

Plant Chromosome Biology

September 15 – 17, 2025, Vienna, Austria



- Genome evolution
- Genome stability
- New tools and applications
- Environmental response
- Chromosome and nucleus architecture
- Cell division, ploidy, and apomixis



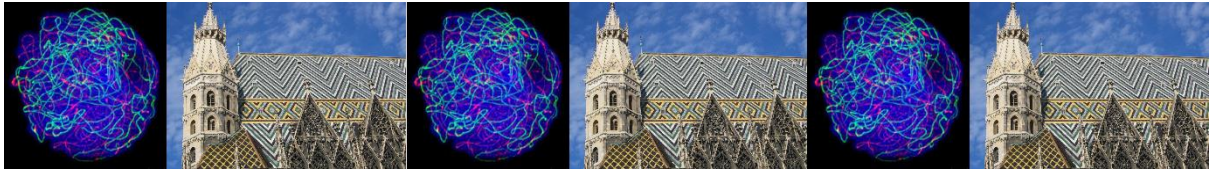
Website: <https://plantcyto2025.univie.ac.at>

Email: chromosomebio2025.botanik@univie.ac.at



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CONFERENCE PROGRAM

BOOK OF ABSTRACTS



This meeting is organized by the University of Vienna and is supported by the Society for Plant Breeding e.V. (GPZ), section “Cytogenetic and Chromosome Analysis”.

Organizer

Hanna Schneeweiss (Department of Botany and Biodiversity Research, University of Vienna, Vienna, Austria)

Scientific co-organizers

Ortrun Mittelsten Scheid (Gregor Mendel Institute, Vienna, Austria)

Andreas Houben (IPK Gatersleben and GPZ, Germany)

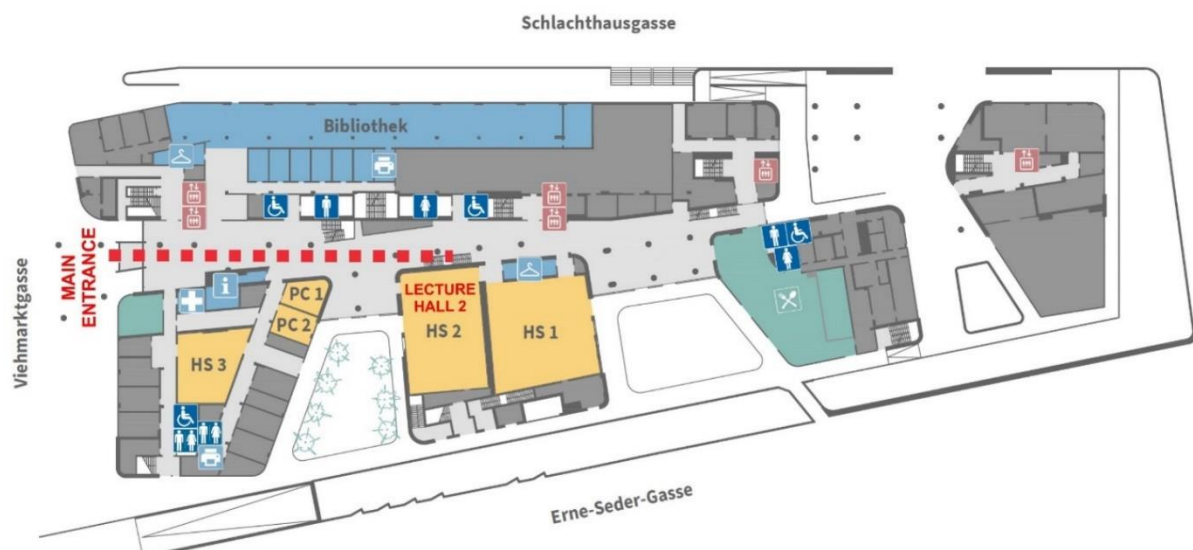
The meeting will take place in the new University of Vienna Biology Building

University Biology Building (UBB)

Lecture Hall 2 (ground floor)

Djerassiplatz 1

1030 Vienna, Austria



How to get to the conference venue

By public transport

- Take railway S7 ("Schnellbahn") to the station *Sankt Marx* (1) and walk c. 5 minutes
- Take subway line U3 to *Schlachthausgasse* (exit "Markhofgasse") (2) and walk c. 9 minutes
- Take bus 74A to the station *Sankt Marx S* (5) and walk c. 2 minutes
- Take tram 71 to the station *Sankt Marx S* (5) and walk c. 2 minutes
- Take tram 18 to the station *Viehmarktgassee* (3) and walk c. 1 minute
Station *Viehmarktgassee* (3) is currently closed due to roadworks; tram 18 operates now only between stations *Burggasse-Stadthalle* and *Sankt Marx*

By plane (Vienna International Airport, VIA)

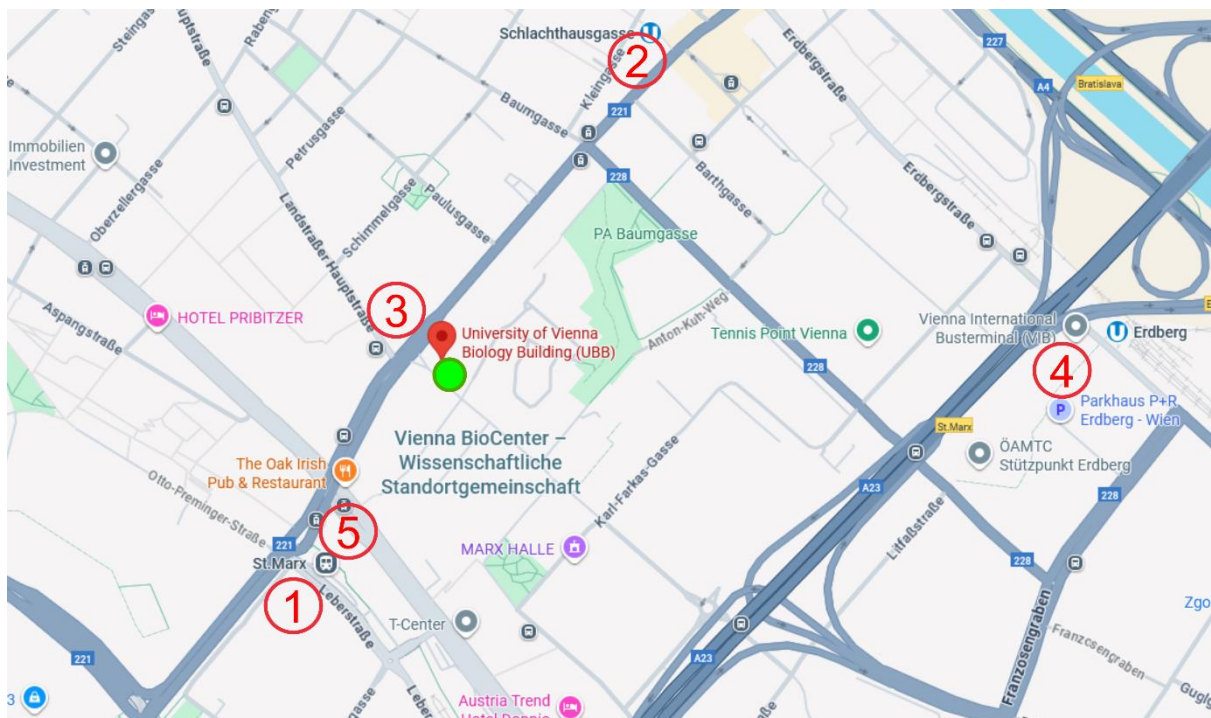
- Take railway S7 ("Schnellbahn"; do NOT board the CAT train as it does not stop at all stations) to the station *Sankt Marx* (1) and walk c. 5 minutes
- Take taxi (c. 30 minutes, depending on traffic); the fare is around € 30-40

By car

- When visiting Vienna, it is recommend to use public transport. In most of Vienna, cars can only be parked for a [short time](#) (max. 2 hours), and only with a parking ticket. When booking a hotel make sure it offers parking space.

By bus (arriving at the Vienna International Busterminal, VIB)

- Take subway line U3 from station *Erdberg* (direction Ottakring) (4) to *Schlachthausgasse* (exit "Markhofgasse") (2) and walk c. 9 minutes
- Walk (c. 20 min)

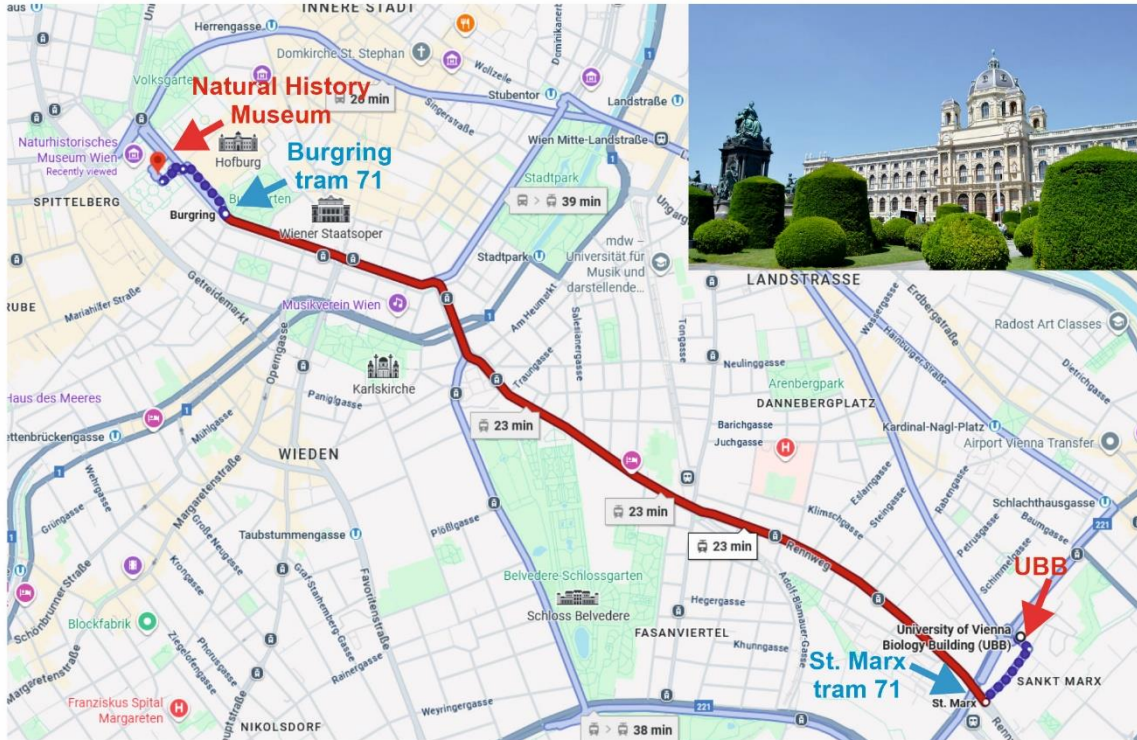


How to get to the Natural History Museum and to conference dinner

Natural History Museum

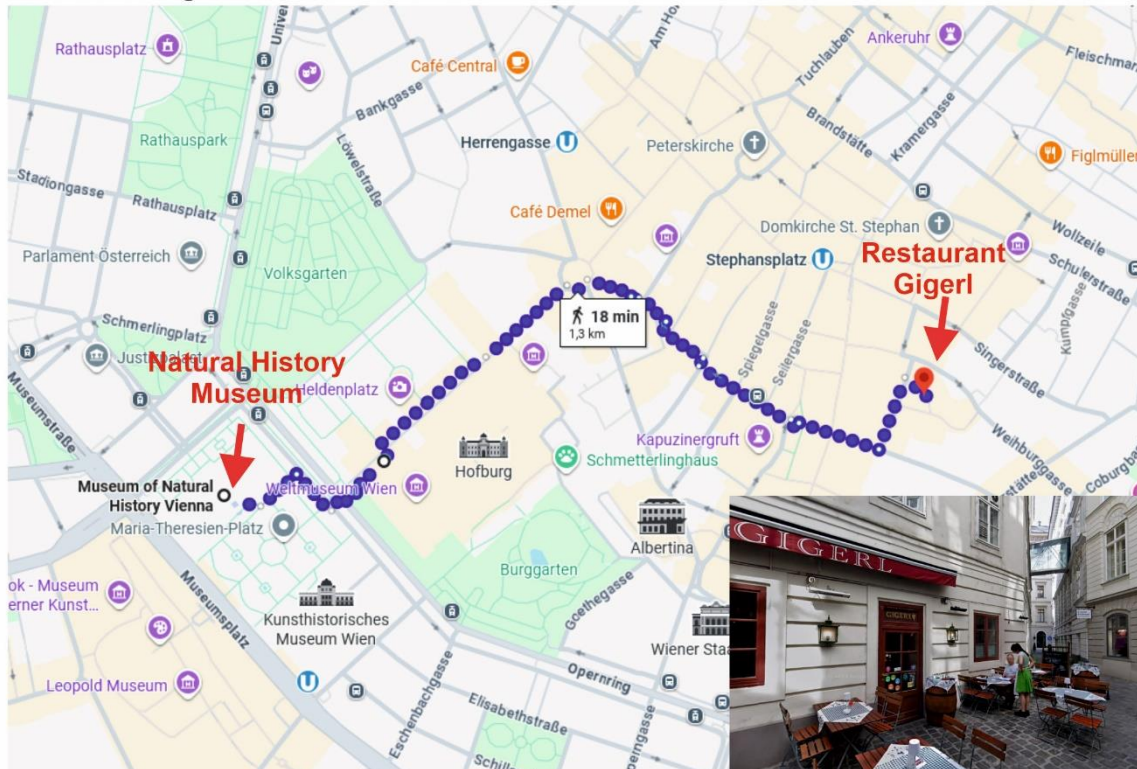
Burgring 7, 1010 Vienna

Tram 71 (station St. Marx to station Burgring - 8 stops)



Restaurant Gigerl – Der Stadtheurige (walk c. 18 min)

Blumenstockgasse 2, 1010 Vienna





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Plant Chromosome Biology Conference September 15-17, 2025, Vienna

Program

Monday 15.09.2025

07:45 - 09:00 Registration open

09:00 - 09:15 **Welcome**

PART 1

Chairperson: **Norman Wickett**

09:15 - 09:50 **Frédéric Berger**: Histone variants: The architects of chromatin organization

09:50 - 10:10 **Yi-Tzu Kuo**: Bigger or more: Divergent evolutionary paths to atypical macro-monocentromeres and holocentromeres

10:10 - 10:30 **Hans-Wilhelm Nützmann**: Somatic mobility of transposons in *Arabidopsis thaliana*

10:30 - 10:50 **Pat Heslop-Harrison**: Cytogenomics of oat (*Avena*) genome evolution: Intra- and inter-genomic chromosomal translocations, repetitive DNA dynamics, and reconstruction of ancestral grass karyotypes show genome evolution in action

10:50 -11:20 Coffee break (non-vegetarian/vegetarian/vegan)

PART 2

Chairperson: **Andreas Houben**

11:20 - 12:10 Poster pitches, part 1, Posters 1 – 28

12:10 - 12:30 **Lucie Hloušková**: The maize B chromosome shapes the transcriptome throughout the entire plant life

12:30 - 12:50 **Teresa Bojdová**: Aspects of B chromosome elimination in *Sorghum purpureosericeum*

13:00-14:00 Lunch (vegetarian/vegan)

PART 3

Chairperson: **Jaroslav Doležel**

14:00 - 14:35 **Daniel Voytas**: From lab to field: The promise of plant gene editing at scale [ONLINE TALK](#)

14:35 - 14:55 **Fabienne Gehrke**: Expanding the toolbox: Prospective uses of the inducible CRISPR-Kill system in plants

- 14:55 - 15:15 **Ondřej Helia:** CRISPR-engineered chromosomal translocations point to cis-regulatory control of arm-specific telomere homeostasis and overall robustness of chromatin structure and phenotype in *Arabidopsis*
- 15:15 - 15:35 **Thomas Buchloh:** Does karyotype change promote species diversity? Revealing hidden patterns in fern diversification
- 15:35 - 15:55 **Nobuko Ohmido:** Plant chromosome architecture revealed by advanced electron microscopy

15:55 - 16:25 [Coffee break \(non-vegetarian/vegetarian/vegan\)](#)

PART 4

- Chairperson: **Trude Schwarzacher**
- 16:25 - 16:45 **Francesca Beclin:** Analyzing the evolutionary history of a uniquely polymorphic chromosome of the genus *Aquilegia*
- 16:45 - 17:05 **Lee Mariault:** Cross-kingdom horizontal gene transfers between plant and fungal genomes: Insights from large-scale comparative genomics
- 17:05 - 17:25 **Veit Herklotz:** Evolutionary tracing of bivalent and univalent subgenomes in pentaploid dogroses (*Rosa* sect. *Caninae*)
- 17:25 - 17:45 **Marco Castellani:** Interdependent HEI10 paralogs and their role in plant meiotic recombination
- 17:45 - 18:05 **Luca Comai:** Most tobacco twin spots result from chromosomal instability and not homologous recombination
- 18:05 - 19:00 [Poster session 1, Posters 1 – 28](#)

Tuesday 16.09.2025

PART 5

- Chairperson: **Ingo Schubert**
- 09:00 - 09:35 **Petra Bulankova:** Uncovering the unexpected features of nuclear division in diatoms
- 09:35 - 09:55 **Amanda Camara:** The chromonema workout
- 09:55 - 10:15 **Valentin Bapteste:** Understanding the role of chromatin mobility in genome integrity
- 10:15 - 10:50 **James Leebens-Mack:** Sequence-based cytogenetics reveals dynamic sex chromosome evolution within the genus *Asparagus*
- 10:50 - 11:00 [Group photo](#)
- 11:00 - 11:30 [Coffee break \(non-vegetarian/vegetarian/vegan\)](#)
- 11:30 - 12:00 [Poster pitches, part 2, Posters 29 – 57](#)
- 12:00 - 13:00 [Poster session 2, Posters 29 - 57](#)
- 13:00 - 14:00 [Lunch \(vegetarian/vegan\)](#)

PART 6

Chairperson:	<u>Ortrun Mittelsten Scheid</u>
14:00 - 14:35	Raphael Mercier: Maximizing meiotic crossover rates
14:35 - 14:55	Stefan Heckmann: The central element protein SCEP3 interlinks crossover and synaptonemal complex formation
14:55 - 15:15	Peter Schlögelhofer: How to establish inter-homologue bias during meiosis
15:30	Departure to the Museum of Natural History
16:00 - 18:30	Guided tour in the Museum of Natural History
19:00 - 22:20	Conference dinner at the Restaurant Gigerl (non-vegetarian/vegetarian)

Wednesday 17.09.2025

PART 7

Chairperson:	<u>Jiří Macas</u>
09:00 - 09:35	Kelly Dawe: Karyotype engineering with synthetic centromeres
09:35 - 09:55	Tetsuji Kakutani: Centrophilic retrotransposon integration via CENH3 chromatin in <i>Arabidopsis</i>
09:55 - 10:15	Handong Su: Genetic diversity and dynamic evolutionary innovations in plant centromeres
10:15 - 10:35	Conny Tränkner: A 2-bp deletion causes the formation of unreduced pollen in <i>Hydrangea macrophylla</i>
10:35 - 10:55	Zhijian Zhang: Evolutionary conservation of the monopolin component Csm1 in sister chromatid mono-orientation during meiosis in <i>Arabidopsis</i>
10:55 - 11:25	Coffee break (non-vegetarian/vegetarian/vegan)

PART 8

Chairperson:	<u>Hanna Weiss-Schneeweiss</u>
11:25 - 11:45	Manikandan Kalidass: Ubiquitin-dependent proteolysis of KNL2 driven by APC/C-CDC20 is critical for centromere integrity and mitotic fidelity
11:45 - 12:05	Jana Fulnečková: Plant telomere-binding proteins
12:05 - 12:25	Nikolai Borisjuk: Deciphering the molecular structure of rDNA loci in the aquatic monocot plants <i>Spirodela polyrhiza</i> and <i>S. intermedia</i>
12:25 - 12:45	Steven Dreissig: Population-wide single-pollen genotyping in rye sheds light on the genetic basis and environmental plasticity of meiotic recombination
12:45 - 13:15	Awards and farewell

Posters

- Maximum poster size: A0 high format (h: 118.9 cm; w: 84.1 cm)
- Mounting: please use only the provided mounting material
- **Posters 1-28 will be displayed and presented on Monday**, September 15th; they can be mounted on Monday morning and should be removed on Monday evening or the latest by Tuesday (early) morning.
- **Posters 29-57 will be displayed and presented on Tuesday**, September 16th; they can be mounted on Tuesday morning and removed during Wednesday morning.

