromosome number (e.g. AA + BB → AABB). In homoploid hybrid speciation, restructuring between the two genomes may establish genome stability, while in allopolyploid speciation, doubling of the genome restores genome stability and fertility. Hybridization can also produce other outcomes: for example, hybridization between allotetraploids sharing a common genome (AABB + BBCC → BBAC). Can new hybrid species arise through this pathway? We created three interspecific hybrid populations by crossing *Brassica juncea* × *B. napus* (F1 = AABC = 2n = 37), *B. juncea* × *B. carinata* (F1= BBAC = 2n = 35) and *B. napus* × B. carinata (F1= CCAB = 2n = 36) to study if new, stable, and fertile species could form in later generations following self-pollination of these hybrids. CCAB and AABC F1 hybrids failed to progress past the S1 generation, as they showed high levels of infertility under self-pollination conditions. BBAC F1 hybrids however showed increased fertility with successive generations. Analysis of these three S1 hybrid populations using the *Brassica* 90K SNP array revealed limited homeologous pairing between the haploid genomes present in both CCAB and AABC hybrids, but a high level of homoeologous pairing between haploid genomes in BBAC hybrids. Our results show homeologous chromosome pairing can also be important in promoting hybrid fertility and stability in some scenarios.

(28) Repeatome dynamics in allopolyploid apomictic hawkweed species

First author | D. Greguraš, Institute of Botany, Czech Academy of Sciences, CZ
Co-authors | J. Chrtek, Institute of Botany, Czech Academy of Sciences, CZ, M. Hartmann, Institute of Botany, Czech Academy of Sciences, CZ, J. Josefiová, Institute of Botany, Czech Academy of Sciences, CZ, P. Mráz, Herbarium and Department of Botany, Charles University, CZ, L. Paštová, Institute of Botany, Czech Academy of Sciences, CZ, J. Pinc, Herbarium and Department of Botany, Charles University, CZ, J. Fehrer, Institute of Botany, Czech Academy of Sciences, CZ

Hybridization and polyploidization in plants can sometimes be followed by a shift to apomixis (asexual seed production). The repeatome, the repetitive fraction of the genome, is often the largest proportion of plant genomes and is positively correlated with genome size. The extent of its restructuring in apomicts is still poorly understood. We investigated two diploid species of the genus Hieracium s.str. and two of their hybridogenous triploid derivatives recognized as different apomictic taxa. They were found to differ in the relative genome dosage obtained from the parents (2:1, 1:2). Genome sizes of both polyploid taxa were additive or nearly so, suggesting only little or no change in genome size followed the allopolyploidization events that created the apomictic lineages. Repeatome analyses using the RepeatExplorer pipeline revealed a surprisingly high similarity of the parental species considering their phylogenetic distance according to multiple molecular markers. Repetitive DNA represented ca. 70% of the genomes; the most prevalent elements were Ty3/Gypsy Chromovirus Tekay and Ty1/Copia Maximus-SIRE. Comparative analyses of allopolyploid repeatomes revealed significant deviations from additivity in several clusters with moderate and low abundance across different repeat categories, which do not, however, contribute significantly to overall genome size. The highest deviations were observed in tandem repeats and rDNA, followed by unclassified repeats and different Ty3/Gypsy elements. These findings point to a relatively low turnover of repetitive DNA during the formation of apomictic lineages in Hieracium compared to other allopolyploid systems and contradict the often claimed genome downsizing in polyploids.

(29) A first look at the effects of whole genome duplication on the phenotype and evolvability of *Chlamydomonas reinhardtii*.

First author: Quinten Bafort, VIB-UGent Center for Plant Systems Biology, BE
Co-authors: Eylem Aydogdu, VIB-UGent Center for Plant Systems Biology, BE, Lucas Prost, VIB-UGent Center for Plant Systems Biology, BE, Olivier De Clerck, UGent, BE, Yves Van de Peer, VIB-UGent Center for Plant Systems Biology, BE

Whole genome duplication (WGD) is a genomic mutation with far-reaching consequences. The increase in genetic material can alter the phenotype considerably e.g. via the well-established correlation between cell and genome size, altering surface area to volume ratios. Additionally the presence of an extra (redundant) genome copy that is free to evolve is supposed to increase the genomic and epigenetic flexibility and therefore the evolvability. Consequently an increased ploidy level creates the potential to adapt faster to changing environments or outside niche conditions. We study the consequences of whole genome duplication using experimental evolution of the unicellular green algae *Chlamydomonas reinhardtii*. Two haploid and two diploid strains of *Chlamydomonas* are evolving in dodecaplicate in both standard benign laboratory conditions and a stressful saline environment. We have followed cell size and growth parameter evolution for over more than 700 generations. After WGD, we observed an increase in cell size (in both environments), an increase in the maximum growth rate (in benign environment) and a reduction in the maximum population size (in both environments). The immediate effect of exposure to a saline environment for both ploidy levels is an increase in cell size, a reduction in maximum growth rate and a reduction in the maximum population size. Over time the size of diploid cells evolving in the saline medium dropped below that of those in a benign environment, the maximum growth rates of both haploid and diploid lines increased in both environments and this increase in growth rate came at least for some lines at the cost of a reduction in the maximum population size.

(30) Chromosomal variation and its relevance to abiotic stress in segmental allotetraploid rice

First author: Ying Wu, Northeast Normal University, CN
Co-authors: Guo Li, Northeast Normal University, CN, Yue Sun, Northeast Normal University, CN, Chunming Xu, Northeast Normal University, CN, Lei Gong, Northeast Normal University, CN, Bao Liu, Northeast Normal University, CN

Polyploidy is a pervasive driving force in both plant and animal evolution. Between the two major types of polyploidy, i.e., autopolyploidy and allopolyploidy, is a continuum which meets the criteria of segmental allopolyploidy. For both segmental and allopolyploidy wherein homeologous chromosomes exist, homoeologous exchanges (HEs) is recognized as a major genetic consequence. Both numerical and structural chromosomal variations may be generated due to HEs and lead to rapid phenotypic diversity. Here, we used whole-genome resequencing to investigate the occurrence of HEs and aneuploidy in a novel segmental allotetraploid rice system derived from inter-subspecies (japonica and indica) hybridization and chromosome doubling in Oryza sativa. The resequencing of 312 randomly selected S5 tetraploid individuals revealed rampant HEs, which converted 30-60% heterozygous genomic regions into homogenized state of either parent. In addition, we identified ca. 40% aneuploids harboring 55 distinct karyotypes in the population under normal conditions. Interestingly, under acute salt stress, the proportion of aneuploidy that survived the stress shifted to ca. 65%, suggesting aneuploidy is overtly more tolerant to the stress than euploidy in our system. We are determining the exact karyotypes of the survived aneuploidy, as well as assessing the effect of HEs under euploidy on abiotic stresses.

(31) Polyploidy and genetic variability in field-collected gametophytes of species of the red algal genus *Porphyra*

First author	E Varela-Álvarez, CCMAR Centro de Ciências do Mar, CIMAR Laboratório Associado, Universidade do Algarve, PT
Presenting author	J Loureiro, Centre for Functional Ecology , Department of Life Sciences, University of Coimbra, PT
Co-authors	J Loureiro, Centre for Functional Ecology , Department of Life Sciences, University of Coimbra, PT, C Paulino, CCMAR Centro de Ciências do Mar, CIMAR Laboratório Associado, Universidade do Algarve, PT, E Serrão, CCMAR Centro de Ciências do Mar, CIMAR Laboratório Associado, Universidade do Algarve, PT

Porphyra is a polyphyletic genus of red algae with similar morphologies and with a wide variety of life history strategies. *Porphyra* species typically have a heteromorphic life cycle consisting of a gametophytic blade phase and a filamentous sporophytic phase called the conchocelis. The commercial cultivation and harvesting of these species (*Porphyra*/*Pyropia* complex) present a retail value of $1.3 billion per year. For decades, it has been assumed than two ploidy levels are involved in the *Porphyra* life cycles with the gametophytic blade being haploid and the sporophytic carpospores/conchocelis being diploid. However with the development of new molecular markers we have detected gametophytic blades possessing heterozygous genotypes with 1–6 alleles per individual in many samples along European coasts. In this study, we assessed the ploidy level, genome size and genetic diversity of three *Porphyra* species: *Porphyra dioica*, *P. umbilicalis*, and *P. linearis* from the Iberian Peninsula. Seven distinct ploidy levels and eight genome sizes were found in these *Porphyra* species, corresponding to three main lineages, triploids (3x), tetraploids (4x) and Mixoploids (2x/3x, 2x/4x, 3x/4x). Overall, the results of the present study reveal that *Porphyra* constitutes a complex polyploid system, composed by autopolyploids, mixoploids, multiploid gametes and probably spontaneous chromosome doubling. In P. linearis, genetic differentiation was found among three polyploid lineages: triploid, tetraploid and mixoploids, representing different evolutionary units. We conclude that the gametophytic phase (n) in *Porphyra* species is not haploid, contradicting earlier theories. New life history strategies for *Porphyra* species are proposed.

(32) QTL analysis in Multiparental Polyploid Populations

First author	A Thérèse, Wageningen University & Research, NL
Co-authors	G Tumino, Wageningen University & Research, NL, R Voorrips, Wageningen University & Research, NL, E van de Weg, Wageningen University & Research, NL, C Maliepaard, Wageningen University & Research, NL

QTL analysis in autopolyploids is more challenging than in diploids, not only due to the different segregation patterns, but also due to a higher number of expected alleles per locus, especially in outcrossing crops. While biallelic SNP markers can easily track parental alleles in a diploid, they become decreasingly informative as heterozygosity and polyploidy increase. For instance, two tetraploid parents can contribute a maximum of eight alleles at a particular locus, while SNP markers will only distinguish two. Since many polyploid crops are highly heterozygous, this approach can drastically decrease statistical power to detect QTLs. To improve this situation, multiple SNPs can be combined to generate haplotype-based markers, which are better at tracking alleles that are Identical-by-Descent (IBD). In a pre-breeding context, where genetic diversity is studied, detection of novel QTLs is generally done by testing multiple biparental populations independently. However, if progenitors are shared between crosses, i.e. those crosses form a multiparental population, they can be analysed together in order to increase statistical power of the study. With such a setting, a larger amount of genetic diversity is analysed than in a biparental cross, while most issues associated with Genome-Wide Association Studies are avoided, such as low frequency alleles and strong population structure. In this study, we used simulated tetraploid multiparental populations with different levels of diversity to show how haplotype-based markers can be applied in a joint QTL analysis. Haplotype-based markers resulted more powerful than biallelic SNP markers, allowing to detect multiple QTLs from different genetic backgrounds in a single analysis.

(33) Modeling microsynteny network data to understand lineage-wide gene retention patterns after polyploidy

First author | Tao Zhao, VIB-UGent Center for Plant Systems Biology, BE
Co-authors | Yves Van de Peer, VIB-UGent Center for Plant Systems Biology, BE

Polyploidy (WGD) is often proposed as a driving force for the origin of novel traits during evolution. In plants, recurrent WGDs often lead to massive genome rearrangements. Shared local gene collinearity (microsynteny) provides valuable information of the common segmental ancestry within and between plant genomes. Here, we attempt to characterize differential fractionation patterns (stable versus unstable) following a WGD event, across different lineages or phylogenetic groups. We have integrated all pairwise microsynteny blocks from ∼150 land plant genomes into a synteny network database. Thereupon, we have combined network metrics (e.g. network size, degree centrality, transitivity, betweenness, etc.) and phylogenetic metrics (e.g. substitution rates, tree building, molecular dating, etc.) to decipher the evolutionary trajectories of gene duplicates (derived from WGD) over large time scales, and to explore the mechanism behind preferential gene retention patterns that are often observed.

(34) Multiple auto- and allopolyploidisations marked the glacial history of the widespread Eurasian steppe plant *Astragalus onobrychis* (Fabaceae)

First author | Božo Frajman, Department of Botany, University of Innsbruck, AT
Co-authors | Eliška Záveská, Department of Botany, University of Innsbruck, AT, Clemens Maylandt, Department of Botany, University of Innsbruck, AT, Ovidiu Paun, Department of Botany and Biodiversity Research, University of Vienna, AT, Peter Schönswetter, Department of Botany, University of Innsbruck, AT

The Eurasian steppes occupy a significant portion of the world-wide land surface and their biota have been affected by specific past range dynamics driven by ice ages-related climatic fluctuations. The dynamic alterations in conditions during the Pleistocene often triggered reticulate evolution and whole genome duplication events. Employing genomic, genetic, cytogenetic and phenotypic tools we investigate the intricate evolution of *Astragalus onobrychis*, a widespread Eurasian steppe plant with diploid, tetraploid and octoploid cytotypes. To analyse the heteroploid RADseq dataset we employ both genotype-based and genotype-free methods that result in highly consistent results, and complement our inference with information from the plastid ycf1 region. We uncover a complex and reticulate evolutionary history, including at least one auto-tetraploidization event and two allo-octoploidization events, involving also genetic contributions from other species, most likely *A. goktschaicus*. The present genetic structure points to the existence of four main clades within *A. onobrychis*, which only partly correspond to different ploidies. Time-calibrated diffusion models suggest that diversification within *A. onobrychis* was associated with ice age-related climatic fluctuations during the last million years. We finally argue for the usefulness of uniparentally inherited plastid markers, even in the genomic era, especially when investigating heteroploid systems.

(35) The impact of whole genome duplication on alternative splicing patterns

First author	Carlos Martí-Gómez, Centro Nacional de Investigaciones Cardiovasculares Carlos III (CNIC), Madrid, Spain, ES
Co-authors	Yamile Marquez, Manuel Irimia, Centre de Regulació Genómica (CRG), Barcelona, Spain, ES, Simen Sandve, Norwegian University of Life Sciences, NO

Whole genome duplication (WGD) events in the vertebrates have been important for the evolution of tissue regulated gene expression. Gene duplication and alternative splicing (AS) are two interconnected mechanisms for increasing proteomic diversity and potentially generating new functions. It has been shown that duplicated genes tend to have fewer splicing isoforms than their unduplicated orthologs, differences that decrease with duplicate age. However, these studies focused on single gene duplications and on the mere detection of splicing isoforms. Therefore, an understanding of the impact of WGD on AS patterns is still lacking. In this study we use a novel exon-orthology pipeline to map ortholog and ohnolog exons across teleost genomes, focusing on the WGD specific to the salmonid fish. We use RNA-seq data for a number of tissues to study the quantitative changes in the mean and tissue regulated splicing rates of homologous exons. Our initial results show a small decrease in the number of tissue-regulated exons in the salmon transcriptome compared with pike and zebrafish. Analysis of the 1:1:2 orthologs suggests that tissue regulation of AS tends to be lost after WGD more often than gained and that evolution of inclusion rates in each tissue proceeds in a highly asymmetric manner, such that one duplicate accumulates most of the divergence from the common ancestor. Whether these AS changes are adaptive or arise from relaxed selective pressure after WGD remains to be elucidated.

