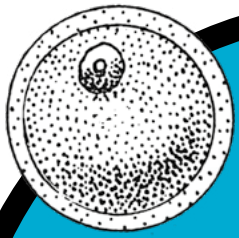
PROGRAMME AND ABSTRACT BOOK

IMPACT OF CHROMATIN DOMAINS ON PLANT PHENOTYPES
9-11 DECEMBER 2019
MADRID, SPAIN



NUCLEUS IMPACT

SOCIETY FOR EXPERIMENTAL BIOLOGY

IMPACT OF CHROMATIN DOMAINS ON PLANT PHENOTYPES

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DELEGATE INFORMATION

BADGES

Badges must be worn for the duration of the conference, both for security purposes and for entry into the scientific sessions and networking events.

CATERING

Lunch and refreshments will be provided for the duration of the conference and will be served in the dining room of the Maria Cristina.

CERTIFICATES OF ATTENDANCE

A certificate of attendance can be requested by attendance or by email from **admin@sebiology.org**

VENUE

Real Centro Universitario Escorial-Maria Christina
Paseo de los Alamillos, 2
28200
San Lorenzo de El Escorial
Madrid
Spain

Web: **https://www.rcumariacristina.com/**

CONFERENCE DINNER

Exe Victoria Palace
Calle Juan de Toledo
4 San Lorenzo De El Escorial
Madrid
28200
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INTERNET ACCESS

Free Wi-Fi is available throughout the Real Centro Universitario Escorial-Maria Christina.

The password is **SEB2019**

LIABILITY

Neither the Society for Experimental Biology nor the Real Centro Universitario Escorial-Maria Christina will accept responsibility for damage or injury to persons or property during the meeting. Participants are advised to arrange their own personal health and travel insurance.

POSTER PRESENTATIONS

Presenters will be able to hang their posters from Sunday 8th December from 20:00 - 21:00.

Posters must be in place for the relevant poster session and can only be removed once the session has taken place. All posters must be removed by 14:00 on December 11. Velcro fastenings for your poster will be provided so please do not use any other method of fastening for your poster.

SOCIAL MEDIA

We're looking to increase the conversation at the meeting using Twitter so please get tweeting! Follow the conversation **#sebiology**

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PROGRAMME MONDAY 9 DECEMBER

MONDAY 9 DECEMBER 2019

09:00 REGISTRATION AND POSTER SETUP WITH COFFEE

10:00 - 10:15
Welcome to El Escorial: Christophe Tatout
Université Clermont Auvergne, France)
& Monica Pradillo (Complutense University of Madrid, Spain

10:15 - 11:00
Opening Keynote: Wendy Bickmore
University of Edinburgh, UK
How important is 3D genome organisation for function?
P19.1

11:00 - 11:30 REFRESHMENT BREAK

SESSION I: EMERGING FUNCTIONS OF CHROMATIN DOMAINS

SESSION CHAIR: SARA FARRONA

11:30 - 12:00
Adam Klosin
Max Planck Institute of Molecular Cell Biology and Genetics, Germany
Phase separation provides a mechanism to reduce noise in cells
P19.2

12:00 - 12:30
Chang Liu
University of Tuebingen, Germany
Marchantia TCP transcription factor regulates target gene expression in the context of 3D chromatin structure
P19.3

12:30 - 12:45
Kalyanikrishna Kalyanikrishna
Institute for Biology Freie Universität Berlin
Elucidating the role of epigenetic regulators at the nuclear periphery in arabidopsis
p19.4

12:45 - 13:00
Emilia Cepowska
Institute of Biochemistry and Biophysics Polish Academy of Sciences
Does your plant remember the last time you watered it? Gene-loops and intra-nuclear gene localization as possible mediators of transcriptional memory
P19.5

PECHA KUCHA TALKS

13:00 - 13:02
Rafal Archacki
University of Warsaw Faculty of Biology Institute of Plant Experimental Biology and Biotechnology
Bromodomains are essential for the functions of Arabidopsis SWI/SNF complex
P19.6

13:02 - 13:04
Fredy Barneche
IBENS – CNRS
Characterizing the chromatin choreography facilitating transcription in plants
P19.7

13:04 - 13:06
Francesco Blasio
Universidad Complutense de Madrid
Modifying the landscape of crossovers in bread wheat meiosis
P19.8

PROGRAMME MONDAY 9 DECEMBER

13:06 - 13:08
Christian Chevalier
INRA Bordeaux
SlInhibitor of Meristem Activity (SlIMA) and At-MIni zinc Finger 2 (At-MIF2): swiss knives for the regulations of floral development in tomato and Arabidopsis
P19.9

13:08 - 13:10
Sandra IM Correia
Centre for Functional Ecology University of Coimbra
Changes in global DNA methylation and chromatin accessibility during somatic embryogenesis induction and long-term multiplication of embryogenic cultures of tamarillo
P19.10

13:10 - 13:12
Sandra Fonseca
Centro Nacional de Biotecnología (CNB-CSIC) Madrid
De-etiolated1 control of transcription factor activity and chromatin states
P19.11

13:12 - 13:14
Michal Franek
Central European Institute of Technology CEITEC
Mapping the chromatin of repeats using extended chromatin fibers
P19.12

13:14 - 13:16
Javier Gallego-Bartolomé
UCLA
Co-targeting RNA Polymerases IV and V Promotes Efficient De Novo DNA Methylation in Arabidopsis.

13:16 - 13:18
Godwin James
National University of Ireland Galway
UBP5, an interactor of PWO1 and PcG proteins that regulates plant development
P19.14

13:18 - 13:20
Martin Groth
Helmholtz Zentrum München
Light-dependent regulation of folate metabolism controls DNA methylation
P19.15

13:20 - 13:22
Aliki Kapazoglou
Hellenic Agricultural Organization - Demeter (HAO-DEMETER)
Population genetic and epigenetic variability and distribution of the endangered Greek endemic Cicer graecum under climate change scenarios
P19.16

13: 22 - 13:24
Marta Koblowska
Faculty of Biology University of Warsaw
H4K16AC is an epigenetic signature of the plant response to abiotic stresses
P19.17

13:24 - 13:26
Francesca Lopez
National University of Ireland Galway
Impact of rDNA copy number depletion on A. thaliana development
P19.18

PROGRAMME MONDAY 9 DECEMBER

13:26 - 13:28
Adéla Machelová
Central European Institute of Technology (CEITEC) and Faculty of Sciences of Masaryk University
Genome Stability in Chromatin Remodelling mutants CHR2, CHR4, CHR5, CHR9 and CHR24
P19.19

13:28 - 13:30
Ewelina Malecka
University of Warsaw
Analysis of genetic interactions between H1 linker histones and SWI/SNF chromatin remodeling complex in Arabidopsis
P19.20

13:30 - 13:32
Anis Meschichi
SLU - Swedish University of Agricultural Sciences
Characterizing chromatin mobility and histone dynamics during DNA damage responses
P19.21

13:32 - 13:34
Aitor Munoz
Centro Nacional De Biotecnologia (CNB-CSIC)
Molecular characterization of the D53-like SMXL repression complexes
P19.22

13:34 - 13:36
Hana Šimková
Institute of Experimental Botany
Changes of chromosome organization during cell cycle in barley
P19.23

13:36 – 14:00
Lauriane Simon
SLU - Swedish University of Agricultural Sciences
SUVH7: New actor in the regulation of endosperm development
P19.24

14:00 - 15:15 LUNCH/POSTERS

SESSION II: ROLE OF HISTONE VARIANTS AND MODIFICATIONS IN CONTROL OF CHROMATIN DYNAMICS

SESSION CHAIR: FREDY BARNECHE

15:15 - 15:45
Xuehua Zhong
University of Wisconsin-Madison, USA
Linking Signaling Pathways to Chromatin Dynamics
P19.25

15:45 - 16:15
Crisanto Gutierrez
Centro de Biologia Molecular Severo Ochoa, Madrid, Spain
Histone variants and chromatin dynamics during organ growth
P19.26

16:15 - 16:30
Aline Probst
GReD Université Clermont Auvergne CNRS Inserm
Histone H3 variant deposition and dynamics in A. thaliana
P19.27

16:30 - 16:45
Pedro Crevillén
Instituto Nacional de Investigación y Tecnología Agraria y Alimentaria (INIA)
Conserved roles of the H3K27me3 epigenome on plant development
P19.28

PROGRAMME MONDAY 9 DECEMBER

16:45 - 17:00
Inna Lermontova
Mendel Centre for Plant Genomics and Proteomics
CEITEC Masaryk University
Generation of haploids in *A. thaliana* based on manipulation of CENH3 assembly factor KNL2
P19.29

17:00 - 17:15
Gianluca Teano
Plant and Diatom Genomics Institut de Biologie de l'Ecole Normale Supérieure (IBENS)
Linker histone H1 dynamics control Arabidopsis nuclear reorganization in response to light signals
P19.30

17:15 - 17:30
Ortrun Mittelsten Scheid
Gregor Mendel Institute of Molecular Plant Biology
Dynamic transcription and DNA methylation patterns in Arabidopsis shoot stem cells
P19.31

17:30 - 18:00 REFRESHMENT BREAK

SESSION III: IMPORTANCE OF CHROMATIN DYNAMICS DURING PLANT REPRODUCTION

SESSION CHAIR: ISABELLE COLAS

18:00 - 18:30
Arp Schnittger
Hamburg University, Germany
Shedding light on chromosome and chromatin behavior in meiosis
P19:32

18:30 - 19:00
Célia Baroux
University of Zürich, Switzerland
Unlocking the route to reproductive lineage initiation in plants – a role for linker histone eviction?
P19:33

19:00 - 19:15
Monica Pradillo
Complutense University of Madrid, Spain
The nuclear envelope is the dance floor for the chromosomes during meiosis
P19:34

19:15 - 19:30
Azahara C Martin
John Innes Centre
ZIP4 and the stabilization of wheat meiosis on polyploidization
P19.35

19:30 - 19:45
Jamie N Orr
The James Hutton Institute
HvST1: an E3 ubiquitin ligase involved in meiosis in barley
P19.36

19:45 - 20:00
Mathilde Grelon
INRA-IJPB
Understanding the mechanisms that shape the distribution of crossovers along chromosomes during meiosis
P19.37

20:00 - 21:00 POSTER SESSION ODD NUMBERS

21:00 DINNER

PROGRAMME TUESDAY 10 DECEMBER

TUESDAY 10 DECEMBER 2019

09:15 - 09:30
Christophe Tatout
Université Clermont Auvergne
Update on INDEPTH Activities

09:30 - 11:15 WORKSHOP WITH INDUSTRIAL PARTNERS

11:15 - 11:45 REFRESHMENT BREAK

SESSION IV: TECHNOLOGICAL ADVANCES FOR CHARACTERISATION OF CHROMATIN DOMAINS

SESSION CHAIR: PAUL FRANSZ

11:45 - 12:15
Dariusz Plewczynski
University of Warsaw, Poland
Three-dimensional GeNOme Modeling Engine for data-driven biophysical simulations ofCTCF and RNAPII-mediatedhigher-order chromatinorganization
P19.38

12:15 - 12:45
James A.H. Murray
University of Cardiff, UK
Chromatin particle spectrum analysis and the dynamics of sub-nucleosomal particles in Arabidopsis
P19.39

12:45 - 13:00
Mio K. Shibuta
Tokyo University of Science
Live imaging system to tracking post-translational modification dynamics in *Arabidopsis thaliana*
P19.40

13:00 - 13:15
Anika Erxleben
University of Freiburg
The European Galaxy server: A platform for accessible, reproducible and collaborative big data analyses
P19.41

13:15 - 13:30
Philippe Andrey
INRA
Statistical modelling of spatial interactions: New insights into the spatial organization of plant heterochromatin
P19.42

13:30 - 13:45
David E Evans
Oxford Brookes University
Probing the structure of the nucleus; live cell imaging revisited
P19.43

13:45 - 14:00 GROUP PHOTO ON EL ESCORIAL TERRACE

14:00 - 15:00 LUNCH/ POSTERS

15:00 - 17:00 MONASTERY TOUR

17:00 - 17:30 REFRESHMENT BREAK

SESSION V: INFLUENCE OF NUCLEAR DOMAINS ON GENE EXPRESSION

SESSION CHAIR: MARTINA DVORACKOVA

17:30 - 18:000
Moussa Benhamed
Institute of plant sciences
Wheat chromatin architecture is organized in genome territories and transcription factories
P19.44

PROGRAMME TUESDAY 10 DECEMBER

18:00 - 18:30
Hans-Wilhelm Nützmann
University of Bath
Three-dimensional chromosomal architecture of metabolic gene clusters
P19.45

18:30 - 18:45
Ales Pecinka
Institute of Experimental Botany
SMC5/6 complex controls ploidy levels of male gametes in Arabidopsis
P19.46

18:45 - 19:00
Takuya Sakamoto
Tokyo University of Science
Involvements of nuclear pore complex proteins in the regulation of spatial arrangement of chromatin domains in Arabidopsis
P19.47

19:00 - 19:15
Tzvetina Brumbarova
Heinrich Heine University
Light-dependent accumulation of an iron uptake transcription factor in nuclear bodies
P19.48

19:15 - 19:30
Stefan Grob
University of Zurich
Transgene Silencing in 3D - How a Chromosomal Knot Can Inactivate Foreign DNA Elements
P19.49

19:30 - 21:00 POSTER SESSION EVEN NUMBERS

21:00 CONFERENCE DINNER AT EXE HOTEL

PROGRAMME WEDNESDAY 11 DECEMBER

WEDNESDAY 11 DECEMBER 2019

09:00 - 09:45
Keynote: Niels Stein
IPK Gatersleben, Germany
The pan-genomes of barley and wheat
P19.50

SESSION VI: ROLE OF CHROMATIN DOMAINS IN RESPONSE TO BIOTIC AND ABIOTIC STRESSES

SESSION CHAIR: ALES PECINKA

09:45 - 10:15
Isabel Bäurle
University of Potsdam
Adaptation to environmental stress by a dynamic chromatin-based stress memory
P19.51

10:15 - 10:30
Alessandra Devoto
Royal Holloway University of London
How jasmonate-mediated distress signals affect plant growth
P19.52

10:30 - 11:00 REFRESHMENT BREAK

11:00 - 11:30
Steven H. Spoel
University of Edinburgh
Dynamic ubiquitination controls transcription dynamics
P19.53

11:30 - 11:45
László Szabados
Biological Research Centre
The MPK4-phosphorylated Heat Shock Factor A4A controls heat and salt stress tolerance
P19.54

11:45 - 12:00
Ana Lopez Sanchez
Spanish National Centre for Biotechnology (CNB-CSIC)
Exploring the effect of mitochondrial changes in the chromatin remodelling mediating priming of immune response.
P19.55

12:00 - 12:15
Serena Varotto
University of Padova
Epigenetics signatures to remember drought stress in maize
P19.56

12:15 - 12:30
Meeting Summary and Poster Prize

12:30 - 14:00 GENERAL MEETING FINISH WITH BOXED LUNCH

12:45 - 15:00
INDEPTH Management Committee Meetings

POSTER SESSIONS MONDAY 9 DECEMBER

Necla Pehlivan
Recep Tayyip Erdogan University, Turkey
Actin Depolymerizing Factor 2 of a halophyte Spartina alterniflora imparts drought tolerance when constitutively overexpressed in tobacco
P19.57

Gulshan G Poladova
National Scientific Academy of Azerbaijan
Institute of Genetic Resources, Azerbaijan
Changes in the functional activity of DNA chromatin as a result inhibition of plant growth
P19.59

Valya Vassileva
Institute of Plant Physiology and Genetics
Bulgarian Academy of Sciences, Bulgaria
Drought stress effects on the chromatin-remodeling factor DDM1 in wheat cultivars with contrasting drought tolerance
P19.61

Nathalie Picault
University of PERPIGNAN, France
Low ribosomal RNA genes copy number provoke genomic instability and chromosomal segment duplication events that modify global gene expression and plant-pathogen response
P19.63

Thorsten Hamann
Norwegian University of Science and Technology, Norway
Coordination of cell wall integrity with cell cycle progression in Arabidopsis thaliana
P19.65

Eva Dvořák Tomaštíková
Institute of Experimental Botany,
Czech Republic
Uncovering SMC5/6 complex DNA damage repair pathway in Arabidopsis
P19.67

Mathieu Ingouff
IRD-University of Montpellier, France
Real-time imaging of DNA Methylation using DYNAMET sensors
P19.69

Dragana Miladinović
Institute of Field and Vegetable Crops,
Serbia and Montenegro
Defensin expression in sunflower undercombined broomrape – downy mildew attack
P19.71

Isabelle Colas
James Hutton Institute, United Kingdom
Chromosome and nucleus organization during Meiotic Recombination in Barley
P19.73

Dionysia A Fasoula
Agricultural Research Institute, Cyprus
Discovery of a unique barley spike phenotype and insights on some associated SNP variation
P19.75