Each poster can be introduced to all participants in a 1–2 minute pitch talk during a session preceding the poster presentations (please see the program). If you wish to give a pitch talk, please prepare 1–2 PowerPoint slides highlighting the major findings of your study. These slides will be combined into one file for a smooth presentation. Your slides must include the name of the presenting author and the poster number as it appears in the *Book of Abstracts*. Please save your file using the following format:

LastName_FirstName_poster_XXX.pptx (e.g., Smith_John_poster_111.pptx).

To be included in the pitch talk session, **send your file no later than Thursday, September 11th**, to:

chromosomebio2025.botanik@univie.ac.at

Talks

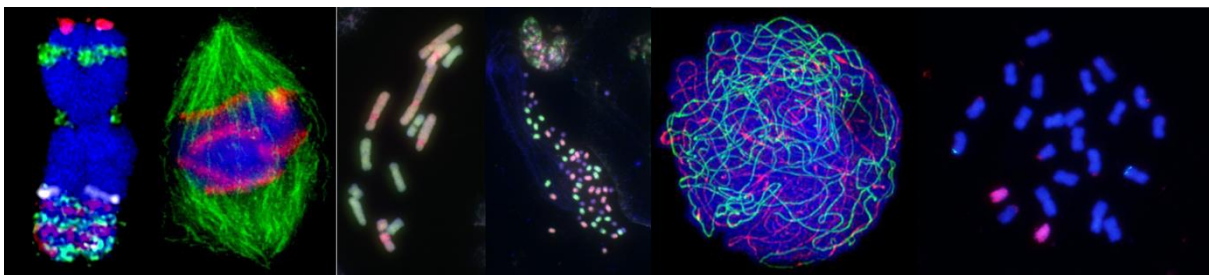
All presentations must be prepared in PowerPoint format and uploaded to a central computer at the conference venue on the morning of the day the talk is scheduled. Please include the full name of the presenting author and the part number (see the program) in the file name:

LastName_FirstName_PX.pptx (e.g., Smith_Jane_P2.pptx).

**Awards will be presented for the best posters
and the best short talks (for early career scientist).**

ABSTRACTS

Talks



INVITED TALK

Histone variants: The architects of chromatin organization

Frédéric Berger¹, Vikas Shukla¹, Facundo Romani²

¹Gregor Mendel Institute, Vienna, Austria, ²Department of Plant Sciences, University of Cambridge, UK

The complexity of chromatin composition is reduced to archetypal combinations called chromatin states that broadly define locally the potential for transcription. The degree of conservation of chromatin states has not been established amongst plants, and how they interact with transcription factors is unknown. We have identified and characterized the chromatin states of the flowering plant *Arabidopsis thaliana* and the bryophyte *Marchantia polymorpha*, showing a large degree of functional conservation over more than 450 million years of evolution of land plants. We analyzed the interplay between transcription factors and chromatin states to understand their influence on gene regulation. Combining binding sites with activity of transcription factors, we have shown their preferential association with specific chromatin states in both species. These associations define distinct groups of transcription factors based on their binding site relative to the transcription start site and broadly associate with distinct functions suggesting co-evolution of the chromatin regulatory mechanisms with transcription factors.

TALK

Bigger or more: Divergent evolutionary paths to atypical macro-monocentromeres and holocentromeres

Yi-Tzu Kuo¹, Pavel Neumann², Jianyong Chen¹, Jörg Fuchs¹, Veit Schubert¹, Katrin Kumke¹, Mariela Analia Sader³, Michael Melzer¹, Zihao Zhu¹, Axel Himmelbach¹, Heiko Hentrich⁴, Jiří Macas², Andreas Houben¹

¹Leibniz Institute of Plant Genetics and Crop Plant Research (IPK Gatersleben), Seeland, Germany, ²Biology Centre, Czech Academy of Sciences, Institute of Plant Molecular Biology, České Budějovice, Czech Republic, ³Multidisciplinary Institute of Plant Biology, National Council for Scientific and Technical Research (CONICET)-National University of Córdoba, Córdoba, Argentina, ⁴Molecular Cell Biology, Joseph Gottlieb Kölreuter Institute for Plant Sciences (JKIP), Karlsruhe Institute of Technology, Karlsruhe, Germany

Centromeres are essential for proper chromosome segregation. While most species possess monocentric chromosomes with a single centromere, a minority exhibit holocentric chromosomes with centromeres distributed throughout the entire chromatid. The sporadic emergence of holocentrics among monocentric lineages suggests multiple independent mono- to holocentric transitions. To explore this, we compared two sister genera with contrasting centromere types, *Chamaelirium luteum* with large macro-monocentromeres and *Chionographis japonica* with holocentromeres. The broad-scale synteny between the two genomes suggests de novo holocentromere formation in *Chi. japonica*. Kinetochore analysis revealed a chimeric Borealin in both species, and the loss of KNL2 and a chimeric NSL1 in *Cha. luteum*. Both species exhibit chromosome-wide (peri)centromeric histone phosphorylation patterns, distinct from typical monocentrics. Although *Cha. luteum*'s macro-monocentromeres share features with both centromere types, they are not a direct intermediate. We propose these atypical centromeres evolved through mutations in kinetochore genes, alterations in histone phosphorylation, and centromeric satellite DNA amplification.

TALK

Somatic mobility of transposons in *Arabidopsis thaliana*

Heena Ambreen¹, Basile Leduque¹, Leandro Quadra¹, Keith Slotkin¹, Alexandros Bousios¹, **Hans-Wilhelm Nützmänn**¹

¹Department of Biosciences, University of Exeter, Exeter, United Kingdom

The activity of transposable elements (TEs) in somatic cells drives genomic and phenotypic heterogeneity across individual cells of eukaryotic organisms. In plants, the implications of somatic transposition are far-reaching because somatic cells have the ability to redifferentiate into germ cells throughout the life cycle of the organism. However, the genome-wide scale and pattern of TE mobilisation in somatic cells of plants remains unknown.

Here, we adapt TE high-throughput sequencing technology for somatic cells and capture de novo TE integrations in the soma of *Arabidopsis thaliana*. Activating TEs and identifying nascent insertion sites in young leaves, we reveal that somatic transposition occurs at unprecedented scales. We detect more than 200,000 somatic transposition events, allowing us to identify distinct genetic and epigenetic features that are associated with TE integration. We show that TE families insert into different niches, spacing themselves out in the genome ecosystem. Notably, we show that environmentally responsive genes are preferred target sites for TEs in somatic cells. Overall, our work provides evidence for the plasticity of plant genomes at the cellular level as a result of TE mobilisation in the soma. We speculate that this enables host genomes to generate diversity at genomic sites of need.

TALK

Cytogenomics of oat (*Avena*) genome evolution: Intra- and inter-genomic chromosomal translocations, repetitive DNA dynamics, and reconstruction of ancestral grass karyotypes show genome evolution in action

Pat Heslop-Harrison^{1,2}, Trude Schwarzacher^{1,2}, Paulina Tomaszewska^{1,3}, Qing Liu²

¹University of Leicester, Institute for Environmental Futures and South China Botanical Garden, ²South China Botanical Garden, Guangzhou, China, ³University of Wrocław, Wrocław, Poland

Chromosome-scale genome assemblies, sequence reads, and fluorescence *in situ* hybridization (FISH) of *Avena* species reveal key evolutionary mechanisms in the Aveneae tribe, complementing and often contrasting with the Triticeae wheats. We demonstrate how *Avena*'s 10-15-fold genome expansion from rice/*Brachypodium* enables visualization of the ancestral grass karyotype, with uniform chromosome arm size expansion, conserved synteny, repetitive DNA divergence with homogenization, and chromosomal rearrangements including nesting. Notably, *Avena* diploid and polyploid species show large (>50Mb) distal translocations during evolution, contrasting with fewer, often centromeric translocations in wheat. *Avena* polyploids exhibit frequent distal intergenomic exchanges, mapped in tetraploids, hexaploids (AACCCDD), and octoploids using repetitive probes and in genome assemblies. Repetitive DNA, comprising some 80% of the genome (dominated by Ty3/Gypsy retroelements), shows constrained family amplification and genome-specific homogenization, driving sequence evolution. Structural variation is critical to characterize, and the patterns have implications for both speciation in Aveneae and the ways the biodiversity in the Tribe can be exploited in breeding, through targeted strategies for introgressing wild *Avena* traits into hexaploid oats.

We are grateful to our co-authors and collaborators; further information and references are at www.molcyt.org

TALK

The maize B chromosome shapes the transcriptome throughout the entire plant life

Lucie Hloušková^{1,2}, Zuzana Tulpová¹, Radim Svačina¹, Kateřina Holušová¹, Petr Cápál¹, Pavla Navrátilová¹, Miroslava Karafiátová¹, Jan Bartoš¹

¹Institute of Experimental Botany AS CR, Olomouc, Czech Republic, ²Department of Cell Biology and Genetics, Palacký University, Olomouc, Czech Republic

Maize (*Zea mays* L.) is one of the world's most important crops and a well-established model for genetic research. The maize B chromosome has been studied cytogenetically for decades, yet its transcriptional activity and broader regulatory effects remain underexplored. Here, we present an improved reference sequence of the maize B chromosome, along with updated gene annotation and a comprehensive transcriptomic atlas covering its expression throughout the maize life cycle. Using RNA-seq across eleven distinct tissues and developmental stages, we identified a set of B chromosome-specific genes with tissue- and stage-specific expression patterns. In contrast to earlier studies limited to leaf samples, our expanded dataset reveals that the B chromosome is transcriptionally active across the entire lifespan, with the highest activity in reproductive organs. Beyond its own gene expression, we also uncover its influence on genes located on the standard A chromosomes, suggesting a broader regulatory role within the genome. These findings deepen our understanding of the maize B chromosome as an active and dynamic element of the maize genome, with implications for chromosome behavior and regulation.

TALK

Aspects of B chromosome elimination in *Sorghum purpureosericeum*

Tereza Bojdová^{1,2}, Lucie Hloušková^{1,2}, Kateřina Holušová¹, Radim Svačina¹, Eva Hřibová¹, Iva Ilíková¹, Johannes Thiel³, Gihwan Kim³, Roman Pleskot⁴, Andreas Houben³, Jan Bartoš¹, Miroslava Karafiátová¹

¹Institute of Experimental Botany of the Czech Academy of Sciences, Centre of Plant Structural and Functional Genomics, Olomouc, Czech Republic, ²Department of Cell Biology and Genetics, Faculty of Science, Palacký University, Olomouc, Czech Republic, ³Leibniz Institute of Plant Genetics and Crop Plant Research (IPK) Gatersleben, Germany, ⁴Institute of Experimental Botany of the Czech Academy of Sciences, Laboratory of Integrative Structural Biology, Praha 6 - Lysolaje, Czech Republic

We present the first contig-level genome assembly of *Sorghum purpureosericeum*, with a focus on elimination of its accessory B chromosome. The B chromosome is composed of multi-A-chromosomal sequences, enriched in repetitive elements, and features a robust (peri)centromeric tandem repeat, SpuCL166. Despite being gene-poor, it harbors a distinct set of upregulated genes enriched in mitosis-related functions. Among 28 candidates likely involved in B chromosome elimination, we identified B-specific variants of key kinetochore proteins (CENH3, CENP-C, MIS12, NUF2), cohesin and condensin components, and checkpoint regulators. Structural modelling revealed that amino acid substitutions in B-encoded CENH3 and CENP-C are positioned at their interaction interface, supporting the possible formation of an altered B-specific kinetochore. Comparative modelling in maize and *Ae. speltoides* revealed similar convergent evolution of B-specific CENH3 variants. These findings support a B chromosome-autonomous elimination mechanism involving both structural elements (e.g., SpuCL166) and co-evolved mitotic regulators. We propose that pre-determination of B chromosome for elimination is mediated through altered kinetochore assembly and specific centromere composition.

INVITED TALK (ONLINE)

From lab to field: The promise of plant gene editing at scale

Daniel Voytas¹

¹University of Minnesota, St Paul, USA

Plant gene editing is usually carried out by delivering reagents such as Cas9 and sgRNAs to explants in culture. Edited cells are then induced to differentiate into whole plants by exposure to various hormones. Creating edited plants through tissue culture is often inefficient, requires considerable time, only works with limited species and genotypes and causes unintended changes to the genome and epigenome. We have been pursuing alternative approaches for plant gene editing that minimize or obviate the need for tissue culture. In one approach, we generate gene edited dicotyledonous plants through de novo meristem induction. Developmental regulators and gene editing reagents are delivered to somatic cells on whole plants. Meristems are induced that produce shoots with targeted DNA modifications, and gene edits are transmitted to the next generation. In a second approach, we use RNA viruses to deliver sgRNAs through infection to transgenic plants that express Cas9. The sgRNAs are augmented with sequences that promote cell-to-cell mobility and movement into the meristem. Gene edited shoots are thus generated that transmit gene edits to the next generation. Because both approaches minimize the need for tissue culture, they promise to help overcome this bottleneck in plant gene-editing.

TALK

Expanding the toolbox: Prospective uses of the inducible CRISPR-Kill system in plants

Fabienne Gehrke¹, Paola Ruiz-Duarte², Angelina Schindele¹, Sebastian Wolf³, Holger Puchta¹

¹Karlsruhe Institute of Technology (KIT), Karlsruhe, Germany, ²Ruprecht-Karls-University Heidelberg, Heidelberg, Germany, ³Eberhard-Karls-University Tübingen, Tübingen, Germany

Since the first application of the CRISPR/Cas system as a sequence-specific programmable nuclease, its use as a biotechnological tool revolutionized molecular biology. Recently, we expanded the repertoire with CRISPR-Kill, offering new possibilities in biotechnology through targeted genome elimination. Using the SaCas9 nuclease, CRISPR-Kill relies on the induction of multiple double-strand breaks in conserved repetitive genomic regions causing targeted cell death. By simply replacing the constitutive promoter of the Cas nuclease with cell type-specific promoters, the process of organogenesis can be effectively blocked in *Arabidopsis*. Complementing these spatial control features, we extended the CRISPR-Kill system with the option for temporal control using the chemically inducible GR LhG4/pOp6 transactivation system to enable the analysis of targeted cell death in an organotypic functional context. The CRISPR-Kill system offers an exciting and powerful platform for precise cell-type ablation in plants, unlocking new possibilities for the fine-tuned manipulation of specialized metabolite production – critical for pharmaceutical and food applications – but also opens new strategies for disease control in agriculture through targeted ablation of infected cells. Not only does it facilitate tissue engineering, its ability to selectively eliminate entire chromosomes also marks a major advancement for plant breeding, highlighting the system's broad potential across plant science and biotechnology.

TALK

CRISPR-engineered chromosomal translocations point to cis-regulatory control of arm-specific telomere homeostasis and overall robustness of chromatin structure and phenotype in *Arabidopsis*

Ondřej Helia^{1,2}, Barbora Matúšová^{1,2}, Kateřina Havlová³, Anna Hýsková², Martin Lyčka¹, Natalja Beying⁴, Holger Puchta⁴, Jiří Fajkus^{1,2,3}, Miloslava Fojtová^{1,2}

¹Mendel Centre for Plant Genomics and Proteomics, Central European Institute of Technology (CEITEC), Masaryk University, Brno, Czech Republic, ²National Centre for Biomolecular Research, Faculty of Science, Masaryk University, Brno, Czech Republic, ³Department of Cell Biology and Radiobiology, Institute of Biophysics, Academy of Sciences of the Czech Republic, Brno, Czech Republic, ⁴Joseph Gottlieb Kölreuter Institute for Plant Sciences – Molecular Biology, Karlsruhe Institute of Technology (KIT), Karlsruhe, Germany

Using targeted CRISPR/Cas-based chromosome engineering, stable *Arabidopsis thaliana* lines with exchanged arms between non-homologous chromosomes were created (Beying et al., 2020, Nature Plants; Schindele et al., 2020, Current Opinion in Biotechnology). Plants with translocated chromosome arms maintained wild-type morphology through multiple generations, as confirmed by the PCA analysis of multiple phenotypic traits (Helia et al., 2025, Plant Journal). Transcriptomic profiling revealed minimal differential gene expression, with affected loci distributed genome-wide rather than clustering near translocation junctions. Chromatin structure was not altered as there were no significant changes in H3K27me3, H3K4me1, or H3K56ac histone marks near breakpoints or genome-wide. Bulk and arm-specific telomere lengths remained stable across multiple plant generations.