(36) Comparative analysis of satellite sequences in allotriploid *A. x cornutum* and its three parental species

First author	I Šamanić, University of Split, Faculty of Science, Deparment of Biology, HR
Presenting author	J Puizina, University of Split, Faculty of Science, Deparment of Biology, HR
Co-authors	J McCann, University of Natural Resources and Biodiversity, Institute of Biotechnology, AT, Ž Fredotović, University of Split, Faculty of Science, Deparment of Biology, HR, J Mlinares, University of Zagreb, Faculty of Science, Department of Biology, Division of Molecular Biology, AT, H Weiss-Schneeweiss, University of Vienna, Department of Botany and Biodiversity Research, AT, H Kalinić, University of Split, Faculty of Science, Deparment of Computer Science, AT, V Besendorfer, University of Zagreb, Faculty of Science, Department of Biology, Division of Molecular Biology, HR

An established minor garden crop, triploid onion *Allium* × *cornutum* (2n=3x=24) is traditionally cultivated in coastal Croatia and is used as spice and condiment. Similar triploid onions are cultivated as garden crops in southeastern Asia, Europe and other parts of the world. Our previous molecular, phylogenetic and cytogenetic studies provided evidence for its unique triparental origin with three putative parental species, *A. cepa, A. pskemense,* and *A. roylei*. Here we performed de novo identification and quantification of repetitive DNA families of *A. x cornutum* and its three parental *Allium* species by application of next generation sequencing and using the RepeatExplorer pipeline (Novák et al., 2013). The genome coverage of three species was about 0.4X. Nuclear repetitive DNA constituted 53,05% of *A. roylei* genome, 57,18% of *A. cepa*, 58,59% of *A. pskemense*. We identified 6 satellite DNA families in *A. cepa,* 5 in *A. pskemense* and 4 in *A. pskemense*. By using PCR, cloning, sequencing and FISH we identified three satellite sequences in both diploid progenitors and A. x cornutum. One satellite sequence is similar to well-known terminal satellite sequence in *Allium cepa* and we mapped it on almost all chromosome ends. The second satellite sequence shows homology to *Allium fistulosum* centromeric sequence and it localizes to the centromeres of several but not all chromosomes. The third satellite sequence is hitherto unknown and it localizes to the terminal and some intercalary positions of several chromosomes of three diploid species and allotriploid. The obtained results in combination with previous data on rDNA phylogeny and mapping will be useful in better understanding of poliploidization and evolutionary history of allotriploid

(37) Small non-coding RNAs might contribute to the success of allopolyploids: insights from *Brassica* neo-allopolyploids

First author: Paulina Martinez Palacios, GQE Le Moulon, FR
Presenting author: Eric Jenczewski, Institut Jean Pierre Bourgin, INRA, FR
Co-authors: Marie-Pierre Jacquemot, GQE Le Moulon, FR, Marion Tapie, GQE Le Moulon, FR, Agnès Rousselet, GQE Le Moulon, FR, Mamoudou Diop, GQE Le Moulon, FR, Carine Remoué, GQE Le Moulon, FR, Matthieu Falque, GQE Le Moulon, FR, Andrew Lloyd, Institut Jean Pierre Bourgin, INRA, FR, Eric Jenczewski, Institut Jean Pierre Bourgin, INRA, FR, Gilles Lassalle, IGEPP, INRA, FR, Anne-Marie Chèvre, IGEPP, INRA, FR, Christine Lelandais, Institute of Plant Sciences Paris-Saclay, FR, Martin Crespi, Institute of Plant Sciences Paris-Saclay, FR, Philippe Brabant, GQE Le Moulon, FR, Johann Joets, GQE Le Moulon, FR, Karine Alix, GQE Le Moulon, FR

Allopolyploidy, combining interspecific hybridization with whole genome duplication is recognised to have a significant impact on plant evolution. Its evolutionary success is related to the rapid and profound genome reorganizations, generated in response to the supposed "Genome Shock", that allow the neo-allopolyploid to adapt efficiently to new environments. Nevertheless, how the neo-allopolyploid genome adapts to regulate its expression remains poorly understood: the hypothesis of a major role for small non-coding RNAs (sRNAs) in mediating the functional response of the neo-allopolyploid genome has progressively emerged. We have characterized the global response of sRNAs to allopolyploidy in *Brassica*, analysing three independent *Brassica napus* neo-allotetraploids in comparison with their diploid progenitors *Brassica oleracea* and *Brassica rapa*, by high-throughput sequencing of sRNAs [*]. Our results suggest an immediate response of specific sRNA populations to the allopolyploidy event, which target non-coding components of the genome and regulate genes implied in stress responses and in plant metabolism. We also identify the early accumulation of both 21- and 24 nt sRNAs involved in the regulation of the same targets, supporting a 'PTGS-to-TGS' shift at the first stages of the neo-allopolyploid formation. We propose that sRNA production is early reorganized in response to allopolyploidy to control the transcriptional reactivation of unexpected non-coding elements as well as of various stress-related genes thus playing a role as guardians of genome stability during the first steps of neo-allopolyploid formation.

[*] Martinez Palacios et al. 2019. Mol Biol Evol 36(4):709–726. doi:10.1093/molbev/msz007

(38) Polyspermy: a triploid bridge with novelty

First author: Y Mao, University of Bremen, DE
Presenting author: D Tekleyoans, University of Bremen, DE
Co-authors: T Nakel, University of Bremen, DE, T Baum, University of Bremen, DE, R Groß-Hardt, University of Bremen, DE

One of the striking features of land plants is their ability to subsist in diverse ecology. Adaptation to challenging environmental conditions demands the physical and physiological strength to survive and the transferring of genetic material to the next generation by producing viable seeds. Such potency often involve changes in genome composition that result in the development of novel traits enabling the species to be more competitive and efficient within a population. Polyploidization is one of the main driving forces behind the evolution such novelties that can bring diversity through promotion of reproductive isolation and adaptive fitness of an organism. In plants, auto- and allopolyploidy can arise from the fusion of unreduced gametes or somatic doubling and often involve a triploid stage that serves as a transitional bridge from a diploid state to polyploidy. Recently, we reported polyspermy to be unprecedented route towards plant polyploidization. Here, we present our new results on the effect of polyspermy in plant polyploidization in general and plant development in particular.

(39) Recent allotetraploid *Capsella bursa-pastoris*: a promising model of post-polyploidization petal reduction

First author: Elina Shnaider, Institute of Information Transmission Problem RAS, RU
Co-authors: Anna Klepikova, Institute of Information Transmission Problem RAS, RU, Dmitry Sokoloff, Lomonosov Moscow State University, RU, Maria Logacheva, Institute of Information Transmission Problem RAS, RU, Aleksey Penin, Institute of Information Transmission Problem RAS, RU

Hybridization and resulting polyploidization is a very frequent phenomenon in plants. The subsequent neo- and subfunctionalization of duplicated genes can increase phenotypic plasticity of polyploid plants. A promising model of post-polyploidization petal reduction is a recent allotetraploid *Capsella bursa-pastoris* (L.) from Cruciferae family. . bursa-pastoris has arisen from interspecific hybridization of two closely related species: ancestor of C. rubella/grandiflora and C. orientalis. In this study we focused on floral structure. For . bursa-pastoris the transformation of petals into stamens was previously described. We have examined the mutants with new type of petal loss - without the increase of stamen number. Such mutants are widespread in natural population of . bursa-pastoris in Eastern Europe. The aim was the localization of genomic regions associated with petal reduction. To achieve this goal, we have analyzed petal number in second progeny from cross of mutant and wild-type plants and then have sequenced the genomes of two pools of F2 plants with contrasting phenotypes. Using the mapping by sequencing, we have found two trait-linked regions located at homeologous chromosomes but not representing homeologous genes. For one loci the inheritance mode of mutant trait is recessive and for the second – codominant. The presented study may shed light on mechanisms of morphological evolution in early polyploids. The research was funded by Ministry of Science and Higher Education RF (0053-2019-0005).

(40) Evolutionary dynamics of meiosis genes in *Brassica* species

First author: SV Schiessl, Justus Liebig University, DE
Co-authors: Y Zhang, The Key Laboratory of Biology and Genetic Improvement of Oil Crops, Chinese Academy of Agricultural Sciences, CN, S Liu, The Key Laboratory of Biology and Genetic Improvement of Oil Crops, Chinese Academy of Agricultural Sciences, CN, I Parkin, Agriculture and Agri-Food Canada, CA, AS Mason, Justus Liebig University, DE

Meiosis is a central process governing both faithful inheritance as well as recombination of the genetic information. Due to its importance and sensitivity, functional genetic control of meiosis is extremely important for the stability of a species. Early generation polyploids often show meiotic failure or mistakes, leading to infertility, seed abortion and reduced seed set, which severely impact the evolutionary fitness of the new species. Established polyploid species, on the other hand, show normal meiosis and fertility. To date, it is unclear if this stabilization process is either due to the breakdown of intragenomic homology, due to genetic control or both. Here, we analyzed homologs of 122 meiosis genes known from *A. thaliana* in the 3 diploid (mesohexaploid) *Brassica* species *B. rapa, B. nigra* and *B. oleracea* (AA, BB and CC genomes, respectively) and compared them to the allotetraploid *B. napus* (AACC genome). We studied copy number, sequence divergence and synteny using the gene sequences from the respective reference genomes, and linked those data to sequence diversity in large populations from publically available resequencing data. We found that more than half of the meiosis genes reverted to a single copy in the diploids, but retained 2 copies in the tetraploid, indicating that reduced redundancy in meiosis genes is beneficial, but may take a long evolutionary time to establish. In the allotetraploid, 36% of the gene groups lost copies, while only 11% gained copies compared to their diploid progenitor.Our results illustrate the evolutionary paths of genes under strong selection pressure and offer a basis for hypothesis testing of the role of specific meiosis genes in *Brassica* allopolyploidisation events.

(41) The origin of duplicates and endogenous constraints underlying long-term genome fractionation in *Biscutella laevigata*

First author	Rimjhim Roy Choudhury, Institute of Plant Sciences, University of Bern, CH
Co-authors	Sandra Grünig, Institute of Plant Sciences, University of Bern, CH, Marc Beringer, Institute of Plant Sciences, University of Bern, CH, Christian Parisod, Institute of Plant Sciences, University of Bern, CH

Biscutella laevigata has colonized alpine ecosystems through recurrent whole-genome duplications (WGD), following a 8MY-old Bl-m-WGD event and then a ca. 1MY-old Bl-n-WGD event that yielded extent diploids and autopolyploids. Thus, it is a model to understand the processes relating to the interaction between duplicated genes and environmental stressors in driving genome evolution through different timescale. We assembled the diploid genome of B. laevigata to infer processes retaining duplicates after long-term genome fractionation subsequent to Bl-m-WGD event using PacBio sequencing. Annotation of genes and transposable elements (TEs) suggests biased fractionation through preferential retention of duplicated genes responding to abiotic stressors and high sequence turnover among other genome fractions due to recently active TEs. Contrastingly, anciently active TEs were observed next to retained genes under purifying selection, highlighting the central role of TEs in duplicate retention. Improvement of assembly contiguity through long-range scaffolding is further assessing mechanisms driving the balance between high sequence turnover and conservation of specific loci. Coupling such evidence of the impact of endogenous factors with an assessment of transcriptional plasticity driven by exogenous factors (Beringer et al.) will link environmental constraints and genome evolution to shed light on the evolution of adaptive gene clusters following WGD events.

(42) Phylo-cytogenomic evidence of independent ancient polyploidization events within a Brassicaceae tribe

First author	Terezie Mandáková, CEITEC – Central European Institute of Technology, Masaryk University, Brno 62500, Czech Republic, CZ
Presenting author	Xinyi Guo, CEITEC – Central European Institute of Technology, Masaryk University, Brno 62500, Czech Republic, CZ
Co-authors	Karolína Trachtová, CEITEC – Central European Institute of Technology, Masaryk University, Brno 62500, Czech Republic, CZ, Barış Özüdoğru, Department of Biology, Faculty of Science, Hacettepe University, Beytepe, Ankara 06800, Turkey, TR, Jianquan Liu, College of Life Sciences, Sichuan University, Chengdu, CN, Martin Lysak, CEITEC – Central European Institute of Technology, Masaryk University, Brno 62500, Czech Republic, CZ

The small tribe Biscutelleae is an early-diverging crucifer (Brassicaceae) tribe consisting of five genera and c. 59 species. The origin of two genera (*Biscutella* and *Ricotia*) has been attributed to independent polyploidization events (Geiser et al. 2016, Plant Cell; Mandakova et al 2018, Plant Journal), however, it remains unknown whether genome evolution in other three genera was shaped by additional whole-genome duplication (WGD) event(s). Here we investigated genome evolution in the Biscutelleae by combining cytogenetic and bioinformatic approaches. Comparative cytogenetic maps were reconstructed in three genera (*Heldreichia, Lunaria* and *Megadenia*). Illumina RNA-seq data was generated for six species, covering all five genera of the tribe. We analyzed the distribution of synonymous substitution rates (Ks) between homeologous and orthologous gene pairs. Phylogenomic analyses of gene families were performed to place the inferred WGD events on a species tree. Except the diploid Megadenia (n=6), the four remaining genera were found to have a tetraploid origin. Heldreichia has experienced recent (less than 3.4 Mya) autopolyploidization from an ancestral n=5 genome. *Biscutella* (n=6, 8, 9), *Lunaria* (n=14, 15) and *Ricotia* (n=13, 14) originated through independent inter-tribal hybridizations between two genomes with n=7 and n=8 chromosomes that diverged around the split of major Brassicaceae lineages (from 20 to 25 Mya). The allopolyploid events were followed by genus- and species-specific descending dysploidies mediated by numerous chromosome rearrangements. We conclude that despite the Biscutelleae represents a monophyletic clade, the origin and diversification of its four genera were preceded by four independent WGDs.

(43) Recent hybrid origin of the narrow endemic *Pulmonaria helvetica*

First author | S Grünig, Institute of Plant Science - University of Bern, CH
Co-authors | C Parisod, Institute of Plant Science - University of Bern, CH

Pulmonaria helvetica is a recently described narrow endemic species presenting the chromosome number (2n = 24). Its current distribution range in the lowlands of Switzerland was entirely covered by ice during the last glacial maximum and indicates a relatively recent origin. In this study, we sequenced a plastid locus and generated 1077 double-digest restriction site associated DNA (ddRAD) loci in 67 individuals from across the distribution range of *P. helvetica* and candidate progenitor species to address whether it represents a genetically differentiated lineage and investigate its origin. Genotypes were assigned to genetic clusters within and among taxa using STRUCTURE and highlighted *P. helvetica* as a distinct genetically homogeneous species showing restricted gene flow with parental lineages in sympatry. It presented clear evidence of admixture between a maternal parent from *P. mollis* s.l. (2n = 18, 22, 24) and a paternal donor from *P. officinalis* (2n = 16), and was thus consistent with a hybrid speciation event after the last ice age. Comparative niche modelling revealed no distinction of environmental niches between *P. helvetica* and its parents, suggesting a key role of intrinsic factors in driving speciation. Homoploid hybrid species documented so far showed constant chromosome numbers among lineages. To what extent karyotype changes having supported the establishment of *P. helvetica* find their origin through a complex scenario of allopolyploid speciation involving dysploid taxa remains elusive.