Ksenija Taski-Ajdukovic
Institute of Field and Vegetable Crops, Serbia and Montenegro
Investigation of drought stress effects on sugar beet in Serbia
P19.77

Ivan Čitaković
Faculty of Biology University of Belgrade, Serbia and Montenegro
Defensin expression in sunflower undercombined broomrape – downy mildew attack
P19.79

Pavla Navratilova
Centre of Plant Structural and Functional Genomics, Czech Republic
Approaching a dwarfing phenotype in wheat by exploring the epigenome
P19.81

Christophe Tatout
Université Clermont Auvergne, France
Development and comparison of segmentation methods for the analysis of the plant's 3D nucleus
p19.83

Vera Inácio
Vera Inácio, Portugal
Cork Oak Young and Traumatic Periderms Show PCD Typical Chromatin Patterns but Different Chromatin-Modifying Genes Expression
P19.85

Paul Fransz
University of Amsterdam, Netherlands
Quantification of morphological features in interphase nuclei of Arabidopsis thaliana
P19.87

Edouard Tourdot
Edouard TOURDOT, France
Spatiotemporal distribution of ploidy levels and ploidy specific transcriptome during Tomato fruit development
P19.89

POSTER SESSIONS TUESDAY 10 DECEMBER

Afet D Mamedova
National Scientific Academy of Azerbaijan
Institute of Genetic Resources, Azerbaijan
Dynamics of changes in chromatin functional activity in plants under drought stress
P19.58

Institute of Experimental Botany CAS Centre of Plant Structural and Functional Genomics, Czech Republic
Towards in vivo analysis of chromatin dynamics in barley
P19.62

Pilar Cubas
Centro nacional de Biotecnologia-CSIC, Spain), Aitor Muñoz (Centro nacional de Biotecnologia-CSIC, Spain), Aitor Muñoz (Centro nacional de Biotecnologia-CSIC, Spain
Molecular characterization of the D53-like SMXL repression complexes
P19.66

Martina Dvorackova
Masaryk University CEITEC MU, Czech Republic
Insight into spatial distribution of nuclear protein interactions
P19.68

Juliette Aubert
RD University of Montpellier, France
Discovering trans-interactions involved in paramutation using Circular Chromosome Conformation Capture
P19.70

Nadia Fernandez-Jimenez
Universidad Complutense de Madrid, Spain
The connection between the nucleoporins SAR1/3 and meiosis: a way to discover chromosome dynamics
P19.72

Geraint Parry
Cardiff University, United Kingdom
GARNet Community Network: Supporting Discovery-led Plant Science in the UK and beyond
P19.74

Paulina Stachula
University of Warsaw Faculty of Biology, Poland
Functional analysis of BRM chromatin remodeler using catalytic point mutations.
P19.76

Ana Paula Santos
Instituto de Tecnologia Química e Biológica António Xavier Universidade Nova de Lisboa, Portugal
Deciphering histone modifications in rice to understand the OsRMC activation by salt stress
P19.78

Mariana Diaz
Tokyo University of Science, Japan
FASCIATA1 is required for de-novo shoot regeneration in Arabidopsis thaliana
P19.80

Tamar Krugman
University of Haifa Institute of Evolution, Israel
Inhibition of jasmonic acid biosynthesis induced powdery mildew resistance in wheat
P19.82

Birsen Cevher Keskin
TUBITAK Marmara Research Center Genetic Engineering Biotech. Inst., Turkey
Epigenetic Responses to Drought Stress and Recovery Period in Olea europaeaL.
P19.84

Eleni Tani
Agricultural University of Athens, Greece
Population genetic and epigenetic variability and distribution of the endangered Greek endemicCicer graecumunder climate change scenarios
P19.86

Tzion Fahima
University of Haifa, Israel
Phylogenetic analysis of the tandem kinase-pseudokinase (TKP) protein family involved in plant immunity
P19.88

ABSTRACTS FOR IMPACT OF CHROMATIN DOMAINS ON PLANT PHENOTYPES

P19.1 HOW IMPORTANT IS 3D GENOME ORGANISATION FOR FUNCTION?

MONDAY 9 DECEMBER **10:15**

WENDY BICKMORE (MRC HUMAN GENETICS UNIT IGMM UNIVERSITY OF EDINBURGH, UNITED KINGDOM)

WENDY.BICKMORE@IGMM.ED.AC.UK

Whilst it is widely appreciated that histone modifications correlate to gene expression states and can directly impact on gene expression and repression, our understanding of chromatin states beyond the level of the nucleosome, and how higher-order chromatin structures contribute to the regulation of gene expression, is more rudimentary.

I will discuss the extent to which the organisation of the human genome within the nucleus may contribute to genome function – especially gene expression. This will include, the organisation of heterochromatin with respect to the nuclear periphery, interactions between heterochromatin domains, and the 3D organisation of enhancers and promoters.

P19.2 PHASE SEPARATION PROVIDES A MECHANISM TO REDUCE NOISE IN CELLS

MONDAY 9 DECEMBER **11:30**

ADAM KLOSIN (MAX PLANCK INSTITUTE OF MOLECULAR CELL BIOLOGY AND GENETICS, GERMANY)

KLOSIN@MPI-CBG.DE

Expression of proteins inside cells is inherently noisy, causing variability in protein concentration among identical cells. A central problem in cellular control is how cells cope with this inherent noise. Compartmentalization of proteins via phase separation has been suggested as a potential mechanism to reduce noise, but systematic studies to support this idea have been missing. Here, by using a physical model that links noise in protein concentration to theory of phase separation, we show that liquid droplets can act as effective noise buffers. We provide experimental support for noise buffering by phase separation using an engineered protein that forms liquid-like compartments in mammalian cells, and an endogenous protein nucleophosmin, a major constituent of the nucleolus. Our data suggest an important role of phase separation in biological signal processing and control.

P19.3 MARCHANTIA TCP TRANSCRIPTION FACTOR REGULATES TARGET GENE EXPRESSION IN THE CONTEXT OF 3D CHROMATIN STRUCTURE

MONDAY 9 DECEMBER **12:00**

CHANG LIU (CENTER FOR PLANT MOLECULAR BIOLOGY (ZMBP), GERMANY)

CHANG.LIU@ZMBP.UNI-TUEBINGEN.DE

Information of the genome is not only encoded with the sequence or epigenetic modification but also found in its folding in 3D space. The formation of self-interacting genomic regions, named Topologically Associated Domains (TADs), is known as a key feature of genome organization beyond the nucleosomal level. Unlike those in animals, TADs formation and function in plants are unknown. Here we show that the genome of Marchantia polymorpha, which is a member of a basal land plant lineage, shares an evolutionary conserved 3D landscape with that of higher plants. By analyzing various epigenetic marks across Marchantia TADs, we find that these regions

generally represent interstitial heterochromatin, and TADs borders are enriched with Marchantia TCP1 protein binding. We also identify a type of TADs that we name TCP1-rich TAD, in which genomic regions are highly accessible and are densely bound by TCP1 proteins. Transcription of genes bound by TCP1 differs according to gene location, that those in TCP1-rich TADs clearly show a lower expression level. In tcp1 mutant lines, neither TCP1-bound TAD borders nor TCP1-rich TADs display altered chromatin organization patterns, suggesting that Marchantia TCP1 is dispensable for TADs formation. However, we find that in tcp1 mutants, genes residing in TCP1-rich TADs have a greater extent of expression fold change as opposed to genes not belonging to these TADs. Our results indicate that, besides standing as spatial chromatin packing modules, plant TADs function as nuclear micro-compartments to shape transcription factor activities.

P19.4 ELUCIDATING THE ROLE OF EPIGENETIC REGULATORS AT THE NUCLEAR PERIPHERY IN ARABIDOPSIS

MONDAY 9 DECEMBER **12:30**

KALYANIKRISHNA KALYANIKRISHNA (INSTITUTE FOR BIOLOGY FREIE UNIVERSITÄT BERLIN, GERMANY), PAWEL MIKULSKI (JOHN INNES CENTRE, UNITED KINGDOM), MAREIKE L. HOHENSTATT (INSTITUTE FOR BIOLOGY HEINRICH-HEINE-UNIVERSITY DÜSSELDORF, GERMANY), DANIEL SCHUBERT (INSTITUTE FOR BIOLOGY FREIE UNIVERSITÄT BERLIN, GERMANY)

KALYANI@ZEDAT.FU-BERLIN.DE

Polycomb group (PcG) proteins plays crucial role in the development of a wide range of eukaryotes, including plants and animals. One of the PcG protein complexes, Polycomb Repressive Complex 2 (PRC2), promotes repressive chromatin formation via tri-methylation of lysine-27 on histone H3 (H3K27me3) (Köhler and Villar, 2008). From previous studies in the lab, we characterized a novel chromatin protein family in Arabidopsis, PWWP INTERACTOR OF POLYCOMBS1 (PWO family) which consists of three members. PWO1 is found to be a plant specific interactor of PRC2 and histone H3 (Hohenstatt et al., 2018). Co-immunoprecipitation experiment coupled with mass spectrometry revealed 109 putative PWO1 interactors (Mikulski et al., 2019) and ~60% of those overlap with components of crude plant nuclear envelope enriched fraction (Sakamoto and Takagi, 2013). Presence of nuclear lamina associated components, especially CROWDED NUCLEI 1(CRWN1) - a coiled coil analog of lamin proteins in Arabidopsis - gained attention as it has prominent role in maintaining nuclear morphology (Wang et al., 2013) and chromocenter organization (Dittmer et al., 2007). Further investigation on PWO1-CRWN1 showed a physical and genetic interaction and similar set of regulated genes (Mikulski et al., 2019). We have identified several chromatin modifiers, which interact with PWO1 and in addition regulate nuclear size and morphology. We are currently characterizing these interactors for studying their interplay in gene regulation and nuclear organization. However, we speculate that PWO1 along with its interacting partners is a putative link between PRC2 mediated gene regulation and the nuclear periphery.

P19.5 DOES YOUR PLANT REMEMBER THE LAST TIME YOU WATERED IT? GENE-LOOPS AND INTRA-NUCLEAR GENE LOCALIZATION AS POSSIBLE MEDIATORS OF TRANSCRIPTIONAL MEMORY

MONDAY 9 DECEMBER **12:45**

EMILIA CEPOWSKA (INSTITUTE OF BIOCHEMISTRY AND BIOPHYSICS POLISH ACADEMY OF SCIENCES, POLAND), KATARZYNA JĘDRZEJOWSKA (INSTITUTE OF BIOCHEMISTRY AND BIOPHYSICS POLISH ACADEMY OF SCIENCES, POLAND), LIEN BRZEŹNIAK (INSTITUTE OF BIOCHEMISTRY AND BIOPHYSICS POLISH ACADEMY OF SCIENCES, POLAND), SZYMON ŚWIEŻEWSKI (INSTITUTE OF BIOCHEMISTRY AND BIOPHYSICS POLISH ACADEMY OF SCIENCES, POLAND)

EMILIA.CEPOWSKA@GMAIL.COM

Plants can remember- it is their evolutionary adaptation to the changing environment. This phenomena is a survival trait that provides thermo-tolerance, drought-resistance, salinity-tolerance or a better response to pathogen attack. But how can plants remember, if they do not have brains? Plants can store information about the past environmental stress in a form of epigenetic (i.e. not through DNA sequence) modification that affects gene expression. We would like to discover what exactly are those modifications. Our project is about describing the role of gene-looping and DNA-anchoring to nuclear pore, as possible explanations. In the study Arabidopsis thaliana was used as a model organism in a repetitive drought assay, to check its learning abilities in different mutant backgrounds that we predict to affect transcriptional memory.

P19.6 MARCHANTIA TCP TRANSCRIPTION FACTOR REGULATES TARGET GENE EXPRESSION IN THE CONTEXT OF 3D CHROMATIN STRUCTURE

MONDAY 9 DECEMBER **13:00**

BROMODOMAINS ARE ESSENTIAL FOR THE FUNCTIONS OF ARABIDOPSIS SWI/SNF COMPLEX

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SWI/SNF subfamily of chromatin remodeleling complexes has been shown crucial for transcriptional control of key developmental processes in both animals and plants. SWI/SNF complexes are composed of a catalytic ATPase subunit and other evolutionarily conserved subunits that are necessary for assembly, activity, and recruitment of SWI/SNF to target loci. A hallmark of SWI/SNF is the presence of bromodomains in the ATPase and some non-catalytic subunits. Bromodomains are capable of interacting with acetylated histones, and it is possible that they facilitate SWI/SNF recruitment to chromatin sites marked with this modification. However, what is their exact contribution to the SWI/SNF functions is currently not clear. Here we characterise bromodomain-containing non-catalytic SWI/SNF subunits (BRDs) in Arabidopsis and show that they are involved in regulation of various processes including leaf development, flowering, and hormonal responses. Genetic analyses and protein interaction studies indicate that three BRD proteins are dedicated to SWI/SNF complexes and that bromodomains present in BRDs and the ATPase subunit BRM are together essential for the SWI/SNF activity.

P19.7 CHARACTERIZING THE CHROMATIN CHOREOGRAPHY FACILITATING TRANSCRIPTION IN PLANTS

MONDAY 9 DECEMBER **13:02**

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The functional determinants of histone H3 Lys-4 trimethylation, their potential dependency on histone H2B mono-ubiquitination (H2Bub) and their contribution to defining transcriptional regimes remain poorly understood in plants. This lack of understanding partly stems from the fact that multiple plant histone methyltransferases can catalyze H3K4me3 deposition in plants, unlike in S. cerevisiae where a single SET1 protein performs this activity as part of COMPASS (COMPlex of proteins ASsociated with Set1). Here, we identify SWD2-like b (S2Lb) as a plant ortholog of yeast Swd2, an axillary subunit that tethers COMPASS on H2Bub-containing nucleosomes in S. cerevisiae. Accordingly, S2Lb co-purifies from plant extracts with the AtCOMPASS core subunit WDR5 within a high-molecular weight complex. S2Lb further co-immunoprecipitates with SDG2, a plant-specific HMT acting as the major H3K4me3 activity in Arabidopsis. S2Lb and SDG2 target the same genomic loci, most notably corresponding to the transcription start sites (TSS) of genes with high RNA Polymerase 2 occupancy and highly expressed. Knocking out either S2Lb or SDG2 triggers overlapping transcriptomic defects and developmental phenotypes at the vegetative and reproductive stages, further confirming their functional association. Determining the epigenomic profiles H3K4me3 and S2Lb protein in hub1 mutant plants lacking histone H2B mono-ubiquitination further revealed that, unlike in budding yeast, most of H3K4me3 deposition and COMPASS recruitment to chromatin does not rely on a trans-histone crosstalk with H2Bub. Collectively, this study unveils that the evolutionarily conserved a COMPASS-like complex has been coopted by a plant-specific HMT and mediates H3K4me3 deposition through an H2Bub-independent pathway in Arabidopsis.

P19.8 MODIFYING THE LANDSCAPE OF CROSSOVERS IN BREAD WHEAT MEIOSIS

MONDAY 9 DECEMBER **13:04**

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Hexaploid wheat Triticum aestivum, (2n=6x=42, AABBDD), also known as bread wheat, is a relatively recent allopolyploid, with three related ancestral genomes sharing high similarity in the coding sequences (>95%). The genome of this species is characterized by its large size (~ 17 Gb) and complexity, with a high proportion of repetitive sequences (~ 80%). During meiosis, as it has been described for other crops, DNA exchanges between homologous chromosomes (crossovers, COs), cytologically visible as chiasmata, display a preferential clustering at distal regions. Indeed, 95% of the recombination events occur in regions covering 29% of the genome. The repression of CO formation in the limits of the remaining regions breeding efforts since agronomically important traits cannot be separated from undesirable ones.

This study aims to increase recombination rates in wheat to reduce linkage-drag and generate new allelic combinations. Since chiasmata are dependent on the formation of DNA double-strand breaks (DSBs), we wondered whether the landscape of crossovers could be modified by artificially increasing the number of DSBs. Here we present our initial results on analyzing the consequences of applying DSB-inducing agents into wheat meiocytes. We have treated spikes from plants corresponding to the Chinese Spring landrace and also from mutant plants for either the Ph1 (Pairing Homologus 1) or the Ph2 gene, which are essential to avoid the pairing between homeologous chromosomes. The analysis of pollen mother cells from these spikes will highlight whether artificial DSBs are processed in a different manner than the subset of programmed DSBs.