These results demonstrate: (i) remarkable phenotypic and genomic stability of *A. thaliana* despite Mb-scale chromosome rearrangements, (ii) telomere length regulation via cis-acting mechanisms rather than the current chromosomal position, (iii) functional independence of chromatin domains from their native chromosomal context.

The findings support the utilization of CRISPR/Cas-based chromosome engineering as a useful approach for studying plant genome evolution and developing plants with enhanced traits. The observed cis-regulation of telomere lengths provides insights for better understanding of genome stability during large-scale DNA rearrangements in plants.

TALK

Does karyotype change promote species diversity? Revealing hidden patterns in fern diversification**Thomas Buchloh**^{1,2}, Norman Wickett¹, Carrie Tribble³, Michael May⁴¹University Of Vienna, Vienna, Austria, ²Clemson University, Clemson, USA,³University of Washington, Seattle, USA, ⁴University of California, Davis, USA

Karyotype change commonly generates reproductive isolation between diverging species. Rapid karyotype evolution, like that seen in plants, may therefore drive increased rates of speciation. Previous tests of this prediction have identified positive correlations between the rate of karyotype change (most often whole genome duplications) and net diversification, but tests have been restricted to relatively small and young clades in angiosperms. In ferns – a karyotype rich clade of vascular plants – speciation is frequently coupled with whole genome duplication (~30% of speciation events). Therefore, diversification in this clade of plants may also be correlated with karyotype change. To investigate the contribution of karyotype change to fern diversification, we extend the Chromosome Number and Hidden State-dependent Speciation and Extinction model (ChromoHiSSE) to include whole genome duplication. Using the fully parameterized ChromoHiSSE model to investigate the rate of karyotype evolution across 962 leptosporangiate ferns, we recover two hidden modes of chromosome number evolution. We find that lineages with high karyotype lability are less likely to persist in the long-term. Our results reinforce the theory that modern fern diversity is shaped substantially by polyploid speciation but challenge the expectation that diversification rates are enhanced by karyotype-driven speciation.

TALK

Plant chromosome architecture revealed by advanced electron microscopy**Nobuko Ohmido**¹, Channarong Sartsanga¹, Astari Dwiranti^{2,3}

¹Graduate School of Human Development and Environment, Kobe University, Kobe, Japan, ²Cellular and Molecular Mechanisms in Biological System (CEMBIOS) Research Group, Department of Biology, Faculty of Mathematics and Natural Sciences, Universitas Indonesia, Depok, Indonesia, ³Institute for Advanced Sustainable Materials Research and Technology (INA SMART), Faculty of Mathematics and Natural Science, Universitas Indonesia, Depok, Indonesia

Chromosomes are essential for the faithful transmission of genetic information, but their higher-order architecture remains only partially understood. This study highlights recent progress in visualizing plant chromosome structure using advanced electron microscopy techniques. Improvements in sample preparation – such as chromosome isolation and ionic liquid coating – have enabled better preservation of native chromatin structures.

Using Helium Ion Microscopy (HIM), Scanning Electron Microscopy (SEM), Focused Ion Beam-SEM (FIB-SEM), Electron Tomography (ET), and High-Voltage Transmission Electron Microscopy (HVTEM), we observed fine-scale chromatin organization in both centromeric and non-centromeric regions. As a model, we used barley (*Hordeum vulgare*), which provides large and well-differentiated chromosomes suitable for ultrastructural analysis. We explored the involvement of axis-associated proteins, topoisomerase II, in chromosome condensation and mechanical stability. Topo II exhibited stage-specific distribution patterns during barley chromosomes condensations. The role of divalent cations in chromatin compaction appears to be conserved in both plant and human.

Comparative observations between plant and human chromosomes reveal both conserved and plant-specific structural features. These insights highlight the diversity of chromosome architecture. These findings illustrate how advanced electron microscopy contributes to our understanding of chromosome architecture across species and will remain essential for advancing our knowledge of chromosome structure.

TALK

Analyzing the evolutionary history of a uniquely polymorphic chromosome of the genus *Aquilegia*

Francesca Beclin¹, Gökçe Aköz¹, Daniele Filiault^{1,2}, Robin Burns^{1,3}, Anna Igoikina¹, Almudena Mollá Morales¹, Viktoria Nizhynska¹, Magnus Nordborg¹

¹Gregor Mendel Institute, Vienna, Austria, ²Bayer Crop Science, St. Louis, USA,

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Evolutionary forces such as selection and introgression can affect sequences differently across the genome, resulting for example in variations in polymorphism, differentiation and local phylogenies. In the genus *Aquilegia*, a unique phenomenon has recently been discovered: one of its chromosomes – chromosome 4 – exhibits higher polymorphism and distinct phylogenetic properties from the other chromosomes. As this discovery was made using only one individual per species, population genomic data was required to control for potential bias in polymorphism estimation. In this study, we sequenced the genomes of 76 European *Aquilegia* accessions, mapped them to a European reference genome, called variants, analyzed the resulting SNP data, and reanalyzed data of 10 worldwide accessions. We described population structure of European *Aquilegia* and confirmed that chromosome 4 indeed exhibits higher nucleotide diversity and distinct phylogenetic clustering. We also found that chromosome 4 differs from the other chromosomes in its distribution of polymorphism along the chromosome, and, curiously, that chromosome 4 seems to share a phylogeny with the arms of the other chromosomes, while their centromeres exhibit a different phylogeny. Our study provides the first population genomic description of European *Aquilegia*, facilitating the investigation of the evolution of a uniquely aberrant chromosome.

TALK

Cross-kingdom horizontal gene transfers between plant and fungal genomes: Insights from large-scale comparative genomics

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Horizontal gene transfer (HGT) is increasingly recognized as a significant driver of eukaryotic genome evolution. In this study, we conducted a large-scale analysis of cross-kingdom HGT between plants and fungi using whole-genome comparisons across hundreds of species. Employing rigorous phylogenetic and sequence-based methods, we identified dozens of novel HGT events, predominantly involving gene transfer from fungi to plants. These transfers were especially prevalent in bryophytes and grasses, with many cases in grasses originating from endophytic fungi of the *Epichloë* genus. Furthermore, we identified a rare case of plant-to-fungi HT involving a 2 kbp plant-derived transposable element (TE) integrated into the genome of a parasitic fungus. Remarkably, this plant TE proliferated extensively within the fungal genome, creating over a hundred copies and significantly altering its genomic landscape. Additionally, our analysis provides evidence suggesting this fungus subsequently mediated secondary transfers of the plant TE back into multiple plant genomes, highlighting its role as a vector for plant-to-plant HT. Collectively, these findings underscore the importance of HGT in promoting genomic plasticity and innovation across the plant kingdom.

TALK

Evolutionary tracing of bivalent and univalent subgenomes in pentaploid dogroses (*Rosa* sect. *Caninae*)

Veit Herklotz¹, Meng Zhang², Thiago Nascimento², Radka Kalfusová³, Jana Lunerová³, Jörg Fuchs⁴, Dörte Harpke⁴, Bruno Huetten⁴, Ursula Pfordt², Volker Wissemann⁶, Aleš Kovařík³, André Marques², Christiane Ritz¹

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Meiotic chromosome pairing is a critical step in sexual reproduction. In allopolyploids, the presence of multiple homoeologous subgenomes can disrupt balanced pairing and segregation, often leading to sterility – especially in odd-numbered ploidy levels. Remarkably, pentaploid dogroses (*Rosa* sect. *Caninae*, $2n = 5x = 35$) circumvent this problem via a unique meiosis: 14 chromosomes form 7 bivalents are inherited biparentally, while the remaining 21 chromosomes remain unpaired and are transmitted solely through the maternal lineage. Consequently, the pentaploid chromosome complement is reconstituted in each generation.

To investigate the genomic basis of this meiosis, we analysed haplotype-resolved, chromosome-level genome assemblies for three dogroses. Subgenome phasing identified one bivalent-forming subgenome consisting of two nearly homozygous chromosome sets, alongside three divergent subgenomes that persist as univalents. Comparative analyses using single-copy orthologues including data from diploid roses revealed that these subgenomes originate from two distinct evolutionary clades. Pollen genome analyses further showed that subgenomes of different evolutionary origins can form bivalents, indicating flexibility in subgenome pairing across lineages. This supports the hypothesis of the multiple hybridogenic origin of dogroses. Our results provide new insights into the genomic architecture and its role in maintaining sexual reproduction in a system with extreme chromosomal asymmetry.

TALK

Interdependent HEI10 paralogs and their role in plant meiotic recombination

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RING finger proteins are essential for meiotic recombination across eukaryotes. Their duplications often lead to new functions. Plants possess a RING finger protein called HEI10, with a key role in regulating meiotic crossovers. We recently discovered two paralogs (HEI10a and HEI10b) in the holocentric *Rhynchospora* plant genus, generated from a duplication 60MYA. Both copies display conserved functional RING and coiled-coil domains. However, their C-terminal regions are divergent. Paralog-specific antibodies revealed distinct immunolocalisation patterns in *Rhynchospora*, with HEI10b restricted only to final crossovers, while HEI10a already visible on early precursors, as observed in other plants. To assess the function of HEI10a and HEI10b, we used them to attempt to restore fertility in an *Arabidopsis* hei10 null mutant. While single transgenes failed to restore fertility, the double insertion partially rescued the sterile phenotype. Folding predictions support a model in which HEI10a and HEI10b form a heterodimer. We also developed a transformation protocol for *Rhynchospora* radicans. We generated multiple CRISPR lines for single knockouts of HEI10a and HEI10b, currently undergoing screening, and double mutants are in preparation. Our data suggest that HEI10a and HEI10b are two subfunctionalised, interdependent RING finger proteins, advancing our understanding of the evolution of recombination proteins in plants.

TALK

Most tobacco twin spots result from chromosomal instability and not homologous recombination

Isabelle J. DeMarco¹, Kirk Amundson¹, Isabelle M. Henry¹, **Luca Comai**¹

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The frequency of homologous recombination (HR) between chromosomes in somatic plant cells remains unclear. In tobacco heterozygous for the yellow sulfur mutation, green and white twin spots have traditionally been attributed to mitotic crossover. To test this hypothesis, we sequenced DNA from twin spots and surrounding tissues, expecting HR to be structurally neutral. Contrary to expectations, 21 of 23 twin spots exhibited structural rearrangements – specifically, reciprocal translocations between the sulfur locus and other chromosomes. Only one twin spot showed a pattern consistent with HR, but the crossover occurred near the centromere, where many DNA breaks accumulated, suggesting a coincidental rearrangement. Among 26 more frequent single spots, most resulted from deletions. Together, these findings indicate that nearly all spots arise from DNA instability – deletions, translocations, and other rearrangements – mediated by nonhomologous repair rather than by HR. These rearrangements altered the dosage of the mutant allele, affecting Mg chelatase function and chlorophyll levels. We conclude that mitotic recombination between homologs is rare, and that genome instability is the dominant driver of somatic sectoring in this system.

INVITED TALK

Uncovering the unexpected features of nuclear division in diatoms

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Cell division is a fundamental process in all living organisms. In eukaryotes, mitosis produces two genetically identical daughter cells through a sequence of DNA replication, nuclear division, and cytokinesis. While mitosis is well studied in model organisms, its regulation remains poorly understood in many non-model species, including diatoms.

Diatoms are a group of unicellular microalgae that thrive in unstable marine and freshwater environments and contribute around 40% of marine primary production. Their ecological success is linked to rapid cell division during massive blooms in marine and freshwater systems.

Although studies dating back to the 19th century suggest that diatoms undergo closed mitosis with unusual cytoskeletal features, the underlying regulatory mechanisms remain largely unknown. In our study, we used cell cycle synchronisation, publicly available transcriptomic data, live-cell imaging and immunofluorescence to examine mitosis in several diatom species. We found that mitotic timing is species-specific and we uncovered evidence that diatoms may possess more than one mode of nuclear division, indicating a previously unrecognised flexibility in their mitotic strategies.

Understanding how diatom mitosis is regulated may provide insights into their genomic organization, phenotypic plasticity, and ability to adapt to diverse environments.

TALK

The chromonema workout

Amanda Camara¹, Claudio Gemsa¹, Alžběta Doležalová², Pavla Navrátilová², Thiago Nascimento³, Kateřina Kaduchová², Martin Mascher¹, Veit Schubert¹, Eva Hřibová², Hana Šimková²

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As a very long polymer, chromatin can fold into a variety of shapes. Which shape depends on the context, affecting and being affected by transcription regulation mechanisms. Through the cell cycle, chromatin gets compacted and decompacted, probably through mechanisms as general as the cell cycle process itself. Loop extrusion is one of them. Recruiting many protein molecules of the Structural Maintenance of Chromosomes family (SMC), each chromosome is compacted into an array of loops, forming a structure that we like to call “chromonema”. The chromonema, with a prophase-like shape, might be flexible enough to coil into a metaphase-like shape, which has been described to follow a general helical path. With polymer simulations, we force the coiled chromonema into a workout – looping, stretching and squeezing it – trying to reproduce HiC results from different stages of the cell cycle of barley chromosomes. Our models suggest that, rather than a scaffold of proteins with specific coiling coordinates, the dynamic exchange of SMC proteins and changes in the chromosome environment are indirectly coiling and uncoiling the chromonema. Though more experiments are needed to test the chromonema’s properties, this hypothesis might lead to a general mechanism of chromosome compaction.

TALK

Understanding the role of chromatin mobility in genome integrity

Valentin Baptiste¹, Anis Meschichi², Myrtille Barrot³, Konstantin Kutashev⁴, Nuria Badia¹, Frederic Pontvianne³, Stefanie Rosa¹

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Chromatin exhibits dynamic structural and physical properties which are critical for the regulation of numerous nuclear processes. In our research, we investigate chromatin mobility in the context of DNA damage repair (DDR) in plants. Plant cells are continuously exposed to exogenous and endogenous factors that threaten genome integrity. While the core components of the DDR pathway are relatively well described, much less is known about how chromatin dynamics, particularly chromatin mobility, is altered in the presence of DNA damage.

To address this, we are examining how double-strand breaks (DSBs) influence chromatin mobility in *Arabidopsis thaliana*. Using DNA-labeling systems (LacO/lacI and ANCHOR) and homologous recombination (HR) reporters, we have found that DSBs lead to increased chromatin mobility both locally at break sites and globally across the genome. Importantly, this enhanced mobility is dependent on SOG1, a key regulator of the plant DDR.

We are currently dissecting the molecular mechanisms driving this response and have identified chromatin remodelers as potential mediators of damage-induced chromatin mobility. Our findings suggest that regulated changes in chromatin mobility are an integral component of the plant DNA damage response, with chromatin remodelers likely playing a central role in coordinating genome dynamics during repair.

INVITED TALK

Sequence-based cytogenetics reveals dynamic sex chromosome evolution within the genus *Asparagus*

James Leebens-Mack¹

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The last common ancestor of all extant flowering plants has been inferred to be hermaphroditic with perfect flowers containing male and female reproductive organs. Since the origin of angiosperms, there have been hundreds of independent shifts from hermaphroditism to dioecy. This situation offers an excellent opportunity to test classical hypotheses for the origin and evolution of sex chromosomes associated with the shift from hermaphroditism to dioecy. As with many dioecious plants, sex chromosomes in dioecious *Asparagus* species are homomorphic. Phylogenomic analyses have confirmed two independent shifts to dioecy within the genus that occurred between 1 and 4 million years ago. Comparative analyses of sequenced and assembled X and Y chromosomes between and within these two dioecious *Asparagus* lineages reveal significant variation in the genes residing in non-recombining sex determination regions and dynamic structural evolution. These findings will be placed in the context of sex chromosome research across the plant tree of life, and prospects for ongoing efforts to understand the molecular and evolutionary processes influencing the origin of dioecy and the evolution of sex chromosomes.

INVITED TALK

Maximizing meiotic crossover rates

Raphael Mercier¹

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Meiotic crossovers are rare, typically one to three per chromosome with common sexual dysmorphism (heterochiasmy). Crossovers tend to be distant from each other, a phenomenon called crossover interference whose mechanisms have been a matter of debate for over a century. Finally, crossovers are non-homogeneously distributed and are notably suppressed in proximity to centromeres.

Multiple mechanisms limit meiotic crossovers. Mutation of the corresponding genes led to a spectacular increase in genome-wide recombination. Additional manipulation of crossover regulators does not further increase crossovers but shifts the balance between crossover pathways, suggesting competition for a limited precursor pool. While wild-type crossover patterns differ between sexes, mutant crossover landscapes converge on a unique distinct profile, which we term crossover potential (COP). We propose that COP reflects the density of eligible recombination precursors, which is determined by genomic features, with sexual dimorphism resulting solely from differential regulation of their maturation into crossovers.

Our data and previous results support a unifying model for crossover control, in which the coarsening dynamic of the HEI10 pro-crossover protein along chromosomes designates crossover position.

Finally, we identified genetic factors that limit crossover in proximity to centromeres, suggesting that crossovers are actively suppressed there and opening new possibilities for plant breeding.

TALK

The central element protein SCEP3 interlinks crossover and synaptonemal complex formation

Chao Feng¹, Jana Lorenz¹, Steven Dreissig^{1,2}, Veit Schubert¹, Baicui Wang¹, Maria Cuacos¹, Nadia Fernández-Jiménez³, Ziliang Zhao^{4,5}, Christian Eggeling^{4,5}, Amanda Souza Camara¹, Axel Himmelbach¹, **Stefan Heckmann**¹

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Genetic variation exploited in breeding relies largely on meiotic crossovers (COs), which arise through homologous recombination. During prophase I, sister chromatids are aligned along chromosome axes, and the synaptonemal complex (SC) links homologs during pachytene. CO formation/patterning depends on the interplay between homologous recombination and axis remodeling, including SC assembly. Despite its structural conservation, a complete picture of the SC's composition and its role in CO formation remains lacking in plants. However, the limited number of meiocytes embedded in floral organs constrains proteomic approaches, to dissect its composition.