(44) Phylogenomic reconstruction of *C. latifolia* to by-pass triploid evolutionary dead-end

First author | P Ollitrault, CIRAD, FR
Co-authors | D Ahmed, INRA, FR, F Curk, INRA, FR, R Morillon, CIRAD, FR, Y Froelicher, CIRAD, FR

Lime is the only citrus horticultural group that include diploid (C. aurantiifolia), triploid (*C. latifolia*) and tetraploid natural germplasm. The lime export-market is based on *C. latifolia* 'Tahiti' lime type with very narrow genetic basis. Genetic diversification of the 'Tahiti' lime type is necessary to promote the durability of its production. However, as in most species, the triploidy of 'Tahiti' lime is an evolutionary dead end. With the objective to diversify *C. latifolia* we analyzed the origin and phylogenomic structure of 'Tahiti' lime and established a breeding program aiming to rebuilt its phylogenomic caryotype from parental species. Nuclear and cytoplasmic diagnostic markers of the four ancestral taxa (*C. maxima, C. medica, C. micrantha* and *C. reticulata*) of cultivated citrus revealed that all were involved in the genesis of 'Tahiti' lime and that it resulted from the fertilization of a haploid ovule of C. limon by a diploid pollen of *C. aurantiifolia*. *C. limon* is an ad-mixture of three ancestors [(*C. maxima x C. reticulata) x C. medica*] while *C. aurantifolia* results from direct hybridization between C. micrantha and C. medica. We established a progeny of several hundred hybrids from fertilization of diploid lemons with pollen from the doubled diploid 'Giant Key' lime. The preferential disomic inheritance of 'Giant key' lime results in diploid gamete with high restitution of the *C. micrantha / C. medica* interspecific heterozygosity, similarly to the diploid gamete which generated 'Tahiti' lime. The greater part of the diversity of the progeny results from lemon gamete segregation. Triploid hybrids with phylogenomic caryotypes and phenotypic traits close to 'Tahiti' lime are identified.

(45) *Calendula officinalis:* is it diploid or tetraploid?

First author	G. Esmaeili, Ferdowsi University of Mashhad, IR
Presenting author	L. Leus, ILVO, BE
Co-authors	K. Van Laere, ILVO, BE, H. Muylle, ILVO, BE, L. Leus, ILVO, BE

Marigold, Calendula officinalis L., is cultivated as ornamental plant but is also grown for its seed oil and the etheric oils in the flowers. The flowers contain high concentrations of faradiol, used in pharmacy and cosmetics. The seed oil contains calendic acid which can be used as a renewable resource in paint industries. Therefore, genotypes with high compound contents these compounds are desired. Inducing variation in the ploidy level is a valuable tool to obtain better performing *Calendula* genotypes. Since natural hybridizations occurred in the evolution of *Calendula*, taxonomy, chromosome number, and ploidy level are unclear for *C. officinalis*. Most data report *C. officinalis* to be tetraploid with 2n=28 or 32. We attempted to confirm the ploidy level of *C. officinalis* by genome size analysis in 8 marigold cultivars and by constructing a FISH-based karyotype using 45S rDNA and 5S rDNA probes. Results showed 32 chromosomes in all genotypes tested and a genome size of 2,78 ±0,06 pg/2C. The morphology of the chromosomes in the karyotype showed sets of 2 homologues rather than 4. The 5S DNA signals were located on 2 homologous chromosomes in the centromeric region, whereas 4 45S signals were observed on 2 pairs of 2 homologous chromosomes on the terminal regions. The number of 2 5S rDNA sites detected are lower than the 4 sites that are expected in tetraploids. This would imply the loss of rDNA sites occurred after polyploidization in *Calendula*. Based on these results we can assume an allotetraploid background in *C. officinalis*. This background is both challenging and interesting for breeders. In further research synthetic chromosome doubling is used to create more variation in seed and flower oil production.

(46) Phylogenetic relationships and repeatome evolution in the mesohexaploid tribe Heliophileae (Brassicaceae)

First author	Mert Dogan, CEITEC - Masaryk University, CZ
Co-authors	Milan Pouch, CEITEC - Masaryk University, CZ, Petra Hloušková, CEITEC - Masaryk University, CZ, Xinyi Guo, CEITEC - Masaryk University, CZ, Terezie Mandáková, CEITEC - Masaryk University, CZ, Ihsan A. Al-Shehbaz, Missouri Botanical Garden, US, Martin A. Lysak, CEITEC - Masaryk University, CZ

The southern African endemic tribe Heliophileae is the morphologically most diverse Brassicaceae lineage. However, there is only little knowledge of its origin, genome evolution and intra-tribal phylogenetic relationships. Here we present an updated phylogenetic framework and summarize our recent progress in analyzing repeatome evolution in Heliophileae species representing different intra-tribal clades. The reconstructed large-scale phylogeny using 48 nuclear protein-coding genes and 48 species established four main crown-group clades (A-D), being sister to the most ancestral species Heliophila circeoides. Maternal phylogeny, reconstructed from de novo assembled chloroplast genomes of 16 species representing the five Heliophila clades, further corroborated the intra-tribal relationships, and served as the basis for divergence time estimates. These estimates dated the divergence of the Heliophileae and the genus Subularia from their most common ancestor to c. 22.9 Mya (early Miocene). Low-coverage genome sequencing (Illumina) was performed in 16 Heliophila species. The repeats were characterized and annotated using RepeatExplorer pipeline. LTR-retrotransposons were predominant (19.07 – 30.12%), whereas the frequency of satellite DNA (satDNA) repeats was relatively low in all genomes (1.32 – 7.67%). The intra- and inter-clade repeatome diversity did not mirror the divergence of five intra-tribal clades. The high number of satDNAs shared among the Clade A species may indicate slower repeatome evolution in this clade and further analyses are in progress. This work was supported by the Czech Science Foundation (grant no. 19-07487S).

(47) Characterisation of de novo induced polyploid Dalmation pyrethrum (Tanacetum cinerariifolium)

First author	N Malenica, Division of Molecular Biology, Department of Biology, Faculty of Science, University of Zagreb, HR
Co-authors	A Jurković, Division of Molecular Biology, Department of Biology, Faculty of Science, University of Zagreb, HR, J Mlinarec, Division of Molecular Biology, Department of Biology, Faculty of Science, University of Zagreb, HR, M Grdiša, Department of Seed Science and Technology, Faculty of Agriculture, University of Zagreb , HR, B Bohanec, Agronomy Department, Biotechnical Faculty, University of LJubljana, SI, V Besendorfer, Division of Molecular Biology, Department of Biology, Faculty of Science, University of Zagreb, HR

Dalmatian pyrethrum (Tanacetum cinerariifolium) is a perennial herb native to Croatia known for the production of insecticidal chemicals known as pyrethrins. Pyrethrum was one of the few natural insecticides before the era of synthetic insecticides. In order to increase pyrethrin yield, we created polypoid pyrethrum by chromosome doubling and subsequent crossing. Tetraploid, triploid, mixoploid and aneuploid plants were recovered and their ploidy level confirmed by karyotyping and flow cytometry. Tetraploid and triploid plants were morphometrically compared showing the 3n plants being more vigorous. A substantial difference in stomata size between 2n, 3n and 4n plants was detected. Furthermore, pollen viability and germination was lower in polyploid plants. FISH analysis with two subterminal satellite DNA families, TcSAT1 and TcSAT2, as probes showed lower variability in number of subtelomeric signals in 3n and 4n plants in comparison to 2n plants. Meiosis in polyploid pollen mother cells showed chromosome pairing that result in formation of chromosome configurations from univalents to multivalents. Increased number of multivalents indicated potentially increased activity of homologous recombination (HR) proteins. In order to support this finding, we analysed the expression of T.cinerariifolium genes involved in HR. By analyzing the published T. cinerariifolium transcriptome, we identified several Tanacetum orthologs of these genes. Their expression profiles in diploid and polyploid pyrethrum will be presented. In addition, we will present diploid and polyploid expression profiles of several pyrethrin biosynthesis genes, as well as our preliminary data of pyrethrin concentrations in diploid and polyploid plants.

(48) Towards stable, genetically diverse *Brassica* allohexaploids

First author	Daniela Quezada-Martinez, Department of Plant Breeding. Justus Liebig University, DE
Presenting author	Daniela Quezada-Martinez, Department of Plant Breeding. Justus Liebig University, DE
Co-authors	Annaliese S. Mason, Department of Plant Breeding. Justus Liebig University, DE

Interspecific hybridization and polyploidization processes are known to confer advantages such as hybrid vigor and increased environmental tolerances. Within the cultivated diploid and allotetraploid *Brassica* species which contain different combinations of the A, B and C genomes, there is no naturally occurring allohexaploid that incorporates all three genomes (AABBCC). In this study we have combined several *Brassica* allohexaploids from different origins: *B. napus* × *B. nigra*, *B. carinata* × *B. rapa*, *B. juncea* × *B. oleracea*, and (*B. napus* × *B. carinata*) × *B. juncea*. These genotypes exhibit different percentages of pollen viability ranging from 1 - 93%. At the same time, the cross compatibility between both species and genotypes varied, with many of the potential hybrid seeds ending up as aborted embryos. Overall, per 100 buds pollinated, the least fertile cross combination produced less than 1 seed, while the most fertile cross combination produced ¿250 seeds. To date, we have produced 97 new combinations comprising 3881 new hybrid seeds. In future work, a subset of these new hybrids will be sown together with the inbred parental lines to compare chromosome pairing behavior during meiosis and seed setting. At the same time, we are in the process of analyzing chromosome complements and chromosome segregation in the progeny of putatively stable, advanced-generation allohexaploid lines using high-throughput SNP genotyping via the Illumina Infinium *Brasssica* 90K array. With our new available hybrid material we aim to identify genotype- or species specific factors related to fertility and genome stability in *Brasssica* allohexaploids that will be useful for producing a novel, stable crop species.

(49) Genome (in)stability in tetraploid and pentaploid *Festulolium* hybrid populations has implications for breeding and selection

First author: K Van Laere, Flanders Research Institute for Agriculture, Fisheries and Food (ILVO), Plant Sciences Unit, BE

Co-authors: S Meeus, Meise Botanic Garden, BE, J Baert, Flanders Research Institute for Agriculture, Fisheries and Food (ILVO), Plant Sciences Unit, BE, J Aper, Flanders Research Institute for Agriculture, Fisheries and Food (ILVO), Plant Sciences Unit, BE

In order to introduce more abiotic stress tolerance in *Lolium* fodder grasses, we carried out interspecific crosses between tetraploid genotypes of *Lolium perenne* (Lp) or *L. multiflorum* (Lm) and tetraploid *Festuca pratensis* (Fp) or hexaploid genotypes of *F. arundinacea* (Fa). Genomic in situ hybridisation (GISH) on the F1 populations showed expected chromosome numbers and genome compositions. Taken together with their good scores for stress tolerance parameters, *Lolium* x *Festuca* hybrids (=*Festulolium Festulolium*) have great potential to combine stress tolerance with good fodder quality. In a next step, two polycrosses were installed, one with 3 selected F1 genotypes of an Lm x Fa population and one with 5 selected F1 genotypes of an Lp x Fp population. F1 seed yield was normal, while in general F2 seed yield was very low. Chromosome analysis revealed a substantial amount of aberrations in chromosome numbers, which can be the cause of low seed yield. In addition, GISH showed no clear shift to one of the composing genomes in the Lp x Fp population, while genome composition in the Lm x Fa clearly shifted towards the *Lolium* genome. Therefore, to validate the potential of *Festulolium* hybrids, in future breeding it will be important to select first for high seed yielding F2 genotypes. In addition, genome (in)stability will be analyzed also in later generations and implications for breeding and selection will be determined.

(50) Did interspecific hybridisation trigger neopolyploidisation in predominantly polyploid genus Hieracium s.str?

First author: Jan Pinc, Department of Botany, Charles University, CZ

Co-authors: Jindřich Chrtek, Institute of Botany, The Czech Academy of Sciences, CZ, Alexander Belyayev, Institute of Botany, The Czech Academy of Sciences, CZ, Ladislava Pastova, Institute of Botany, The Czech Academy of Sciences, CZ, Patrik Mráz, Department of Botany, Charles University, CZ

It is assumed that interspecific hybridisation might stimulate the formation of neopolyploids. Despite the crucial importance of this speciation process, experimental studies focused on frequency of neopolyploidization following interspecific hybridization are scare. We used control crosses among strictly self-incompatible diploid species of the genus Hieracium s.str. (Compositae), which absolute majority of taxa are polyploid apomicts, to study incidence of neopolyploidization. We performed series of homoploid crosses and produced F1, F2 BC1 and BC2 plants. Subsequently, we estimated seed sets as a proxy of interspecific crossability, evaluated origin of arisen progeny morphologically (true hybrids versus autogamously formed progeny) and determined its ploidy level. In the cases of neopolyploid progeny, we applied cytogenetic approach (GISH and FISH) to determine the genomic dosage of parent species. We observed reduced crossability in crosses where maternal plant or both parents were interspecific hybrids compared to crosses where maternal or both parents were not hybrids. In certain crosses, we also observed a high frequency of induced autogamy (mentor effect). We found 8 neopolyploids out of 3,735 analysed progeny (0.2%) arisen in 6 out of 432 crosses (1.4%). Contrary to our expectation, we did not find increased production of neopolyploids in hybrids and introgressants when compared to parental diploid species. Our results thus showed that neopolyploidization in Hieracium s.str occurs in natural conditions in relatively low rate independently on the origin (hybrid or non-hybrid) of a plant. Nevertheless, the neopolyploids when crossed to diploid taxa frequently produced polyploid progeny.

(51) Identification of PRC1 and PRC2 core components in hexaploid wheat

First author	Beáta Strejčková, Institute of Experimental Botany, Centre of the Region Haná for Biotechnological and Agricultural Research, Olomouc, Czech Republic, CZ
Presenting author	Marek Glombik, Institute of Experimental Botany, Centre of the Region Haná for Biotechnological and Agricultural Research, Olomouc, Czech Republic, CZ
Co-authors	Radim Čegan, Department of Plant Developmental Genetics, Institute of Biophysics, Academy of Sciences of the Czech Republic, 61200 Brno, Czec, CZ, Zbyněk Milec, Institute of Experimental Botany, Centre of the Region Haná for Biotechnological and Agricultural Research, Olomouc, Czech Repub, CZ, Marek Glombik, Institute of Experimental Botany, Centre of the Region Haná for Biotechnological and Agricultural Research, Olomouc, Czech Repub, CZ, Aleš Pečinka, Institute of Experimental Botany, Centre of the Region Haná for Biotechnological and Agricultural Research, Olomouc, Czech Repub, CZ, Jan Šafář, Institute of Experimental Botany, Centre of the Region Haná for Biotechnological and Agricultural Research, Olomouc, Czech Repub, CZ

Polycomb repressive complex 1 and 2 (PRC1, PRC2) are multiprotein complexes which play an important role in developmental stages of plant life through epigenetic gene regulation by chromatin remodelling. PRC1 catalyzes histone H2A monoubiquitination and is able to bind trimethylated lysine 27 of histone H3 whereas PRC2 has methyltransferase activity for lysine 27 of histone H3. In plants, the PRC1 and PRC2 core subunits have been firstly identified in model organism Drosophila melanogaster and *Arabidopsis* thaliana. Here, we describe individual components of these complexes in bread wheat for the first time. In silico identification of PRC1 and PRC2 subunits in hexaploid wheat was supported by gene expression data from RNAseq. Phylogenetic analysis revealed strong sequence conservation of PRC2 subunits among Triticeae wild relatives such as wild emmer wheat, Triticum urartu and Aegilops tauschii. Protein alignment of Triticeae (monocots) and *Arabidopsis* (dicots) PRC2 subunits showed high conservation of protein domains across the plant kingdom.