P19.9 MARCHANTIA TCP TRANSCRIPTION FACTOR REGULATES TARGET GENE EXPRESSION IN THE CONTEXT OF 3D CHROMATIN STRUCTURE

MONDAY 9 DECEMBER **13:06**

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Tomato fruit weight and shape depends on a complex regulatory network occurring during multiple stages throughout plant development. This network is initiated as early as in the floral meristem (FM). Since the gynoecium is the last structure to develop within the flower, the number of carpel primordia directly depends on the maintenance of stem cell activity controlled by the master-regulator SlWUSCHEL (SlWUS). Sl-Inhibitor of Meristem Activity (SlIMA) and At-MIni zinc Finger 2 (AtMIF2) are two members of the MIni zinc Finger family (MIF) involved in the regulation of floral and ovule developments. MIF proteins possess a unique non-canonical zinc-finger domain, which confers to MIF the capacity to interact with other proteins. The characterization of SlIMA and AtMIF2 gain- and loss-of-function transgenic lines in Solanum lycopersicum and Arabidopsis thaliana respectively, allowed the demonstration of their functional homology in the termination of floral stem cell maintenance. During early floral development, the SlIMA and AtMIF2 genes are induced by the MADS-Box transcription factors Tomato AGAMOUS1 (TAG1) and AGAMOUS (AG) respectively. Then, AtMIF2 and SlIMA proteins recruit the C2H2 zinc finger KNUCKLES (KNU) and SlKNU respectively, in a transcriptional repressor complex together with TOPLESS (TPL) and HISTONE DEACETYLASE19 (HDA19). This complex binds to the WUSCHEL (WUS) and SlWUS loci leading to their repression. Our results provide novel insights into the molecular mechanisms

governing FM termination and highlight the essential role of AtMIF2/SlIMA as adaptor proteins during this developmental step, which determines carpel number and therefore fruit size.

P19.10 CHANGES IN GLOBAL DNA METHYLATION AND CHROMATIN ACCESSIBILITY DURING SOMATIC EMBRYOGENESIS INDUCTION AND LONG-TERM MULTIPLICATION OF EMBRYOGENIC CULTURES OF TAMARILLO

MONDAY 9 DECEMBER **13:08**

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Plant regeneration through somatic embryogenesis (SE) is the expression of plant cell totipotency and it has been widely used in plant biotechnology as an efficient tool for plant cloning and as a model system for studying plant embryogenesis and morphogenesis. At cellular level, the balance between stability and plasticity is accomplished through temporal and spatial control of gene expression, chromatin organization and adequate response to external stimuli, which might induce cell fate changes. In the present study we analyzed the variations in DNA methylation, one of the most relevant epigenetic marks, in different cell lines and throughout the first stages of somatic embryo development in indirect SE of tamarillo (Solanum betaceum Cav.), a solanaceous tree with well-established SE protocols. Assays with trichostatin A and 5-azacycitide were also performed, and their effect evaluated on SE efficiency and gene expression changes in different embryogenic lines.The results revealed that long-term embryogenic lines exhibited higher multiplication capacity and a loss of embryogenic competence, associated with accumulation of DNA methylation. Non-embryogenic cell masses, even when recently induced,showed high methylation levels. An initial DNA hypomethylation was detected upon auxin removal, especially in groups of cells of the embryogenic masses that showed a typical meristematic-like organization. In contrast, embryo differentiation was accompanied by a progressive increase of DNA methylation levels. Trichostatin A treatment allowed higher rates of embryo conversion and showed a correlation with the expression levels of genes related to somatic embryo quality.

P19.11 DE-ETIOLATED1 CONTROL OF TRANSCRIPTION FACTOR ACTIVITY AND CHROMATIN STATES

MONDAY 9 DECEMBER **13:10**

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Light is a powerful stimulus that controls a multitude of developmental responses through massive gene expression reprograming. This requires a fine control of the activity of specific transcription activators and the modification of chromatin marks, as well as an intense protein turnover regulated by the ubiquitin-proteasome system. DE-ETIOLATED1 (DET1) is a DDB1-CULLIN4 Associated Factor that, together with COP10 and DDA1, constitutes the CDDD substrate adaptor module within CRL4 E3 ubiquitin ligases. DET1 is a classic photomorphogenesis repressor whose mode of action is not completely understood. In etiolated seedlings, DET1 facilitates CRL4-COP1-SPA activity promoting HY5 destabilization. Yet, DET1 can also directly interact with Histone 2B (H2B) and regulates histone H2B monoubiquitination (H2Bub). In this context, we recently found that the CDDD subunit DDA1 directly interacts with SAGA-INTERACTING FACTOR (SGF11), which is part of a deubiquitination module (DUBm). Recognition of SGF11 by DDA1 recruits the CRL4-CDDD module to ubiquitinate and degrade the DUBm in a DET1- and dark- dependent manner. Therefore, Arabidopsis det1 mutants display reduced levels of

H2Bub mark as a consequence of the accumulation of DUBm and increased H2Bub deubiquitination activity. Noticeably, a mutation in HY5 seems to be sufficient to partially restore H2Bub deficient levels of det1 mutants. This might occur because direct HY5-mediated recruitment of SGF11 to the light transcribed genes during photomorphogenesis. Here we will present recent results suggesting that the CRL4-CDDD complex limits the function of potent transcription factors by controlling their binding to chromatin as well as their stability.

P19.12 MAPPING THE CHROMATIN OF REPEATS USING EXTENDED CHROMATIN FIBERS

MONDAY 9 DECEMBER **13:12**

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Functional and non-functional repeats are important constituents of eukaryotic genomes. Just like single copy genes and regulatory elements, different repeats (or a subset of them) will have particular epigenetic profiles. While chromatin immunoprecipitation has yielded a comprehensive map of chromatin modifications in non-repeat regions, important functional repeats such as telomeres and ribosomal genes are difficult to probe by biochemical approaches. At best, these offer an average profile of all the repeats in a mixed population of cells. An alternative approach that we present is the optical mapping of epigenetic modifications in repeat regions. This is based on the extension of chromatin fibers from isolated nuclei on a microscopic slide and combining immunofluorescence and fluorescent in-situ hybridization detection. Using FISH probes directed at either ribosomal or telomeric repeats, it is possible to detect either histone marks (e.g. H3K4me3 or H3K27me3), histone variants (e.g. H3.1 vs H3.3) and DNA methylation patterns in individual repeats by microscopy. Restrictions on the physical stretching of a particular fiber limit the resolution to approximately 16 kBPs per micron. We show that super-resolution microscopy approaches (e.g. STED, SIM, STORM) can be integrated into the analysis and provide resolution on the scale of a few kBPs.

P19.13 CO-TARGETING RNA POLYMERASES IV AND V PROMOTES EFFICIENT DE NOVO DNA METHYLATION IN ARABIDOPSIS.

MONDAY 9 DECEMBER **13:14**

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The RNA-directed DNA methylation (RdDM) pathway in plants controls gene expression via cytosine DNA methylation. The ability to manipulate RdDM would shed light on the mechanisms and applications of DNA methylation to control gene expression. Here, we identified diverse RdDM proteins that are capable of targeting methylation and silencing in Arabidopsis when tethered to an artificial zinc finger (ZF-RdDM). We studied their order of action within the RdDM pathway by testing their ability to target methylation in different mutants. We also evaluated ectopic siRNA biogenesis, RNA polymerase V (Pol V) recruitment, targeted DNA methylation, and gene-expression changes at thousands of ZF-RdDM targets. We found that co-targeting both arms of the RdDM pathway, siRNA biogenesis and Pol V recruitment, dramatically enhanced targeted methylation. This work defines how RdDM components establish DNA methylation and enables new strategies for epigenetic gene regulation via targeted DNA methylation.

P19.14 UBP5, AN INTERACTOR OF PWO1 AND PCG PROTEINS THAT REGULATES PLANT DEVELOPMENT

MONDAY 9 DECEMBER **13:16**

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PWWP-DOMAIN INTERACTOR OF POLYCOMBS 1 (PWO1) is a new component of the PcG pathway, which are master regulators of development through the epigenetic regulation of gene expression (Hohenstatt et al., 2018), and a novel component of the nuclear lamina, a protein mesh that interacts with the inner nuclear envelope (Mikulski et al, 2019). PWO1 belongs to a small subfamily of three PWWP proteins highly conserved through the plant kingdom which play an important role in the regulation of plant development. The PWWP domain is required for the nuclear localization of PWO1 in speckles where it recruits PcG proteins. In addition, the PWWP domain interacts with histone (H3) peptides and this binding is impaired by phosphorylation of H3S28 (Hohenstatt et al., 2018).

To further characterize PWO1's functions, we analyzed PWO1 putative complex(es) in planta by immunoprecipitation (IP) experiments and subsequent liquid chromatography-mass spectrometry (LC-MS/MS). The experiments yielded more than hundred potential PWO1 interactors (Mikulski et al, 2019). Among them, we are presenting UBIQUITIN PROTEASE5 (UBP5), which is a member of the plant UBP family (March and Farrona, 2017).

Here, we will show UBP5 nuclear localisation and co-localisation with PWO1. We will also present protein-protein data indicating a further link between UBP5 and the PcG pathway, but not to the nuclear lamina. In addition, we will show data from the phenotypic analysis of anubp5line generated by CRISPR/Cas suggesting a key role of UBP5 in the regulation of plant development.

P19.15 LIGHT-DEPENDENT REGULATION OF FOLATE METABOLISM CONTROLS DNA METHYLATION

MONDAY 9 DECEMBER **13:18**

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Changes in DNA methylation can lead to phenotypic variation, which is well-documented in plants. In addition to the striking developmental changes displayed by some natural epimutants or caused by somaclonal variation, DNA methylation dynamics have been linked to various environmental responses, including pathogen defence, nutrient starvation, and drought acclimation, but the underlying mechanisms are largely unknown. Here were show that light-dependent dynamics in folate metabolism affect genome-wide DNA methylation patterns and gene silencing in Arabidopsis.

DNA and histone methylation depend on S-adenosylmethionine (SAM) as methyl donor, which is produced in the methionine cycle. We have previously shown the methionine cycle in turn depends on METHYLENETETRAHYDROFOLATED-EHYDROGENASE1 (MTHFD1), a central enzyme in the folate cycle. Using a reverse genetic approach to further elucidate the role of MTHFD1, we identified a new regulator of MTHFD1-dependent DNA and histone methylation named SUMD (SUPPRESSOR OF MTHFD1). Knock out of SUMD in the mthfd1-1 mutant background suppressed the mthfd1-1 phenotype and restored DNA methylation and gene silencing. Moreover, we observed that the mthfd1-1 phenotype is sensitive to day length. Accordingly, MTHFD1 and SUMD are differentially regulated during day and night and depending on day length, as shown by expression analysis and enzymatic assays. Our results suggest that SUMD regulates folate metabolism and chromatin methylation in response to light and that misregulation of SUMD in mthfd1-1 blocks the methionine cycle and DNA methylation. In summary, this work illustrates how metabolic dynamics are involved in epigenetic regulation in plants, connecting chromatin modification and environmental response.

P19.16 POPULATION GENETIC AND EPIGENETIC VARIABILITY AND DISTRIBUTION OF THE ENDANGERED GREEK ENDEMIC CICER GRAECUM UNDER CLIMATE CHANGE SCENARIOS

MONDAY 9 DECEMBER **13:20**

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Crop wild relatives (CWR) are an incredible resource for crop improvement due to their high level of genetic diversity. Many of their beneficial traits for plant breeding have the potential to help the adaptation of crops to new stressful conditions that they will arise due to climate change. The Mediterranean hot-spot includes numerous endemic and socio-economically important plant species, seriously threatened by climate change and habitat loss. On the other hand, epigenetic variation in natural populations with different habitats might be an important component, in addition to the genetic variation, in plant adaptation to environmental stress. In this study, the genetic and epigenetic diversity of five populations ofCicer graecum, an endangered endemic species from Northern Peloponnese, Greece, wild relative of the cultivatedC. arietinum, was investigated using ISSR, AFLP and methylation-sensitive AFLP (MSAP) markers. Nei's gene diversity (GD) for ISSR markers ranged from 0.191 to 0.228 and for AFLP markers from 0.120-0.148, indicating medium to high genetic diversity at the population level despite of their small size. The ecological adaptation ofC. graecumpopulations was also investigated by correlation of their genetic diversity with certain environmental variables. Aridity arose as the dominant factor positively affecting the genetic diversity ofC. graecumpopulations. Furthermore, we used species distribution modeling in order to predict present habitat suitability for the species, using distribution data and several environmental variables. The possible effects of climate change in habitat suitability were also calculated according to different climate change scenarios.

P19.17 H4K16AC IS AN EPIGENETIC SIGNATURE OF THE PLANT RESPONSE TO ABIOTIC STRESSES

MONDAY 9 DECEMBER **13:22**

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Acetylation at H4 lysine 16 (H4K16ac) was recognized as an epigenetic mark involved in gene regulation in both animals and plants. It was demonstrated that in Arabidopsis and rice the positive correlation exists between H4K16ac and gene expression level with preferential enrichment of this post-translational histone modification around the transcription start sites (TSS) (Lu at al., 2015).

Stress-related changes in plant chromatin epigenetic landscape are important elements of transcriptional regulation responsible for plant adaptation to stress. Using Arabidopsis T87 cell culture we show that lysine 16 of histone H4 undergoes rapid and transient acetylation in response to high salinity and cold and belongs to epigenetic signatures of plant stress response. Combining microarray expression profiling of T87 Arabidopsis cells responding to high salinity with genome-wide distribution of H4K16ac we have shown that in those cells H4K16ac also positively correlates with the level of gene transcription. This modification is enriched especially around the TSS and first exones.

Our data indicates that prior to stress majority of both up- and down- regulated genes are enriched in acetylated H4K16 comparing to transcribed to a similar level but not stress responsive genes.

Li Lu , Xiangsong Chen, Dean Sanders , Shuiming Qian , and Xuehua Zhong (2015) High-resolution mapping of H4K16 and H3K23 acetylation reveals conserved and unique distribution patterns in Arabidopsis and rice. Epigenetics 10:11, 1044--1053

P19.18 IMPACT OF RDNA COPY NUMBER DEPLETION ON A. THALIANA DEVELOPMENT

MONDAY 9 DECEMBER **13:24**

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45s rDNA genes are ubiquitous to all eukaryotes, however the number of repeats and their chromosomal locations are highly polymorphic across different phyla. The role of transcriptionally active and inactive 45s copies has been amply characterized in cellular biology by numerous studies linking loss of 45s copies to abnormal proliferation and cancer (Xu et al., 2017), and also to genome integrity and senescence (Obayashi and Kobayashi, 2014).

In plants however, it hasn't yet been possible to formulate a direct link between variation in 45s copy number and their function in development. Previous studies employed the Chromatin Assembly Factor-1 (CAF-1) mutants which are knock-out alleles of histone chaperonesFASCIATA 1/2, which lead to a drastic reduction in rDNA copy number (Kaya et al., 2001; Mozgova et al., 2010), however it has not yet been possible to draw a causative association between the phenotypes observed and the reduction of copy number. Here, we show how CRISPR-Cas9 mutagenesis has been employed to generateA. thalianaplants with a stably inherited reduction of up to 80% of 45s rDNA genes. We report that decrease of 45s copies is linked with aberrant seedling and seed development, which is also associated with deregulated mechanisms of cell proliferation and meristematic activity.

Currently we are investigating potential chromatin rearrangements which may have been caused by the drastic reduction of rDNA Copy Number, as well as mechanisms that regulate transcription of rDNA genes which in plants remain uncharacterised.

P19.19 GENOME STABILITY IN CHROMATIN REMODELLING MUTANTS CHR2, CHR4, CHR5, CHR9 AND CHR24

MONDAY 9 DECEMBER **13:26**

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Chromatin remodelling affects genome stability as it is necessary for DNA replication, transcription and double-strand breaks repair. Repetitive sequences - telomeres and ribosomal rRNA genes (45S rDNA), belong to the least stable genomic sites, thus representing the good elements to study mechanisms responsible for genome integrity maintenance. We investigate Chromatin Remodelling (CHR) genes, ATPases from the SNF2 complex, affecting accessibility of chromatin by sliding, nucleosome eviction or spacing. CHR genes are classified into several groups. PICKLE (PKL) related genes - CHR4 (PICKLE RELATED 1, PKR1) and CHR5 - from CHD group (Chromodomain Helicase DNA-binding) have been only partially characterised so far.CHR2 (BRAHMA, BRM) belongs to the SWI/SNF group and affects many plant development processes. Another possible DNA repair genes belong to the ERCC6-like group (Excision Repair Cross-Complementation group 6) – CHR9 and CHR24. We show that among these chromatin remodelling factors, CHR2, CHR4, CHR9 and CHR24 participate in the telomere maintenance since mutants in these genes show shorter telomeres than in WT. Two subsequent generation of mutant plants were analysed. In addition, the rDNA copy number also dropped inchr2, chr4, chr9andchr24. Chr5in comparison, showed WT-like telomeres and heterogenous rDNA levels. Our data indicate that CHR genes are involved in the telomere and rDNA stability maintenance and it will be further investigated whether they influence overall genome integrity and epigenetic landscape, or whether the observed effect is specific to the repeats.

This work was supported by The Czech Science Foundation (grant 16-04166Y), COST INDEPTH, INTERCOST (LTC18048).