In *Arabidopsis*, using TurboID-based proximity proteomics, we identified novel meiotic proteins, including SCEP3, which localizes to the central region of the SC. Loss of SCEP3 disrupts SC formation and abolishes both CO assurance (ensuring that each chromosome pair receives at least one CO) and CO interference (the tendency for COs to be spaced non-randomly along chromosomes). Its absence also increases female CO frequency, eliminating sex-specific differences in CO patterning, known as heterochiasmy. Additionally, SCEP3 is critical for some COs arising in SC-deficient mutants, and it interacts with the transverse filament protein of the SC, ZYP1, but loads independently of ZYP1 and other SC components.

TALK

How to establish inter-homologue bias during meiosis**Peter Schlögelhofer¹**¹Max Perutz Labs, University of Vienna, Vienna, Austria

During meiosis, SPO11 mediated DNA double strand breaks (DSBs) are processed yielding recombinogenic ssDNA overhangs. These are loaded with the recombinases DMC1 and RAD51 and are subsequently guided to invade repair templates located at the homologous chromosomes (rather than at the sister chromatids), a process also known as inter-homologue bias (IH-bias). Multiple factors, identified in different organisms, were shown to support IH-bias: HOP2 and MND1 emerged as common denominators across kingdoms, but factors like MEI5, SAE3, MEK1 or HED1 were only found in some species and not in plants. Irrespective of the involved players or organisms, a comprehensive model for IH-bias is still missing.

We propose that interhomolog (IH) bias arises from DSB-induced, spatially restricted post-translational modifications (PTMs) that locally alter protein interactions, abundance, or localization. Such PTMs may deplete IH-promoting factors and recruit IH-antagonizing ones near DSBs, creating a distinct microenvironment favoring strand invasion at homologues over sister chromatids. Supporting this model, phospho-mimicking HOP2T25E and MND1S08E variants with reduced dsDNA binding fail to form bivalents, consistent with inter-sister repair. Additionally, our Cryo-EM analyses of ssDNA/DMC1/HOP2/MND1 complexes inform a mechanistic model of meiotic strand invasion and biased repair that we are currently testing.

INVITED TALK

Karyotype engineering with synthetic centromeres

Kelly Dawe¹

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Karyotype engineering is a form of genome editing that involves altering large segments of a genome, joining chromosomes, or potentially increasing the number of chromosomes. Creating a new chromosome is particularly challenging because it requires engineering a new centromere. We have demonstrated that a LexA-CENH3 tethering approach can activate new centromeres at maize synthetic repeat arrays containing LexO binding sites (CENH3 is a key centromere protein). Chromosome breakage of the initially dicentric chromosomes liberates neochromosomes composed of pieces of chromosome 4L. The neochromosomes can be maintained as partial trisomics for multiple generations. In recent work we have identified truncated forms of normal chromosome 4 and matched them with neochromosomes to create diploid lines with 11 chromosomes instead of the usual 10. We call these new chromosomes 4a and 4b, where chromosome 4b contains a synthetic centromere. Chromosome 4b is accurately transmitted through both mitosis and meiosis when propagated in the 11-chromosome line, demonstrating that synthetic centromeres can be fully functional.

TALK

Centrophilic retrotransposon integration via CENH3 chromatin in *Arabidopsis*

Sayuri Tsukahara¹, Alexandros Bousios², Estela Perez-Roman², Sota Yamagichi¹, Basile Leduque³, Aimi Nakano¹, Matthew Naish⁴, Akihisa Osakabe¹, Atsushi Toyoda⁵, Hidetaka Ito⁶, Alejandro Edera³, Sayaka Tominaga¹, Juliarni Juliarni¹, Kae Kato⁵, Shoko Oda¹, Soichi Inagaki¹, Zdravko Lorković⁷, Kiyotaka Nagaki⁸, Fred Berger⁷, Akira Kawabe⁹, Leandro Quadrana³, Ian Henderson⁴, **Tetsuji Kakutani**¹

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In organisms ranging from vertebrates to plants, major components of centromeres are rapidly-evolving repeat sequences, such as tandem repeats (TRs) and transposable elements (TEs), which harbor centromere-specific histone H3 (CENH3). Complete centromere structures recently determined in human and *Arabidopsis* suggest frequent integration and purging of retrotransposons within the TR regions of centromeres. Despite the high impact of “centrophilic” retrotransposons on the paradox of rapid centromere evolution, the mechanisms involved in centromere targeting remain poorly understood in any organism. Here we show that both Ty3 and Ty1 LTR retrotransposons rapidly turnover within the centromeric TRs of *Arabidopsis* species. We demonstrate that the Ty1/Copia element Tal1 (Transposon of *Arabidopsis lyrata* 1) integrates de novo into regions occupied by CENH3 in *A. thaliana*, and that ectopic expansion of the CENH3 region results in spread of Tal1 integration regions. The integration spectra of chimeric TEs reveal the key structural variations responsible for contrasting chromatin-targeting specificities to centromeres versus gene-rich regions, which have recurrently converted during the evolution of these TEs. Our findings reveal the impact of centromeric chromatin on TE-mediated rapid centromere evolution, with relevance across eukaryotic genomes.

TALK

Genetic diversity and dynamic evolutionary innovations in plant centromeres**Handong Su¹**¹Huazhong Agricultural University, Wuhan, China

Centromeres are indispensable for accurate chromosome segregation, but are subject to rapid sequence turnover while maintaining conserved functions. To unravel this dichotomy, we integrated 190 fully resolved centromeres from eight diploid angiosperms spanning 150 million years of divergence, along with 436 pan-genomic assemblies, 581 pan-genomic and 284 congeneric whole-genome sequencing datasets. Our study shows that centromere organization is determined by lineage- or species-specific combinations of satellite repeats and TEs, which in turn shape distinct epigenetic landscapes and evolutionary trajectories within centromeres. In particular, TE insertion patterns are found to be key drivers of structural diversification and positional shift of angiosperm centromeres. Intriguingly, population-level analyses also uncovered considerable plasticity in centromere sequences within species, with satellite repeats acting as focal points of evolutionary change and displaying species-specific heterogeneity patterns. Temporal reconstructions in congeneric species revealed the initial emergence and differentiation of centromeric repeats and chart a dynamic continuum ranging from a gradual sequence diversification to a complete turnover during speciation, often accompanied by karyotype reorganization. By integrating intra- and inter-species comparisons, we propose a unifying framework in which centromere innovation is governed by a delicate interplay between genome evolution, chromosomal shuffling and selection constraints, resulting in phylogenomic signatures of centromere-driven speciation.

TALK

A 2-bp deletion causes the formation of unreduced pollen in *Hydrangea macrophylla***Conny Tränkner¹**, Helmut Volk¹, Katharina Weigl¹, Anke Müller¹, Katja Krüger¹¹University of Applied Sciences Erfurt, Erfurt, Germany

Polyploidization through unreduced gametes is a key player in evolution. Furthermore, it is a powerful tool for the production of polyploid offspring. Crosses between diploid cultivars of the ornamental plant *Hydrangea macrophylla* resulted in 50-98% triploid offspring due to spontaneous polyploidization through unreduced pollen. The production of unreduced pollen follows a monogenic dominant-recessive inheritance. Genetic and physical mapping approaches located this so-called UNREDUCED POLLEN (UP) locus within a 3.9 Mbp interval on chromosome 13. Fine-mapping reduced the interval to 105,528 bp, containing four gene models. One gene showed 43% protein identity to a meiosis gene of *Arabidopsis thaliana*, whose mutant phenotype was similar to *H. macrophylla* plants that were homozygous for a 2-bp deletion at the 5th exon. This deletion results in a truncated, putative loss-of-function protein and explains the phenotypic variation within 93 hydrangea cultivars. Pollen mother cells of mutated plants formed dyads, triads and tetrads, whose gametes displayed both homozygosity and heterozygosity within the genome, indicating disordered cytokinesis after pollen meiosis I/II. Hence, the unreduced pollen is genetically diverse, resulting in offspring variations. Introducing this mutation into other species can facilitate the breeding of triploid, seedless varieties of crop plants such as banana, melon, or grape vine.

TALK

Evolutionary conservation of the monopolin component Csm1 in sister chromatid mono-orientation during meiosis in *Arabidopsis*

Zhijian Zhang¹, Franziska Böwer¹, Shinichiro Komaki², Aladár Pettkó-Szandtner³, Eelco Tromer⁴, Zoltán Magyar³, Maren Heese¹, Arp Schnittger¹

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Unlike mitosis, during which sister chromatids are separated, the kinetochores of the sister chromatids are mono-orientated in the first meiotic division causing the segregation of entire chromosomes to opposite cell poles. In budding yeast, mono-orientation is achieved by the Monopolin complex, which includes Csm1. However, outside of yeast, little is known how kinetochore mono-orientation is accomplished. Here, we characterized TITAN9 (TTN9), a Csm1-related gene, in *Arabidopsis*. The loss of TTN9, as previously shown, causes embryonic lethality. However, its molecular function has not been clear especially since TTN9 does not share some of the residues with Csm1 that have been suggested to be crucial for its meiotic function. We find that TTN9 is present during the entire cell cycle in *Arabidopsis* and directly binds to kinetochores. Correspondingly, we find that TTN9 binds to kinetochore components and itself in affinity purification experiments and in Y2H assays. Strikingly, knock-down of TTN9 leads to defects in mono-orientation of sister chromatids, providing evidence for a conserved role of a monopolin-like complex outside of yeast. Given the evolutionary distance of yeast and plants, we postulate that already the last common ancestor of all eukaryotes had a monopolin-like complex, which co-oriented sister chromatids in meiosis.

TALK

Ubiquitin-dependent proteolysis of KNL2 driven by APC/C-CDC20 is critical for centromere integrity and mitotic fidelity

Manikandan Kalidass¹, Venkata Ganesh Jarubula¹, Jothipriya Ramakrishnan Chandra¹, Samuel Le Goff², Aline V Probst², Klaus D Grasser³, Jérôme F Gagneux⁴, Florian A Steiner⁴, Dmitri Demidov¹, Pascal Genschik⁵, Inna Lermontova¹

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Kinetochores are large protein complexes that serve as attachment sites for spindle microtubules, ensuring proper chromosome segregation during cell division. KINETOCHORE NULL2 (α KNL2) is a key kinetochore protein required for the incorporation of the centromeric histone variant CENH3. The precise regulation of α KNL2 levels is crucial, but the molecular mechanisms controlling this process remain largely unexplored. In this study, we demonstrated that the Anaphase-Promoting Complex/Cyclosome (APC/C) mediates the ubiquitin-dependent proteolysis of α KNL2 during mitosis. Our findings revealed that α KNL2 accumulates in the presence of 26S proteasome inhibitors, and our yeast 2-hybrid and proteomic screens showed that proteins from the ubiquitin-proteasome pathway interact with KNL2 in *Arabidopsis thaliana* and nematode (*Caenorhabditis elegans*). *Arabidopsis* α KNL2 directly interacts with Anaphase-Promoting Complex subunit 10 (APC10) and Cell Division Cycle 20.1 (CDC20.1), 2 substrate recognition components of the APC/C. RNAi-mediated depletion of APC/C resulted in the accumulation and mislocalization of endogenous α KNL2.

Additionally, mutation or deletion of the D-box1 region, or substitution of residues K336 and K339, impaired α KNL2 degradation. The expression of a proteasome-resistant α KNL2 variant in planta caused severe defects in growth, fertility, and mitotic division. These findings show that APC/C-CDC20-mediated degradation of α KNL2 is critical for proper kinetochore function and centromere integrity.

TALK

Plant telomere-binding proteins**Jana Fulnečková**¹, Jana Faturová¹, Jaroslav Fulneček¹, Karel Říha¹¹CEITEC, Masaryk University, Brno, Czech Republic

Although sought after for over 30 years, a functional plant equivalent of the shelterin complex that protects eukaryotic telomeres has remained elusive. We employed a proteomic approach in *Arabidopsis thaliana* to identify nuclear proteins that preferentially bind telomeric DNA. Among the putative candidates, we identified three phylogenetically related proteins of the TRF-like (TRFL) family that have not yet been characterized. These proteins form homo- and heterodimers and bind telomeric DNA in vitro. Their localization, observed in transiently transformed mesophyll protoplasts as well as in the roots of stably transformed *Arabidopsis* lines, revealed nuclear speckles closely associated with the nucleolus. This pattern is reminiscent of nucleolus-associated telomere clustering typical of *Arabidopsis* somatic cells. Furthermore, we validated their binding to telomeres using chromatin immunoprecipitation. Notably, simultaneous disruption of all three TRFL proteins leads to telomere lengthening, whereas a mutation producing a truncated protein lacking the C-terminal Myb domain results in telomere shortening. Together, these findings suggest that the identified TRFL proteins are canonical and functionally relevant components of plant telomeres and their further characterization will shed light on the mechanisms governing chromosome-end protection in plants.

TALK

Deciphering the molecular structure of rDNA loci in the aquatic monocot plants *Spirodela polyrhiza* and *S. intermedia*

Anton Stepanenko^{1,2}, Veit Schubert¹, Guimin Chen³, Phuong Hoang⁴, Todd Michael⁵, Eric Lam⁶, Ingo Schubert¹, **Nikolai Borisjuk**²

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The genome surveys of the two most ancient duckweed species, *Spirodela polyrhiza* and *S. intermedia*, revealed an unusually low copy number of 35S and 5S rRNA genes, compared to other plant species. Taking advantage of this specific feature of *Spirodela* genomes, we revealed the detailed molecular organization of the species' rDNA loci through a combination of molecular cytology, extra-long DNA reads and conventional sequencing of rDNA gene units. In both species, the GC-rich rDNA repeats of 35S and 5S rDNA genes are surrounded by highly AT-enriched sequences with possible regulatory functions. Further detailed analysis of the 5S rDNA gene clusters demonstrated that they are localized on two different chromosomal loci, and are composed of locus-specific repeat units, differing in length and nucleotide composition of their non-transcribed spacers. We also demonstrated haplotype specificity of 5S rDNA arrays in *S. polyrhiza*, manifested in copy number variation between homologous chromosomes and sequence divergence for the locus with longer type rDNA repeats. In summary, this study advances our understanding of the basic principles of rDNA organization in plants by revealing the molecular architecture and evolutionary dynamics of the 35S and 5S rDNA loci, which are extendable to other eukaryotic genomes.

TALK

Population-wide single-pollen genotyping in rye sheds light on the genetic basis and environmental plasticity of meiotic recombination

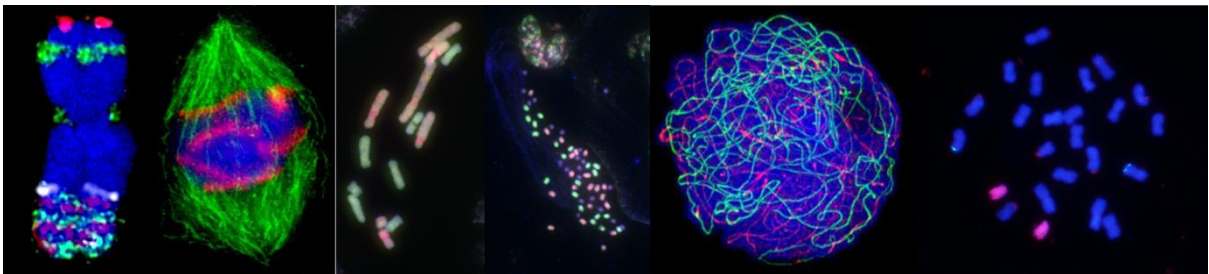
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The core molecular machinery of meiosis is conserved deep across eukaryotic lineages. Nevertheless, meiotic genes and proteins show sequence variation even within species. Patterns of meiotic recombination vary at multiple scales. In order to improve our understanding of the causes and consequences of this variation, we need to identify the underlying genetic architecture. Here, we explored the genetic basis and environmental plasticity of meiotic recombination in a large rye population grown under control and nutrient deficiency conditions. We used single-pollen nuclei genotyping to directly measure male meiotic crossovers in 2539 pollen nuclei from 476 individuals, and detected a significant reduction of crossover frequency in response to nutrient deficiency (-8%). Genome-wide association scans for crossover count, crossover interference, and intra-chromosomal shuffling, revealed a complex oligogenic architecture of these traits. Most loci associated with crossover traits were unique to control or nutrient deficiency conditions, although the underlying allelic distribution was the same, suggesting that alleles regulating crossover traits act in response to nutrient availability. Furthermore, we found that large effect crossover modifiers are kept under purifying selection. Finally, we uncovered differences in recombination landscapes measured in pollen and plants, which may be explained by a survivorship bias in meiosis.

ABSTRACTS

Posters



POSTER 1

Oligo-based chromosome painting for specific chromosome identification and chromosome rearrangement confirmation in *Brassica* species

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Polyploidization and interspecific hybridization are important processes for crop improvement. The *Brassica* genus is a good model to study and understand these events, including diploid species and their allotetraploid hybrids. Interspecific hybridization has been used to produce *Brassica* plants with different subgenome compositions where chromosome rearrangements between subgenomes are common and can make substantial contributions to crop traits. Here, we developed and tested specific chromosome probes to differentiate the highly similar homoeologous chromosomes A01 and C1, which undergo frequent translocation events in *Brassica* hybrids. Specific signals for one single chromosome pair for A01 oligos in *B. rapa* and *B. napus* species and one for C1 oligos in *B. oleracea* and *B. napus* species were observed. In addition, a reciprocal translocation between the A01 and C1 pairs in *B. napus* Surpass400_024DH and a non-reciprocal translocation of an A01 fragment into a C1 pair in an allohexaploid *Brassica* hybrid (*B. carinata* × *B. rapa*) were confirmed. Identification of A01 and C1 chromosomes during meiosis is possible, allowing to observe the pairing behavior of these chromosomes. These probes can be applied to answer breeding and research-related questions related to genome dynamics, chromosome rearrangements and meiotic behavior in this agriculturally important taxon.