(52) Expression partitioning and retention of whole genome duplication-derived genes in *Biscutella laevigata*

First author	Marc Beringer, University of Bern, CH
Co-authors	Rimjhim Roy Choudhury, University of Bern, CH, Christian Parisod, University of Bern, CH

Whole genome duplication (WGD) events provide a large amount of duplicated sequences whose evolutionary significance remains elusive. Subsequent long-term genome evolution tends to pseudogenize or delete neutral and maladaptive duplicates in a process known as fractionation. Retained duplicates may thus be expected to represent essential loci involved in adaptive functions through either subfunctionalization, or neofunctionalization. Here we investigate duplicated genes retained in the diploid Biscutella leavigata subsp. austriaca (Brassicaceae) after a WGD event that occurred some 8-MYA to address mechanisms of genome fractionation. Duplicates having evolved under purifying selection appear enriched in functions related to environmental stresses and here we further address to what extent expression partitioning may have supported their retention. Using a high quality transcriptome atlas, comprised of seven individually RNA-sequenced tissues, we identify retained duplicates showing differential expression pattern and assess whether particular functional categories are more likely to show such evidence of subfunctionalization. Such resources for our non-model system support the characterization of mechanisms shaping genomes following recurrent WGD events.

(53) Application of CRISPR/Cas9 to Tragopogon (Asteraceae), an evolutionary model for the study of polyploidy

First author | S Shan, University of Florida, US
Co-authors |

Tragopogon (Asteraceae) is an excellent natural system for studies of recent polyploidy. Development of an efficient CRISPR/Cas9 [U+2010] based genome editing platform in Tragopogon will facilitate novel studies of the genetic consequences of polyploidy. Here, we report our initial results of developing CRISPR/Cas9 in Tragopogon. We have established a feasible tissue culture and transformation protocol for Tragopogon. Through protoplast transient assays, use of the TragCRISPR system was capable of introducing site [U+2010] specific mutations in Tragopogon protoplasts. Agrobacterium [U+2010] mediated transformation with Cas9 [U+2010] sgRNA constructs targeting the phytoene desaturase gene (TraPDS) was implemented in this model polyploid system. Sequencing of PCR amplicons from the target regions indicated simultaneous mutations of two alleles and four alleles of TraPDS in albino shoots from Tragopogon porrifolius (2x) and Tragopogon mirus (4x), respectively. The average proportions of successfully transformed calli with the albino phenotype were 87% and 78% in the diploid and polyploid, respectively. This appears to be the first demonstration of CRISPR/Cas9 [U+2010] based genome editing in any naturally formed neopolyploid system. Although a more efficient tissue culture system should be developed in Tragopogon, application of a robust CRISPR/Cas9 system will permit unique studies of biased fractionation, the gene [U+2010] balance hypothesis and cytonuclear interactions in polyploids. In addition, the CRISPR/Cas9 platform enables investigations of those genes involved in phenotypic changes in polyploids and will also facilitate novel functional biology studies in Asteraceae. Our workflow provides a guide for applying CRISPR/Cas9 to other nongenetic model plant systems.

(54) Analysis of mutations accumulated within the first generations after polyploidization

First author | Vidya Oruganti, Max Planck Institute for Plant Breeding Research, DE
Co-authors | Helena Toegelová, Institute of Experimental Botany of the Czech Academy of Sciences, CZ, Hana Šimková, Institute of Experimental Botany of the Czech Academy of Sciences, CZ, Aleš Pečinka, Institute of Experimental Botany of the Czech Academy of Sciences, CZ, Andreas Madlung, University of Puget Sound, US, Korbinian Schneeberger, Max Planck Institute for Plant Breeding Research, DE

We investigated the genomic changes that occurred during the first ∼15 generations after a spontaneous whole-genome duplication event. This unexpected WGD occurred in a triploid hybrid plant produced by crossing the natural allotetraploid *Arabidopsis* suecica (ÁÁBB) with *Arabidopsis* thaliana Col-0 (AA) (Matsushita et al., 2012). This generated a plant with a complex autoallopolyploid karyotype (AAÁÁBB; 2n=6x=36; 4n=20, 2n=16) consisting of two copies of the *A. arenosa* genome (BB) and four copies of the *A. thaliana* genome (AAÁÁ). The progeny of this hexaploid were used to initiate a few dozen mutation accumulation lines which were propagated via single seed descent of up to 17 generations. First we generated a chromosome-level assembly of the allopolyploid genome of *A. suecica* using long reads, optical mapping and homology to existing reference sequences of the subgenomes (scaffold N50 of 5.71 Mb). This reference was then used for a resequencing analysis of progeny genomes sampled from five mutation accumulation lines. A coverage-based optimization algorithm was used to determine chromosome gain or loss events in the subsequent generations of hexaploids and was independently validated by the frequency of SNP alleles. Already within the first generations we observed large-scale mutations including whole chromosome loss/gain, genetic rearrangements and exchanges between subgenomes. Some of these changes were fixed in later generations, however, we could not observe genome stabilization as some of the large-scale mutations still occurred after 15 generations. Current analyses include mutation rate estimation and transposable element movement.

(55) Variation for fertility and chromosome rearrangements in a diverse set of resynthesized *Brassica napus* allotetraploid lines

First author: Elizabeth Ihien, Department of plant breeding Justus Liebig University Giessen, DE
Co-authors: Schierholt Antje, Department of crop sciences Georg-August University Goettingen, DE, Heiko C. Becker, Department of crop sciences Georg-August University Goettingen, DE, Rod Snowdon, Department of plant breeding Justus Liebig University Giessen, DE, Annaliese S. Mason, Department of plant breeding Justus Liebig University Giessen, DE

Rapeseed (*Brassica napus*, AACC) is a young allotetraploid species formed by the hybridization of *Brassica rapa* (turnip, AA) and *Brassica oleracea* (European cabbage, CC). However, resynthesized *B. napus* lines are often highly unstable and infertile, unlike natural *B. napus*. Meiotic stability in natural *B. napus* may have arisen through allele inheritance from the progenitor species or via one or more de novo mutations post-polyploidisation. We aimed to test these hypotheses by characterizing a diverse set of resynthesized lines for chromosome rearrangements, allele inheritance, fertility and meiotic behaviour. SNP genotyping was performed using the Illumina Infinium *Brasssica* 60K array, and allele copy number used to infer translocation events between the A and C genomes. Approximately 52% of lines (91/174) with SNP genotyping information were homozygous as expected. Average pollen viability in the population was 87% (ranging from 0 to 100%). Self-pollinated seed-set (average 611, range 0 – 3876 per plant) was significantly influenced by both parental genotypes and their interaction. Genome-wide across the population, 663 copy number variants were observed, with loss or gain of a single copy of a chromosomal region roughly twice as common as loss or gain of both copies (duplication or deletion events). Overall, 39% of lines showed clear evidence of unbalanced translocations between the A- and C-genomes. These results suggest that allelic variation present in both of the diploid parents interacts to affect the chance of chromosome rearrangement and copy number variation events, but that the presence of such events may not always be detrimental to fertility in synthetic *B. napus* lines.

(56) Polyploidy in lime confers better tolerance to Huanglongbing

First author: G Sivager, CIRAD, GP
Presenting author: R Morillon, CIRAD, GP
Co-authors: L calvez, CIRAD, GP, S Bruyère, CIRAD, GP, R Boisne Noc, CIRAD, GP, P Brat, CIRAD, GP, O Gros, Université des Antilles, GP, P Ollitrault, CIRAD, FR

Huanglongbing (HLB) is nowadays the major threat of the citrus industry. Because of this disease, millions of trees are currently dying worldwide. The causative agent is a motile bacteria belonging to Candidatus Liberibacter spp. which is transmitted by psyllids. HLB is responsible of the synthesis of callose at sieve plate of the phloem leading to the obstruction of the pores that provide connection between adjacent sieve elements, thus limiting the symplastic transport of sugars and starch synthesized in leaves to the other organs of the plants. Diploid (2x, Citrus aurantiifolia) and triploid (3x, Citrus latifolia) limes were investigated when infected by HLB. Leaf petiole was analyzed using Scanning Electron Microscope (SEM) to observe callose deposition at sieve plate of the phloem. Leaf starch contents and detoxification enzyme activities in 2x and 3x leaves were investigated. In the field, leaves of 3x lime present more limited symptoms due to HLB than 2x. Photosynthesis, stomatal conductance and transpiration decreased compared to control plants but values remained greater in 3x than in 2x. Analysis of the petiole sieve plate in control petiole samples shown that pores were about 30% larger in 3x than in 2x. SEM analysis of petiole samples of symptomatic leaves, shown important deposition of callose onto 2x and 3x pores, while biochemical analysis revealed similar behavior regarding detoxification in 3x and in 2x. SEM analysis of infected petiole samples of asymptomatic leave showed much larger callose deposition onto 2x than in 3x pores while biochemical traits revealed much better behavior in 3x than in 2x. Our results provide the first insights regarding the better tolerance of citrus polyploid to HLB.

(57) Heteroarthrocarpic fruit development in allotetraploid *x Brassicoraphanus*, an intergeneric hybrid of *Brasssica rapa* and *Raphanus sativus*

First author: Jeong Eun Park, Seoul National University, KR
Co-authors: Gibum Yi, Seoul National University, KR, Hosub Shin, Seoul National University, KR, Hye Rang Park, Seoul National University, KR, Soo-Seong Lee, BioBreeding Institute, KR, Jin Hoe Huh, Seoul National University, KR

Hybridization between different species or genera is a rare event, but occasionally produces novel plant species with neomorphic phenotypes. Although hybrids from interspecific or intergeneric crosses are generally hard to obtain, xBrassicoraphanus was successfully produced as a newly synthesized intergeneric allotetraploid from a cross between Chinese cabbage (*Brasssica rapa* L.) and radish (*Raphanus sativus* L.). One of the notable characteristics of xBrassicoraphanus is a composite structure of the pistil and silique, in which distinct features of both Chinese cabbage and radish are expressed together. Chinese cabbage has a dehiscent silique with two valves, which are shattered to disperse seeds when dried. By contrast, radish has an indehiscent silique with no valves, and thus shattering does not occur. The proximal segment of xBrassicoraphanus silique resembles that of Chinese cabbage, which consists of the valves attached to the replum, whereas the distal segment has a radish-like structure without valves. A transverse cleft appears at the junction between proximal and distal segments, forming a joint region structure. Therefore, the xBrassicoraphanus fruit is regarded as heteroarthrocarpic, with both dehiscent and indehiscent features on a single silique. In this study, we identified dehiscence genes in both Chinese cabbage and radish, and comparatively analyzed their expressions in xBrassicoraphanus. Significant alterations in expression of duplicate genes occurred in xBrassicoraphanus fruits, conceivably responsible for heteroarthrocarpy. This work shows an example of how genome hybridization contributes to the advent of neomorphic trait by reconstruction of transcription profiles in the hybrid genome.

(58) Combined effects of salinity and inundation on phosphoenolpyruvate carboxylase activity in two *Spartina* species and its hybrid

First author: Blanca Gallego-Tevar, University of Sevilla, ES
Co-authors: Procopio Peinado-Torrubia, University of Sevilla, ES, *Rosa*rio Álvarez, University of Sevilla, ES, Brenda J. Grewell, USDA-University of California Davis, US, Jesús M. Castillo, University of Sevilla, ES

Interspecific hybridization and polyploidy are linked to improved invasiveness of plant species in relation to higher competitiveness, tolerance to abiotic factors and phenotypic plasticity. However, little is known about their effect on enzymatic activities in response to environmental stress factors. Polyploid plant taxa resulting from natural hybridization represent an opportunity for the study of these evolutionary mechanisms in situ. Sea Level Rise (SLR) associated with climate change is expected to intensify the period of permanent submersion and salinity in coastal salt marshes. In this context, the study of the responses of native and invasive halophytic plant species to SLR is essential to maintain their conservation status. With this aim, we assessed the effect of inundation depth and salinity on phosphoenolpyruvate carboxylase (PEPC) for three species of the polyploid cordgrass taxa of *Spartina* (S. densiflora ($2n = 7x = 70$); S. foliosa ($2n = 6x = 60$) and S. densiflora x foliosa ($2n = 6.5x = 65$) in a greenhouse experiment. PEPC is key enzyme involved in the photosynthetic C4 metabolism,. We also measured oxidative stress by recording malondialdehyde (MDA) and free proline contents in flag leaves of the three taxa. A general improvement of PEPC phosphorylation in vivo was found at deep flooding conditions. A different behaviour for S. densiflora in comparison with another ecotype from SW Iberian Peninsula was observed. These results pointed out the relevance of heterosis and non-additive gene regulation in the process of hybridization of these *Spartina* taxa, bringing about transgressive traits in S. densiflora x foliosa.

(59) Consequences of polyploidy on the floral polymorphism of Linum suffruticosum s.l.

First author: Ana Afonso, Centre for Functional Ecology, Department of Life Sciences, University of Coimbra, Portugal; Departamento de Biología Vegetal y Ecología, Universidad de Sevilla, Spain, PT

Co-authors: Erika Olmedo-Vicente, Departamento de Biología Vegetal y Ecología, Universidad de Sevilla, Spain, ES, Juan Arroyo, Departamento de Biología Vegetal y Ecología, Universidad de Sevilla, Spain, ES, João Loureiro, Centre for Functional Ecology, Department of Life Sciences, University of Coimbra, Portugal, PT, Marie Cordier, Centre for Functional Ecology, Department of Life Sciences, University of Coimbra, Portugal, PT, Sílvia Castro, Centre for Functional Ecology, Department of Life Sciences, University of Coimbra, Portugal, PT

The consequences of polyploidy on gene expression may lead to morphological, reproductive and physiological shifts. Among the direct effects of polyploidy is an increase in cell size and, potentially, in the overall size of the plant's organs. This may have a significant effect on the structure of flowers and its sexual organs, with possible effects in the reproductive success of polyploid individuals. Such changes in reproductive traits due to polyploidy will be particularly relevant in species with complex breeding systems, such as heterostylous species. Linum suffruticosum s.l., is diploid-polyploid complex distributed through the Mediterranean Basin, bearing five main cytotypes (diploids, tetraploids, hexaploids, octoploids and decaploids), and comprising heterostylous individuals with a strong self-incompatible reproductive system. The objective of this study was to evaluate if there are differences in floral traits among cytotypes, and if these differences might mediate reproductive isolation among each cytogenetic entity. For that, we sampled flowers of 90 populations comprising all the cytogenetic entities and measured several floral traits (e.g., corolla, anthers and pistil lengths). We found that the size of sexual organs and the length differences between styles and anthers increase with increased ploidy level; nevertheless, such increase was more evident for the long style morph than in the short style morph. A high variation in sexual organs size was found in all cytotypes which is reflected in lower reciprocity indexes. The differences in flower morphology and the occurrence of polymorphisms within and among all cytogenetic entities is discussed in the context of a highly complex polyploid system.