P19.20 ANALYSIS OF GENETIC INTERACTIONS BETWEEN H1 LINKER HISTONES AND SWI/SNF CHROMATIN REMODELING COMPLEX IN ARABIDOPSIS

MONDAY 9 DECEMBER **13:28**

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SWI/SNF chromatin remodeling complexes (CRCs) and linker histones H1 are essential components of chromatin, affecting its structural and functional dynamics. SWI/SNF CRCs can regulate transcription by changes in nucleosome occupancy and chromatin accessibility, and in plants they have been implicated in regulation of development, growth, stress responses and hormonal signaling pathways. Plant linker histones H1 provide stabilization of nucleosomes and play pivotal role in developmental processes, stress adaptation and regulation of DNA methylation. Previous studies in animals suggested functional antagonism between H1 and SWI/SNF CRCs, but the exact mechanism of this relationship remains unknown. Interestingly, comparison of the global binding profiles of SWI/SNF catalytic subunit BRM and H1s in Arabidopsis revealed strong anticorrelation between binding of BRM and the occurrence of major histone H1 variants H1.1 and H1.2, but not stress-inducible H1.3 variant. In this study we investigated possible antagonism between H1 linker histones and BRM remodeler by genetic approach in Arabidopsis. We generated double, triple and quadruple mutant lines carrying different combinations of mutations in H1 variants and BRM gene, and performed phenotypic and gene expression analyses. Our results shed new light on functional relationship between linker histones and SWI/SNF CRC.

This work was supported by National Science Centre grant 2017/26/E/NZ2/00899.

P19.21 CHARACTERIZING CHROMATIN MOBILITY AND HISTONE DYNAMICS DURING DNA DAMAGE RESPONSES

MONDAY 9 DECEMBER **13:30**

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Plant cells are subject to DNA damage such as double-strand breaks (DSBs), which are a deleterious type of damage. DSBs can be repaired by two different pathways: nonhomologous end joining (NHEJ) and homologous recombination (HR). Double-strand break repair by HR requires a homology search between the DSB ends and a homologous locus used as a template for repair. We have developed a protocol to measure global chromatin mobility as well as DSB site mobility, byin vivoimaging of a locus tagged using thelacO/LacI system and RAD51-GFP foci, respectively. Our results have shown an increase in mobility at DSB sites as well as at the level of the global chromatin upon genotoxic stress using zeocin as a DSB-inducer agent. In order to identify the molecular mechanisms underlying this increase in mobility, we tested the susceptibility of several chromatin mutants to zeocin. We identified two chromatin remodelers (INO80, ARP6) with altered response to DNA damage. Since these remodelers affect histone composition and nucleosome accessibility, we are currently testing the effect of zeocin treatment on histone-DNA interactions in living cells by performing Fluorescence Recovery After Photobleaching (FRAP). Altogether, our preliminary data suggests that general changes in chromatin accessibility may increase chromatin mobility upon DSBs in order facilitate DNA repair by homologous recombination.

P19.22 MOLECULAR CHARACTERIZATION OF THE D53-LIKE SMXL REPRESSION COMPLEXES

MONDAY 9 DECEMBER **13:32**

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The control of branch formation is a crucial aspect of plant development and a relevant agronomic trait. This process is tightly regulated by BRANCHED1 (BRC1), a TCP transcription factor that negatively controls shoot branching. BRC1 acts as an integrator of different developmental, hormonal and environmental signals. Some of these signals promote BRC1expression, like a low red to far-red (R:FR) light ratio, apical dominance and strigolactone signalling, whereas other repress it, such as a high R:FR light ratio, decapitation, and cytokinin and sugar treatments. Although some of these regulatory processes begin to be characterized, the molecular mechanisms underlying the transcriptional regulation of BRC1 remain largelyunknown.In this context, we are studying a group of transcriptional repressors of the strigolactone pathway that repress BRC1 expression: the D53-like proteins SMXL. SMXLs are nuclear, bind the BRC1 promoter (and others), and negatively regulate BRC1 mRNA levels. However, they lack a DNA motif-specific binding domain, which suggests that SMXLs interact with DNA through other cofactors and form part of larger transcriptional repressing complexes. Remarkably, SMXLs form very characteristic speckles in the nucleus, but it is still unknown whether they correspond to transcriptional repressing complexes. Our aim is to characterize these speckles and the components of the putative complexes to understand their involvement in BRC1 regulation.

P19.23 CHANGES OF CHROMOSOME ORGANIZATION DURING CELL CYCLE IN BARLEY

MONDAY 9 DECEMBER **13:34**

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Higher-order structure of chromosomes changes dramatically during the cell cycle. The transcriptionally active interphase requires a different mode of chromatin packaging than the transfer of genetic information during mitosis. Chromosome dynamics has been studied by several approaches, including advanced microscopic techniques and chromatin conformation capture (3C) methods, Hi-C in particular. The greatest amount of knowledge on mitotic chromosomes and their shaping has been obtained from studies in vertebrates, yeasts and animal models. On the other hand, information on plants has been limited and comes predominantly from microscopic studies.

Aiming to study chromatin dynamics during the cell cycle in barley (Hordeum vulgare L.), we coupledin situHi-C protocol with flow sorting, which enabled purifying metaphase chromosomes and nuclei at G1, S and G2 phase,respectively. Outcomes of a Hi-C study on metaphase chromosomes revealed a helical folding of the DNA with ~35 Mb per turn. Our estimate was supported by FISH with several oligo probes, covering continuously a part of barley 5H chromosome. This experiment, employing 3D super-resolution microscopy, confirmed the model computed from the Hi-C data. Analysis of Hi-C data for G1, S and G2 revealed dynamics of the chromosome shaping and indicated slightly different organisation of interphase chromosomes between leaves and root-tip meristems.

Acknowledgements: This research has been supported by a DFG-GACR project, award No. 18-14450J.

P19.24 SUVH7: NEW ACTOR IN THE REGULATION OF ENDOSPERM DEVELOPMENT

MONDAY 9 DECEMBER **13:36**

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The endosperm of most angiosperms is a triploid tissue containing two maternal and one paternal genome. This tissue transfers nutrients to the embryo and is essential for its development, similar to the placenta in mammals. Genomic imprinting is an epigenetic phenomenon taking place predominantly in the endosperm, causing parental alleles to be differentially expressed. Changing the balance of parental genomes causes deregulation of imprinted genes and endosperm collapse, a phenomenon referred to as “triploid block”. Mutations in several imprinted genes can bypass the triploid block, revealing the central importance of balanced imprinted gene expression.

One strong suppressor of the triploid block is the paternally expressed geneSU(VAR)3-9 HOMOLOG 7(SUVH7) (Wolff et al., 2015). Here, we show that increased expression ofSUVH7in the endosperm of triploid seeds correlates with increased deposition of the heterochromatic modification H3K9me2 on transposable elements (TEs), mainly helitrons and MuDR elements. Conversely, triploid seeds lacking SUVH7 function are depleted of H3K9me2. We furthermore demonstrate that SUVH7 interacts with ADMETOS (ADM), a DNAJ protein that previously was shown to cause increased H3K9me2 deposition in triploid seeds (Jiang et al., 2017). Importantly, loss of ADM and SUVH7 affects the same set of TEs, strongly suggesting that they act together to regulate H3K9me2 deposition in the endosperm. We propose that SUVH7/ADM deposit H3K9me2 on TEs that are localized next to imprinted paternally expressed genes (PEGs), causing PEG overexpression and the triploid block phenotype.

P19.25 LINKING SIGNALING PATHWAY TO CHROMATIN DYNAMICS

MONDAY 9 DECEMBER **15:15**

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The genomes of plants are under constant threat ranging from jumping genetic elements to solar UV irradiation. A fundamental question is how cells sense intrinsic and extrinsic cues and reprogram chromatin landscapes to generate adaptive responses. The overarching goal of our research is investigate how epigenetic modifications regulate genome expression, organization, stability, interaction with environment, and inheritance. We address these questions at the whole genome level by combining functional genomics, genetic, proteomic, biochemical, cell biological, and structural approaches. Our lab recently discovered a novel functional connection between light signaling pathway with DNA methylation. I will discuss the data describing our latest understanding of the underlying mechanism.

P19.26 HISTONE VARIANTS AND CHROMATIN DYNAMICS DURING ORGAN GROWTH

MONDAY 9 DECEMBER **15:45**

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Organ development depends on a finely regulated cellular homeostasis that involves cell production and cell differentiation. Stem cells produce derivatives that constitute the transient amplifying compartment where cell cycle exit and cell production are in equilibrium to maintain the organ steady-state size. This involves a significant and continuous reprogramming of gene expression to control cell fate acquisition in proliferating cells before they take different differentiation lineages.

Chromatin dynamics is crucial to establish gene expression patterns, which can be affected by changes

in DNA methylation, post-translational modification of histone and the presence of histone variants in the nucleosome. We are focusing on the role played by histone variants during normal development as well as in response to a variety of stress stimuli.

The root consists of well-defined anatomical and functional zones: the meristem, where cell actively proliferate, the transition zone, where cells undergo endoreplication, and the differentiation zone. Detailed analysis of the balance between canonical and variant histone H3 proteins, H3.1 and H3.3, respectively, we have identified cell populations with different proliferation potential. This is associated with changes in cell cycle phase progression and we are analyzing its molecular basis. The Arabidopsis genome also encodes other histone H3 proteins that share features of canonical H3.1 and variant H3.3, and their role has not been elucidated yet. We will discuss our ideas on how the atypical H3 variant proteins play a role during plant development to accommodate their growth to environmental challenges.

P19.27 HISTONE H3 VARIANT DEPOSITION AND DYNAMICS IN A. THALIANA

MONDAY 9 DECEMBER **16:15**

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Histones are essential components of the nucleosome, the basic subunit of chromatin that structures linear DNA molecules and regulates access of other proteins to DNA. Chromatin organization and thereby DNA accessibility can be modulated through incorporation of different variants of the core histones H3, H2A and H2B into the nucleosome. A sophisticated network of histone chaperones ensures the well-timed deposition of these variants. While H3 histone chaperones are evolutionary conserved, H3 histone variants have emerged several times in different lineages.

To better understand the dynamics of H3 histone variants in plants, we have characterized Arabidopsis CenH3, H3.1 and H3.3 histone chaperones and investigated how imbalanced histone variant deposition affects chromatin organization and gene expression. We provide evidence that the Nuclear autoantigenic sperm protein (NASP) escorts CenH3 in plants and that Histone Regulator A (HIRA) and the Arabidopsis Alpha Thalassemia-mental Retardation X-linked (ATRX) homologue function in complementary pathways of histone H3.3 deposition. Indeed, loss of ATRX reduces cellular histone H3.3 pools and in consequence modulates the H3.1/H3.3 balance in the cell. At the genome-wide scale, our data indicate that ATRX loss-of-function leads to altered H3.3 deposition at a set of genes characterized both by elevated H3.3 occupancy and high expression levels. Instead, hira mutants show reduced nucleosomal occupancy both a genes and in heterochromatin as well as reactivation of transposable elements, altogether emphasizing the role of histone chaperones in regulating chromatin organization and fine-tuning genome expression.

P19.28 CONSERVED ROLES OF THE H3K27ME3 EPIGENOME ON PLANT DEVELOPMENT

MONDAY 9 DECEMBER **16:30**

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The study of the epigenome and its relation with the underlying genome sequence has become a central question in Biology nowadays. Genome-wide maps of histone modifications have been obtained for several plant species. However, most studies focus on model systems and do not enforce FAIR data management principles. We have developed a Reproducible Epigenomics Analysis (REA) pipeline combining a Galaxy environment for data analysis and Jupyter notebooks for results visualization composed into Docker. We successfully applied this REA pipeline to study of the H3K27m3 epigenome on leaves and

flowers of plant model (Arabidopsis thaliana) and crop (Brassica rapa) species. The comparative analysis between leaves and inflorescences genomic datasets suggested that the expression of various floral regulatory genes during development is controlled by H3K27 methylation. H3K27me3 is a repressive epigenetic mark set by Polycomb protein complexes and counteracted by the histone demethylase activity of specific Jumonji domain proteins. We are also studying the role of these epigenetic factors in A. thaliana and B. rapa. Mutant analyses have shown that these epigenetic factors regulate a wide range of developmental responses including flowering time. The similarities and differences between model and crop systems on the role of H3K27me3 regulating plant development will be discussed.

P19.29 GENERATION OF HAPLOIDS IN A. THALIANA BASED ON MANIPULATION OF CENH3 ASSEMBLY FACTOR KNL2

MONDAY 9 DECEMBER **16:45**

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The generation of haploids is one of the most powerful means to accelerate plant breeding. Two main methods, i.e. explantation of gametophytic tissuesin vitroand the selective losses of parental chromosome setin vivovia inter- or intraspecific hybridization, are widely used for DH generation. However, both methods can be applied only to a limited number of genotypes, while depend on tissue culture and regeneration ability or crossability of the species of interest. A recently developed approach based on the manipulation of the centromeric histone H3 (CENH3) results, when applied to crop species, only with a very low efficiency in haploids. To increase the efficiency of this method we employed CENH3 assembly factor KNL2 as further haploid inducer. Our data show that theknl2mutant functions as a maternal haploid inducer in crosses with wild typeA. thalianaplants. We demonstrate that application of stress (high temperature) toknl2mutants has a positive effect on the frequency of haploidization (increase from 1-3% to up to 12%). To identify further potential candidate genes that are involved in the regulation of CENH3 assembly RNAseq experiments using seedlings and flower buds ofknl2mutant and wild type were performed. Additionally, IP-MS analysis with the N- and C-terminal parts of KNL2 helped us to identified proteins forming complex with KNL2. It is interesting to note that CENH3 was precipitated only with the C-terminal part of KNL2 containing the CENPC-k motif required for the targeting of KNL2 to centromeres.

P19.30 LINKER HISTONE H1 DYNAMICS CONTROL ARABIDOPSIS NUCLEAR REORGANIZATION IN RESPONSE TO LIGHT SIGNALS.

MONDAY 9 DECEMBER **17:00**

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Being capable of rapid phenotypic adaptations in response to environmental cues, plants are characterized

by a remarkable developmental plasticity. Specially, plants have the ability to sense light conditions by multiple photosensory receptors and use this information to adapt their morphology and physiology accordingly to a changing environment. For example, the first perception of light by young plantlets emerging from the soil induces radical changes in gene expression that launch growth and photosynthetic activity. During this transition, genome expression reprogramming is accompanied by massive rearrangements of chromatin organization. In dark-grown cotyledon cells, a large part of heterochromatin containing silent and condensed repeated elements is scattered within multiple foci in the nucleoplasm. We refer them as nano-chromocenters. Upon exposure to light, cotyledon de-etiolation triggers the rapid condensation of heterochromatic domains into 8-to-10 large chromocenters that form around centromeres. This phenomenology has led us to the identification of a light-regulated linker histone as a key molecular player in triggering light-induced chromocenter dynamics. Linker histones are conserved structural components of eukaryotic chromatin that contribute to both local and higher-order chromatin organization by restricting DNA accessibility. Here we first report that heterochromatin rearrangements in response to light signals are largely mediated by a fine modulation of linker histone variant H1.3. We further characterize the influence of H1s in defining 3D structures by cytological and Chromosome Conformation Capture (Hi-C) based approaches, aiming at assessing how changes in chromatin topology may adapt the chromosomal landscape for a new transcriptional program.

P19.31 DYNAMIC TRANSCRIPTION AND DNA METHYLATION PATTERNS IN ARABIDOPSIS SHOOT STEM CELLS

MONDAY 9 DECEMBER **17:15**

ORTRUN MITTELSTEN SCHEID (GREGOR MENDEL INSTITUTE OF MOLECULAR PLANT BIOLOGY, AUSTRIA), KLAUS REMBART (GREGOR MENDEL INSTITUTE OF MOLECULAR PLANT BIOLOGY, AUSTRIA), THOMAS NUSSBAUMER (HELMHOLTZ CENTER MUNICH, GERMANY), FALKO HOFFMANN (GREGOR MENDEL INSTITUTE OF MOLECULAR PLANT BIOLOGY, AUSTRIA), RAHUL PISUPATI (GREGOR MENDEL INSTITUTE OF MOLECULAR PLANT BIOLOGY, AUSTRIA), GABRIELE BRADAMANTE (GREGOR MENDEL INSTITUTE OF MOLECULAR PLANT BIOLOGY, AUSTRIA), VU NGUYEN (GREGOR MENDEL INSTITUTE OF MOLECULAR PLANT BIOLOGY, AUSTRIA), NICOLE LETTNER (GREGOR MENDEL INSTITUTE OF MOLECULAR PLANT BIOLOGY, AUSTRIA), MATTIA DONÀ (GREGOR MENDEL INSTITUTE OF MOLECULAR PLANT BIOLOGY, AUSTRIA), RUBEN GUTZAT (GREGOR MENDEL INSTITUTE OF MOLECULAR PLANT BIOLOGY, AUSTRIA)

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Changes in chromatin-based gene regulation, like those induced by cold treatment during vernalization, are usually reset during sexual propagation. There is little evidence that environmentally induced epigenetic modifications are passed on to the progeny across many generations. If such directed changes are inherited, they must pass the bottleneck of the stem cells in the shoot apical meristem (SAM), from which all postembryonic aerial organs are continuously formed, and descendants of these stem cells are progenitors of germ cells that give rise to male and female gametes. Activity of transposable elements (TEs) in the stem cells challenges genome integrity across generations. TEs are epigenetically controlled by several pathways, and we wanted to investigate how this is achieved in these important cells. SAM stem cells are low in number and deeply embedded in the surrounding meristem tissue. To investigate their molecular characteristics, we isolated their nuclei from Arabidopsis SAMs by fluorescence-activated nuclei sorting (FANS) and analysed stage-specific gene expression and DNA methylation at different developmental time points. Stem-cell expression signatures are largely defined by developmental stage but include a core set of stem cell-specific genes, among which are genes implicated in epigenetic silencing. Transiently increased expression of transposable elements in meristems prior to flower induction correlates with dynamic methylation changes at TE genes. The results suggest that epigenetic reprogramming may occur early and could contribute to genome protection in stem cells during germline development.