POSTER 2

Chromothripsis induced by distant hybridization regulates genome stability and salt tolerance in wheat-barley additional line**Yiqian Chen**¹¹Huazhong Agricultural University, Wuhan, China

Structural variations are hallmark genomic features of distant hybridization progeny. In this study, we identified localized chromothripsis on chromosome 7D in descendants of wheat-barley 4H additional lines (WBA4). Genomic analyses revealed complex structural variations in the chromothripsis-affected regions, including localized copy number variations (CNVs) and inversions. Mechanistic investigations demonstrated that transposable element activity and repeat sequence-mediated non-homologous recombination likely act synergistically to induce chromatin fragmentation, facilitating large-scale genome rearrangements. Notably, chromothripsis events persisted in the S1 generation, with dynamic alterations observed in both the composition and size of ring chromosomes in selected S1 progeny. Transcriptome profiling showed significant enrichment of differentially expressed genes within chromothripsis regions, potentially attributable to CNV-associated gene dosage effects. Most importantly, salt stress screening of S2 generation yielded lines exhibiting substantially enhanced tolerance compared to the parental WBA4. These findings provide compelling evidence that chromothripsis serves a dual function in polyploid crops: while inducing genomic instability, it simultaneously generates selectable structural variations that facilitate rapid environmental adaptation. Our study establishes the first causal link between crop chromothripsis and stress resilience, offering novel insights into genome evolution and potential applications for developing stress-tolerant crops through targeted genomic rearrangement induction.

POSTER 3

Unveiling the hidden genetic exchanges between lianas and trees

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Horizontal transfer (HT) refers to the exchange of genetic material between species outside of sexual reproduction. While most known cases involve parasitic plants and their hosts, little is known about the extent and routes of HT in non-parasitic species. We recently demonstrated that two non-parasitic climbers, *Hedera helix* (common ivy) and *Dioscorea communis* (black bryony), have acquired genetic material from multiple tree species. In this study, we performed a comprehensive genome-wide screen for HT events among 11 climbing plant species from a natural ecosystem, using a comparative analysis against over 700 plant genomes. High-quality de novo genome assemblies of these lianas were generated using PacBio long-read sequencing and optical mapping. Our analyses uncovered an unexpectedly high frequency of HTs, not only between lianas and trees but also among different liana species. These transfers predominantly involved LTR-retrotransposons of the Copia lineage and varied significantly across liana species. These findings reveal that HT is more common in non-parasitic plants than previously thought and highlight lianas as key players in interspecies genetic exchange in plants.

POSTER 4

The *Crocus* panrepeatome: Dynamics, dysploidy, and diversification

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Dysploidy is a crucial driver of species diversification, as it alters karyotypes through diploidization following whole-genome duplication (WGD). WGD can trigger repeat bursts; however, our understanding of the evolutionary relationships between WGD, repeat bursts, and descending dysploidy is limited. Using *Crocus* as a model, we performed a panrepeatome analysis to gain insights into the influence of WGD on repeat bursts and of repeat dynamics in descending dysploidy. We identified a WGD event before the initial divergence of *Crocus* (Cr- β) and nested WGD events (Cr- α) before the divergence of some series. Repeat dynamics corresponded with cladogenesis. One satellite repeat (Crosat003) indicated an involvement in descending dysploidy in the series Verni. This work demonstrated the links between WGD, repeat bursts, and descending dysploidy, which have shaped the species diversification in *Crocus*.

POSTER 5

Dynamic patterns of repeats and retrotransposons in the centromeres of *Humulus* species

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Centromeres are essential chromosomal regions required for accurate segregation during cell division. They are typically composed of large arrays of tandem repeats and centrophilic retrotransposons, characterized by unique chromatin and epigenetic features. While centromere function is highly conserved across eukaryotes, centromeric DNA sequences exhibit remarkable diversity, even among closely related species, with rapid centromere turnover reported, for example, in *Arabidopsis* and *Drosophila*. The dioecious plants common hop (*Humulus lupulus*) and Japanese hop (*Humulus japonicus*), both members of the Cannabaceae family, diverged approximately 3.7-10.7 mya and differ in genome size, chromosome number, and sex chromosome systems. Despite the increasing availability of genomic resources and high-quality genome assemblies, the structure and organization of *Humulus* centromeres remain poorly understood.

Utilizing a combination of bioinformatics, molecular, and cytogenetic approaches, we investigated the centromere organization in both *Humulus* species. We identified and characterized major centromeric repeats, revealing pronounced reorganization of centromeric regions between the two related species. Specifically, we observed satellite DNA invasion and expansion, both indicating dynamic evolutionary processes shaping the centromeric landscape in *Humulus* species. Our findings contribute to a deeper understanding of centromere dynamics in plants and provide new insights into the complexity of centromere organization within the Cannabaceae.

POSTER 6

Unveiling the role of PRC2 beyond developmental gene silencing in the green lineage

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Polycomb Repressive Complex 2 (PRC2) plays a crucial role in the establishment of facultative heterochromatin regions in the genome of multicellular organisms such as flowering plants. It typically targets inactive genes and catalyses the deposition of the H3K27me3 histone mark over these regions. This is associated with a compaction of the underlying chromatin leading to a relatively stable silencing of the genes. By controlling the loci that are accessible to the transcription machinery, PRC2 plays a crucial role in cell differentiation, organogenesis and life-stage transitions. Nevertheless, the role of PRC2 appears diverse in the green lineage. In *Arabidopsis*, it is also likely to participate in the modulation of stress-responsive and metabolic genes; in the livewort, it is rather associated with constitutive heterochromatin and transposon elements. In this study, we have characterized the chromatin landscape defined by 7 chromatin marks in a unicellular chlorophyte alga, *Chlorella sorokiniana*. We present the distribution of H3K27me3 on genes and transposon elements, and reveal remarkable patterns, including the overlap of H3K27me3 and H3K9me2, by contrast with flowering plants. Finally, we compare H3K27me3-associated states in *Chlorella* with *Arabidopsis thaliana*, with particular focus on non-developmental and active PRC2-targets that we identified by extensive re-analyses of public data.

POSTER 7

Endogenous pararetroviruses in tomatoes and their hybrids: Exploring their role from silencing to subgenome regulation

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Endogenous pararetroviruses (EPRVs) are remnant DNA fragments of past viral infection, widely distributed across the plant kingdom. Despite being typically silenced by small RNAs (sRNAs; e.g., Schmidt et al. 2021), genomic perturbances may reactivate them. Studies on F4 hybrids between cultivated and wild tomatoes (*Solanum lycopersicum* × *S. pennellii*) demonstrated that EPRV-derived sRNAs are upregulated with impacts on gene expression (Lopez-Gomollon et al. 2022), highlighting the need to characterize SolanumEPRVs for understanding their role in genome regulation.

In a genome-wide analysis, we identified EPRVs from four distinct genera in the tomato genome, including the Solendo-, Florendo-, Xendo- and Yendoviruses, along with several structural variants. Using high-quality genome assemblies, we found SolanumEPRVs to be conserved across tomatoes, with Solendo- and Xendovirus-divergent 3' regions that are specifically associated to sRNAs. Similar to other transposable elements, SolanumEPRVs are usually truncated and dispersed throughout the genome. However, Xendo- and Yendoviruses in particular tend to be associated with ribosomal DNA (rDNA).

The association of SolanumEPRVs with rDNA and sRNA-mediated silencing may contribute to their long-term persistence and endogenization, putatively promoting their exceptional diversity. These findings, combined with the impact of EPRV-derived sRNAs, contribute to our understanding of how EPRVs shape subgenome regulation, especially after hybridization.

POSTER 8

The puzzling duckweed genomes hold the key for unlocking infrageneric biodiversity and evolution

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The monocot duckweeds (Lemnaceae) are the fastest growing flowering plants, free floating on freshwater surfaces and a new crop without need for arable land. The family is characterized by progressive evolutionary reduction of roots and body size, mainly asexual propagation and infrageneric morphological similarity between species. Variable chromosome numbers (20-126) and genome sizes 160 - >2000 Mbp/1C (even within presumed species) have been reported. The taxonomic assignment and phylogenetic relationship are unclear for many accessions of the genera *Lemna*, *Wolffiella* and *Wolffia*.

Multiple genomic and cytogenetic approaches (plastid and nuclear sequence polymorphism, genome size, chromosome counts, and genomic *in situ* hybridization) enabled elucidating biodiversity and evolution of accessions of genus *Lemna*. We postulate the species complexes *Le. aequinoctialis* and *Le. minor* which in addition to diploid species represent hitherto undetected polyploids, interspecific hybrids and backcrosses.

POSTER 9

Cytogenomics of pasture grasses identifies rapidly evolving repetitive DNA to reveal features of chromosome evolution and polyploidy for phylogenetics and exploitation of biodiversity

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Grasslands represent the majority of the world's farmland, often growing in a poorer environment than crops, with low inputs and high biodiversity. Within the cultivated grass genera, there is a wide range of genome sizes, ploidies and hybrids. We overview cytogenomics findings from our collaborative work in the broadly-defined genera *Elymus* (Triticeae), *Urochloa*, and *Cenchrus*. High-throughput survey sequencing enables identification of the major repetitive components of genomes including retroelements and satellite sequences. Comparisons of these most rapidly evolving, abundant, distinct genomic components from multiple species let us characterize relationships, refine phylogenies, show patterns of evolution at the species, chromosome and genome level. We found repeats that are genome specific, or show defined chromosomal distribution patterns, either dispersed along chromosomes, or enriched near centromeres or telomeres. In *Urochloa*, our work showed evolutionary divergence from ancestors, and can inform on the selection wild species biodiversity in breeding programs. In *Elymus*, we identified chromosomal evolution in the reticulate polyploid complex. We also looked at epigenetic effects and genome interactions, including DNA methylation in *Cenchrus* and *Urochloa*, and are extending our research to combine cytogenetics and genome assemblies.

We thank other co-authors on grass project publications. Further information and references are at www.molcyt.org

POSTER 10

The *Crocus* chronicles: Decoding the triploid puzzle of clonal saffron crocus

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Saffron crocus (*Crocus sativus* L.), a sterile triploid ($2n=3x=24$), is a clonally propagated crop valued for producing saffron, the world's most expensive spice. The evolutionary origin of triploid *C. sativus* was long debated, but recent cytogenetic and genomic studies (Schmidt et al. 2019; Nemati et al. 2019; Kazemi-Shahandashti et al. 2022) suggest an autotriploid origin from diverse cytotypes of *C. cartwrightianus* in ancient Greece.

Here, we target the process before and after emergence of saffron to look into the past, present and future of this clonal line:

Past: How is chromosomal variability in the progenitor *C. cartwrightianus*? We detected high haplotype diversity in certain chromosomes, suggesting uneven genetic diversity in ancestral populations. This diversity became fixed in today's triploid saffron karyotype, reflecting the genomic state 5,000 years ago.

Present: Are there any somaclonal variability in the saffron clone? Comparing saffron accessions across the globe revealed chromosomal variability in three independent instances. We conclude that several saffron lineages evolved somaclonally post-emergence and are likely spread via vegetative propagation.

Future: Can saffron serve as a model for epigenetics of adaptation? To support comparative and epigenetic analyses, we are assembling reference genomes for triploid *C. sativus* and diploid *C. cartwrightianus*, with ongoing sequencing, assembly, and chromosomal anchoring.

POSTER 11

Distribution and diversity of the maize B chromosome in Latin America

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B chromosomes are dispensable genetic elements found in representatives of plants, animals and fungi. In a population, they are present only in some individuals. The maize B chromosome is one of the first discovered (a hundred years ago) and most studied. Recently, we combined available genomics tools to assemble its reference sequence. Subsequently, we have developed molecular markers to detect the presence of the B chromosome and screened its presence in a collection of 770 landraces. The screening revealed that the B chromosome is widespread in Latin Americas being found in 307 (40%) accessions. We further sequenced the genome of 85 lines carrying the B chromosome and identified polymorphisms for both the B and A chromosomes. Their analysis provided information on the evolution of the B chromosome as well as the conservation of the individual genes encoded by the B chromosome.

POSTER 12

Contribution of repeats to genome size evolution and species diversification of the genus *Capsicum* (Solanaceae)

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Capsicum, a monophyletic genus within the Solanaceae family, comprises approximately 43 species distributed across South and Central America, with five taxa cultivated worldwide. All chile pepper species are exclusively diploid with base chromosome numbers of either $x = 12$ or 13 , although the genome sizes within the genus vary nearly fourfold. Repetitive DNA fractions of 39 *Capsicum* taxa and sister genus *Lycianthes* were characterized using Illumina genome skimming and RepeatExplorer to analyze their evolution in a phylogenetic framework. In the absence of polyploidy, genome size evolution of chile peppers has largely been driven by the amplification and removal of repetitive DNAs. Repetitive elements accounted for a significant portion of the genomes, ranging from 52% in the smallest genome to 83% in the largest. Comparative repeat profile analyses identified the Tekay lineage of Ty3/gypsy LTR retrotransposons and various families of satellite DNAs as key contributors to genome size variation. Overall repeat profiles, especially of satellite DNAs, correlated with clade diversification and recurrent ascending dysploidy. Species in clades with $x = 13$ exhibited higher levels of satellite DNAs diversity, whereas clades encompassing domesticated species had fewer satellite DNA families.

POSTER 13

Chromosomal diversity in *Hydrangea*: Insights from repeatome and FISH-based chromosomal analysis

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Hydrangea is a widely cultivated ornamental plant known for its morphological diversity. Despite its horticultural importance, taxonomic classification within the genus remains challenging due to frequent hybridization, polyploidy, and morphological convergence. The cytogenetic diversity and genome organization of *Hydrangea* are still poorly understood. In this study, we investigated the chromosomal diversity across seven representative *Hydrangea* species by integrating repeatome analysis with fluorescence in situ hybridization (FISH)-based cytogenetics.

Species-specific satellite DNAs identified among the seven *Hydrangea* species were designated “*Hydrangea* tandem repeat” (*Hydrangea*TR). FISH mapping using major *Hydrangea*TR probes enabled the visualization of chromosomal localizations and revealed previously unrecognized karyotypic differences among morphologically similar species. Chromosomal localization of TR02 and 10 revealed the existence of extra chromosomes for *H. serrata* and *H. yesoensis* within section *Macrophyllae*. These extra chromosomes exhibited variable numbers and frequency across individuals, a pattern characteristic of B chromosome (Bs). The distinct distribution patterns of the repeats specific to the Bs of *H. yesoensis* and *H. serrata* suggest potential roles in buffering genome structural variation or in accumulating repeats, which may have contributed to genome evolution within the genus.

Overall, our findings provide new insights into genome organization of *Hydrangea*, contributing to the refinement of its taxonomic classification.

POSTER 14

Genus-wide evolution of genome size and rDNA loci in chile peppers (*Capsicum*, Solanaceae)

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The genus *Capsicum* of the nightshade family comprises approximately 40 exclusively diploid species, five of which are cultivated worldwide. Chromosome numbers, genome sizes, and the positions of 5S and 35S rDNA loci were analyzed in 23 wild and cultivated species. Two base chromosome numbers ($x = 12$ and 13) were observed, corresponding to major phylogenetic clades. Genome size (1C) values varied nearly fivefold, despite the absence of polyploidy. Mapping of 5S and 35S rRNA genes revealed divergent evolutionary patterns. A single interstitial 5S rDNA locus was present in all species, except *C. tovarii*, where 5S rDNA localized to subtelomeric regions of all chromosomes due to 5S rDNA-derived satellite DNA expansion. In contrast, 35S rDNA loci exhibited a higher level of variation, ranging from one in basal Andean species to 16 in crown clades, which included all cultivated taxa. The number of major active 35S rDNA loci ranged from one to three, and some additional minor loci represented novel satellite DNAs. The ancestral *Capsicum* genome was reconstructed as small, carrying a single 5S and 35S rDNA loci. Diversification of the genus was inferred to be accompanied by 35S rDNA loci number increases and recurring genome size expansions and reductions.

POSTER 15

Interspecific hybridization is sufficient to establish nucleolar dominance in recently formed *Spartina* homoploid hybrids and allopolyploids

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Allopolyploid *Spartina anglica* C.E. Hubbard ($2n = 12x = 120-124$, genomic composition AAMM) arose by interspecific hybridisation between *S. alterniflora* ($2n = 6x = 62$, A-genome) introduced from North America and the native British *S. maritima* ($2n = 6x = 60$, M-genome) in UK about 150 years ago. Sterile first-generation homoploid hybrids are still extant at the original hybridisation zones allowing to study impacts of hybridization and genome duplication on gene expression. Nucleolar dominance is an epigenetic phenomenon where one of the parental rDNA locus (encoding 18S, 5.8S and 26S ribosomal RNAs) is epigenetically silenced in allopolyploids. Here, we carried out a population-level study of nucleolar dominance in *Spartina* system. Using locus-specific qRT-PCR analysis we observed strong transcriptional dominance of A-genome rDNA in 75 (100%) individuals of *S. anglica* and 36 (100%) individuals of *S. x townsendii* and *S. x neyrautii* homoploid hybrids. Active A-genome rDNAs were nearly devoid of CWG methylation while silenced M-genome rDNAs were hypermethylated at these sites. At the cytogenetic level, the A-genome loci were decondensed in interphase while those of M-genome were highly condensed in interphase. We conclude that interspecies hybridization is sufficient to immediately reprogram epigenetic patterns of parental NORs in the first generation *Spartina* homoploid hybrids. Established patterns are stably transmitted to *S. anglica* allopolyploids.

POSTER 16

Transposon induced somaclonal variation in tissue culture of a variety of crop species

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Tissue culture is an important plant multiplication method used widely in plant breeding programs. This form of clonal propagation is used because of efficiency, scalability and the preserved genotype and phenotype. It allows for rapid multiplication and generally results in uniform plantlets. Prolonged tissue culture can lead to deviating phenotypes however. This phenomenon is called somaclonal variation and the underlying causes are not yet fully understood, partially because there is a wide variety of phenotypes. It is understood genome stability plays a role and hypothesized that loss of epigenetic silencing results in increased transposable element activity and novel expansions of transposons. In our study we will monitor epigenetic silencing and transposon activation and insertion during tissue culture in eight different commercially important crop species. This screening may elucidate the progression of somaclonal variation during prolonged tissue culture and reveal common transposons active across the species boundary. Finally we intend to provide insight on how to decrease somaclonal variation occurrence through better understanding of the underlying processes.