(60) Parental genome divergence allows the stabilization of hybrid genome of xBrassicoraphanus

First author: Hosub Shin, Seoul National University, KR

Co-authors: Gibum Yi, Seoul National University, KR, Jeong Eun Park, Seoul National University, KR, Hye Rang Park, Seoul National University, KR, Soo-seong Lee, Biobreeding institute, KR, Hyun Hee Kim, Sahmyook University, KR, Jin Hoe Huh, Seoul National University, KR

The Brassicaceae family includes many vegetable crops with great morphological diversity. It is speculated that interspecific hybridization and subsequent genome duplication have facilitated the evolution of many plant species, while producing novel characteristics to increase fitness to the new environment. xBrassicoraphanus ($2n = 4x = 38$, AARR) is an intergeneric allotetraploid synthesized from a cross between Chinese cabbage (*Brasssica rapa*; $2n = 2x = 20$, AA) and radish (*Raphanus sativus*; $2n = 2x = 18$, RR). Unlike most neoallopolyploid plants, xBrassicoraphanus is fertile and genetically stable, with many characteristics displayed as mixed phenotypes between the parents due presumably to incomplete dominance and enormous changes in transcriptome profiles. We showed that both genomes of B. rapa and R. sativus reside in xBrassicoraphanus without apparent chromosome rearrangement. In addition, a large portion of duplicated genes was shown to undergo significant changes in expression levels, indicating that massive reconstruction of transcription control network has occurred after a merger of two divergent genomes. Intergeneric hybridization also involves significant changes in genome-wide DNA methylation, small RNA, and histone modification profiles. Our work demonstrates that a certain level of parental genome divergence is helpful to suppress genome shuffling, and that, after hybridization, the hybrid genome is stabilized by the adjusted expressions of duplicated genes and silencing of transposable elements while ameliorating both transcriptome and genomic shocks.

(61) Genome dominance in allopolyploid *Festulolium* (Festuca × *Lolium*) revealed by gene expression analysis

First author: Marek Glombik, [1]Institute of Experimental Botany, Centre of the Region Haná for Biotechnological and Agricultural Research, Olomouc, Czech Republic, CZ

Co-authors: Štěpán Stočes[1], Steven Yates, Institute of Agricultural Sciences, Molecular Plant Breeding, ETH Zurich,8092 Zurich, Switzerland, CH, Tom Ruttink, Institute for Agricultural and Fisheries Research (ILVO), Plant Sciences Unit – Growth and Development, Caritasstraat 39, 9090 M, BE, Bruno Struder, Institute of Agricultural Sciences, Molecular Plant Breeding, ETH Zurich,8092 Zurich, Switzerland, CH, Jan Bartoš[1], Zbigniew Zwierzykowski, Laboratory of Cytogenetics and Molecular Biology, Institute of Plant Genetics, Polish Academy of Sciences, Strzeszyńska 34, 60–4, PL, Tomasz Ksiazczyk, Laboratory of Cytogenetics and Molecular Biology, Institute of Plant Genetics, Polish Academy of Sciences, Strzeszyńska 34, 60–4, PL, Jaroslav Doležel[1], David Kopecký[1]

Interspecific hybridization offers a unique opportunity to combine traits of agricultural interest from two species, or even genera, into a single organism. However, the merge of two or more genomes from different species via interspecific hybridization causes major changes at the genetic and epigenetic levels, with important consequences for the expression patterns of genes from both genomes. It was evidenced in several cases that one of the parental genomes in interspecific hybrids takes a lead. This "genome dominance" can be observed in various forms and in all cases leads to the elimination of the "weaker" parental genome from the hybrid. *Festulolium* (*Festuca* × *Lolium*) hybrids display gradual replacement of Festuca chromosome by those of *Lolium* in successive generations. The aim of our project was to analyze potential genome dominance on the level of RNA. Using RNAseq, we were able to identify SNPs distinguishing the parental alleles of orthologous genes. This enabled us to analyze expression level dominance (overall expression in hybrids compare to the expression in their parents) and homoeologous expression bias (the contribution of both parental alleles to the overall expression of the particular gene).

(62) Climatic niche comparison among ploidal levels in the classic autopolyploid system, Galax urceolata

First author: Michelle Gaynor, University of Florida, US

Co-authors: D. Blaine Marchant, University of Florida, US, Douglas E. Soltis, University of Florida, US, Pamela S. Soltis, University of Florida, US

Polyploidy, or whole-genome duplication, is a major evolutionary force in angiosperms. Polyploidy may result from genome duplication within a species (autopolyploidy) or via duplication associated with hybridization between species (allopolyploidy). Unlike allopolyploids, autopolyploids were historically considered very rare in nature and maladaptive. However, recent studies have increased awareness of the importance and frequency of autopolyploids, which may actually be as common as allopolyploids. Despite the increased awareness, autopolyploids remain drastically understudied. In particular, although autopolyploids may often form recurrently from the same diploid parent, almost nothing is known about the frequency of recurrent formations or the factors that drive them or the populations resulting attributes. That is, how do populations of independent formation compare in genomic attributes and niche space? To address these major current gaps in the literature, we investigated the ecological consequences associated with recurrent formations of autopolyploids in populations of mixed ploidy. An ideal study organism for this objective is the flowering plant Galax urceolata, an endemic to the southern Appalachians which includes diploid, triploid, and autotetraploid cytotypes. Multiple independent origins of the autotetraploid has resulted in morphologically indistinguishable populations. We found minimal niche divergence among cytotypes, but further work is needed to unravel the evolutionary repercussions their recurrent formations. In regard to climate change, we predict all cytotypes will face extensive range loss. Galax urceolata may therefore be under extreme threat due to loss of suitable habitat.

(63) Do reproductive barriers facilitate cytotype coexistence in Gladiolus communis contact zones?

First author	Mariana Castro, CFE, Centre for Functional Ecology and Department of Life Sciences, University of Coimbra, PT
Presenting author	Mariana Castro, CFE, Centre for Functional Ecology and Department of Life Sciences, University of Coimbra, PT
Co-authors	João Loureiro, CFE, Centre for Functional Ecology and Department of Life Sciences, University of Coimbra, PT, Brian Husband, Department of Integrative Biology, University of Guelph, CA, Sílvia Castro, CFE, Centre for Functional Ecology and Department of Life Sciences, University of Coimbra, PT

Polyploidy is considered an important mechanism of sympatric speciation. However, to successfully establish, the new polyploids must overcome strong frequency-dependent selection. Assortative mating is known to increase polyploid fitness within parental populations, allowing it to overcome the numerical disadvantage. To understand how assortative mating influences cytotype coexistence, we assessed several reproductive barriers (phenological, morphological, behavioural, selfing and gametic) between tetraploid and octoploid Gladiolus communis in a contact zone in Western Iberian Peninsula and quantified the reproductive isolation (RI) between cytotypes. Controlled pollinations were performed to assess self-incompatibility differences and quantify the production of hexaploid hybrids under pure- and mixed-ploidy pollen loads. We discovered that, the pre-pollination barriers were weak enabling pollen flow between cytotypes in mixed-ploidy arrays; by opposition, post-pollination isolation depended on the pollen load composition. While high rates of hexaploid formation were detected after inter-cytotypes crosses, high post-pollination isolation was observed after mixed-ploidy pollinations. Post-pollination RI was always higher in tetraploids than in octoploids, with self-pollination playing an important role in driving RI. Overall, a high cumulative RI was observed for both cytotypes. The production of intermediate hexaploids and unreduced gametes and the higher reproductive success after selfing of the octoploids are some of the factors that may enable the its successful establishment. Furthermore, cytotype coexistence in sympatry could be favoured by strong post-pollination barriers, resulting in dynamic contact zones.

(64) Nonsyntenic homoeologs contribute to the functional repertoire of Gossypium hirsutum

First author	Natasha Glover, University of Lausanne, CH
Co-authors	Shaoline Sheppard, University of Lausanne, CH, Christophe Dessimoz, University of Lausanne, CH

Homoeologs are corresponding genes between subgenomes in an allopolyploid, and studying them can provide insight into the evolutionary consequences of polyploidization. Here, we focus on the allotetraploid Gossypium hirsutum, or upland cotton, in which the progenitors diverged ∼5 MYA, followed by a hybridization event ∼1-2 MYA.

We used homoeolog predictions from Orthologous MAtrix (OMA) to classify pairs of homoeologs based on their synteny. Nonsyntenic homoeologs are those pairs where one gene has been translocated to a different area of the genome, whereas syntenic homoeologs are those that have stayed in their ancestral positions. In this comparative genomics study, we found that nonsyntenic homoeologs have characteristics of pseudogenes compared to syntenic homoeologs (higher synonymous and nonsynonymous substitution rates, shorter protein length, fewer exons, more likely to be duplicated, located in recombination coldspots, and have fewer orthologs). However, these nonsyntenic pairs also have a low dN/dS, indicating they are under purifying selection. Additionally, most nonsyntenic pairs are highly and broadly expressed, and are enriched for GO functions associated with protein translation.

These results show that nonsyntenic and duplicated genes are contributing to the functional repertoire of cotton and provide insight into gene movement in an allopolyploid.

(65) Interploidy crosses in Viola L. enrich biodiversity by formation of hybrids, introgressants and new species

First author: J Żabicka, Jagiellonian University, Department of Plant Cytology and Embryology, Cracow, Poland, PL

Co-authors: E Sliwinska , UTP University of Science and Technology, Laboratory of Molecular Biology and Cytometry, Bydgoszcz, Poland, PL, A Słomka, Jagiellonian University, Department of Plant Cytology and Embryology, Cracow, Poland, PL, G Migdałek, Pedagogical University of Cracow, Institute of Biology, Cracow, Poland, PL, P Żabicki, Jagiellonian University, Department of Plant Cytology and Embryology, Cracow, Poland, PL, T Marcussen, University of Oslo, CEES, Department of Biosciences, Oslo, Norway, NO, E Kuta, Jagiellonian University, Department of Plant Cytology and Embryology, Cracow, Poland, PL

Interploidy crosses are common in Viola L. genus. We investigated pairs of tetraploids and octoploids commonly hybridizing in nature: V. reichenbachiana Jordan ex Bor. (2n=4x=20), V. uliginosa Bess. (2n=4x=20) with V. riviniana Rchb. (2n=8x=40), all belonging to subsect. Rostratae (Kupffer)W.Becker of Viola section and also hybrids between V. epipsila Ledeb. (2n=4x=24) and V. palustris L. (2n=8x=48) from Plagiostigma Godr. section. First generation of hexaploid hybrids have reduced fertility due to disturbances in female gametophyte and pollen development and in embryo formation. The vegetative propagation of hybrids by stolons or rhizomes compensates impaired generative reproduction. For rare and endangered species it is an escape from total gene loss. Their genes are conserved in hybrids which are more tolerant to environmental changes and loss of species niches. Hexaploid hybrids (2n=6x=36) between V. epipsila and V. palustris have average genome size (DNA=3.4 pg/2C) being the sum of haploid genomes of both parental species (2.49 and 4.19 pg/2C, respectively), indicating lack of genome upsizing or downsizing in their evolution. Hybrid derived species Viola pubifolia (Kuta)G.H.Loos (2n=8x=48) likely originated via unidirectional introgression (V. epipsila x V. palustris) towards V. palustris, has the same genome size as V. palustris. Evolution of V. palustris was not accompanied by changes in genome size, its both subspecies (ssp. palustris and juressii) with different geographical distribution, represent the same genome size (4.19 pg/2C, 4.22 pg/2C, respectively). This is in contra to the general statement that hybridization and polyploidization might induce rapid genomic changes - DNA gain or loss.

(66) MEIOTIC BEHAVIOR OF DOUBLED-DIPLOID CITRUS PLANTS

First author: Pablo Aleza, IVIA, ES

Co-authors: Houssem Rouiss, IVIA, ES, Jose Cuenca, IVIA, ES, Andres Garcia-Lor, IVIA, ES, Luis Navarro, IVIA, ES, Patrick Ollitrault, CIRAD, FR

The two main mechanisms of citrus polyploid formation are somatic doubling of chromosomes and meiotic nuclear restitution leading to unreduced gamete. In citrus, doubled-diploid (DD) plants were identified in seedlings of diploid apomictic genotypes and they arise from spontaneous duplication of chromosomes in nucellar cells. Artificial DD plants have also been produced with antimitotic chemicals like colchicine. Genomic studies revealed that most of the modern citrus varieties resulted from admixture of four ancestral taxa: Citrus reticulata (mandarin), C. maxima (pummelo), C. medica (citron) and C. micrantha (papeda). DD plants arising from direct interspecific crosses are allotetraploid while duplication from more complex admixture genomes results in segmental allotetraploidy. With DD parents, heterozygosity restitution depends on the extent of preferential chromosome pairing. Allo- and autotetraploids (with disomic and tetrasomic inheritance, respectively) are the extremes of the range. In cases where parents are divergent but have retained enough homology to prevent exclusive preferential pairing, inheritance patterns intermediate between di- and tetrasomic can be expected. We have analyzed the meiotic behavior of two contrasted DDs genotypes, Clementine mandarin with large genome regions fully inherited from mandarin and Mexican lime resulting from direct hybridization between C. micrantha and citron. DD clementine displayed predominant tetrasomic inheritance although three linkage groups (LG) presented intermediate inheritance and one LG predominant disomic inheritance. Mexican lime had predominantly disomic segregation but interspecific recombinations were not precluded, even strongly limited.

(67) Cytonuclear coordination and evolution in allopolyploids

First author: Joel Sharbrough, Colorado State University, US
Co-authors: Justin L. Conover, Iowa State University, US, Corrinne E. Grover, Iowa State University, US, Emma R. Miller, Iowa State University, US, Jonathan F. Wendel, Iowa State University, US, Daniel B. Sloan, Colorado State University, US

Whole-genome duplications (WGDs), in which the number of nuclear genome copies is elevated as a result of autopolyploidy or allopolyploidy, underlie many of the major episodes of diversification in eukaryotes. The genetic and evolutionary forces that WGD imposes upon cytoplasmic genomes are not well understood, despite the central role that cytonuclear interactions play in eukaryotic function and fitness. In particular, cellular respiration and photosynthesis depend upon successful interaction between the 3000+ nuclear-encoded proteins destined for the mitochondria or plastids and the gene products of cytoplasmic genomes in multi-subunit complexes such as Rubisco, OXPHOS, Photosystems I and II, and organellar ribosomes. Allopolyploids are thus faced with the critically important task of maintaining successful interactions and coordinated gene expression between nuclear and cytoplasmic genes that were inherited from different species. Because maternal homoeologs are expected to be more closely "matched" to cytoplasmic genomes than paternal homoeologs, incompatibilities between the organelle genomes and paternal subgenomes of allopolyploids may lead to relaxed selection on paternal vs. maternal homoeologs of genes targeted to the mitochondria or plastids. To test this hypothesis, we compared rates of molecular evolution in maternal vs. paternal homoeologs of organelle-targeted genes in the allotetraploids Gossypium hirsutum (Malvaceae) and Triticum dicoccoides (Triticeae). This global assessment of cytonuclear coevolution in diploid vs. polyploid angiosperms provides powerful insights into the molecular dynamics of cytonuclear incompatibilities that are likely to influence the success and evolution of hybrid polyploids.