P19.32 SHEDDING LIGHT ON CHROMOSOME AND CHROMATIN BEHAVIOR IN MEIOSIS

MONDAY 9 DECEMBER **18:00**

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Meiosis is a fundamental source of genetic variation and the basis of nearly all genetics. With this, meiosis

is also key to evolution and breeding. The exchange of chromosome segments is accomplished in meiosis and involves tight control of chromosome behavior, i.e. pairing and synapsis of homologous chromosomes, the formation of DNA double strand break and subsequent repair through cross-overs followed by an intricate mechanism to equally distribute the chromosomes to daughter cells. Our team is interested in molecular mechanisms that govern the progression through meiosis and how chromosome dynamics are spatially and temporally controlled. To this end, we have developed a live cell imaging system that allows us to follow chromosome dynamics throughout meiosis. Here, I will present the latest data from this approach focusing in particular on chromosome and chromatin behavior in meiosis.

P19.33 UNLOCKING THE ROUTE TO REPRODUCTIVE LINEAGE INITIATION IN PLANTS – A ROLE FOR LINKER HISTONE EVICTION?

MONDAY 9 DECEMBER **18:30**

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The separation of the germline from the soma is central to sexual reproduction in multicellular organisms: dedicated cells are entrusted the mission of transmitting an intact, yet unique, genetic makeup to the next generation. Animals and plants evolved different strategies in that the germline is set aside early during embryogenesis for the first, and late during adult growth for the latter. Despite distinct developmental trajectories, the conservation of large-scale chromatin reprogramming between animal PGC (primordial germ cells) and plant SMC (spore mother cells) is remarkable, as shown in mouse and Arabidopsismodel organisms (Hajkovaet al.,Nature 452, 2008; our work: Sheet al., Dev. 140,2013). One striking similarity is the prime eviction of somatic linker histones that precedes a breadth of changes in chromatin structure (heterochromatin content, chromatin compaction) and composition (histone modifications and DNA methylation). Linker histones are fine-scale architects of chromatin structures in eukaryotes and have a broad impact on the epigenetic landscape, as demonstrated as well inArabidopsis(Wierzbicki and Jerzmanowski,Genetics 104, 2005; Zemach et al.Cell 153,2013; our recent work: Rutowicz, Lirskiet al,Genome Biol. 20, 2019). We are investigating the role of H1 eviction and its mechanisms involving notably specific post-translational modifications.

P19.34 THE NUCLEAR ENVELOPE IS THE DANCE FLOOR FOR THE CHROMOSOMES DURING MEIOSIS

MONDAY 9 DECEMBER **19:00**

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Chromosome movements are driven by forces in the cytoplasm, and the nuclear envelope (NE) transduces these forces to telomeric regions that are attached to the nuclear periphery. During meiosis, these movements help in the dynamics of homology searching in, not only facilitating the identification of the homologous sequences on the homologous chromosome but also removing the unwanted contacts between non-homologous chromosomes. We previously reported that two inner Arabidopsis thaliana NE proteins, AtSUN1 and AtSUN2, are required for proper meiotic recombination and synapsis. Additional observations in our laboratory have provided a meiotic function for the plant specific nucleoporin AtNUP136, located in the nuclear basket subcomplex of the nuclear pore complexes (NPCs). The absence of this protein produces alterations in nuclear morphology, interlocks among non-homologous chromosomes, univalents, and reduced fertility. In addition, other Arabidopsis nucleoporins seem to be important for normal meiotic chromosome behaviour. To get insights into the essential function of the NE during meiosis we have analysed the distribution of SUN proteins and NPCs in male meiocytes, from both wild-type and mutant plants deficient in NE-associated proteins. Our results reveal a non-random distribution pattern of these proteins during early prophase I that could be related to chromosome dynamics.

P19.35 ZIP4 AND THE STABILIZATION OF WHEAT MEIOSIS ON POLYPLOIDIZATION

MONDAY 9 DECEMBER **19:15**

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The foundation of western civilization owes much to the high fertility of bread wheat, which results from the stability of its polyploid genome. Despite possessing multiple sets of related (homeologous) chromosomes, hexaploid wheat (AABBDD, 2n=6x=42) behaves as a diploid during meiosis. Ph1 is the major locus controlling this behavior. Ph1 ensures that only homologous chromosomes recombine, which although necessary for successful segregation, is an obstacle for the exploitation of wheat's wild relatives as donors for economically important traits.

Using immunolocalization of key meiotic proteins, combined with FISH (fluorescence in situ hybridization), we demonstrated that independently of Ph1, only homologous chromosomes can synapse at the telomere bouquet stage. However, in wheat lacking Ph1, overall synapsis is delayed, with more synapsis occurring after the telomere bouquet, when synapsis between related chromosomes is also possible. Next, we used RNA-seq to reveal that only two genes located in the Ph1 region were expressed during early meiotic prophase I, one of which is the duplicated ZIP4 gene (TaZIP4-B2). Finally, using both TILLING mutants and CRISPR-Cas9 technology, 60 years since the discovery of Ph1, we could verify that a single gene, ZIP4, inside this locus, is responsible for the Ph1 effect on recombination.

Apart from the Ph1 effect on synapsis and crossover formation, an effect on centromere behaviour has also been reported. We are currently exploring whether ZIP4 is also responsible for this effect. If so, this would suggest that ZIP4 have a more central role in meiosis than originally suspected from studies on model species.

P19.36 HVST1: AN E3 UBIQUITIN LIGASE INVOLVED IN MEIOSIS IN BARLEY

MONDAY 9 DECEMBER **19:30**

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Post translational modifications (PTMs) play a regulatory role in virtually all cellular processes, including mitotic and meiotic cell division. One of the most abundant and versatile PTMs of proteins is ubiquitination, the covalent attachment of ubiquitin to target proteins. The versatility of ubiquitination derives from the diversity of ubiquitin configurations: eight possible C-terminal ubiquitin chain linkages which can be homo- or heterotypic, as well as unlinked ubiquitin. The transfer of ubiquitin to a target protein is governed by a cascade involving E1 activating, E2 conjugating, and E3 ligating enzymes; increasing in specificity and decreasing in conservation from E1 to E3. The canonical function of ubiquitination is targeting of proteins for degradation by the proteasome. However, varied configurations of attached ubiquitin can serve unique functions such as activation or cellular trafficking. Cytological investigation of a spontaneous semi-sterile barley mutant revealed defects in chromatin condensation and male meiotic progression leading to an intra-chromosomal polycomplex which prevented complete assembly of the synaptonemal complex (SC) during prophase I. Fine mapping led to the identification an insertion in the mutant resulting in a truncated RING zinc finger domain containing gene, Hordeum vulgare Sticky Telomeres 1 (HvST1). Expression and enzyme activity assays of the wild type protein show that it acts as an E3 ubiquitin ligase. Through comparison of ubiquitinated proteins in HvST1 mutant and wild type barley we hope to elucidate the substrate specificity of HvST1, and uncover its role in SC formation, recombination, and DNA repair.

P19.37 UNDERSTANDING THE MECHANISMS THAT SHAPE THE DISTRIBUTION OF CROSSOVERS ALONG CHROMOSOMES DURING MEIOSIS

MONDAY 9 DECEMBER **19:45**

MATHILDE GRELON (INRA-IJPB, FRANCE), AURÉLIE HUREL (INRA-IJPB, FRANCE), AURÉLIE CHAMBON (INRA-IJPB, FRANCE)

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Meiotic recombination and the formation of crossovers (COs) is a crucial step of sexual reproduction that reshuffles genetic characters and thus generates new combinations of alleles. Meiotic recombination is also essential to the good segregation of homologous chromosomes at the first meiotic division, hence ensuring fertility. Despite the stochastic occurrence of COs, their distribution in the genome is under strong constraints, whose mechanisms and functions are still obscure. First, the number of COs is almost always very limited, typically 1-3 per chromosome pair (in average 10 per cell inArabidopsis thaliana). Second, the localisation of COs is not homogeneous among the genomes, with large domains devoid of events, notably the peri-centromeric regions that can represent a very large part of the chromosomes (e.g. 80% of wheat chromosomes). Third, the presence of a CO along a chromosome pair inhibits the occurrence of another one close by, a phenomenon called interference. In order to gain insights into the forces that govern these CO patterns, we have recently developed a protocol that allows studying meiosis progression without modifying the tri-dimensional organization of chromosomes and nuclei (Hurel et al., Plant J. 2018). By combining immuno-localization of chromosome axes and CO events, we can now trace individual chromosome pairs and determine the complete pattern of COs along each of them. I will present data on CO pattern analyses during A. thaliana meiosis in wild-type plants and in mutants that modify the number of recombination events.

P19.38 THREE-DIMENSIONAL GENOME MODELING ENGINE FOR DATA-DRIVEN BIOPHYSICAL SIMULATIONS OFCTCF AND RNAPII-MEDIATEDHIGHER-ORDER CHROMATINORGANIZATION

TUESDAY 10 DECEMBER **11:45**

DARIUSZ PLEWCZYNSKI (UNIVERSITY OF WARSAW, POLAND)

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Chromosomal folding are important features of genome organization, which play critical roles in genome functions, including transcriptional regulation. ChIA-PET is unique in its ability to generate multiple datasets (in a single experiment), including binding sites, enriched chromatin interactions (mediated by specific protein factors, like CTCF), as well as non-enriched interactions that reflect topological neighborhoods of higher-order associations.

ChIA-PET experimental strategy combined with computational modelling can comprehensively map higher-order chromosome folding and specific chromatin interactions mediated by CTCF and RNAPII with haplotype specificity and nucleotide resolution in different human cell lineages. CTCF/cohesin-mediated interaction anchors serve as structural foci for spatial organization of constitutive genes concordant with CTCF-motif orientation, whereas RNAPII interacts within these structures by selectively drawing cell-type- specific genes towards CTCF-foci for coordinated transcription.

I will present the foundations of the 3D GeNOme Modeling Engine (3D-GNOME) - a web service which generates 3D structures from 3C data and provides tools to visually inspect and annotate the resulting structures, in addition to a variety of statistical plots and heatmaps which characterize the selected genomic region. 3D-GNOME simulates the structure and provides a convenient user interface for further analysis. Alternatively, a user may generate structures using published ChIA-PET data for the GM12878 human cell line by simply specifying a genomic region of interest. 3D-GNOME is freely available athttp://3dgnome.cent.uw.edu.pl/providing unique insights in the topological mechanism of human variations and diseases.

P19.39 CHROMATIN PARTICLE SPECTRUM ANALYSIS AND THE DYNAMICS OF SUB-NUCLEOSOMAL PARTICLES IN ARABIDOPSIS

TUESDAY 10 DECEMBER **12:15**

JAMES A.H. MURRAY (SCHOOL OF BIOSCIENCES CARDIFF UNIVERSITY MUSEUM AVENUE CARDIFF, UNITED KINGDOM), DANIEL A. PASS (SCHOOL OF BIOSCIENCES CARDIFF UNIVERSITY MUSEUM AVENUE CARDIFF, UNITED KINGDOM), EMILY SORNAY (SCHOOL OF BIOSCIENCES CARDIFF UNIVERSITY MUSEUM AVENUE CARDIFF, UNITED KINGDOM), NICHOLAS KENT (SCHOOL OF BIOSCIENCES CARDIFF UNIVERSITY MUSEUM AVENUE CARDIFF, UNITED KINGDOM)

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Whilst details of nucleosome position in Arabidopsis have been previously analysed, there is less understanding of their relationship to more dynamic protein particles that protect sub-nucleosomal fragments of DNA (subNSPs), such as transcription factor complexes. We combined differential micrococcal nuclease (MNase) digestion and a modified paired-end sequencing protocol to reveal the chromatin structure landscape of Arabidopsis cells across a wide particle size range (Pass et al., 2017). Linking this data to RNAseq expression analysis provides detailed insight into the relationship of DNA-bound particles with transcriptional activity, including the more labile -1 nucleosome positioned upstream of the transcription start site (TSS) of active genes which we found sensitive to higher levels of MNase digestion. We investigated the response of the chromatin landscape to changes in environmental conditions using light and dark growth, given the large transcriptional changes that result. Shifts in the suites of expressed and repressed genes show little correspondence to changes in nucleosome positioning, but led to significant alterations in the profile of subNSPs upstream of TSS both globally and locally. We found specific changes upstream of known light responsive genes, and when we examined previously mapped positions for the light responsive transcription factors (PIF3, PIF4 and CCA1) which regulate light responses, these show striking changes in subNSPs co-localized with these binding sites. However this small particle structure is detected only under low MNase digestion and lost on more complete digestion of chromatin to nucleosomes. In contrast, further examining chromatin under genetic conditions where cellular differentiation is inhibited due to overexpression of the cell cycle regulator CYCLIN D3;1 (Dewitte et al., 2003) showed differences in nucleosome positioning around transcription start sites. Wide-size spectrum analysis by differential MNase digestion therefore allows detection of binding potentially linked to active processes such as transcription and comparisons of genome-wide subNSP profiles reveals dynamic changes in response to environmental conditions at distinct genomic locations such as transcription start sites. The approach allows insight into the complex influence of genetic and extrinsic factors in modifying the sub-nucleosomal landscape in association with transcriptional changes.

P19.40 LIVE IMAGING SYSTEM TO TRACKING POST-TRANSLATIONAL MODIFICATION DYNAMICS IN ARABIDOPSIS THALIANA

TUESDAY 10 DECEMBER **12:45**

MIO K. SHIBUTA (TOKYO UNIVERSITY OF SCIENCE, JAPAN), MEGUMI MATSUOKA (TOKYO UNIVERSITY OF SCIENCE, JAPAN), MAYU YOSHIKAWA (TOKYO UNIVERSITY OF SCIENCE, JAPAN), KAZUKI KURITA (TOKYO UNIVERSITY OF SCIENCE, JAPAN), TAMAKO YAMAOKA (TOKYO UNIVERSITY OF SCIENCE, JAPAN), TAKUYA SAKAMOTO (TOKYO UNIVERSITY OF SCIENCE, JAPAN), HIROSHI KIMURA (TOKYO INSTITUTE OF TECHNOLOGY, JAPAN), SACHIHIRO MATSUNAGA (TOKYO UNIVERSITY OF SCIENCE, JAPAN)

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Post-translational modifications such as histone modification and RNA polymelase modification play a crucial role in tissue differentiation and cellular responses to environmental stimuli. To track dynamics of these modifications, we introduced a genetically encoded system, termed modification-specific intracellular antibodies (mintbodies) into Arabidopsis thaliana. To confirm if mintbody can act correctly as an antibody in living cells, we conducted ChIP-seq using RNA polymelase II ser2ph (the elongation form of PolII)- specific mintbody, and found that distribution patterns over genebody and target genes were mostly overlapped between native RNA polymelase II ser2ph and mintbody. These results suggest that we can use RNA polymelase II ser2ph-specific mintbody as a reliable monitoring tool. Fluorescent microscopy showed that RNA polymelase II ser2ph-mintbody localized in euchromatin but not in heterochromatin. We also observed RNA polymelase II ser2ph foci in

nuclear, and these may indicate highly transcription active regions. For quantitative measurement of endogenous modification levels, mintbody and standard fluorescence protein were co-expressed using 2A peptide-mediated co-expression system, and calculated the ratio of intensity of mintbody to that of standard fluorescence protein. This method enable the accurate measurement because the two proteins were expressed within single ORF at the even levels through a co-translational cleavage event. We also observed H3K9ac-, H3K4me3- and H3K27me3-specific mintbodies in several tissues. In this poster, we plan to organizing these and reporting them.