POSTER 17

The repetitive DNA sequence landscape in *Epipactis*: Chromosome and genome evolution defined by major repeat classes

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Epipactis is a diverse genus of terrestrial orchids distributed primarily across temperate regions of Europe and Asia, with some species extending into North Africa and North America. Characterized by their rhizomatous growth and symbiotic mycorrhizal associations, *Epipactis* species occupy a wide range of ecological niches, from woodlands and calcareous soils to wet dune slacks. Species within *Epipactis* display considerable reproductive plasticity, exhibiting varying degrees of autogamy and allogamy, often influenced by ecological pressures and pollinator availability. The genus is notorious for its taxonomic complexity, driven by high morphological similarity among species, hybridization and adaptation to local environments. As a result, species delimitation remains contentious, and the current systematics of *Epipactis* is considered provisional by many taxonomists, with species numbers varying significantly depending on the adopted classification criteria. Given these characteristics, *Epipactis* represents a valuable model for investigating plant evolution, ecological adaptation, and mechanisms of speciation. In our study, we employed low-coverage whole-genome sequencing data combined with bioinformatic analyses and molecular cytogenetic approaches to investigate the landscape of repetitive DNA sequences across selected *Epipactis* species. Repetitive elements, due to their rapid evolutionary dynamics and structural roles in genome organization, offer a complementary perspective to gene-based phylogenies.

POSTER 18

A holistic multi-omics approach to elucidate chromatin-mediated drought stress memory in *Solanum lycopersicum*

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Chromatin modifications can provide a basis for epigenetic memory, integrating environmental cues with developmental programs. To investigate drought-induced epigenetic memory, we combine transcriptomic analysis with genome-wide profiling of histone modifications in two tomato genotypes subjected to severe-recurrent and mild-prolonged drought stress, respectively.

The effects of recurrent severe drought on chromatin and its correlation with gene expression were analyzed by monitoring H3K4me3 enrichment dynamics via ChIP-Seq in leaf tissues for identification of distinct categories of stress-memory genes. A mild prolonged drought stress was applied to identify stress-responsive epigenetic targets. We focused on gene loci showing coordinated changes in transcription and different chromatin marks.

To further explore the impact of drought on chromatin organization and histone modification patterns, immunolocalization assays were performed on nuclei isolated from leaf tissues and revealed an enrichment of H3K27me3 in nuclei from drought-stressed leaves, under both mild and severe conditions. H3K27me3 dynamics were further examined by ChIP-Seq during stress exposure and recovery.

To functionally validate the role of chromatin remodeling in drought response, expression analyses of selected candidate memory genes were conducted in *ddm1* and *hda19* mutants. Our findings underscore the role of chromatin dynamics as a crucial component in the establishment of stress memory.

POSTER 19

Pollen and anther morphological variation was shaped by domestication in rye

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Pollen and sperm morphology vary widely across species. In plants, cross-pollination enables genetic recombination, aiding adaptation. While pollen diversity across species is well studied, within-species variation remains less understood.

Our study examines quantitative variations in pollen and anther morphology in rye (*Secale cereale*), a wind-pollinating grass. We analyzed 339 individuals from 64 rye accessions, including domesticated (221), wild-like (91), and wild (4) types. A PCA based on 56,713 SNPs clustering by degree of domestication.

Pollen morphology in 286 individuals was assessed via multispectral imaging flow cytometry, and anther length in 314 individuals via light microscopy, displaying pronounced within-species diversity. Genome-wide association scans identified five and eight genomic regions linked to pollen and anther length, respectively. Some loci overlapped with known domestication regions of previously unknown function.

PST-FST analysis suggested selection for pollen and anther traits during rye domestication. A population genomic analysis detected selection signals at one pollen-length and three anther-length loci. Underlining that, we found significantly higher pollen and anther length in domesticated rye.

Overall, our findings indicate selection for these traits during domestication, advancing our understanding of the genetic basis of pollen and anther diversity in rye.

POSTER 20

Phylo- and cytogenetics unravel the complex evolutionary history of autumn-flowering Iberian crocuses**Ruifang An**¹, Frank Blattner¹, Nomar Waminal¹, Doerte Harpke¹¹Leibniz Institute of Plant Genetics and Crop Plant Research (IPK Gatersleben), Seeland, Germany

The Iberian autumn-flowering *Crocus serotinus* group complex encompasses five recognized species with numerous populations of uncertain taxonomic status. Variable genome sizes and chromosome numbers indicate extensive hybridization and dysploidy events, while the group's recent origin (~3.5 Mya) suggests rapid evolutionary diversification. These factors have created significant challenges for species delimitation and contributed to ongoing taxonomic confusion.

We employed an integrative taxonomic approach combining molecular phylogenetics, population genomics, and cytogenetic analyses to resolve these complexities. Our comprehensive sampling included 110 populations across the Iberian Peninsula, with genotyping-by-sequencing (GBS) of 282 individuals and whole-genome sequencing of 26 representative samples. We conducted repeatome analysis using RepeatExplorer2, estimated genome sizes via flow cytometry, performed chromosome counts, and analyzed karyotypes using fluorescent *in situ* hybridization (FISH).

Our analyses revealed 13 previously unrecognized diploid species within the complex. Diploid taxa exhibited genome sizes of $2C = 3.3\text{--}4.6$ pg, with the notable exception of *C. salzmännii*, which showed significantly larger genomes ($2C = 5.9\text{--}7.2$ pg), likely resulting from ancient polyploidization followed by rapid diploidization. Polyploid lineages displayed genome sizes of $2C = 6.7\text{--}9.2$ pg with chromosome numbers of $2n = 36, 44$, and 48 , indicating multiple independent polyploidy events and subsequent chromosomal rearrangements.

POSTER 21

Small meets smaller: siRNA mediated TE silencing in the smallest flowering plant

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Transposable elements (TEs) are mobile DNA sequences that shape their host genome architecture, gene regulation and epigenome. In angiosperms, TEs are silenced partly through small RNAs (sRNAs) through two key pathways: post-transcriptional gene silencing (PTGS) via 21-22 nucleotide (nt) sRNAs and Transcriptional Gene Silencing (TGS) via 24nt-sRNAs. The latter, through the RNA-directed DNA methylation (RdDM) pathway, targets the deposition of DNA methylation on the TE loci. Duckweeds (Lemnaceae) appear to be an exception, as they show no expression of RdDM components, and low 24nt-siRNAs and RdDM-associated DNA methylation. To fully understand TE silencing mechanisms in duckweeds, we assembled and analysed the genome and epigenome of *Wolffia brasiliensis*, a recently derived species within the family.

The *W. brasiliensis* genome is TE-rich (74%), with TEs broadly distributed and affecting gene structure and expression. As in other duckweeds, *W. brasiliensis* shows reduced RdDM activity. However, sRNA profiling showed abundant 24nt and 22nt-sRNAs arising from TEs, often mapping to the same loci. We further investigated different TE features that could explain this observation and its impact on *W. brasiliensis* epigenome. Our findings position duckweeds, and *W. brasiliensis*, as a model for studying TE dynamics in a context where silencing pathways show differences with those in well-studied model organisms.

POSTER 22

Role of RNA polymerases in centromeric transcription in *Arabidopsis thaliana*

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The centromeres are essential chromosomal regions where the kinetochore protein complex assembles, ensuring accurate chromosome segregation. Despite their conserved function, centromeric DNA sequences vary widely among species. In *Arabidopsis thaliana*, centromere identity is marked by the histone variant CENH3, whose deposition may be epigenetically regulated through centromeric DNA transcription. This loading process requires licensing factors such as KNL2 and CENP-C. Although RNA Polymerase II (RNA Pol II) mediates centromeric transcription in many organisms, its role in plants remains unclear. In vivo evidence shows that KNL2-C binds to CEN180 repeats, and yeast two-hybrid assays suggest a potential interaction between KNL2 and a subunit of RNA Pol II in *Arabidopsis*, hinting at a regulatory relationship. Additionally, plants possess RNA Polymerases IV and V, which function in the RNA-directed DNA methylation (RdDM) pathway, maintaining heterochromatin and transcriptional silencing. Affinity purification-mass spectrometry (AP-MS) data further reveal KNL2's association with subunits of RNA Pol IV and V. These findings highlight the need to examine the transcriptional interplay between KNL2, RNA Pol II, RNA Pol IV, and RNA Pol V to better understand the regulation of CEN180 transcription and the maintenance of centromere stability in plants.

POSTER 23

At least two trans-acting factors control the non-Mendelian drive of the rye B chromosome**Jianyong Chen**¹, Taoran Liu¹, Jochen Kumlehn¹, Andreas Houben¹¹The Leibniz Institute of Plant Genetics and Crop Plant Research (IPK), Seeland OT Gatersleben, Germany

B chromosomes are dispensable genomic elements found in many eukaryotes that persist through non-Mendelian chromosomal drive. The rye B chromosome (B) undergoes nondisjunction during the first pollen mitosis, preferentially segregating into the generative nucleus. This mechanism also functions efficiently when the rye B chromosome is introduced into wheat. The drive is controlled by a B-located Drive Control Region (DCR). We previously narrowed the DCR to a ~40 Mb region and identified five candidate genes, including the microtubule-binding protein DCR28 (Chen et al. 2024, Nat. Commun.). CRISPR/Cas9 knockout of the DCR28 gene family (>15 copies in a 2-Mb region) caused B chromosomal deletions in wheat. Three independent deletion lines lacking all DCR28 copies and downstream regions of the B showed reduced drive frequencies (42-69%), compared to 89-92% in wild-type Bs. In contrast, a deletion line retaining most DCR28 copies but lacking a downstream region showed a drive frequency of 76-82%, indicating that loss of DCR28 significantly impairs B chromosome drive. We conclude DCR28 plays a key role in controlling drive. However, loss of the entire DCR results in a drive frequency of 0%, suggesting the existence of a second B trans-acting drive controlling factor in a ~10 Mb region upstream of DCR28.

POSTER 24

The mutational dynamics of the *Arabidopsis* centromeres

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Centromeres are specialized chromosome regions essential for sister chromatid cohesion and spindle attachment during mitosis. Many centromeres comprise highly variable, megabase-scale satellite DNA arrays, yet the mutation spectrum driving this variability remains poorly understood. Using replicated genome assemblies of six *Arabidopsis* mutation accumulation lines, we identified centromeric mutations consisting almost exclusively of point mutations and structure-preserving, in-frame indels spanning a few kilobases. Centromeric point mutations occurred at a ninefold higher rate (6.1×10^{-8} /bp/gen) than in chromosome arms, frequently introduced by non-allelic gene conversions from closely linked repeat units. Forward-in-time simulations based on the observed mutation spectrum recapitulated the emergence of megabase-scale higher-order repeat (HOR) structures, including long-range sequence similarities, without requiring large-scale rearrangements, closely mirroring centromeric divergence among natural genomes. Our results show that centromere evolution is driven by a unique mutational spectrum, providing a quantitative framework to understand how small-scale mutations shape and maintain the large-scale architecture of centromeric DNA.

POSTER 25

When the B chromosome matters: Pollen developmental switch driven by sorghum B chromosome

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The development of pollen grains involves two divisions resulting in the formation of one vegetative cell and two sperm cells. This three-cellular structure constitutes the male-germ unit. The model of pollen development is conserved among angiosperms, with deviations occurring only in exceptional cases. One such case is the pollen of the wild sorghum, which contain B chromosomes.

As the B chromosomes are dispensable with no benefit to the host, the accumulation mechanisms are critical for their existence. In sorghum, the B chromosome accumulates through nondisjunction during the first pollen division. Interestingly, the segregation of the B chromosomes has frequently been observed. Thus, in order to survive, the B chromosome alters the developmental programme of the pollen. It initiates an additional pollen mitosis, leading to the production of pollen grains containing a higher than normal number of cells. This extra cell is an independent unit separated from the vegetative cell by a membrane. Consequently, it does not interfere with the pollen function and such pollen is viable and capable of pollination.

POSTER 26

Identification of genes controlling the process of root-specific B chromosome elimination in *Aegilops speltoides*

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Genome stability is critical for eukaryotic development, yet certain organisms selectively eliminate specific chromosomes. In *Aegilops speltoides*, B chromosomes are eliminated specifically in root tissues during early embryogenesis. While the cytological process is well-characterized, the molecular regulators remain unknown. To enable functional studies, we first generated a high-quality *Ae. speltoides* genome assembly using PacBio HiFi, Nanopore, and Hi-C sequencing, allowing confident identification of B chromosome sequences. To discover candidate genes, we performed comparative RNA-seq on tissues undergoing B chromosome elimination: young embryos (6-8 days post-pollination), embryonic roots from mid-stage embryos (17-20 days, isolated via laser-capture microdissection), and adventitious root buds, in B-positive versus B-negative plants. Subtraction of transcripts from tissues not exhibiting elimination (embryonic leaves via LCM, leaves, primary roots) further refined the candidate list.

Differential expression analysis revealed SYN2-B, a B chromosome-encoded homolog of SYN2 (SCC1/RAD21), as a promising candidate involved in elimination. Functional validation is underway in *Arabidopsis thaliana*. In parallel, we identified a CENH3 gene encoded on the B chromosome and are analyzing its potential role in elimination, alongside differences in centromere sequence composition between A and B chromosomes. These findings advance our understanding of the genetic control of chromosome elimination and plant genome plasticity.

POSTER 27

Centromere plasticity with evolutionary conservation and divergence uncovered by Wheat 10+ Genomes**Huan Ma**¹, Wentao Ding¹¹Huazhong Agricultural University, Wuhan, China

Centromeres (CEN) are essential chromosomal regions critical for maintaining genomic stability. Despite their highly repetitive and rapidly evolving DNA sequences, which drive karyotype diversity in eukaryotes, the mechanisms ensuring centromere functional homeostasis remain poorly understood. In this study, we explored the genetic and epigenetic architecture of CEN in a diverse population of wheat lines from global breeding programs. High-resolution analysis revealed extensive sequence, positional, and epigenetic variation within the large and complex wheat centromeres. We identified Cereba retrotransposons as the predominant CENH3-associated repeats, exhibiting phylogenetic homogenization across wheat lines, while less-associated repeats diverged lineage-specifically. This suggests selective mechanisms prioritize certain repeats as functional core CEN elements. Additionally, CENH3 nucleosomes exhibited looser DNA wrapping at their termini on complex centromeric repeats, including repositioned CEN regions. Strict nucleosome positioning and intrinsic DNA features further contributed to centromere identity, with specific non-B-form DNAs enriched at repositioned CENH3 loci. These findings highlight multiple adaptive mechanisms stabilizing CENH3 nucleosomes, ensuring centromere function amid genomic diversity. We propose that centromere chromatin exhibits remarkable epigenetic plasticity, with robustness shaped by historical breeding selection, ultimately safeguarding genome stability in wheat. This study advances our understanding of centromere evolution and stability in polyploid crops, offering insights into their adaptive mechanisms in complex genomes.

POSTER 28

Is LTR retrotransposon removal more uniform in holocentric than in monocentric chromosomes?

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Long terminal repeat retrotransposons (LTR-RTs) are the most abundant group of transposable elements in plant genomes. They transpose via a copy-and-paste mechanism, generating high sequence similarity that can lead to ectopic recombination. However, recombination is often suppressed in pericentromeric and other heterochromatic regions of monocentric chromosomes. As a result, full-length LTR-RTs may accumulate in these recombination coldspots, while solo-LTRs mark their removal from recombination hotspots in the arms. In contrast, holocentric chromosomes exhibit a more uniform chromatin organization without evident euchromatic or heterochromatic clusters. Therefore, it may be more difficult for LTR-RTs to “hide” somewhere, and both full-length LTR-RTs and solo-LTRs are expected to be more evenly distributed along holocentric chromosomes. Using a combined bioinformatic and cytogenetic approach, we investigated LTR-RT family abundance and elimination dynamics in holocentric cyperids and compared them with closely related monocentric grasses. Our findings provide insights into how chromosomal architecture shapes LTR-RT distribution and turnover and their roles in plant genome evolution.

POSTER 29

Fast evolution of Interstitial Telomeric Repeats in *Arabidopsis thaliana*

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Telomeric repeats are found at terminal telomeres but also at internal chromosomal positions, either as individual gene regulatory motifs (telobox) or as clusters within heterochromatic domains referred to as Interstitial Telomeric Repeats (ITRs). Despite being identified in many eukaryotes, including plants, animals, and humans, the structure, function, and origin of ITRs remain elusive. Using long-read genome assemblies of 169 *A. thaliana* natural accessions, we describe the identity, structure and diversity of ITRs in a model plant species. Although displaying a conserved ~500bp elemental unit, which surprisingly consists of both telomeric and centromeric-like repeats, and similar positions on chromosomes 1 and 4 pericentromeres, ITRs show a remarkable diversity in structure and size, ranging from 0.2 to over 2 Mb. Analysis of the internal and chromosomal organization, and comparisons to telomere length or population structure shed light on rapid evolution facilitated by mechanisms operating across a range of dimensions, from local amplification of the elemental units to segment duplications spanning hundreds of kilobases, while the proximity to centromeres and presence of centromeric-like sequence hint at possible centromeric origin of ITRs. In future studies, I will explore the impact of ITR evolution on epigenome homeostasis and nuclear organization.

POSTER 30

Does chromoanagenesis play a role in the origin of B chromosomes?

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B chromosomes (Bs) exist in addition to the standard (A) chromosomes in a wide range of species. The process underlying their origin is still unclear. We propose pathways of intra- and interspecific origin of B chromosomes based on known mechanisms of chromosome evolution and available knowledge of their sequence composition in different species. We speculate that a mitotic or meiotic segregation error of one or more A chromosomes initiates, via chromoanagenesis, the formation of a proto-B chromosome. In the second step, proto-B chromosomes accumulate A chromosome- and organelle-derived sequences over time, most likely via DNA double-strand break (DSB) mis-repair. Consequently, the original structure of the early-stage proto-B chromosomes becomes masked by continuous sequence incorporation. The similarity between A chromosome sequences integrated into B chromosomes and the original sequences on the donor chromosomes decreases over time if there is no selection pressure on these sequences on B chromosomes. However, besides chromoanagenesis, other mechanisms leading to the formation of B chromosomes might exist.

POSTER 31

Beyond genes: Exploring repeatome diversity in *Marchantia polymorpha*

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Bryophytes, one of the earliest land plants, are classified into three groups: hornworts, liverworts, and mosses. *Marchantia polymorpha* (287 Mbp/1C) is a model species of liverworts. In this study, we analyzed repeatome profiles of eight *M. polymorpha* populations collected in Central Europe as well as GenBank data for all three *Marchantia* subspecies and closely related species of Marchantiopsida.