(68) Eco-genetic additivity of diploids in allopolyploid wild wheats

First author: C. Parisod, University of Bern, CH
Co-authors:

Underpinnings of the distribution of diploids and allopolyploids in space and along ecological gradients are elusive. Allopolyploidy expectedly yields species with divergent ecological niches to escape competition from diploid progenitors, but departure from genetic and ecological additivity remains to be tested. Here, four diploid wild wheats that differentially combined into four allopolyploid species are used to assess the impact of historical and ecological constraints on species ranges. Genetic variation relating diploid progenitors to allopolyploids supports their genetic additivity. Comparative phylogeography and modelling of climatic niches further support ecological additivity of locally adapted diploid progenitors into allopolyploid species that expanded to become widespread. Diploids further occupy only a small fraction of their potential distribution, whereas allopolyploids largely fill suitable range with specific lineages. Genetic and ecological additivity thus promote the expansion of such polyploid species under environmental changes. The apparent paradox between such conservative evolution and the patent diversification of wild wheats under the influence of transposable elements will be discussed.

(69) Distinguishing successive ancient polyploidy levels based on genome-internal syntenic alignment

First author	David Sankoff, University of Ottawa, CA
Presenting author	Yue Zhang, University of Ottawa, CA
Co-authors	Yue Zhang, University of Ottawa, CA

Background: A basic tool for studying the polyploidization history of a genome, especially in plants, is the distribution of duplicate gene similarities in syntenically aligned regions of a genome. Often there are two or more peaks, each representing a different polyploidization event. These distributions may be generated by means of a discrete time, non-homogeneous branching process, followed by a standard sequence divergence model. While the similarities data allows for inference of fractionation rates and other parameters they usually cannot pin down the ploidy level of each event. Methods: For a sequence of two events of unknown ploidy, either tetraploid or hexaploid, we base our analysis on high-similarity triples of genes – triangles. We calculate the probability of the four triangle types with origins in one or the other event, and impose a mutational model so that the distribution resembles the original data. Using a ML transition point in the similarities between the two events as an discriminator for the hypothesized origin of each similarity, we calculate the predicted number of triangles of each hypothesized type for each model combining hexaploidization and/or tetraploidization. This yields a profile of triangle type for each model. Results: We use the model profiles to identify the polyploidization history of a number plant genomes.

(70) Cytogeographic Distribution of *Spartina* (Poaceae) Taxa in the European Wadden Sea Area

First author	Dirk Granse, Institute of Plant Sciences and Microbiology, Universität Hamburg, DE
Co-authors	Sigrid Suchrow, Institute of Plant Sciences and Microbiology, Universität Hamburg, DE, Kai Jensen, Institute of Plant Sciences and Microbiology, Universität Hamburg, DE

Whole genome duplication (WGD) is one of the most striking mechanisms of evolution in plants, particularly in the radiation of angiosperms. Polyploids were shown to be more vigorous in comparison to diploids and capable of inhabiting more extreme environments with generally increasing relative frequency from the equator to the poles. However, the effect of polyploidy on the ecology and the geographic distribution is often not clearly recognizable or confusing because in older polyploid taxa, the effect of ploidy can not be distinguished from the effect of selective evolutionary forces. The objectives of this study are to identify niche-responses and cytogeographic distribution pattern in a relatively young taxa-group of a hexaploid F1-hybrid (*Spartina* × townsendii H. Groves & J. Groves) and the allododecaploid descended of the hybrid (*Spartina* anglica C.E. Hubbard). Both taxa hardly underwent speciation since they emerged. We investigated the geographic distribution of more than 400 *Spartina* clones in more than 1000 vegetation plots in salt marshes along the coast of the European Wadden Sea. We measured elevation in each plot as a niche-proxy to account for the stress gradient from mudflats, via pioneer zones to high marsh communities. Flow-cytometry was used to differentiate between hexaploid and dodecaploid cytotypes and HOF-modeling to describe the ecological niches of the two taxa. The dodecaploid cytotype was evenly distributed along the whole coast of the study area. Its niche optimum according to elevation was in the pioneer zone. By contrast, the hexaploid cytotype was only present at a few sites along the coastline and restricted to elevational ranges of the mudflat with more severe abiotic conditions.

(71) Interspecific hybridisation facilitates niche adaptation in beer yeast

First author: B Gallone, VIB, BE
Co-authors: J Steensels, VIB, BE, S Mertens, VIB, BE, MC Dzialo, VIB, BE, JL Gordon, VIB, BE, R Wauters, VIB, BE, FA Theßeling, VIB, BE, F Bellinazzo, VIB, BE, V Saels, VIB, BE, B Herrera-Malaver, VIB, BE, T Prahl, White Labs Inc., US, C White, White Labs Inc., US, M Hutzler, TU München, DE, F Meußdoerffer, TU München, DE, P Malcorps, AB-InBev SA/NV, BE, B Souffriau, AB-InBev SA/NV, BE, L Daenen, AB-InBev SA/NV, BE, G Baele, Rega Institute, BE, S Maere, VIB, BE, KJ Verstrepen, VIB, BE

Hybridisation between species often leads to inviable or infertile offspring, yet examples of evolutionary successful interspecific hybrids have been reported in all kingdoms of life. However, many questions on the ecological circumstances and evolutionary aftermath of interspecific hybridisation remain unanswered. In this study, we sequenced and phenotyped a large set of interspecific yeast hybrids isolated from the brewing environments to uncover the influence of interspecific hybridisation in yeast adaptation and domestication. Our analyses demonstrate that several hybrids between Saccharomyces species originated and diversified in industrial environments by combining key traits of each parental species. Newly formed hybrids experienced extensive genome reorganization resulting in severe aneuplodies: we found significant variation within and between hybrid types in overall ploidy, copy number of large chromosomal fragments and full chromosomes, as well as in the degree of parental species contribution to the hybrid genome. Furthermore, post-hybridisation evolution within each hybrid lineage reflects sub-specialisation and adaptation to specific beer styles, a process that was accompanied by extensive chimerisation between subgenomes. Our results reveal how interspecific hybridisation provides an important evolutionary route that allows swift adaptation to novel environments.

(72) Polyploidy in a phylogenetic context and correlation with the geography distribution of Cactaceae species in South America

First author: karen Bauk, IMBIV-CONICET, AR
Co-authors: Maria Laura Las Peñas, IMBIV-CONICET, AR

Cactaceae are an important floristic component of arid and semiarid areas of America and the hybridization and polyploidy are the major evolutionary forces in the family. Ploidy levels are often correlated with morphological and geographic distribution, and are crucial to the evolution and systematics of the family. The great diversification in Cactaceae has been associated with polyploidy and few chromosome rearrangements visible with conventional staining, i.e., large duplications, pericentric inversions, and reciprocal translocations of segments of unequal size. In this sense, cumulative small and cryptic structural changes are proposed to play an important karyoevolutionary role in Cactaceae. The objective of this work is to see the origin and how polyploidy events relate to the species distribution. The ancestor would have these characters: diploid (2n=22, x=11), with a small genome. Chromosome numbers in Cactaceae are nearly always a multiple of eleven (x=11). We also present a review of chromosome counts reported for Cactaceae species in South America. Ploidy in these taxa ranged from diploid, 2n=2x=22 to nonaicosaploid, 2n=29x=319. Of the 875 species of Cactaceae in South American, only chromosome counts have been carried out for 23 %, (wich 26.2% are diploid, 13.4% are both diploid and polyploid, and 60.4% are polyploidy) confirming that the frequency of genome duplication in the group is far more common than diploidy. From the ecological point of view, the ability of polyploids to thrive under rigorous conditions is known. A correlation between polyploidy and aridity was determined in each of the subfamilies. Finally, a greater number of polyploid species was observed in extreme environments of aridity and cold

(73) Identification of genome-dosage sensitive genes controlling seed size in *Arabidopsis* thaliana

First author	R Castillo Bravo, Genetics & Biotechnology Lab, Plant & AgriBiosciences Research Centre (PABC), Ryan Institute, National University of Ireland Galway, University Road, Galway H91 REW4, Ireland., IE
Presenting author	R. Castillo Bravo, Genetics & Biotechnology Lab, Plant & AgriBiosciences Research Centre (PABC), Ryan Institute, National University of Ireland Galway, University Road, Galway H91 REW4, Ireland., IE
Co-authors	A. Fort, Genetics & Biotechnology Lab, Plant & AgriBiosciences Research Centre (PABC), Ryan Institute, National University of Ireland Gal, IE, G. Brychkova, Genetics & Biotechnology Lab, Plant & AgriBiosciences Research Centre (PABC), Ryan Institute, National University of Ireland Gal, IE, C. Spillane, Genetics & Biotechnology Lab, Plant & AgriBiosciences Research Centre (PABC), Ryan Institute, National University of Ireland Gal, IE

Heterosis (or hybrid vigour) refers to improvement in yield or other characteristics in the F1 offspring relative to parents. Heterosis is an essential tool for increasing yields per unit area for the world's crops. Despite its economic importance, the mechanistic basis of heterosis remains largely unknown. Heterosis for F1 seed size can occur when genetically different parental lines of *Arabidopsis* thaliana are crossed together. Heterosis-like genome dosage effects on F1 seed size can also be elicited by crossing genetically identical parental lines that differ only according to ploidy. However, the genes underlying such genome dosage effects on F1 seed size are largely unknown. To identify loci responsible for maternal genome dosage effects on triploid F1 seed size, a genome wide association study (GWAS) mapping experiment was performed with a diverse panel of F1 hybrid triploids. To generate the panel of 182 different F1 hybrid triploids, 182 *Arabidopsis* thaliana diploid (2x) accessions were used as pollen donors to pollinate a tetraploid (4x) tester line in a Ler-0 genetic background. Each of the 182 F1 hybrid triploid progeny had a maternal genome dosage excess consisting of 2 sets of maternal chromosomes and 1 paternal set (2m:1p). The Genome-Wide Association mapping conducted with the 182 F1 hybrid triploids identified a number of genomic regions that are being pursued by functional experiments to identify genome-dosage sensitive loci controlling F1 seed size.

(74) Speciation events, not time, explain patterns of gene loss following whole genome duplication in catostomid fishes

First author	Trevor Krabbenhoft, University at Buffalo, US
Presenting author	Trevor Krabbenhoft, University at Buffalo, US
Co-authors	Tianying Lan, University at Buffalo, US, Thomas Dowling, Wayne State University, US

Catostomid fishes (suckers) diverged from diploid cyprinid relatives following an ancestral whole genome duplication event approximately 100 million years ago. Catostomids retain a duplicated karyotype (2n=100 in suckers versus 2n=50 in most cyprinids), and, in striking contrast to other polyploid fishes (e.g., salmonids), tetrasomy has not been observed. Based on these observations, previous researchers have suggested that suckers are allotetraploids resulting from inter-specific hybridization, a common occurrence in cypriniform fishes. We assembled reference-quality genomes of Chinese sucker (Myxocyprinus asiaticus) – the sister lineage of all other catostomids, along with a morphologically-derived species (razorback sucker, Xyrauchen texanus) using nanopore long-read sequencing. We compare patterns of retention of duplicated genes across these species to test whether derived lineages express fewer genes in duplicate, despite having identical time since polyploidization. We also examine whether there are genomic hot spots of gene loss as compared to diploid relatives (zebrafish, Danio rerio) and discuss the evolutionary significance of non-random gene loss across the catostomid phylogeny. Catostomid fishes represent a largely overlooked clade for studying the effects of polyploidy on patterns of phenotypic diversification, speciation, sex-determination, and hybridization dynamics.

(75) Mechanisms of unreduced ovule and pollen formation in *Citrus*

First author: J Cuenca, Instituto Valenciano de Investigaciones Agrarias, ES
Co-authors: H Rouiss, Instituto Valenciano de Investigaciones Agrarias, ES, A Garcia-Lor, Instituto Valenciano de Investigaciones Agrarias, ES, P Aleza, Instituto Valenciano de Investigaciones Agrarias, ES, L Navarro, Instituto Valenciano de Investigaciones Agrarias, ES, P Ollitrault, CIRAD, FR

The formation of unreduced (2n) gametes is a major component in polyploid plant evolution. Sexual polyploidization can be achieved mainly through meiotic restitution (First/Second Division Restitution –FDR/SDR–) or pre-/post-meiotic genome doubling (PRD or PMD). The genetic outcomes for each mechanism are very divergent, based on the different level of heterozygosity transmitted from the parents to the offspring. In citrus (Aurantioideae, Rutaceae; x=9), 2n gametes can be exploited for triploid and tetraploid breeding. Triploid citrus plants are generally sterile, thus potentially resulting in new seedless citrus varieties, while tetraploid genotypes are used for triploid breeding in interploid crosses and have proven to be useful for rootstock breeding. We have used citrus as a model species for genetic studies of sexual polyploidization, developing statistical methods for determining the mechanism underlying 2n ovule and pollen formation at both population and individual level. The individual LOD analysis from centromeric and telomeric loci genotyping and the analysis of parental heterozygosity restitution patterns along a linkage group allowed us to distinguish among the different mechanisms of 2n gamete formation. Our studies revealed that citrus sexual polyploidization can occur by different mechanisms. Spontaneous triploids mainly results from unreduced ovule formation, and predominantly through SDR mechanism (99%) as identified in seventeen mandarins and mandarin hybrids. In lemons, we identified three mechanisms of 2n female gamete formation, i.e. SDR (88%), FDR/PRD (7%) and PMD (5%). Unreduced pollen formation was observed in a diploid

(76) Mutual exclusion between sexual and asexual cytotypes of Potentilla puberula accounts for most of their geographical distribution in a sympatric area.

First author: Flavia Domizia Nardi, University of Salzburg, AT
Co-authors: Karl Hülber, University of Vienna, AT, Dietmar Moser, University of Vienna, AT, Henar Alonso-Marcos, University of Vienna, AT, Andreas Tribsch, University of Salzburg, AT, Christoph Dobeš, Austrian Research Centre for Forest, AT

Differences in the geographical distribution of cytotypes can depend on several not mutually-exclusive factors. First, they may reflect a differentiation in the ecological niches of cytotypes which correlates to geographical features. Secondly, they may be associated to differences in dispersal and colonisation histories. Third, they can be determined by direct or indirect mutual exclusion or population dynamics among cytotypes (e.g., minority cytotype exclusion, reproductive interference, competition...). Moreover, such differences and interactive processes among cytotypes could be related not only to polyploidy itself, but also to asexuality and to hybridity, traits that often co-occur in plants. The aims of our research are: a) to weigh the three factors underlying the ecogeographical distribution of cytotypes by means of variance partitioning analysis; and b) to identify the main trait (ploidy or reproductive mode) determining such differential distribution. We thus studied 238 Eastern Alpic populations of Potentilla puberula Krašan (*Rosa*ceae), a species reproductively differentiated between sexual tetraploids and mostly apomictic penta- to octoploids. After assessing the non-hybrid origin of asexuals, we determined mutual exclusion among cytotypes as the most relevant factor explaining the distribution of cytotypes. This may depend primarily on asymmetric reproductive interference. An ecological differentiation was found as well, with sexuals tendentially occurring to more pristine, drier habitats than asexuals. Finally, our results support the thesis that reproductive mode plays a stronger role than ploidy in the biology of this species, given the little genetic and ecological differentiation among asexual cytotypes.