P19.41 THE EUROPEAN GALAXY SERVER: A PLATFORM FOR ACCESSIBLE, REPRODUCIBLE AND COLLABORATIVE BIG DATA ANALYSES

TUESDAY 10 DECEMBER **13:00**

ANIKA ERXLEBEN (UNIVERSITY OF FREIBURG, GERMANY), BJOERN A. GRUENING (UNIVERSITY OF FREIBURG, GERMANY)

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Analysis of large biomedical datasets produced by high-throughput data generation technologies requires sophisticated statistical and computational methods, as well as substantial computational power. Researchers want to be involved in their computation-dependent data analyses but therefore they need a suitable computational infrastructure and in-depth bioinformatics training. The Galaxy project (1) tries to solve this by providing a framework for accessible, reproducible and collaborative data analysis to ensure the researcher participation on their own data analysis.

Galaxy is an open-source web-based scientific data analysis platform used nowadays by hundreds of thousands of scientists across the world to analyze large biomedical datasets such as those found in epigenetics, genomics, proteomics, metabolomics, and imaging. In this talk we will present the newest release of the Galaxy framework, new features of the European Galaxy server (2) and highlights from the community. Furthermore, we will provide updates on recent developments of the Galaxy Training Network (GTN) (3). The GTN provides a dozen of high-quality hands-on tutorials for Galaxy, e.g. on big data from epigenetics, imaging, genome annotation, and multi-omics analyses such as transcriptomics, metabolomics, and proteomics. For analyzing the nuclear architecture and chromatin organization, we will present the Galaxy HiCExplorer (4) that facilitates the study of the 3D conformation of chromatin by allowing Hi-C data processing, analysis and visualization.

(1) Galaxy project https://galaxyproject.org

(2) Galaxy Europe https://usegalaxy.eu

(3) Galaxy Training material https://training.galaxyproject.org

(4) Galaxy HiCExplorer https://hicexplorer.usegalaxy.eu

P19.42 STATISTICAL MODELING OF SPATIAL INTERACTIONS: NEW INSIGHTS INTO THE SPATIAL ORGANIZATION OF PLANT HETEROCHROMATIN

TUESDAY 10 DECEMBER **13:15**

PHILIPPE ANDREY (INRA, FRANCE), JAVIER ARPÒN (INRA, FRANCE), KAORI SAKAI (INRA, FRANCE), VALÉRIE GAUDIN (INRA, FRANCE)

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The spatial organization in the cell nucleus is tightly linked to genome functions such as gene regulation. Similarly, specific arrangements of macromolecular complexes, organelles and cells are involved in many biological functions. Deciphering organization principles is thus essential to the understanding of biological systems. Spatial interactions, such as attraction or repulsion, are key determinants of spatial patterns and are typically analyzed using spatial statistical methods for point patterns. However, these methods have been developed in contexts, such as ecology, where data are single observations of 2D points distributed within sampling windows, and are generally not appropriate for analyzing biological spatial data. Furthermore, spatial studies frequently rely on complete spatial randomness as reference model, which only represents a first step in the analysis of spatial interactions. Methods are thus needed for analyzing repeated observations of 3D patterns with objects of variable size distributed within domains of arbitrary shapes, and new spatial models are required to finely dissect spatial interactions. To this aim, we develop a computational framework combining unbiased statistical tests, novel spatial descriptors and models more elaborate than the classical random model. We used constitutive heterochromatin, a dynamic, structural and functional nuclear compartment, as a model system to demonstrate the potential of our

framework and its ability to reveal complex organization principles. We show that chromocenters are organized along two directions in the nuclear space and obey a multiscale organization. The proposed framework can be used to identify determinants of spatial organizations and to question their interplay with biological functions.

P19.43 PROBING THE STRUCTURE OF THE NUCLEUS; LIVE CELL IMAGING REVISITED

TUESDAY 10 DECEMBER **13:30**

DAVID E EVANS (OXFORD BROOKES UNIVERSITY, UNITED KINGDOM)

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Live cell imaging using fluorescent probes has provided a powerful tool to study the plant nucleus. Using AiryScan technology, the location of nuclear envelope, nucleosketon and associated proteins can be reassessed and deeper insights into protein location and underlying structure explored. This presentation will focus on components of the LINC complex from several studies and on the methodological challenges posed by live-cell imaging of the nuclear periphery at high resolution.

P19.44 WHEAT CHROMATIN ARCHITECTURE IS ORGANIZED IN GENOME TERRITORIES AND TRANSCRIPTION FACTORIES

TUESDAY 10 DECEMBER **17:30**

MOUSSA BENHAMED (INSTITUTE OF PLANT SCIENCES, FRANCE)

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Polyploidy is considered as a major factor in successful plants domestication. However, how polyploidy challenges chromosome folding and its functional organization is poorly understood. In this context, we examined the hexaploid wheat nuclear architecture by integrating RNA-seq, ChIP-seq, ATAC-seq, Hi-C and Hi-ChIP data. Our results highlight the presence of three levels of large-scale spatial organization: (i) the arrangement into genome territories, (ii) the diametrical separation between facultative and constitutive heterochromatin, and (iii) the organization of RNA polymerase II around transcription factories. We found that transcription factories are established through the micro-compartmentalization of transcriptionally active genes, which is determined by physical interactions between genes with specific euchromatic histone modifications. Both intra- and interchromosomal RNAPII-associated contacts involve multiple genes displaying similar expression level. Our results provide new insights into the relationship between epigenetic marks and chromosome conformation to determine a 3D spatial organization of gene expression, a key factor governing gene transcription in polyploids.

P19.45 THREE-DIMENSIONAL CHROMOSOMAL ARCHITECTURE OF METABOLIC GENE CLUSTERS

TUESDAY 10 DECEMBER **18:00**

HANS-WILHELM NÜTZMANN (UNIVERSITY OF BATH, UNITED KINGDOM), DANIEL DOERR (UNIVERSITY OF BIELEFELD, GERMANY), AMÉRICA RAMÍREZ-COLMENERO (CINESTAV, MEXICO), JESÚS EMILIANO SOTELO FONSECA (CINESTAV, MEXICO), SELENE L. FERNANDEZ-VALVERDE (CINESTAV, MEXICO), STEVEN WINGETT (BABRAHAM INSTITUTE, UNITED KINGDOM), EVA WEGEL (JOHN INNES CENTRE, UNITED KINGDOM), ANNE OSBOURN (JOHN INNES CENTRE, UNITED KINGDOM)

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Recent genetic, genomic and biochemical research has established a new feature of plant genomes – the re-occurring co-localisation of genes for specialised metabolic pathways. The ability to synthesise a diverse cocktail of specialised metabolites is essential for all plants to interact and communicate with the environment and provides humans with a plethora of chemical scaffolds to combat illnesses and develop higher-value molecules. The co-localisation of the functionally related biosynthesis genes contrasts the general gene order in eukaryotes and raises numerous questions about the genetic organisation, evolution and regulation of these so-called metabolic gene clusters.

Characteristically, genes within plant metabolic clusters are co-expressed and show concerted expression in response to changing environmental conditions and during cellular and developmental differentiation. Recently, we have discovered that metabolic gene clusters are delineated by signatures of chromatin marks. Here, we present cluster-specific chromosome conformation capture and high-resolution fluorescencein situhybridisation data inArabidopsis thaliana. We show that metabolic gene clusters reside in local interactive domains within the three-dimensional nuclear space and demonstrate that the cluster-associated chromosome conformation displays variable pattern in expressing and non-expressing organs. These changes are accompanied by a repositioning of clusters within the nucleus.

Our work offers an unprecedented view on the three-dimensional organisation of co-regulated gene clusters in plants. It demonstrates a unique and flexible chromosome architecture at clusters that may be crucial in their tight transcriptional regulation and the metabolic output of plants.

P19.46 SMC5/6 COMPLEX CONTROLS PLOIDY LEVELS OF MALE GAMETES IN ARABIDOPSIS

TUESDAY 10 DECEMBER **18:30**

ALES PECINKA (INSTITUTE OF EXPERIMENTAL BOTANY, CZECH REPUBLIC), FEN YANG (INSTITUTE OF EXPERIMENTAL BOTANY, CZECH REPUBLIC), NADIA FERNANDEZ JIMENEZ (UNIVERSIDAD COMPLUTENSE DE MADRID, SPAIN), MARTINA TUČKOVÁ (INSTITUTE OF EXPERIMENTAL BOTANY, CZECH REPUBLIC), JAN VRÁNA (INSTITUTE OF EXPERIMENTAL BOTANY, CZECH REPUBLIC), MARIANA DIAZ (MAX PLANCK INSTITUTE FOR PLANT BREEDING RESEARCH, GERMANY), MONICA PRADILLO (UNIVERSIDAD COMPLUTENSE DE MADRID, SPAIN)

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Flowering plants undergo a series of complex developmental transitions including production of haploid gametes and seeds. Seed development starts with double fertilization, where the first haploid sperm fuses with the haploid egg nucleus and the second haploid sperm fuses with the diploid central cell nucleus leading to the development of diploid embryo and triploid endosperm, respectively. Structural maintenance of chromosomes 5/6 (SMC5/6) complex has a crucial function in the control of genome stability and DNA damage repair. However, the role of SMC5/6 complex in plant development and stress responses is little known. We will demonstrate that loss of function from HPY2, E3 SUMO ligase subunit of the SMC5/6 complex, causes uniparentally-inherited abnormal seed development, characterized by poor embryo development and liquid endosperm. By searching for the cause of this defect, we noticed that hpy2 plants produce pollen of variable sizes. Because large size of the plant organs is often associated with higher polyploidy, we measured ploidy of the hpy2 offspring and found that some plants are triploid. We traced the origin of these defects into meiotic division II. This suggested that SMC5/6 complex plays important function in gametogenesis and seed development, and contributes to the maintenance of the normal ploidy levels across generations.

P19.47 INVOLVEMENTS OF NUCLEAR PORE COMPLEX PROTEINS IN THE REGULATION OF SPATIAL ARRANGEMENT OF CHROMATIN DOMAINS IN ARABIDOPSIS

TUESDAY 10 DECEMBER **18:45**

TAKUYA SAKAMOTO (TOKYO UNIVERSITY OF SCIENCE, JAPAN), YUKA OKO (TOKYO UNIVERSITY OF SCIENCE, JAPAN), NANAMI ITO (TOKYO UNIVERSITY OF SCIENCE, JAPAN), YUKI SAKAMOTO (OSAKA UNIVERSITY, JAPAN), SACHIHIRO MATSUNAGA (TOKYO UNIVERSITY OF SCIENCE, JAPAN)

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Specific chromatin domains such as centromeres, telomeres, and rDNA arrays have the regularity of spatial arrangement in interphase nuclei of somatic cells. In Arabidopsis, centromeres are fixed proximally to nuclear periphery and show scattered distribution. Although 45S rDNA arrays are located away from centromeres on chromosomes 2 and 4, the spatial location of 45S rDNA arrays basically show an association with centromeres. So far, we proposed a two-step regulatory mechanism of spatial arrangement of centromeres mediated by a large complex composed by nuleoplasmic protein condensin II and nuclear envelope proteins SUNs and KASHs, and CRWNs. In addition, we found that condensin II is required for the association of rDNA arrays with centromeres. Here, to further understand the regulatory mechanism of spatial arrangement of chromatin domains, we focused on the nuclear pore complex, a subunit of which has been shown to interact with pericentromeric chromatin. Nuclear pore complex is composed of 28 subunits. We isolated the mutants for 23 subunits and analyzed the spatial arrangement of centromeres and the association of rDNA arrays with centromeres. As a result, we found that some mutants showed uneven distribution of centromeres and some showed disassociation of rDNA arrays from centromere, indicating the involvement

of nuclear pore complex proteins in the regulation of spatial arrangement of chromatin domains. We will discuss how nuclear pore complex proteins work in the two-step regulatory mechanism of centromere distribution and the relationships between condensin II and nuclear pore complex proteins in terms of positioning of rDNA arrays.

P19.48 LIGHT-DEPENDENT ACCUMULATION OF AN IRON UPTAKE TRANSCRIPTION FACTOR IN NUCLEAR BODIES

TUESDAY 10 DECEMBER **19:00**

TZVETINA BRUMBAROVA (HEINRICH HEINE UNIVERSITY, GERMANY), KSENIA TROFIMOV (HEINRICH HEINE UNIVERSITY, GERMANY), RUMEN IVANOV (HEINRICH HEINE UNIVERSITY, GERMANY), YVONNE STAHL (HEINRICH HEINE UNIVERSITY, GERMANY), PETRA BAUER (HEINRICH HEINE UNIVERSITY, GERMANY)

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Iron (Fe) is of crucial importance for the growth and development of plants, and hence for human food security. In order to utilize the highly abundant but poorly bioavailable Fe from the soil, Arabidopsis employs a strategy for active solubilization and acquisition of Fe. The bHLH transcription factor FER-LIKE IRON DEFICIENCY-INDUCED TRANSCRIPTION FACTOR (FIT) is a key regulator of this process and undergoes post-translational activation upon low Fe availability. However, the molecular mechanism discriminating between active and inactive FIT forms in response to internal and external signals is still poorly understood. Previously, it has been shown that Fe uptake and FIT transcription are light-dependent and circadian clock-regulated. In order to put the light regulation of FIT activity in a biological context, the effect of different light qualities on several physiological and molecular read-outs of FIT activity is investigated. We have discovered that blue light triggers the formation of FIT-GFP-containing nuclear bodies (NBs) which coincides with FIT protein activation. Co-localization studies between FIT and marker proteins for different types of subnuclear structures aim to uncover the nature of these FIT-containing NBs. The functional relevance of their appearance is targeted by FRET-APB and FRET-FLIM studies on the prominence of FIT hetero-dimer formation with activating protein partners in NBs versus the nucleoplasm. The fact that a specific light quality can influence the protein activity of a root expressed transcription factor regulating nutrient uptake opens up new venues of research in this field.

P19.49 TRANSGENE SILENCING IN 3D – HOW A CHROMOSOMAL KNOT CAN INACTIVATE FOREIGN DNA ELEMENTS

TUESDAY 10 DECEMBER **19:15**

STEFAN GROB (UNIVERSITY OF ZURICH, SWITZERLAND)

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Cells require elaborate mechanisms to efficiently pack chromosomes in the nucleus, while still allowing access to the genetic information. In addition, three-dimensional (3D) chromosome architecture is linked to epigenetic processes and transcriptional activity. Despite progress in the field, well-established cases of functional relationships between transcription and 3D chromatin architecture remain rare. We previously identified a 3D chromatin structure in Arabidopsis termed the KNOT, in which ten genomic regions (KEEs) physically contact each other. Here we show that KEEs are involved in the silencing of transgenes. Transgenes integrated in the genome can fold towards the KNOT, coinciding with their transcriptional silencing. Thus, transgene integration can lead to significant perturbation of 3D chromosome architecture. Regions adjacent to the insertion sites are not subjected to silencing, despite their dislocation within the nucleus. This novel silencing mechanism, termed KNOT-linked Silencing (KLS) may act independently of previously described silencing mechanisms, as we cannot observe any significant contribution of small RNAs and DNA methylation. KLS is heritable across generation and shows trans-silencing effects, as the introduction of KNOT-silenced transgenes can lead to the silencing of previously active transgenes.

P19.50 THE PAN-GENOMES OF BARLEY AND WHEAT

WEDNESDAY 11 DECEMBER **09:00**

NILS STEIN (IPK GATERSLEBEN, GERMANY)

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By the effort of international consortia first reference sequences of barley and wheat have recently become public. With no doubt, there is great value of these reference sequences, providing a new knowledge-base for research and breeding in these global crop species, however, single reference sequences by far do not provide deep insight into the functional genome diversity representative of an entire crop species. This requires the deep analysis of genome sequences of different haplotypes that are representative for individual sub-populations, an approach referred to as pan-genome analysis. In wheat and barley pan-genome projects are underway. While in wheat elite genotypes representative for different global wheat breeding programs were selected to maximize the value for breeding research, in barley a more comprehensive approach was chosen. Based on a diversity dataset obtained from sequence analysis of 22,000 international barley accessions, a panel of twenty highly diverse genotypes, representing different diversity sub-populations was selected for whole genome sequencing and assembly. First results on pan-genome diversity in barley and wheat, including large scale structural variation analysis by Hi-C, will be presented demonstrating the impact for research and application.

P19.51 ADAPTATION TO ENVIRONMENTAL STRESS BY A DYNAMIC CHROMATIN-BASED STRESS MEMORY

WEDNESDAY 11 DECEMBER **09:45**

ISABEL BÄURLE (UNIVERSITY OF POTSDAM, GERMANY)

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In nature, plants often encounter chronic or recurring stressful conditions. An increasing number of observations suggest that plants can be primed by exposure to stress, thereby activating a stress memory that enables a more efficient response upon a recurring stress incident. My lab studies heat stress memory in plants as a model case for environmental stress memory. Seedlings acquire thermotolerance through a heat treatment at sublethal temperatures (priming heat stress) that enables them to survive an otherwise lethal heat stress. This thermotolerance is actively maintained for several days as indicated by the existence of mutants which are able to establish thermotolerance, but fail to maintain it.