Using clustering-based approach, we characterized the repetitive DNA landscape and explored variation in repeatome composition across taxa. Repetitive elements represent approximately 21% of the *Marchantia* nuclear genome. The most abundant repeats were Ty3/gypsy (10,18% genome proportion (GP)), Ty1/copia (0,24% GP), and rDNA (9,1% GP). Out of retroelements the *M. polymorpha* genome was dominated by Ty3/gypsy/Athila (8,92% GP) and Ty1/copia/Tork (0,24% GP) lineages. Other lineages were either absent or only marginally represented, indicating that *M. polymorpha* possesses a restricted spectrum of retroelements compared to angiosperms.

Repeatome fingerprint analysis revealed high similarity among repeatomes of *M. polymorpha* populations, with exception of the Austrian population, which shifted position in coordinate analysis depending on the cluster set used, which may indicate possible intersubspecific hybridization. Our findings demonstrate that repeatome profiling can support traditional morphological and ecological methods, especially where subspecies identification is challenging.

POSTER 32

The puzzle of sex determination in spice nutmeg (*Myristica fragrans*)

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Sex determination mechanisms and sex chromosome evolution remain poorly understood in dioecious trees. This study explores the sex-determining region in the holocentric *Myristica fragrans* (true nutmeg), a commercially valuable dioecious tropical spice tree. Unlike well-characterized monocentric model plants, *M. fragrans* offers a unique and valuable system for advancing our understanding of sex chromosome biology in holocentrics.

We investigated the sex-determining region (SDR) using whole-genome resequencing data from 18 individuals (11 females, 7 males) and a male reference genome. Following stringent preprocessing, reads were mapped to the reference genome, and genetic variants were called. Coverage analysis, fixation index (*F_{st}*) between sexes, and nucleotide diversity were used to identify the location of the SDR on Chromosome 6. We then developed 15 primer pairs and tested their specificity. We found that one primer pair amplified exclusively in males.

This work offers one of the first insights into sex chromosome biology in a holocentric tree species and provides a foundation for developing sex-linked markers to support marker-assisted breeding in nutmeg.

POSTER 33

Antibodies against kinetochore proteins as highly versatile markers of centromeres in flowering plants (angiosperms)**Pavel Neumann**¹, Ludmila Oliveira¹, Jiří Macas¹¹Biology Centre CAS, České Budějovice, Czech Republic

The precise identification of functional centromere domains depends on the detection of centromere-associated proteins, which are usually structural or regulatory components of the kinetochore. This multiprotein complex assembles at the centromeres and mediates the attachment of chromosomes to the microtubules of the mitotic spindle. While the overall structure of the kinetochore is conserved in eukaryotes, the sequences of kinetochore proteins differ considerably between species. This divergence complicates the development of broadly reactive antibodies. In our previous projects, we have developed antibodies targeting proteins from different kinetochore subcomplexes. Most of the recognised epitopes were only conserved within the same genus or between closely related genera. However, some antibodies such as those against KNL1, NDC80, BUB3, CENPX and CENPO targeted domains that were conserved across a wider phylogenetic range, suggesting broader applicability.

The anti-KNL1 antibody proved to be particularly versatile, successfully detecting KNL1 in most seed plant species. This cross-species recognition is due to a short, highly conserved motif within its target domain. Taking advantage of this feature, we sought to develop an anti-KNL1 monoclonal antibody that could serve as a renewable and consistent tool for studying plant centromeres and improve reproducibility in different laboratories and experimental systems.

POSTER 34

5S rDNA is dynamic and centromere-linked: Population-level insights from *Arabidopsis thaliana*

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Ribosomal DNA (rDNA) represent the second (after centromeric satellites) largest group of tandemly arranged repeats in the *Arabidopsis thaliana* genome. Contrast to 35S rDNA, 5S rDNA is relatively well represented in chromosome assemblies, allowing its structural and evolutionary analysis. We combined high-resolution cytogenetic techniques with advanced bioinformatic tools to investigate the organization and variability of 5S rDNA loci across 60 high-quality *A. thaliana* genome assemblies (PRJEB62038). Using a suite of computational methods (TRASH, ModDotPlot, DANTE) alongside fluorescence *in situ* hybridization and fine-scale microscopy, we mapped the chromosomal positions, array structures and sequencing diversity of 5S rDNA. Major loci on chromosomes 4 and 5 contained hundreds to thousands of regularly arranged units, while minor, more heterogeneous loci, were identified on chromosomes 3 and 1. Substantial inter- and intrapopulation variation in 5S rDNA copy number was observed, including near-complete loss of major loci in c. 5% individuals. Phylogenetic analysis revealed that arrays on chromosomes 4 and 5 are closely related, whereas those on chromosomes 3 and 1 are more divergent. Notably, all 5S rDNA loci – regardless of transcriptional activity – were embedded in transposon-rich, centromere-associated chromatin. These centrophilic tendencies may explain the frequent association of 5S rDNA with chromocenters during interphase.

POSTER 35

Trifluoromethanesulfonamide (TFMSA) induce male sterility in plants via inhibition of proline transport and phenylpropanoid biosynthetic pathway

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Physical removal of the anthers is labor-intensive and crucial process in breeding and genetic study in plants. TFMSA has emerged as an effective chemical hybridizing agent (CHA) inducing male sterility in various species. This study investigated the broader applicability of plants and elucidated the mechanism of male sterility using metabolome, transcriptome and cytogenetic analyses. TFMSA applied to diploid and tetraploid *Arabidopsis thaliana*, *A. suecica*, cowpea, soybean, pea, tomato, petunia, pepper, eggplant, pearl millet, and wheat. Alexander staining and fruit set assessments confirmed induction of male sterility. In *A. thaliana*, accumulation of phenylalanine and histidine in TFMSA treated flower buds were observed. The increased expression of genes involved in glucosinolate biosynthesis, such as CYP79F1 and MAM1, reduced the activity of PAL in the phenylpropanoid pathway. Proline levels in cowpea anthers declined, suggesting that both the disruption of proline transport and the reduction of PAL induced male sterility in plants. Observation of meiotic cells revealed unusual chromatin condensation during meiosis. TFMSA was an effective inducer of male sterility in a number of different plants, which highlights its strong potential for use in plant breeding and the plant genetic study.

POSTER 36

Shoot apical meristem bombardment enables stable *in planta* transformation in pearl millet (*Pennisetum glaucum* L.)**Grace Emmanuel Arikpo**¹, Kentaro Yoshida², Takayoshi Ishii^{3,4,5}

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Pearl millet (*Pennisetum glaucum* L. $2n = 2x = 14$) is a resilient cereal crop essential for food security in arid regions, yet its improvement via genetic transformation has been limited by regeneration-dependence. This study aimed to develop a regeneration-independent *in planta* transformation method by bombarding the shoot apical meristem (SAM) of pearl millet mature embryos. The germination potential of 12 genotypes after SAM exposure was evaluated. Genotype IP22281 was selected for bombardment for its high post-SAM exposure germination rate. A plasmid encoding (tGFP) under the ubiquitin promoter was delivered to the SAMs via gold particle bombardment. The effect of Helium pressure (1100 or 1500 psi), particle amount (750 or 1080 μ g), and TransIT-2020 coating method were accessed. The highest transformation rate (21-31 %) was recorded when 1100 psi, 750 μ g gold and TransIT-2020 were combined. From 275 embryos bombarded, 68 germinated (25%), and PCR confirmed the transgene in 20 T0 plants (~30%). Two T0 plants transmitted the transgene to T1 (10%), and Mendelian segregation was observed in T2. GFP expression was confirmed cytologically in the nuclei of T0 and T1 leaves, however, no T2 leaves expressed GFP. This study offers a promising platform for pearl millet genetic improvement via regeneration-independent transformation system.

POSTER 37

Plant genome annotation with DANTE/TideCluster toolset**Petr Novak**¹, Pavel Neumann¹, Nina Hošťáková¹, Jiří Macas¹¹Biology Centre CAS, Ceske Budejovice, Czech Republic

The RepeatExplorer Galaxy web server has evolved from its original focus on repeat annotation in unassembled genomes to provide comprehensive tools for analyzing genome assemblies. The platform now offers the DANTE toolset (DANTE, DANTE_LTR, and DANTE_TIR) for transposable element annotation and TideCluster for tandem repeat analysis, enabling complete characterization of repetitive elements in plant genomes.

The DANTE suite provides domain-based annotation of transposable elements using the REXdb database. DANTE identifies and classifies LTR-retrotransposons beyond superfamily level, revealing lineage-specific distribution patterns. DANTE_LTR pinpoints complete LTR retrotransposons including structural features, while DANTE_TIR detects DNA transposons with terminal inverted repeats through analysis of sequences flanking conserved transposase domains.

TideCluster offers advanced tandem repeat annotation using TideHunter (Gao et al. 2019) for initial detection, followed by similarity-based clustering and consensus reconstruction via TAREAN. Recent enhancements include higher-order repeat detection through the KITE method, which analyzes monomer size variability across individual tandem repeat arrays. A second enhancement enables comparative analysis of tandem repeats across multiple genomes, making the tool suitable for pangenome annotations.

All tools generate GFF3-formatted annotations compatible with standard genome browsers and support repeat analysis from complete chromosomes to short sequences. This comprehensive approach advances our understanding of repetitive element evolution and distribution.

POSTER 38

Enhancing GT efficiencies via SunTag-mediated exonuclease recruitment**Seda Yasar**¹, Niklas Capdeville¹, Laura Merker¹, Holger Puchta¹¹Joseph Gottlieb Kölreuter Institute for Plant Sciences (JKIP) – Molecular Biology, Karlsruhe Institute of Technology (KIT), Karlsruhe, Germany

CRISPR/Cas-mediated DNA double-strand break (DSB) induction has proven to be a valuable tool for targeted mutagenesis by non-homologous end joining (NHEJ) but site-specific insertion of longer sequences into plant genome is still difficult. Therefore, tools that enable insertions such as homologous recombination (HR)-based gene targeting (GT) are of particular interest for plant breeding. However, since in somatic plant cells HR plays only a minor role, efficient GT remains a major challenge.

Recently, enhanced GT was achieved by fusing Cas nucleases with the herpes simplex virus 1 exonuclease (HSVUL12), or its ortholog from Papiine alpha herpesvirus 2 (PapExo). Additionally, SunTag-mediated recruitment of up to ten exonuclease copies was shown to outperform direct fusion in targeted mutagenesis by NHEJ, with human TREX1 showing the strongest effect.

In this study we combined these approaches to enhance GT in *Arabidopsis* through SunTag-mediated recruitment of HSVUL12, PapExo or HsTREX1 to ttLbCas12a-i. While recruiting HsTREX1 showed no significant impact, recruitment of PapExo and HSVUL12 slightly enhanced and even doubled GT efficiency, respectively. Furthermore, GT experiments in the NHEJ mutant ku70 revealed higher GT frequencies regardless of exonuclease recruitment, suggesting that the observed effect is based on the exonuclease-mediated removal of KU from DSB ends.

POSTER 39

Breaking the silence: Understanding recombination suppression in wheat wild gene introgressions**Floriane Chéron**¹, Luxi Yan¹, Isabelle Nadaud¹, Pierre Sourdille¹¹INRAE, UMR1095 INRAE - Université Clermont Auvergne, Genetics, Diversity & Ecophysiology of Cereals, Clermont-Ferrand, France

Introgression of beneficial genes from wild species like *Aegilops ventricosa* is a valuable strategy for improving disease resistance in wheat (*Triticum aestivum*). However, crossovers are suppressed in these introgressions preventing reduction of their size and often leading to linkage drag. Understanding the origin of this suppression is key to unlock their full potential in breeding. We evaluated three hypotheses that may explain this phenomenon, focusing on two already fixed introgressions coming from *Ae. ventricosa* in the cultivar Renan. First, we tested if the rate of double-strand breaks (DSBs) which are further repaired as crossovers is reduced in introgressions. We developed a ChIP-seq approach targeting DMC1, a protein marking early recombination sites to count and locate DSB sites within introgressions. Second, we established immuno- and oligo-FISH to observe chromosome pairing and synaptonemal complex formation in hybrids between Renan (carrying *Ae. ventricosa* segments) and Chinese Spring (without introgressions). This will allow us to assess potential synapsis defaults.

Finally, we explored whether epigenetic mechanisms or chromatin structure contribute to the suppression of recombination by rendering these regions inaccessible during meiosis. By integrating these complementary approaches, we will elucidate the causes of recombination failure within introgressions, supporting more effective and precise wheat breeding strategies.

POSTER 40

Structural variation within triploid bananas revealed by oligo painting FISHJana Cizkova¹, Ines Van den Houwe², Rony Swennen^{3,4}, **Eva Hribova**¹

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Auto and allopolyploidization played important roles in the evolution of edible banana cultivars. Combination of A- and B-subgenomes derived from *M. acuminata* and *M. balbisiana*, gave rise to triploid clones with AAA, AAB, and ABB genome constitutions. Polyploidy and hybridization are often accompanied by chromosomal rearrangements, one of the key players in plant speciation. On the other hand, structural heterozygosity can lead to suppressed recombination and reduction of pollen fertility, which is typical for edible bananas. Large genome structural changes can be identified by application of long-read sequencing technologies or cytogenetically. Recently, we have developed chromosome arm-specific oligo-painting probes, which provide a powerful tool for identification of large chromosomal translocations in banana. In this study, comparative chromosome painting was used to reveal karyotype structure of triploid banana clones, including misclassified accessions and clones with unknown origin. We showed that karyotypes of the mislabeled or unclassified banana accessions correspond to their phylogenetic position within *Musa*, when closely related accessions share the same chromosome structures. Moreover, we enlarged the information of general karyotype structure within *Musa*, and supported the assumptions about a complex mode of evolution in edible cultivars, which was most probably accompanied by repeated introgressions and backcrosses.

POSTER 41

Bimodal architecture of centromeres in bivalents and univalents of pentaploid dogroses sect. *Caninae* harbouring asymmetric meiosis

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Allopentaploid ($2n = 5x = 35$) dogroses (*Rosa* sect. *Caninae*) evolved an unusual mode of reproduction called canina meiosis with asymmetrical distribution of the genetic material to gametes. One subgenome is paternally transmitted via pollen while four subgenomes are maternally inherited through the egg cell. In both pollen and egg cells, two homologous subgenomes (14 chromosomes) pair forming seven bivalents, the three non-pairing univalent subgenomes (21 chromosomes) are transmitted only through the egg cell. In this way, the pentaploid genome is restored in a zygote after fertilisation. Here we analyzed three haplotype-resolved chromosome TdT-level assemblies: two *Rosa canina*, sect. *Caninae* individuals and one *R. agrestis*, sect. *Rubigineae* including their close diploid relatives. A comprehensive study of centromeric regions revealed a bimodal architecture of centromeres on bivalent and univalent chromosomes. Using comparative genomics, centromeric histone (CENH3) ChIP and high-resolution cytogenetics, we reveal that bivalent-forming centromeres are enriched with ATHILA retrotransposons, contrasting with larger tandem-repeat-based centromeres mainly found in univalents. We hypothesize the structural bimodality of dogrose centromeres may account for asymmetrical division of dogrose chromosomes in meiosis leading to their meiotic drive in the maternal germline.

POSTER 42

Understanding mechanisms of chromosome elimination in plant interspecific hybrids

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Interspecific hybridization and allopolyploidy are key drivers of plant evolution, and vital tools in breeding to introduce valuable traits into elite cultivars. However, non-Mendelian inheritance, whereby chromosomes are not transmitted equally to successive generations, occurs in some interspecific hybrids, causing the elimination of chromosomes from one parent. This can lead to issues with trait inheritance. The responsible mechanisms remain poorly understood.

Our results in *Festuca x Lolium*, commercially important grass hybrids, suggest that preferential chromosome elimination is associated with allele-specific silencing of kinetochore genes during meiosis. To further study the underlying mechanisms, we expanded our focus to include *Arabidopsis thaliana* x *A. lyrata* hybrids. We employ protein modeling and protein-protein interaction assays to investigate the compatibility of kinetochore components from different parental genomes. Additionally, we examine the chromosome composition of F2 hybrid generations both cytogenetically and via hiplex amplicon sequencing to assess the extent of chromosome elimination.

So far, our data suggest that *Arabidopsis* hybrids do not naturally exhibit chromosome elimination, but this can be induced using *A. thaliana* kinetochore gene mutants.

These findings offer new insights into the mechanisms of chromosome elimination in hybrids and have practical implications for plant breeding, aiding in the selection of parental lines.

POSTER 43

A Polyploid Paradox: Does polyploid diversity arise from maladapted DNA replication and repair?

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Whole genome duplication (WGD) occurs in all kingdoms and can boost adaptation. But early on, WGD is traumatic to the cell, and often fatal. In autopolyploids (within-species WGD), many processes must adapt to the new cellular landscape. We showed that DNA repair and cell division can nimbly shift, allowing natural polyploids to survive: but what explains the bigger question: how do some polyploids thrive? I suggest that the very challenges that WGD brings provide a hint: these processes control the generation of genomic structural variation. Here I suggest that the post-WGD shakeup – the stumbling process of adaptation to WGD – is itself a generative engine for evolutionary success. That is, WGD-mediated success may arise from early maladaptation to the very same challenges WGD brings. We are now testing mechanisms: in particular, if repair and division errors and centromere/kinetochore coevolution are key missing pieces in the century-long quest to understand polyploid adaptation.

POSTER 44

Structural rearrangements and synapsis variation drive recombination diversity in holocentric beaksedges**Meng Zhang**¹, Stefan Steckenborn¹, Marco Castellani¹, André Marques¹¹Max-Planck-Institute for Plant Breeding Research, Cologne, Germany

Holocentric chromosomes, with multiple centromeres distributed along their length, allow structural rearrangements without disrupting segregation. This study explores how such architecture enables rapid karyotype evolution and shapes meiotic recombination in *Rhynchospora*, a holocentric genus with high chromosome number variation ($2n = 4-36$). Using genome assemblies from 20 species (52 haplotypes), we detected extensive chromosomal rearrangements and identified centromere satellite DNA (Tyba repeats) as frequent fusion/fission breakpoints, suggesting their potential role in ectopic recombination and double-strand break-induced fragmentation.

We constructed recombination maps in seven species via single-cell sequencing of pollen nuclei, revealing diverse crossover (CO) patterns, ranging from strong distal bias to irregular distributions, with no correlation to broad genomic or epigenomic features. Cytological analyses linked these recombination landscapes to synapsis dynamics: telomere-led, clustered synapsis correlated with distal COs, while flexible synapsis initiation yielded irregular COs. We observed that higher chromosome numbers correlated with increased COs, and smaller chromosomes exhibited higher recombination rates. Hi-C analysis further indicated that smaller chromosomes have shorter chromatin loops, which should result in longer axis length and thus higher recombination rates, suggesting that physical chromosome organisation modulates recombination frequency. Together, our findings provide a mechanistic framework linking genome plasticity, chromosome structure, meiotic behaviour and recombination.