(77) Investigating homoeologous recombination in *Arabidopsis*.

First author | AH Lloyd, Aberystwyth University, GB
Co-authors | JH Doonan, Aberystwyth University, GB

A valuable and under-utilised source of beneficial traits for plant breeding resides in the thousands of living wild crop relatives. This diversity can be accessed either via hybrid introgression lines or by regenerating allopolyploid crops from their diploid progenitors, however for both of these processes it is necessary to optimise the degree to which homoeologous chromosomes recombine. While a good understanding of the molecular machinery involved in homologous recombination and how to manipulate it has recently emerged, our understanding of homoeologous recombination and its regulation lags behind. *Arabidopsis* allopolyploids and interspecific hybrids represent a powerful model system to further address this knowledge gap. We are currently introgressing a series of fluorescent pollen-based recombination reporter lines (FTLs) from diploid *Arabidopsis* thaliana into allotetraploid *Arabidopsis suecica*, and can use these lines to rapidly assess levels of homoeologous recombination. We can also assess the level of homoeologous recombination cytologically in both inter-specific hybrids and allopolyploids. We will use these tools to identify genes that regulate the level of homoeologous recombination using both forward and reverse genetic approaches including i) CRISPR/CAS9 mutagenesis of key candidate genes (ZIP4, MSH2 and RECQ4A/B), and ii) mapping loci responsible for differences in homoeologous recombination between synthetic neo-tetraploid *A. suecica* and an evolved natural accession. Using these approaches, we aim to further our understanding of homoeologous recombination and its regulation, and develop approaches to manipulate the level of homoeologous recombination to accelerate plant breeding.

(78) The OneTwoTree phylogeny reconstruction webserver

First author | M Drori, Tel-Aviv University, IL
Co-authors |

Phylogeny reconstruction is a key instrument in numerous biological analyses, ranging from evolutionary and ecology research, to conservation and systems biology. The increasing accumulation of genomic data makes it possible to reconstruct phylogenies with both high accuracy and at increasingly finer resolution. Yet, taking advantage of all sequence data available requires the use of computational tools for efficient data retrieval and processing, or else the process could quickly become an error-prone endeavour. Here, we present the OneTwoTree webserver that allows fully automated phylogeny reconstruction. Given a list of taxa names of interest as the sole input requirement, OneTwoTree retrieves all available sequence data from NCBI GenBank, clusters these into orthologue groups, identifies the most informative set of markers, searches for an appropriate outgroup, and assembles a partitioned sequence matrix that is then used for the final phylogeny reconstruction step. We show that the use of OneTwoTree results in substantially higher data coverage in terms of both taxon sampling and the number of informative markers assembled compared to those manually reconstructed.

(79) Transcriptome and Alternative Splicing Analysis of *Brassica napus* in Response to *Sclerotinia* Infection

First author	Grant de Jong, University of British Columbia, CA
Presenting author	Keith L Adams, University of British Columbia, CA
Co-authors	

Brassica napus is an allotetraploid that was formed by hybridization of *Brasssica rapa* and *Brassica oleracea* along with chromosome doubling roughly 5000 years ago. There have been several studies of gene expression in *Brassica napus* on a transcriptome-wide scale to examine global patterns of gene expression. *Brassica napus* is a good system for studying homoeologous gene expression in polyploid plants in response to biotic stresses. We have infected natural and resynthesized *Brassica napus*, *Brassica rapa*, and *Brassica oleracea* with the fungal pathogen *Sclerotinia sclerotiorum* and performed RNA-sequencing using Illumina NovaSeq 6000 to obtain transcriptome data from infected and uninfected control plants. We analysed patterns of gene expression and alternative splicing to do comparisons between infected and uninfected plants, comparisons between diploids and polyploids, and comparisons between natural and resynthesized polyploids. Data analysis is in progress at the time of abstract submission. Our analyses include examination of expression and alternative splicing of the homoeologs compared with each other and to their corresponding diploids. We have identified many genes that respond to *Sclerotinia* infection in their expression and alternative splicing. We also classified expression changes in response to *Sclerotinia* into categories: additive, dominant, transgressive, and no change. We will present various findings from the study and their implications.

(80) Using digital organisms to investigate the effects of whole genome duplication on (artificial) evolution

First author	Yao Yao, VIB-UGent Center for Plant Systems Biology, BE
Co-authors	Lorenzo Carretero-Paulet VIB / Ghent University Evolutionary Systems Biology, ES, Yves Van de Peer, VIB-UGent Center for Plant Systems Biology, BE

The potential role of whole genome duplication (WGD) in evolution is controversial. Whereas some view WGD mainly as detrimental and an evolutionary 'dead end', there is growing evidence that the long-term establishment of polyploidy might be linked to environmental change, stressful conditions, or periods of extinction. However, despite much research, the mechanistic underpinnings of why and how polyploids might be able to outcompete non-polyploids at times of environmental upheaval remain elusive. Here, we improved our recently developed bio-inspired framework, combining an artificial genome with an agent-based system, to form a population of so-called Digital Organisms (DOs) and examine the impact of WGD on evolution under different environmental scenarios mimicking extinction events of varying strength and frequency. We found that, under stable environments, DOs with non-duplicated genomes formed the majority, if not all, of the population, whereas the numbers of DOs with duplicated genomes increased under dramatically challenging environments. We also observed that fewer mutational changes were allowed to accumulate in DOs with duplicated genomes than in DOs with non-duplicated genomes, suggesting the former are under stronger purifying selection. Also, DOs with non-duplicated genomes need to accumulate more mutations than polyploid genomes to reach adaptation (using homeostasis - as measured by gene expression - and the energy level as proxies). Thus, after tracking the evolutionary trajectories of individual artificial genomes in terms of sequence and encoded gene regulatory networks (GRNs), we propose that increased complexity, modularity, and redundancy of duplicated GRNs might provide DOs with increased adaptive potential under extinction events, while ensuring mutational robustness of the whole GRN. Our results confirm the usefulness of our computational simulation in studying the role of WGD in evolution and adaptation, helping to overcome the traditional limitations of evolution experiments with model organisms, and provide some additional insights into how genome duplication might help organisms to compete for novel niches and survive ecological turmoil.

(81) Multi-omics resolving the genetic and epigenetic differences of polyploidization in cotton

First author: Xiongming Du, Institute of Cotton Research of Chinese Academy of Agricultural Sciences (ICR, CAAS), CN

Co-authors: Zhen Peng[1], Hua Chen[1,2], Shoupu He[1], Panhong Dai[1], Gaofei Sun[2], Wenfang Gong[1], Zhaoe Pan[1], Yinhua Jia[1] [1]State Key Laboratory of Cotton Biology, Institute of Cotton Research, the Chinese Academy of Agricultural Science, Anyang 455000, China [2]Department of Computer Science and Information Engineering, Anyang Institute of Technology, Anyang 455000, China

Cotton is one of the most important economic crops worldwide. Presently, almost all the cottons cultivated are allotetraploids. Comparing with diploids, cultivated allotetraploids (*G. hirsutum* and *G. barbadense*) possess numerous advantages including higher fiber yield and better fiber quality. Allotetraploid cottons contained two complete sets of sub-genomes, their independent function and mutual interaction is always the hotspot in research field of polyploidy. For better understanding genetic basis of fiber property variations during cotton polyploidization, we analyzed the transcriptional and epigenetic differences between diploid (*G. raimondii* and *G. arboreum*) and tetraploid (*G. hirsutum*) cottons by integrating multi-omics approaches. Our major concerns including: 1. Gene expression patterns (total 12 models) in ovule and fiber after polyploidization. 2. The dominantly expressed orthologous genes in tetraploid cotton. 3. The subfunctionalization and neofunctionalization of orthologous genes in tetraploid cotton and their bias expression in subgenomes. Our results indicated that the majority of the orthologous genes showed the consistent expression patterns among diploids cottons and tetraploid cotton, which implied that the regulation mechanism was conserved between subgenome. We further found that most of the differentially expressed genes were non-addictive genes. Therefore, we comprehensively analyzed the TEs and 24nt-siRNAs data around differentially expressed genes, the whole genomic DNA methylation level bias of CG, CHG and CHH at gene body and TE region, and the identification of differential DNA methylation level region. The results demonstrated that TEs and siRNAs were enriched around up/downstream of differentially expressed genes among diploid and tetraploid genomes, which could buffer the impact of polyploidization.

(82) Comparative effect of allopolyploidy on transposable element composition and gene expression between *Gossypium hirsutum* and its two diploid progenitors

First author: Hua Cheng, State Key Laboratory of Cotton Biology, Institute of Cotton Research, the Chinese Academy of Agricultural Science, Anyang 455000, China, CN

Presenting author: Shoupu He, State Key Laboratory of Cotton Biology, Institute of Cotton Research, the Chinese Academy of Agricultural Science, Anyang 455000, China, CN

Co-authors: Gaofei Sun[2], Shoupu He[1], Wenfang Gong[1], Zhen Peng[1], Xiongming Du[1] [1] State Key Laboratory of Cotton Biology, Institute of Cotton Research, the Chinese Academy of Agricultural Science, Anyang 455000, China [2] Department of Computer Science and Information Engineering, Anyang Institute of Technology, Anyang 455000, China

An allopolyploidization event formed allotetraploid *Gossypium* species from an A-genome diploid species and a D-genome diploid species. To explore the responses of transposable elements (TEs) to allopolyploidy, we assembled parallel TE datasets from *G. hirsutum, G. arboreum* and *G. raimondii* and analyzed the TE types and the effects of TEs on orthologous gene expression in the three Gossypium genomes. Gypsy was the most abundant TE type and most TEs were located ~500 bp from genes in all three genomes. In *G. hirsutum*, 35.6% of genes harbored TE insertions, whereas insertions were more frequent in *G. arboreum* and *G. raimondii*. *G. hirsutum* had the highest proportion of uniquely matching 24nt small interfering RNAs (siRNAs) that targeted TEs. TEs, particularly those targeted by 24nt siRNAs, were associated with reduced gene expression, but the effect of TEs on orthologous gene expression varied substantially among species. Orthologous gene expression levels in *G. hirsutum* were intermediate between those of *G. arboreum* and *G. raimondii*, which did not experience TE expansion or reduction resulting from allopolyploidization. This study underscores the diversity of TEs co-opted by host genes and provides insights into the roles of TEs in regulating gene expression in *Gossypium*.

(83) Phenotypic and Fitness Effects of Aberrant Dosage of Single-Copy Genes

First author: Eylem Aydogdu, Department of Plant Biotechnology and Bioinformatics, Ghent University, Ghent, Belgium, VIB Center for Plant Systems Biology, VIB, Gent, Belgium, , BE

Co-authors: Eylem Aydogdu*[1,2], Wout Boerjan[1,2], Yves Van de Peer[1,2,3] [1]Department of Plant Biotechnology and Bioinformatics, Ghent University, Ghent, Belgium, [2]VIB Center for Plant Systems Biology, VIB, Gent, Belgium, [3]Department of Biochemistry, Genetics and Microbiology, University of Pretoria, Pretoria 0028, South Africa

Gene loss following whole genome duplication is a non-random process, for instance, particular sets of gene families were shown to be primarily single-copy (SC) across species. Persistent restoration of these genes to single-copy status post-duplication during (plant) evolution suggests potential negative fitness consequences when they are duplicated. To our knowledge, no studies to date have experimentally validated the significance of the copy number status of these SC genes for plant phenotype and fitness, or experimentally investigated potential mechanisms underlying their intolerance to duplication. We engineered two large sets of *Arabidopsis thaliana* lines, one set over-expressing a selected group of SC genes, and the other over-expressing a selected group of multi-copy (MC) genes, serving as a control group. Plant viability, phenotype(s) and fitness of the over-expression lines were assessed and compared via a large-scale phenotyping experiment, consisting of over 1200 plants. As expected, over-expression lines, in general, showed a reduced fitness compared to wild-type. However, the increased expression of MC genes had a higher detrimental impact on plant fitness than increased expression of SC genes. This indicates that dosage balance sensitivity may not be the driver of the gene-loss process for the SC genes, especially in the case of small-scale duplications. The effect on plant fitness was distinctive based on the functional annotation of the gene families, with thylakoid related MC genes showing the highest negative impact while their SC counterparts showed no significant effect on fitness. Further exploration of available datasets in *Saccharomyces cerevisiae* gave results supporting our findings in *Arabidopsis*, showing a conserved pattern across plants and fungi.

(84) Following the tracks of triploid asexual Prussian carp (Carassius gibelio) – one of the most successful invasive vertebrates in Europe

First author: Dunja K. Lamatsch, University of Innsbruck, Research Institute for Limnology Mondsee, Austria.

Co-authors: Manuel Deinhardt[1,2], Roman Fuchs[3], Robert Schwarz[1], Maria Pichler[1], Jouni Taskinen[2], Daniel L. Jeffries [4], Jukka Jokela[5,6], Matthias Stöck[7]; [1] University of Innsbruck, Research Institute for Limnology Mondsee, Austria. [2] University of Jyväskylä, Aquatic Sciences, Department of Biological and Environmental Science, PO 35, FIN-40014, Finland. [3]University of Salzburg, Austria. [4] University of Lausanne, Department of Ecology and Evolution, Lausanne, Switzerland [5] EAWAG, Swiss Federal Institute of Aquatic Science and Technology, CH-8600 Dübendorf, Switzerland. [6] ETH-Zürich, Institute of Integrative Biology, 8092 Zürich, Switzerland. [7] Leibniz-Institute of Freshwater Ecology and Inland Fisheries, Berlin, Germany

Biological invasions present a major threat to freshwater ecosystems in past (e.g. Vitousek et al. 2017) and future (Sala et al. 2000). Several teleosts of the genus *Carassius* are global invaders; particularly *C. gibelio* is one of the most successful invasive vertebrates in Europe, with the highest ecological and economic impact of all established invasive fish (van der Veer Nentwig 2015). Yet, we neither have enough information to provide tools for its management nor to fully understand its impact on the genomic integrity of native cyprinid fish species. Morphological similarity and variability make species recognition in *Carassius* difficult (Kalous et al. 2012). *C. gibelio* are widely distributed, and occur as diploid sexuals (2n = 100) and triploid invasive asexuals (3n≈150) (Kalous et al. 2012), and even tetraploids (Knytl et al. 2013). Triploids are clonal female lineages that reproduce asexually by sperm-dependent parthenogenesis (= gynogenesis; e.g. Gui Zhou, 2010). We compared the genetic structure of the triploid and tetraploid invasive *C. gibelio* in Finland, Germany and Austria. We used flow cytometry for ploidy determination, nuclear microsatellites, and mitochondrial d-loop sequences for lineage identification. We also elucidated the high percentage of paternal introgression in imperfect gynogenesis by artificial crosses and inferences of the parental species that contributed

genomes to form tetraploids. This contributes to a better understanding of the conservation requirements of the threatened native diploid *C. carassius* and the control of invasive polyploid *C. gibelio* in Europe.