We have found that heat stress induces sustained histone methylation at heat stress memory-related loci that outlasts the transcriptional activity of these loci and marks them as recently transcriptionally active. In a forward genetics approach, we have found that regulation of nucleosome occupancy is also required for sustained activation of memory gene expression. Sustained low nucleosome occupancy is mediated by the FORGETTER1 (FGT1) protein through interaction with chromatin remodeling proteins. From the same genetic screen, we have identified additional components that positively regulate HS memory and indicate further involvement of transcriptional regulation, but also a crosstalk with membrane dynamics. Details on the characterization of these novel components will be presented. In summary, the physiologically defined phenomenon of HS memory has a molecular equivalent in the transcriptional memory and associated changes in chromatin structure and protein dynamics.

P19.52 HOW JASMONATE-MEDIATED DISTRESS SIGNALS AFFECT PLANT GROWTH

WEDNESDAY 11 DECEMBER **10:15**

ALESSANDRA DEVOTO (ROYAL HOLLOWAY UNIVERSITY OF LONDON, UNITED KINGDOM)

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Plant development and stress responses are regulated by complex signalling networks that mediate specific and dynamic plant responses upon activation by various types of signals. Together with other plant hormones, jasmonates (JAs) regulate responses to stress, mediate defence and a plethora of processes while acting like growth inhibitors. The latest work has identified new regulatory nodes in the transcriptional network that regulates several diverse plant responses to developmental and environmental cues.

In my laboratory, we are interested in discovering the cellular components linking plant stress responses to growth processes with the aim to improve yield and adaptation of plants to their environment. An update will be provided on how JAs modify growth, by altering cell proliferation and expansion. The work presented provides tools to uncover novel mechanisms coordinating cell division and post-mitotic cell expansion also in the absence of organ developmental control. The studies also contribute to the understanding of the production of metabolic resources by JAs. We also exploit the ability of JAs to induce protective secondary metabolites to develop novel functional screenings to improve the understanding of key pathways leading to the production of economically important compounds.

P19.53 DYNAMIC UBIQUITINATION CONTROLS TRANSCRIPTION DYNAMICS

WEDNESDAY 11 DECEMBER **11:00**

STEVEN H. SPOEL (UNIVERSITY OF EDINBURGH, UNITED KINGDOM)

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Gene expression plays pivotal roles in the development of eukaryotic cells and their response to the environment. Failure to precisely program cellular gene expression often has pathological or deleterious consequences. In plants, hormone-responsive transcriptional programs are controlled by nuclear E3 ubiquitin ligases that often function as both hormone receptors and as transcription cofactors. Indeed, E3 ligase-mediated ubiquitination events at or near the chromatin have been reported to regulate the stability of transcription activators and repressors, providing an ON/OFF switch for the control of gene expression. Our latest findings, however, challenge the simplicity of this model and instead demonstrate that dynamic ubiquitination by various ubiquitin-modifying enzymes determines transcription regulator activity. Upon subsequent arrival at the proteasome, transcription regulators undergo further ubiquitin chain remodelling, which determines their final fate. We will discuss how these diverse ubiquitination events may be linked to changes in the chromatin landscape and control hormone-responsive gene expression programmes.

P19.54 THE MPK4-PHOSPHORYLATED HEAT SHOCK FACTOR A4A CONTROLS HEAT AND SALT STRESS TOLERANCE.

WEDNESDAY 11 DECEMBER **11:30**

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Heat shock factors are principal regulators of responses to high temperatures and to other stresses such as salinity, water deprivation or heavy metals. Their function in stress combinations is however not known. The Arabidopsis heat shock factor A4A (HSFA4A) is implicated in salt and oxidative stress tolerance and is a substrate of MAP kinases MPK3 and MPK6. Here we show, that theHSFA4Agene is induced by salt, elevated temperature and combination of these conditions. Fast translocation of HSFA4A-YFP protein from cytosol to nuclei takes place in salt-treated cells. HSFA4A can be phosphorylated not only by MAP kinases MPK3 and MPK6 but also by MPK4 and Ser309 is the dominant MAPK phosphorylation site.In vivophosphorylation data suggest that HSFA4A can be substrate of other kinases as well. Changing Ser309 to Asp or Ala has altered intramolecular multimerisation. Chromatin immunoprecipitation assays could confirm binding of HSFA4A to promoters of various target genes encoding the small heat shock protein HSP17.6A and transcription factors WRKY30 and ZAT12, key regulators of responses to biotic and oxidative stresses. HSFA4A overexpression enhanced tolerance not only to individual stresses but also to simultaneously applied heat and salt stresses through reduction of oxidative damage. Our results suggest that this heat shock factor is a component of a complex stress regulatory pathway, connecting upstream signals mediated by MAP kinases MPK3/6 and MPK4 with transcription regulation of a set of stress-induced target genes. Funding:OTKA NN-110962, NKFI NN-118089 and GINOP 2.3.2-15-2016-00001 grants.

P19.55 EXPLORING THE EFFECT OF MITOCHONDRIAL CHANGES IN THE CHROMATIN REMODELLING MEDIATING PRIMING OF IMMUNE RESPONSE.

WEDNESDAY 11 9TH DECEMBER **11:45**

ANA LOPEZ SANCHEZ (SPANISH NATIONAL CENTRE FOR BIOTECHNOLOGY (CNB-CSIC), SPAIN), CARMEN CASTRESANA (SPANISH NATIONAL CENTRE FOR BIOTECHNOLOGY (CNB-CSIC), SPAIN)

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In the course of an infection plants induce immune responses, which include production of oxylipins and activation of defence signalling. In the case of a recurrent infection, the responses activated are faster and stronger. This is referred as acquired resistance or priming and involves a certain memory of the stress. Different priming processes have been described in terms of durability. When transmitted to the next generations it is known as "transgenerational acquired resistance" (TAR). Epigenetic mechanisms controlling nuclear DNA compaction, such as histone modifications and DNA methylation, play a role in priming. However, the signalling cascade between the pathogen perception and the changes in the epigenetic marks acting as priming fingerprints still elusive.

In this work, we present our results pointing to a role of the mitochondria in this process. We found that mutants altered in synthesis or signalling of 9-LOX-derived oxylipin are impeded in TAR and display differences in DNA methylation at specific nuclear loci. Given that a) oxylipin signalling involves mitochondrial changes, b) oxylipin defective mutants are altered in mitochondrial proteins and c) the production of methyl-groups donors for epigenetic marks partially take place at the mitochondria; we are currently assessing whether the signalling triggered by the pathogen recognition and mediated by oxylipins, induces mitochondrial changes affecting the deposition of epigenetic marks. This work would position chromatin compaction as part of the retrograde signalling, assigning by the first time, a role of the mitochondria as an interface integrating external signals and coordinating the plant responses to environmental changes.

P19.56 EPIGENETICS SIGNATURES TO REMEMBER DROUGHT STRESS IN MAIZE

WEDNESDAY 11 DECEMBER **12:00**

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Plants are exposed to stressful factors that affects their development and reproductive fitness. Dynamic changes in chromatin structure and concomitant transcriptional variations play an important role in stress response and are involved in epigenetic memory mechanisms. Histone marks and gene expression patterns could be stably maintained once the triggering stimulus has been removed.

To understand whether environmental memories are created and propagated, we integrated transcriptome and genome-wide histone modification analyses in maize plants subjected to a mild and prolonged drought stress before flowering transition. Stress was followed by a recovery period to evaluate drought memory mechanisms. We found that the extensive transcriptional and chromatin changes present soon after the stress application were only partially reset after the recovery. Three categories of stress-memory genes were identified: i) "transcriptional memory" genes, with stable transcriptional changes persisting after the recovery; ii) "epigenetic memory candidate" genes in which stress-induced chromatin changes persist longer than the stimulus, in absence of transcriptional changes; iii) "delayed memory" genes, not immediately affected by the stress, but perceiving and storing stress signal for a delayed response. This last memory mechanism is described for the first time in drought response. Applied drought stress altered floral patterning, possibly by affecting expression and chromatin of flowering regulatory genes. Drought stress was also applied to five subsequent generations of maize plants, to assess the transgenerational stress memory. We are evaluating drought stress response and expression of stress-memory genes in the offspring of these plants, which genome was re-sequenced at the sixth generation.

POSTER ABSTRACTS

P19.57 ACTIN DEPOLYMERIZING FACTOR 2 OF A HALOPHYTE SPARTINA ALTERNIFLORA IMPARTS DROUGHT TOLERANCE WHEN CONSTITUTIVELY OVEREXPRESSED IN TOBACCO

MONDAY 9TH DECEMBER

NECLA PEHLIVAN (RECEP TAYYIP ERDOGAN UNIVERSITY, TURKEY), VENKATA MANGU (UNIVERSITY OF PENNSYLVANIA, UNITED STATES), KANNIAH RAJASEKARAN (USDA-ARS, UNITED STATES), NIRANJAN BAISAKH (LOUISIANA STATE UNIVERSITY AGRICULTURAL CENTER, UNITED STATES)

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Current climate fluctuations leading to prolonged drought episodes result in drastic yield losses in tobacco. Thus, breeding new tobacco varieties with minimal yield loss under low water input is of great significance. Here, we report on the physiological and biochemical response of tobacco (Petit Havana) transgenics overexpressing Spartina alterniflora ADF2(SaADF2) under controlled greenhouse conditions. Five independent transgenic lines showed significantly higher tolerance phenotype than non-transgenic controls under drought that was imposed by withholding water for 3 days (D3) and 7 days (D7). The transgenics were characterized by less MDA accumulation but higher MSI and water status (RWC) under drought. The results implicated that SaADF2 has the potential in imparting enhanced drought tolerance in plants via modulation of cytoskeletal architecture.

P19.58 DYNAMICS OF CHANGES IN CHROMATIN FUNCTIONAL ACTIVITY IN PLANTS UNDER DROUGHT STRESS

TUESDAY 10 DECEMBER

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The functional activity of the chromatin of the nucleus is associated with its structural state.Euchromatin DNA is not saturated with histones, has many free groups and metastable easily denatured sites.The predominant part of the DNA is firmly linked to histones and is a component of heterochromatin. To determine the activity of the genetic apparatus in resistant and stress-sensitive plants, it is important to evaluate the functional activity of interphase chromosomes (or DNA gene activity). Therefore, we studied the change in fractional composition, the ratio of labile and stable DNA during drought incotton varieties characterized by varying degrees of resistance to stress.In varieties with high drought resistance, an increase in labile DNA content is noted, the amount of stable DNA decreases.For example, under stress in a drought-resistant sample 5010-V, the increase in the share of labile DNA was 11.1%, the percentage of stable DNA decreased by 8.0%.In samples of cotton, which in stressful conditions are characterized by a decrease in physiological indicators, a decrease in the amount of labile and an increase in stable DNA are observed compared to control plants.DNA transitions from one state to another underlie the regulation gene functions of DNA and morphogenetic processes in the cell. Labile chromatin is mainly associated with metabolic processes occurring in growing cells or in differentiated cells with an active physiological function. It is known that DNA stabilization is promoted by factors inhibiting growth and metabolic processes.

P19.59 CHANGES IN THE FUNCTIONAL ACTIVITY OF DNA CHROMATIN AS A REsULT INHIBITION OF PLANT GROWTH

MONDAY 9TH DECEMBER

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Morphologically, in the chromatin, there is a diffuse part – euchromatin and a compact part – heterochromatin. Growth inhibitors can cause numerous structural and functional changes in plants. Among these changes, a significant role is played by the reaction of the genetic apparatus, on which depends largely on what, ultimately, proteins, with what intensity and sequence will be synthesized by the cell in this situation. As a result, of the research it was found that the maleic hydrazide altered the germination of triticale seeds, wheat and rye, exerting an inhibitory effect at a concentration of 0.6%. In all experimental variants, under the influence of growth inhibition, a decrease in the amount of DNA in a plant cell is observed due to a decrease in labile and stable fractions. Inhibition of growth processes is accompanied by a decrease in the proportion of euchromatin DNA in the nucleus and a decrease in the ratio of the labile fraction of DNA to the stable. For example, in control rye plants, the labile percentage of total DNA was 45%. After 24 hours of growth inhibition, this ratio in experimental plants was 40.2%. The ratio of labile to stable DNA decreased from 0.90 to 0.74%. Thus, under the influence of plant growth inhibition, the proportion of labile to total DNA decreases and the ratio of labile DNA to stable, which probably leads to the suppression of division processes, as a result – to the inhibition of plant growth.

P19.61 DROUGHT STRESS EFFECTS ON THE CHROMATIN-REMODELING FACTOR DDM1 IN WHEAT CULTIVARS WITH CONTRASTING DROUGHT TOLERANCE

MONDAY 9TH DECEMBER

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DNA methylation is a key epigenetic modification that contributes to gene regulation and maintenance of genome integrity via transposable element silencing. DNA methylation affects many aspect of plant development and responses to adverse environment. In addition to multiple methyltransferase enzymes involved in the establishment and maintenance of DNA methylation, the methylation machinery is controlled by the Snf2 family nucleosome-remodeling factor DECREASE IN DNA METHYLATION1 (DDM1). We characterized the effect of dehydration on DDM1 expression in the leaves and roots of wheat cultivars with contrasting drought tolerance after severe dehydration and a subsequent recovery. The cultivars behaving as drought tolerant tend to have a higher expression level of DDM1 in leaves, whereas more susceptible cultivars displayed significantly less abundant DDM1 transcripts. Root expression of DDM1 in all the studied varieties was higher compared to that in the leaves, but also showed cultivar-dependent changes. These results imply for the direct or indirect involvement of DDM1 in the cultivar-specific response to drought stress, probably by modifying chromatin structure.

P19.62 TOWARDS IN VIVO ANALYSIS OF CHROMATIN DYNAMICS IN BARLEY

TUESDAY 10 DECEMBER

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The organization of chromatin in cell nuclei is dynamic and undergoes changes during cell cycle and cell tissue differentiation. This is necessary for correct segregation of genetic information, regulation of gene expression, DNA replication etc. While there is growing information about in vivo dynamics of nuclear domains in plant species with small genomes represented mainly by Arabidopsis thaliana, such data are practically missing in plants with large and complex genomes. We will present our efforts in developing a series of Hordeum vulgare (2n = 2x = 14; 5 Gbp/1C) marker lines carrying fluorescently labelled fusion proteins indicative of specific chromosome and nuclear domains such as centromere, telomere and nucleolus. Production of multi-marker lines will enable comprehensive analysis of chromatin dynamics in both wild type and mutant plants under ambient and stress conditions.

P19.63 LOW RIBOSOMAL RNA GENES COPY NUMBER PROVOKE GENOMIC INSTABILITY AND CHROMOSOMAL SEGMENT DUPLICATION EVENTS THAT MODIFY GLOBAL GENE EXPRESSION AND PLANT-PATHOGEN RESPONSE

MONDAY 9TH DECEMBER

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Among the hundreds to thousands of ribosomal RNA (rRNA) genes copies in the genome present in the nucleolus organizer regions (NORs), only a portion are usually actively expressed and participate in the ribosome biogenesis process in the nucleolus. The role of these extra-copies remains elusive, but previous studies suggest their importance in genome stability and global gene expression. Since the nucleolus was also shown to be an important platform for the 3D genome organization, we tested the impact of having a low amount of rRNA genes copies in theA. thalianagenome using a line only displaying 20% of the copy number found in wild-type (20rDNA line). Although the 3D genome organization and the identity of nucleolus-associated chromatin domains remains similar, we found important signs of genome instability and changes in global gene expression in the 20% rDNA line. Strikingly, we identified using genomic and microscopic approaches up to 7 large duplication events (DEs) from 60 kb to 1,44 Mb. In consequence, more than 500 genes are now duplicated, sometimes provoking changes in their expression

profile. Among them, we found genes implicated in the plant-pathogen responses, whose up-regulation modified the 20rDNA line to resist to both bacterial and nematode infections. Finally, we show that the DEs provoke gene fusions and/or truncations and discuss their potential implication in plant genome evolution.

P19.65 COORDINATION OF CELL WALL INTEGRITY WITH CELL CYCLE PROGRESSION IN ARABIDOPSIS THALIANA

MONDAY 9TH DECEMBER

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Plant cell wall metabolism must adapt to developmental processes like cell elongation and division. This is exemplified by the tightly regulated cellulose deposition during cytokinesis when a new cell wall must be generated to separate the two daughter cells forming. However, processes like the cell cycle are dependent on the functional integrity of the cell wall to allow successful cell cycle progression and completion. This is illustrated by results from Saccharomyces cerevisiae where the cell wall integrity (CWI) maintenance mechanism influences cell cycle regulation progression.

Here we investigated in Arabidopsis thaliana seedlings both how the cell cycle responds to cell wall damage (CWD) impairing CWI and the mode of action of the regulatory mechanism responsible. We found that CWD generated by cellulose biosynthesis inhibition leads to osmo-sensitive inhibition of root growth and cell cycle progression. Genetic analysis showed that none of the known CWI signaling components are required, instead intact NIA1 NIA2, encoding nitrate reductases implicated in NO metabolism, are essential. A phenotypic characterization showed that co-treatments with zeatin could rescue the observed inhibition in a concentration dependent manner. Quantification of cytokinin levels detected CWD-induced, osmo-sensitive changes in certain cytokinins. These results were followed up with additional genetic and gene expression profiling studies suggesting that CWD-induced cytokinin degradation is responsible for the observed effects on cell cycle progression. The available data suggest a NIA1 NIA2-dependent process may be responsible for changes in the levels of certain cytokinins, which in turn seem to regulate cell cycle activity.