POSTER 45

Sex chromosome configuration and (a)synapsis during meiosis I in the dioecious plant *Silene latifolia*

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Sex chromosome structure has profound implications for chromosomal behaviour during meiosis. In most eukaryotes, homologous chromosomes undergo synapsis and crossover formation to ensure faithful segregation and fertility. However, in species with heteromorphic sex chromosomes, non-homologous regions often fail to fully assemble the synaptonemal complex (SC), resulting in asynapsis and transcriptional silencing. Despite major advances in animal systems, the genetic and structural features that in plants affect the sex chromosome meiotic behaviour remain largely unexplored. In this study, we examined the synapsis of the Y chromosome in the dioecious model *Silene latifolia*. Using volumetric quantification and 3D reconstruction of synaptonemal complexes, we assessed the pairing capacity of XY chromosomes in individuals with single and increased sex chromosome dosage. The localization of class I crossovers (HEI10) revealed that *S. latifolia* has a relatively high number of COs and DSBs, comparable to those observed in animals or *Arabidopsis* meaning similar genetic determinants of DSBs induction. These findings provide novel insights into the structural constraints of sex chromosome synapsis and underscore how partial homology and chromatin context may influence XY pairing.

POSTER 46

How to make the process of mitotic nondisjunction B chromosome-specific?**Lulu Bao**¹, Jianyong Chen¹, Andreas Houben¹¹IPK Gatersleben, Seeland, Germany

Mitotic chromosome nondisjunction of standard A chromosomes produces aneuploidy, which is often fatal to organisms. However, supernumerary B chromosomes (B), present in a wide range of organisms, utilise the process of mitotic nondisjunction to boost their own inheritance without harming the organism. The rye B performs drive during the first pollen mitosis, leading to their accumulation in the next generation. The underlying mechanism involves nondisjunction and lagging of the B chromatids towards the generative nucleus. Recently, we identified the rye B chromosome-specific gene DCR28. This candidate gene is only active during the first pollen mitosis and encodes a microtubule-associated protein. However, DCR28 alone is unable to regulate the B drive. Now, we aim to test whether B-specific satellite DNA is an additional component of the B-specific nondisjunction process. Barley stripe mosaic virus-induced genome editing (BSMVIGE) will be used to downsize the copy number of the B-specific repeats Cl11 and Sc26C38, by transiently delivering single guide RNAs (sgRNAs) into Cas9-expressing wheat lines containing the rye B. The drive frequency of the resulting B chromosome variants will be determined by pollen-FISH. Live imaging of the first pollen mitosis will be performed to compare the segregation dynamics of B chromosome variants.

POSTER 47

Assessing non-Mendelian inheritance patterns in *Arabidopsis thaliana* x *A. lyrata*

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Non-Mendelian inheritance is a phenomenon where chromosomes are not transmitted equally to successive generations, often leading to the partial or complete elimination of one parent's chromosome set. This uniparental chromosome elimination, which has already been described in commercially important hybrids, can be challenging for breeders, as it disrupts normal patterns of trait inheritance and affects the stability of hybrid offspring. Although the mechanisms involved in this process are still not fully understood, results in *Lolium* × *Festuca* suggest a link with allele-specific silencing of kinetochore genes from the submissive parent during meiosis. In this study we aimed to investigate whether similar mechanisms operate in other interspecific hybrids. We generated hybrids between *Arabidopsis thaliana* and *A. lyrata*, including lines with knockout mutations in *A. thaliana* kinetochore complex genes. Using fluorescence *in situ* hybridization with species-specific probes, we analyzed meiotic chromosome behavior in both wild-type and mutant F1 hybrids. Preliminary results indicate that these hybrids do not naturally exhibit chromosome elimination. However, we observed differences in univalent and lagging chromosome frequencies between the wild-type and mutant lines. These findings improve our understanding of chromosome elimination/retention mechanisms in hybrids and bring new insights into strategies to manipulate non-Mendelian inheritance in interspecific plant hybrids.

POSTER 48

Meiotic temperature resilience in wild barley**Yixuan Gao**¹, Steven Dreissig¹¹IPK Gatersleben, Seeland, Germany

Meiosis is an important process in germ cell maturation, influencing genetic diversity and the faithful segregation of chromosomes is also essential for fertility, and in case of cereal crops, grain yield.

Environmental stresses, such as extreme temperatures, can disrupt meiotic processes, leading to sterility due to spindle or synaptonemal complex (SC) disruption. The aim of our experiment is to examine the impact of short-term heat stress on meiotic chromosome structure and crossover frequency, and to explore natural variation in meiotic temperature resilience in wild barley (*Hordeum vulgare* ssp. *spontaneum*). To address this question, we characterised meiotic progression and chromosome structure in genetically diverse wild barley accessions subjected to short-term heat stress during meiotic prophase (30°C for 24h). Results revealed varying degrees of temperature resilience among the accessions, with different responses to high temperatures. Most chromosomal aberrations were univalents, correlated with a significant reduction in SC length. Three accessions exhibited high temperature resilience, with a maximum 4.26% univalent frequency, while accession HID_004 was highly temperature sensitive, showing 80.36% univalents. In response to this phenotypically significant difference, we performed RNA sequencing of the contrasting accessions, through which we identified differentially expressed genes that may affect meiotic stability.

POSTER 49

Quantitative cytogenetic characterisation and structural analysis of REC8 proteoforms in plant meiosis

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Meiosis is a specialized cell division essential for eukaryotic reproduction, ensuring balanced homologous chromosome segregation. This process is highly plastic, influenced by chromatin environment, genetic divergence, and environmental factors while maintaining conserved structural progression across eukaryotes. Understanding molecular mechanisms driving meiotic variations is key to elucidating its evolution. Here, we investigate functional consequences of allelic variation in REC8, a meiosis-specific α -kleisin protein crucial for chromosome organization. REC8 is part of the cohesin complex, facilitating sister chromatid cohesion and chromatin loop extrusion, influencing recombination rates.

Recent analyses identified allelic variation in HvREC8.2 between wild and domesticated barley (*Hordeum vulgare*). Barley and other Triticeae members possess two paralogous REC8 variants (REC8.1 and REC8.2) from an ancient whole-genome duplication. Non-synonymous substitutions between wild and domesticated HvREC8.2 occur in domains mediating DNA interactions and cohesin complex binding. Near-isogenic lines with wild or domesticated HvREC8.2 alleles show differences in meiotic chromosome compaction during prophase I, suggesting proteoform divergence.

A comprehensive approach investigates HvREC8.2 variation in meiosis, including cytogenetic analysis, functional assays, and structural studies using computational and experimental methods. Fluorescent immunolabelling, heterologous protein expression, biochemical assays, molecular dynamics simulations, and cryo-EM reveal how allelic variation influences cohesin complex function.

POSTER 50

Exploring the role of H4 acetylation in shaping patterns of recombination**Maxie Sophie Lippold**¹, Steven Dreissig¹¹Leibniz Institute of Plant Genetics and Crop Plant Research (IPK Gatersleben), Seeland, Germany

Meiosis is a specialised form of cell division that drives evolution by generating genetic variation. Meiotic recombination events are initiated by DNA double-strand breaks. This can lead to the formation of crossovers, which can result in allele reshuffling. The recombination landscape is shaped by an interplay of environmental, genetic and epigenetic factors. While H3 acetylation promotes crossovers in *Arabidopsis*, the role of H4 acetylation remains unclear. This study explores how H4 acetylation affects meiotic recombination in rye (*Secale cereale* L.) and *Arabidopsis thaliana*.

Recently we found that recombination landscapes differ between wild and domestication rye. A major QTL linked to low recombining regions identified a H4 acetyltransferase as the most likely candidate gene (ScESA1). Due to its similarity to *Arabidopsis* HAM1 and yeast ESA1, ScESA1 likely has H4 acetyltransferase activity. To analyse the influence of ScESA1 on meiotic recombination, a knockout mutant will be generated. Moreover, a conserved transposon-like insertion was identified in the ScESA1 promoter region of domesticated accessions. This negatively correlates with HEI10 count indicating a decreased meiotic recombination rate. This factor will also be analysed in regards to relative transcriptional and protein expression as well as cytological characteristics.

POSTER 51

Pollen FISH and tracking of chromosome fate during achiasmatic inverted meiosis in the holocentric plant *Rhynchospora tenuis*

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The *Rhynchospora tenuis* has the lowest known chromosome number in plants ($n=2$) and features holocentric chromosomes, where centromeric activity is distributed along their length. This species undergoes inverted meiosis, a unique mechanism in which sister chromatids separate equationally in the first meiotic division, followed by reductional segregation of homologs in the second division. Previous studies have reported complete achiasmy (no cytological manifestation of crossovers) during male meiosis, implying an absence of meiotic recombination. Single-gamete sequencing of pollen nuclei has also revealed strong segregation distortion, favoring specific haplotype combinations that vary between accessions.

In this study, we used fluorescent *in situ* hybridization (FISH) with 35S rDNA and haplotype-specific probes to track chromosome behavior during meiosis, pollen development, and pollen tube growth. Despite the low chromosome number and inverted meiosis, which theoretically reduce mis-segregation, all accessions displayed non-Mendelian segregation among viable pollen grains. These results support a model in which meiotic drive and post-pollination selection eliminate non-viable haplotype combinations, leading to offspring genetically identical to the mother plant. This represents a unique case of sexual reproduction that functionally mimics clonal propagation offering new insights into the evolutionary dynamics and genetic mechanisms that bridge sexual and clonal reproductive strategies.

POSTER 52

Identifying genomic regions in tetraploid wheat involved in unreduced gamete formation by QTL-seq

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Hexaploid common wheat (*Triticum aestivum* L.) is established through hybridization between tetraploid *Triticum turgidum* L. and diploid wild species *Aegilops tauschii* Coss. and subsequent genome doubling in their triploid hybrid. The mechanism of genome doubling in the triploid wheat relies on the formation of unreduced gametes via non-reductional meiosis. We previously found QTLs on chromosomes 1A, 2A, and 4B in a population of trihaploid segregants generated by crossing the F1 hybrids between *T. turgidum* subsp. *durum* cv. 'Langdon' and *T. pyramidale* KU-9882 with *Ae. tauschii* KU-2103. In this study, we conducted bulk-segregant QTL-seq analysis and confirmed the previously identified QTLs. The QTL on chromosome 1A was narrowed down to the 34Mbp region including meiosis-related genes encoding MSH5, BRCA1, and β -AURORA kinase. Between 'Langdon' and KU-9882, missense variants for MSH5 and BRCA1 were detected but variants potentially causing functional change for AURORA were not predicted based on SnpEff. In addition, we present cytological observations of the non-reductional meiosis and discuss the potential underlying mechanisms.

POSTER 53

Heat stress interferes with male meiotic cell division in European pear (*P. communis*)**Lucas Philips**¹, Han Palmers¹, Nico De Storme¹¹Laboratory for Plant Genetics and Crop Improvement (PGCI), Department of Biosystems, KU Leuven, Heverlee, Belgium

European pear (*Pyrus communis*) is an economically important fruit crop in Europe and other temperate regions. As demonstrated in other plant species, heat stress can disrupt meiotic cell division at various stages, often leading to induction of meiotic restitution and ectopic formation of $2n$ pollen. To investigate reproductive genome stability in *P. communis*, we characterized male sporogenesis in multiple cultivars both under regular temperature conditions as well as upon heat shock treatments (ranging from 28°C to 40°C) using a cut-branch experimental set-up. Cytological analysis revealed that elevated temperatures induce abnormalities in homologous chromosome pairing and segregation dynamics, compromising meiotic stability. These defects are associated with the occurrence of meiotic restitution, as shown via tetrad analysis and the detection of enlarged diploid ($2n$) pollen grains. Higher temperatures progressively increased meiotic restitution, indicating a shift toward mitotic-like division.

Our findings provide new insights into the effects of temperature stress on reproductive stability in pear and align with previous reports in other plant species, where heat similarly caused ectopic induction of $2n$ pollen due to meiotic restitution. From an applied perspective, these findings provide a promising strategy for inducing $2n$ gametes in perennial crops, for facilitating polyploidy-based trait introgression in pome fruits.

POSTER 54

Simultaneous disruption of ZMM complexes does not alter dynamics of pro-crossover protein HEI10

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Meiosis is a hallmark of sexual reproduction which enables ploidy reduction of parental gametes. Central to this process is recombination via crossover formation (CO). Homologous repair of programmed DNA double-stranded breaks (DSBs) promotes CO formation catalyzed by a meiotic specific group of proteins known as ZMMs. Despite an excess of CO precursors in form of DSBs, the number of COs are limited. The mechanisms underlying crossover designation remains largely elusive. Recent evidence suggests that the pro-crossover ZMM protein HEI10, plays a central role in CO designation through a diffusion-mediated coarsening process along the synaptonemal complex (SC). Here we show that simultaneous disruption of the ZMM protein complexes Mer3, MutSy and ZZS does not perturb synapsis in *Arabidopsis thaliana*. Further we show that these ZMM complexes are dispensable for the coarsening dynamics of HEI10 and recruitment of MutLy resolvase MLH1, which is downstream of HEI10. This highlights that the SC is sufficient to drive the coarsening dynamics of HEI10 with ZMMs being important for stabilization of late HEI10 foci and CO implementation. Finally, we show that the ZZS complex plays an important role in controlling HEI10 dynamics in synapsis defective *zyp1* mutants.

POSTER 55

Impact of wild introgressions on meiotic chromosome dynamics and stability in wheat**Luxi Yan**^{1,2}, Isabelle Nadaud¹, Floriane Chéron¹, Dengcai Liu², Pierre Sourdille¹¹INRAE, UMR 1095 INRAE - Université Clermont Auvergne, Genetics, Diversity & Ecophysiology of Cereals, Clermont-Ferrand, France, ²Triticeae Research Institute, Sichuan Agricultural University, Chengdu Sichuan, China

To meet human needs by 2050, bread wheat (*Triticum aestivum* L.; $2n = 6x = 42$) production needs to be significantly improved but considering the context of a sustainable agriculture. One effective approach is to better utilize the extensive but underexploited genetic resources in cereals. Disease resistance genes from wild species have been introduced into wheat, such as the rust-resistance locus Lr37/Yr17/Sr38 (2A/2N translocation) and the eyespot resistance gene Pch1 (7D/7Dv translocation) from *Aegilops ventricosa* (DvDvNN). These introgressed segments alter meiotic chromosome dynamics and stability. Cytogenetic analyses of Chinese Spring (CS), Renan (harboring the two introgressions), and their hybrids revealed that the hybrids exhibited an increased frequency of rod bivalents and higher levels of chromosomal fragmentation, indicating disrupted meiotic chromosome configurations and reduced stability. Oligo-FISH targeting chromosome 2A revealed stable ring bivalents in CS and Renan, but a mixture of ring and rod bivalents in hybrids, suggesting that *Ae. ventricosa* segments compromise chromosome integrity and behavior during meiosis. To our knowledge, this is the first systematic investigation of meiotic behavior involving *Ae. ventricosa* introgressions in wheat. These findings provide essential insights for optimizing alien gene introgression and improving wheat breeding strategies.

POSTER 56

Distribution of 5-methylcytosine, 5-hydroxymethylcytosine and 6-methyladenine in *Secale* chromosomes**Anna Kalinka**¹, Magdalena Achrem¹¹Institute of Biology, University of Szczecin, Szczecin, Poland

The rye genome is exceptionally rich in repetitive sequences, which constitute up to 90% of its content. Some of these sequences form compact blocks of constitutive heterochromatin. According to the widely accepted view, constitutive heterochromatin regions are rich in 5-methylcytosine (5mC). However, 5mC is not detectable at the chromosomal level in rye subtelomeric heterochromatin, centromeric regions, or intercalary heterochromatin blocks. A comparative distribution analysis of 5mC, 5-hydroxymethylcytosine (5hmC), and 6-methyladenine (6 mA) revealed similar patterns across chromosomes in four rye species: *Secale cereale*, *Secale strictum*, *Secale sylvestre*, and *Secale vavilovii*. All three modifications co-localized within the same chromosomal regions and were dispersed along the chromosome arms. Immunofluorescence (IF) signals for these modifications were absent in centromeres, telomeres, and subtelomeric heterochromatin, with minor differences observed in secondary constrictions, certain intercalary bands, and in the extent of IF signal occurrence within pericentromeric heterochromatin. These findings suggest that the analyzed DNA modifications may not function as epigenetic regulators of constitutive heterochromatin blocks in rye, or alternatively, that their regulatory roles may operate below the detection threshold of chromosome-level analyses.

POSTER 57

Epigenetic modifications of histones in the centromeric and telomeric regions of *Secale cereale* chromosomesAnna Kalinka¹, **Magdalena Achrem**¹¹Institute of Biology, University of Szczecin, Szczecin, Poland

In this study the distribution of selected epigenetic histone modifications (PTMs) in the centromeric region (the core centromere and pericentromeric heterochromatin) and the telomeric region (telomeres and subtelomeric heterochromatin) of *Secale cereale* chromosomes was analyzed. The localization of histone PTMs was examined on metaphase chromosomes and extended chromatin fibers. To determine the epigenetic status of the centromeric and telomeric regions, modifications characteristic of heterochromatin (H3K9me2, H3K27me3, H3K9me3) and euchromatin (H3K4me3, H3K9ac, H3K36me3) were analyzed. Immunofluorescence signals for the histone modifications H3K4me3, H3K9ac, and H3K9me2 were detected in small amounts in the centromere, the pericentromeric and subtelomeric heterochromatin, but were not observed in telomeres. The H3K36me3 modification was present in greater amounts in the pericentromeric region, while it was not detected in centromeres or in some of the telomeres. These findings were confirmed by the analysis of extended chromatin fibers. H3K9me3, a marker of constitutive heterochromatin, was present in the centromeric region, primarily in the pericentromere. In contrast, the H3K27me3 modification was observed mainly in subtelomeric regions. Analysis of extended fibers indicated the presence of this modification also in small amounts in the centromeres and some telomeres. The results indicate a high complexity and heterogeneity of the rye heterochromatin epigenetic status.

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