Participants

List of participants

International Conference on Polyploidy

R. Shawn Abrahams	University of Missouri, Columbia, USA
Ana Afonso	Top Atlântico Coimbra, PT
Malika Ainouche	Université de Rennes 1, FR
Reiko Akiyama	University of Zurich, CH
Gökce Aköz	Gregor Mendel Institute - Institute of Population Genetics, AT
Dirk Albach	Carl von Ossietzky-University, DE
Victor Albert	University at Buffalo, USA
Warren Albertin	ISVV - UR Oenologie, FR
Pablo Aleza	IVIA, ES
Tia-Lynn Ashman	University of Pittsburgh, USA
Eylem Aydogdu Lohaus	VIB-UGent Center for Plant Systems Biology, BE
Franz Boideau	Institut national de la recherche agronomique (INRA), FR
Quinten Bafort	VIB-UGent Center for Plant Systems Biology, BE
Wei-Ning Bai	Beijing Normal University, CN
Mike Barker	University of Arizona, USA
Romana Bartošová	Faculty of Science, Charles University, CZ
Karen Bauk	IMBIV-CONICET, AR
Yves Bawin	Ugent, Department of Plant Biotechnology and Bioinformatics, BE
Marc Beringer	Institute of Plant Sciences at the University of Bern, CH
Visnja Besendorfer	University of Zagreb, HR
Paul Blischak	University of Arizona, USA

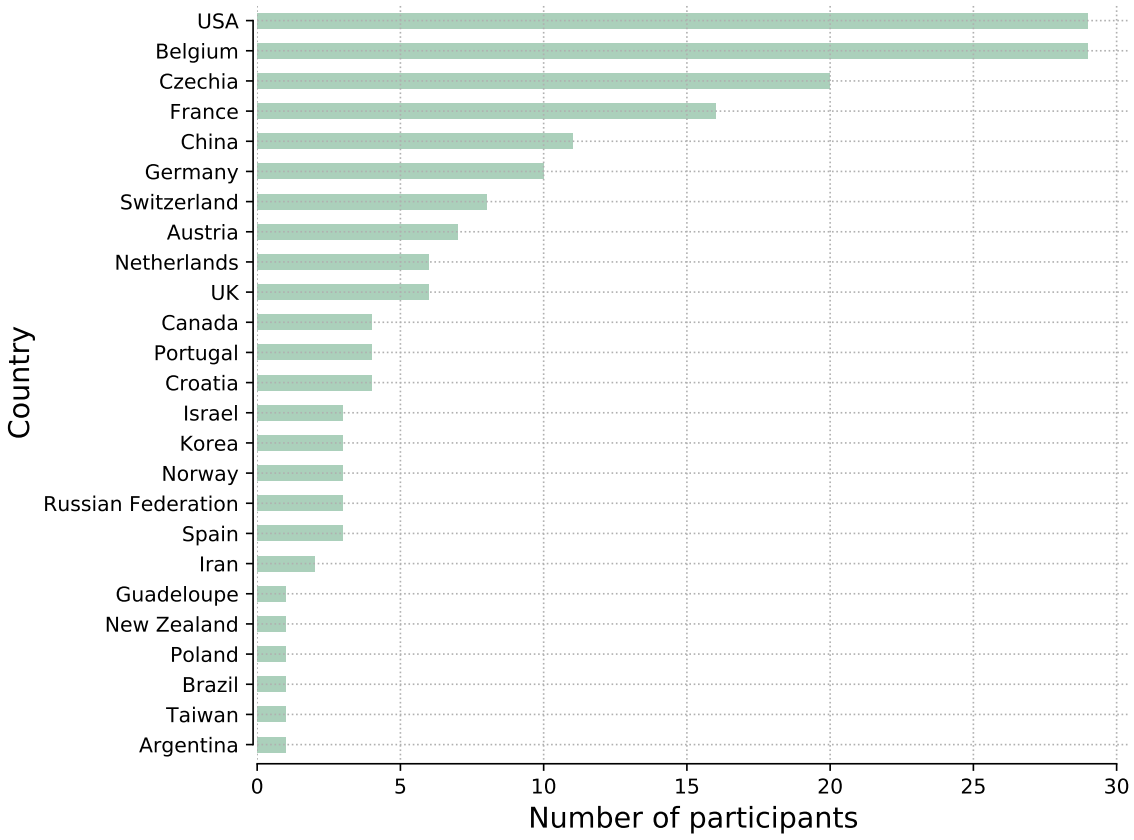

Figure 1: Number of participants in the international conference on polyploidy (2019 - Ghent, Belgium) by country.

Lander Briels	VIB Conferences team, BE
Robin Burns	Vienna Biocenter, Gregor Mendel Institute, AT
Jose Cuenca	Instituto Valenciano De Investgiationes Agrarias, ES
Rosa Castillo Bravo	PABC, Ryan Institute, National University of IR Galway, IR
Mariana Castro	Top Atlântico Coimbra, PT
Sílvia Castro	Centre for Functional Ecology, Department of Life Sciences, University of Coimbra, PT
Jen-Tsung Chen	Department of Life Sciences, National University of Kaohsiung, TW
Duo Chen	Institute of Ecology College of Life Sciences Beijing Normal University, CN
Rimjhim Roy Choudhury	University of Bern, CH
Anne-Marie Chèvre	INRA France, FR
Justin Conover	Iowa State University, USA
Britt De Pelsmaeker	Merck, BE
Kevin Debray	UNIV ANGERS, FR
Rob Denton	University of Minnesota Morris, USA
Angélique Dhondt	CIRAD, FR
Mert Dogan	CEITEC-Masaryk University, CZ
Michal Drori	Tel Aviv University, IL
Mimmi Eriksson	University of Vienna, AT
Julie Ferreira de Carvalho	IGEPP INRA, FR
Don Fox	Duke University Medical Center, US
Božo Frajman	Department of Botany, University of Innsbruck, AT
Andres Garcia Lor	Instituto Valenciano De Investgiationes Agrarias, ES
Brigida Gallone	VIB-KU Leuven Center for Microbiology, BE
Emanuel Gaquerel	Institute of Plant Molecular Biology (CNRS / University of Strasbourg), FR
Michelle Gaynor	University of Florida, USA
Danny Geelen	UGENT, BE
Delphine Giraud	UMR-CNRS 6553 ECOBIO, University of Rennes 1, FR
Marek Glombik	Institute of Experimental Botany, AS CR, CZ
Natasha Glover	University of Lausanne, CH
Lei Gong	Northeast Normal University, CN
Jeroen Goorden	Sysmex, BE
Olivier Grasmeur	CIRAD, FR
Danijela Greguras	Institute of Botany, Czech Academy of Sciences, CZ
Cortland Griswold	University of Guelph, CA
Corrinne Grover	Iowa State University, USA
Sandra Grünig	University of Bern / Ecological Genomics, CH
Wenjia Gu	Queen Mary University of London, UK
Xinyi Guo	CEITEC, Masaryk University, CZ
Yan-Ping Guo	College of Life Sciences, Beijing Normal University, CN
Christopher Hamel	California State University, Sacramento, USA
Matthias Hartmann	Czech Academy of Science Institute of Botany, CZ
Rebecca Heald	University of California, USA
Marie Henniges	RBG Kew, UK
Elvira Hoerandl	Georg-August-University of Goettingen, DE
Guanjing Hu	Iowa State University, USA
Jin Hoe Huh	Seoul National University, KR
Elizabeth Ihien	Justus liebig university (institute of plant breeding), BE
Eric Jenczewski	Institut National de la recherche Agronomique, FR
Chunlian Jin	Ghent University, Belgium, BE
Thomas Jungas	CNRS - CBI toulouse, FR
Elvis Katche	Instutute of Plant Breeding, DE

Anna Klepikova	The Institute for Information Transmission Problems RAS, RU
Abbas Koaik	Merck, BE
Samantha Koehler	Universidade Estadual de Campinas, BR
Filip Kolář	Charles University, CZ
Veronika Konecna	Charles University, CZ
Trevor Krabbenhoft	University at Buffalo, USA
Dunja K Lamatsch	University of Innsbruck, Research Institute for Limnology Mondsee, AT
Ilia Leitch	Royal Botanic Gardens, Kew, UK
Andrew Leitch	University of London, UK
Kelley Leung	University of Groningen, USA
Leen Leus	ILVO, BE
Linfeng Li	School of Life Sciences Fudan University, CN
Ning Li	Northeast Normal University, CN
Zhen Li	VIB-UGent Center for Plant Systems Biology, BE
Mingrui Li	School of Life Sciences Fudan University, CN
Yanlin Liao	Wageningen University and Research, NL
Jinsong Liu	Texas Medical Center, USA
Bao Liu	Northeast Normal University, CN
Andrew Lloyd	Aberystwyth University, UK
João Loureiro	Centre for Functional Ecology, Department of Life Sciences, University of Coimbra, PT
Jana Lunerova	Institute of Biophysics of the CAS, v. v. i., CZ
Martin Lysak	Central European Institute of Technology (CEITEC), Masaryk University, CZ
Sophie Maebe	VIB-UGent Center for Plant Systems Biology, BE
Steven Maere	VIB-UGent Center for Plant Systems Biology, BE
Nenad Malenica	University of Zagreb, HR
Terezie Mandakova	CEITEC, Masaryk University, CZ
Jian-Feng Mao	Beijing Forestry University, CN
Carlos Marti	Centro Nacional de Investigaciones Cardiovasculares Carlos III, Madrid, ES, NO
Annaliese Mason	Justus Liebig University, DE
Itay Mayrose	Tel Aviv University, Plant Evolution, bioinformatics comparative genomics, IL
Jelena Mlinarec Novosel	University of Zagreb, HR
Emma Jane Morgan	Charles University in Prague, CZ
Raphaël Morillon	CIRAD, GP
Patricio Munoz	University of Florida, USA
Flavia Domizia Nardi	University of Salzburg, AT
Maurine Neiman	The University of Iowa, USA
Polina Novikova	VIB-UGent Center for Plant Systems Biology, BE
Patrick Ollitrault	CIRAD, FR
Stan Oome	Averis Seeds B.V., NL
Vidya Oruganti	Max-Planck Institute for Plant Breeding Research, DE
Christian Parisod	University of Bern, CH
Jeong Eun Park	Seoul National University, KR
Ovidiu Paun	University of Vienna, Botany and Biodiversity Research, AT
Clara Peeters	Sysmex, BE
Jan Pinc	Charles University in Prague, Botany department, CZ
Nicolas Pompidor	CIRAD, FR
Lucas Prost	VIB-UGent Center for Plant Systems Biology, BE
Jan Ptáček	Charles University, Department of Botany, CZ
Jasna Puizina	University of Split, HR
Daniela Quezada-Martinez	Department of Plant Breeding, Justus Liebig University, DE
Ludmila Rejlová	Institute of Botany, The Czech Academy of Sciences, Průhonice, Czech Republic, CZ

Anna Rice	Tel-Aviv University, IL
Tom Ruttink	ILVO, BE
Line Lieblein Røsæg	Norwegian University of Life Sciences, Ås, Norway, NO
Armel Salmon	Université de Rennes 1 / CNRS ECOBIO, FR
Simen Sandve	Norwegian University of Life Sciences, Aas, NO
David Sankoff	University of Ottawa, CA
Sarah Schiessl-Weidenweber	Justus Liebig University, DE
Eric Schranz	Wageningen University and research, NL
Emily Sessa	University of Florida, USA
Shengchen Shan	University of Florida, USA
Tim Sharbel	Global Institute of Food Security, CA
Joel Sharbrough	Colorado State University, USA
Kentaro Shimizu	University of Zurich, Department of Evolutionary Biology and Environmental Studies, CH
Rie Shimizu-Inatsugi	University of Zürich, CH
Hosub Shin	Seoul National University, KR
Ramin Shirali Hossein Zade	Delft University of Technology, NL
Elina Shnaider	Institute of Information Transmission Problem RAS, RU
Doug Soltis	University of Florida, Florida Museum of Natural History, Gainesville, USA
Pamela S Soltis	University of Florida, USA
Jonathan Spoelhof	University of Florida, USA
Zuzana Storchová	Technical University Kaiserslautern, DE
Yue Sun	Northeast Normal University, CN
Limin Sun	Ghent university, BE
Brittany Sutherland	University of Arizona, USA
Radim Svačina	Institute of Experimental Botany AS CR, CZ
Dawit Girma Tekleyohans	University of Bremen, DE
Susan Thomson	The New Zealand Institute for Plant Food Research LTD, NZ
Alejandro Thérèse	Wageningen University and Research, NL
Katrijn Van Laere	ILVO - Plant Sciences Unit, BE
Yves Van de Peer	VIB-UGent Center for Plant Systems Biology, BE
Irina Van der Vlies	Merck, BE
Zoe Vance	Trinity College Dublin, IR
Clayton Visger	California State University Sacramento, USA
Radka Vozarova	Institute of Biophysics of the CAS, and Masaryk University Faculty of Science, CZ
James Watkins	Colgate University, USA
Jonathan Wendel	Iowa State University, USA
Ying Wu	Northeast Normal University, CN
Tao Wu	Baylor College of Medicine, USA
Linlin Xu	Beijing Normal University, BE
Jiayu Xue	VIB-UGent Center for Plant Systems Biology, BE
Levi Yant	University of Nottingham, UK
Kira Zadesenets	ICG SB RAS, RU
Yue Zhang	University of Ottawa, CA
Tao Zhao	VIB-UGent Center for Plant Systems Biology, BE
Arthur Zwaenepoel	VIB-UGent Center for Plant Systems Biology, BE
Laurens de Haas	Sysmex, BE
Kars de Wildt	Sysmex, BE
Dirk Granse	Institute of Plant Sciences and Microbiology, University of Hamburg, DE
Mathieu Rousseau-Gueutin	INRA, FR
Peter van Dam	Genetwister Technologies B.V., NL
Jana Čížková	Institute of Experimental Botany, CZ

Denisa Šimoníková — Institute of Experimental Botany AS CR, v.v.i., CZ
Gabriela Šrámková — Faculty of Science, Charles University, CZ
Justyna Żabicka — Department of Plant Cytology and Embryology, Institute of Botany, Jagiellonian University, PL

Design and typesetting by Arthur Zwaenepoel

Prof. Dr. Yves Van de Peer
Department of Plant Biotechnology and Bioinformatics, Ghent University
VIB - UGent Center for Plant Systems Biology
Technologiepark 71, B-9052 Ghent, Belgium
yves.vandepeer@psb.vib-ugent.be