P19.66 MOLECULAR CHARACTERIZATION OF THE D53-LIKE SMXL REPRESSION COMPLEXES

TUESDAY 10 DECEMBER

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The control of branch formation is a crucial aspect of plant development and a relevant agronomic trait. This process is tightly regulated by BRANCHED1 (BRC1), a TCP transcription factor that negatively controls shoot branching. BRC1acts as an integrator of different developmental, hormonal and environmental signals. Some of these signals promote BRC1expression, like a low red to far-red (R:FR) light ratio, apical dominance and strigolactone signalling, whereas other repress it, such as a high R:FR light ratio, decapitation, and cytokinin and sugar treatments1–4 . Although some of these regulatory processes begin to be characterized, the molecular mechanisms underlying the transcriptional regulation ofBRC1 remain largely unknown.

In this context, we are studying a group of transcriptional repressors of the strigolactone pathway that repressBRC1expression: the D53-like proteins SMXL. SMXLs are nuclear, bind theBRC1promoter (and others), and negatively regulateBRC1mRNA levels. However, they lack a DNA motif-specific binding domain, which suggests that SMXLs interact with DNA through other cofactors and form part of larger transcriptional repressing complexes. Remarkably, SMXLs form very characteristic speckles in the nucleus, but it is still unknown whether they

correspond to transcriptional repressing complexes. Our aim is to characterize these speckles and the components of the putative complexes to understand their involvement inBRC1regulation.

P19.67 UNCOVERING SMC5/6 COMPLEX DNA DAMAGE REPAIR PATHWAY IN ARABIDOPSIS

MONDAY 9TH DECEMBER

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Structural maintenance of chromosomes (SMC) complexes include cohesin (SMC1/SMC3), condensin (SMC2/SMC4) and SMC5/6. All three are key players involved in chromosome structure and function by modulating chromatin dynamics and genome stability throughout all stages of cell cycle. Canonical functions of SMC5/6 involve multiple roles in maintenance of genome integrity. However, it remains unclear, in which DNA damage repair pathway(s) SMC5/6 complex takes place.

To better uncover SMC5/6 pathway(s) in plants, we set up reverse genetic screen, where mutants in SMC5/6 complex subunitsSMC6B,NSE4AandHPY2were crossed with mutants representing different DNA damage repair pathways, including e.g. homologous recombination, non-homologous end joining or base excision repair. We will report on the double homozygous mutant phenotypes including plant development, sensitivity to DNA damaging agents causing various types of lesions and molecular and cellular markers of DNA damage. Furthermore, we present our efforts to reveal chromosomal localization of SMC5/6 and its role in chromatin dynamics during DNA damage repair process. This will provide novel link between dynamic chromatin organization during DNA damage repair process.

This work was supported from grant nr. CZ.02.1.01/0 .0/0.0/16_019/0000827 and 19-13848S.

P19.68 INSIGHT INTO SPATIAL DISTRIBUTION OF NUCLEAR PROTEIN INTERACTIONS

TUESDAY 10 DECEMBER

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Plant nucleus and its subdomains represent fascinating cellular entities that open many scientific questions, further driving the development of innovative approaches. In our work we focus on the distribution of histones and ribosomal genes inside the nucleolus and the changes in the chromatin structure caused by histone chaperone gene mutations. Mutual interactions of histone chaperones and their histone binding specificityin vivohave not been fully understood in plants. We focus on the H3/H4 chaperones - Chromatin Assembly Factor 1 (CAF-1) and H2A/H2B chaperones NUCLEOSOME ASSEMBLY PROTEIN 1 (NAP1), and NAP1-RELATED PROTEIN 1 and 2 (NRP1, NRP2) and determine their interaction potential in plants using yeast two hybrid system or Bimolecular Fluorescence Complementation assay (BiFC). We optimized this method for use in protoplasts isolated fromArabidopsisseedlings. We will also present alternative approaches suitable to study protein-protein interactions, such as Fluorescence Lifetime Imaging and the use of Super-resolution microscopy on isolated nucleoli, facilitating studies of epigenetic profile inside the nucleolus.

This work was supported by GACR 19-11880Y, INTER-COST LTC18048

P19.69 REAL-TIME IMAGING OF DNA METHYLATION USING DYNAMET SENSORS

MONDAY 9TH DECEMBER

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DNA methylation is a well-studied epigenetic mark in many organisms including plants, but remains difficult to study dynamically with available tools, particularly for individual cells. This generates important limitations when studying reproductive cells, which undergo very dramatic and rapid epigenetic changes during gametogenesis, fertilization, and subsequently early embryo development. We recently developed genetically encoded fluorescent reporters of DNA methylation called DYNAMET in the model plant Arabidopsis (Ingouff et al, 2017). They can be used to follow for the CG and CHH methylation patterns in poorly accessible cells in real time, with very high temporal resolution. Here we will present an expanded set of DYNAMETs that are more sensitive and a new reporter for CG hemi-methylation.

P19.70 DISCOVERING TRANS-INTERACTIONS INVOLVED IN PARAMUTATION USING CIRCULAR CHROMOSOME CONFORMATION CAPTURE

TUESDAY 10 DECEMBER

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Reprogramming of epigenetic information has been described in both plants and mammals. Paramutation is a rare exception where reprogramming is both mitotically and meiotically stable over many generations. An example is located at the booster1 gene (b1) in Zea mays, where the weakly expressed Booster' (B') allele stably decreases the expression of the Booster-Intense (B-I) allele, and changes it into a new B' allele. These two alleles are genetically identical, and only differ by the chromatin status of 7 tandem repeats inserted 100 kb upstream of the b1 gene (called b1-TR). Previous work using 3C technology (Chromosome Conformation Capture) has shown that b1-TRs are involved in the formation of loops with different loci upstream of b1. These loops are tissue-specific and differ depending on the epiallele studied (B' or B-I). These changes in cis-interactions are potential regulators of b1 expression in both B' and B-I alleles. To further answer this question, we are currently developing a protocol to identify trans-interactions that are associated with the B' and B-I alleles in maize. Circular Chromosome Conformation Capture (4C) experiments will be held on leaves from plants that are either capable or not capable of paramutation. We aim to find consistent patterns in the detected trans-interactions, and highlight those that are candidates to regulate paramutations.

P19.71 DEFENSIN EXPRESSION IN SUNFLOWER UNDERCOMBINED BROOMRAPE – DOWNY MILDEW ATTACK

MONDAY 9TH DECEMBER

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• Through the evolution cultivated sunflower (Helianthus annuus L.) has developed a wide range

of pathogen defense mechanisms. In the last few decades, the pests that coexist with this crop went through fast evolution as a result of its massive commercial exploitation and use of different protection methods. Great efforts are being made to produce new genotypes that are resistant, or at least tolerant to new, aggressive pathogen races.

• Previous research that has been done on H. annuus was mainly focused on studying the model of separate diseases effect on this crop and rarely to the dualistic effect of different biotic factors. The aim of our study was to explore the possibility of sunflower immune response caused by one disease to defend the plant from another.

• In our experiment we were focused on two widespread sunflower pests Plasmopara halstedii and Orobanche cumana. P. halstedii-resistant genotype was inoculated with the pathogen and planted into O. cumana-inoculated soil. Defensin expression was analyzed using RT-PCR in leaf samples taken at different stages from plants with combined broomrape-downy mildew infection, plants only infected with broomrape, and control plants.

P19.72 THE CONNECTION BETWEEN THE NUCLEOPORINS SAR1/3 AND MEIOSIS: A WAY TO DISCOVER CHROMOSOME DYNAMICS

TUESDAY 10 DECEMBER

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The Nuclear Envelope (NE) entails a barrier between the cytoplasm and the nucleus. While clearly essential in maintenance of nuclear integrity, the NE is a highly dynamic organelle. The integrity of the NE relies on different complexes, among which are Nuclear Pore Complexes (NPCs). These complexes regulate the macromolecule transport, and physically interact with chromatin and the transcriptional machinery. It is well established that the NE undergoes a dramatic breakdown and reformation during plant cell division. In addition, this structure has a specific function in meiotic prophase I as it anchors and positions telomeres and contributes to facilitate the pairing between homologous chromosomes. We hypothesise that NPCs could be involved in telomere attachment to the NE. To highlight a potential function of the structural components of the NPCs in meiosis, we have isolated several Arabidopsis lines with mutations in genes coding for nucleoporins. Plants defective for either SUPPRESSOR OF AUXIN RESISTANCE1 (SAR1) or SAR3 show pleiotropic growth and developmental defects and exhibit reduced fertility. The cytological characterisation of pollen mother cells (PMCs) has revealed condensation abnormalities in around 15% of prophase I meiocytes. Chromosome fragmentation is also present in some cells at second meiotic division. To further analyse this meiotic phenotype, we have obtained different double mutants to describe possible genetic interactions and functional relationships with known meiotic proteins. Additionally, we have detected an abnormal distribution of NE proteins in these mutants. These experiments could provide new insights into the role of plant nucleoporins in meiotic chromosome behaviour and fertility.

P19.73 CHROMOSOME AND NUCLEUS ORGANIZATION DURING MEIOTIC RECOMBINATION IN BARLEY

MONDAY 9TH DECEMBER

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In cereals, such as wheat and barley, crossing overs (CO) are distributed mainly at the end of chromosomes so that centromeric and pericentromeric regions including up to 30% of the genes rarely, if ever, recombine, preventing the generation of novel gene combinations and useful variation that could be exploited in breeding programmes. Therefore an ability to modify the pattern of recombination in these species would have a profound impact on the breeding of these crops. In order to investigate means of altering the patterns of CO in barley, we have utilised SNP technology and cytological procedures, to investigate a collection non-allelic desynaptic barley mutants. All the desynaptic mutants exhibited perturbed meiosis and semi-sterility compared to wild type with some exhibiting unexpected phenotypes during synapsis (Colas et al, 2016) as

visualized through the use of 3D structure illumination microscopy (OMX). Our studies have suggested a tighter control of meiotic progression of chromatin/meiotic axes compared to the model Arabidopsis but also chromatin modification as suggested by our latest identified Desynaptic mutant. A mutation in HvST1, a novel grass specific E3 ubiquitin ligase, affects meiosis progression and crossover formation. But instead of being detrimental to recombination, knocking down HvST1 results in an increase recombination despite changes in chromatin organization. Here we discuss the potential relationship between chromatin and recombination and further use for plant breeding improvement.

P19.74 GARNET COMMUNITY NETWORK: SUPPORTING DISCOVERY-LED PLANT SCIENCE IN THE UK AND BEYOND

TUESDAY 10 DECEMBER

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GARNet is a UKRI-BBSRC funded research network that supports the UK discovery-led plant science community. We aim to ensure that UK plant science remains competitive and productive at the national and international level by helping researchers make the best use of available funding, tools and resources.

This is achieved in a number of important ways:

- Organisation of meetings and workshops that bring together plant scientists in order to encourage knowledge exchange and collaboration. GARNet will bring the International Conference on Arabidopsis Research (ICAR) to Belfast in the UK in 2021
- Disseminating information about conferences, published material and grant funding in order to keep the plant science community up to date with the state of the art
- Promoting UK excellence amongst global research communities such as COST Actions (INDEPTH, PlantEd) and the Multinational Arabidopsis Research Community (MASC).

We will highlight current GARNet activities as well as introduce our plans for the next grant period.

For more information:

www.garnetcommunity.org.uk

blog.garnetcommunity.org.uk

arabidopsisresearch.org/

https://www.brookes.ac.uk/indepth/

https://plantgenomeediting.eu/

http://icar2021.arabidopsisresearch.org/

P19.75 DISCOVERY OF A UNIQUE BARLEY SPIKE PHENOTYPE AND INSIGHTS ON SOME ASSOCIATED SNP VARIATION

MONDAY 9TH DECEMBER

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During the barley breeding activities at the Agricultural Research Institute in the 2010-2011 growing season, we discovered in the field a novel spike phenotype not previously described in the literature. This unique phenotype consisted of one or more additional minor spikes growing at the base of the main spike and appeared only in certain plants of one specific line among the multiple tested. In order to characterize an apparent potential genetic base for the phenotype, we proceeded with additional field trials spanning a period of 7 years, where the unstable, but persistent, nature of the phenomenon was confirmed, along with an apparent environmental influence. During these trials, hundreds of individual plants and sibling lines were phenotyped for the mutation and also precisely characterized in terms of yield and stability according to the standards of the Honeycomb Selection Designs and the associated Prognostic breeding paradigm. Taking advantage of the existing barley genomic resources, we are using the newly developed Barley 50K SNP chip to characterize some selected lines. The bioinformatics pipelines used to analyse and interpret the data as well as some first preliminary analysis results will also be presented.

P19.76 FUNCTIONAL ANALYSIS OF BRM CHROMATIN REMODELER USING CATALYTIC POINT MUTATIONS.

TUESDAY 10 DECEMBER

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SWI/SNF remodeling complex is an crucial component of chromatin structure which can influence its functional role. It can regulate transcription by changes in nucleosome occupancy and chromatin accessibility.

BRM (BRAHMA) is a core ATPase subunit of Arabidopsis SWI/SNF chromatin remodeling complex, known to be involved in regulation of plant growth and development, as well as stress response and hormone signaling.

As BRM is a chromatin remodeler, it is thought to act by moving nucleosomes using the energy from ATP hydrolysis. Indeed, we showed that transcription from a BRM-targeted promoter requires ATPase activity of BRM, as point catalytic BRM mutant is unable to efficiently activate transcription in a transient transactivation system. Here we show more detailed analysis of the role of BRM catalytic domain in transcription regulation. We generated/created a model of BRM ATPase domain by homology modeling and predicted amino acid positions both within and outside ATP-binding pocket that might be important for BRM catalytic activity.

We introduced a set of point mutations into BRM coding sequence and applied transient transactivation system, FAST (FastAgro-mediatedSeedlingTransformation), both in Tabaco and Arabidopsis, to analyze transient activation of SCL3 promoter, which is known to be directly controlled by BRM.

Importantly, our results provide new insights into the mechanism through which BRM as being chromatin remodeler regulates transcription.

P19.77 INVESTIGATION OF DROUGHT STRESS EFFECTS ON SUGAR BEET IN SERBIA

MONDAY 9TH DECEMBER

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Drought is the prime abiotic factor that limits sugar beet (Beta vulgaris L.) production in Serbia and other regions where the crop is not usually irrigated. As increased irrigation is not an economically viable solution, the most effective one is development of varieties adapted for successful growth in drought-prone environments. Within the framework of ongoing projects, in Institute of Field and Vegetable Crops, Novi Sad was performed research with aim to select drought tolerant sugar beet genotypes, improve production under water deficit conditions, and clarify the physiological processes of drought tolerance in sugar beet. Genotypic diversity for drought-related tolerance indices were assessed in the field trials and the strength of association between them and crop performance was measured. As the drought tolerance is a complex trait, very difficult to evaluate in the field, the study was also conducted through the greenhouse experiments and in vitro screening. The plant material was studied for morphological and physiological parameters of water regime and the expression of genes that are known to respond to osmotic stress. There are reasons to believe that fullerenol ability to form hydrogen bonds with water molecules makes this nanoparticle a potential intracellular water depot, which can be used if osmotic stress occurs. In collaboration with colleagues from the Faculty of Science, Novi Sad we have started to analyze the influence of fullerenol on sugar beet plants exposed to drought stress. Results indicate that application of fullerenol can modified intracellular water metabolism and enabling adaptation of plants to drought stress.

P19.78 DECIPHERING HISTONE MODIFICATIONS IN RICE TO UNDERSTAND THE OSRMC ACTIVATION BY SALT STRESS

TUESDAY 10 DECEMBER

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Climate change is increasing soil salinity which is a major environmental constrain for crops with negative impact on plant growth, development and sustainable production levels. Rice cultivation is particularly sensitive to salinity stress and this stress has been associated with a concerted flexibility of chromatin structure, DNA methylation, histone modifications marks and gene expression. A more decondensed interphase chromatin landscape under salinity conditions was revealed by in situ immunodetection with specific histone modification antibodies. The regulation of OsRMC, a gene highly induced by salt stress, is likely to involve epigenetic modulation since its expression was increased when seedlings were treated with the hypomethylating drug 5-azacytidine or when epigenetic backgrounds were modified due to the presence of specific mutations for epigenetic regulators as it is the case of the rice mutant lines oshac704 (lacking one acethyltransferase) and the osdrm2 (a knockout for a DNA methyltransferase). To further investigate the epigenetic regulation of OsRMCgene, chromatin immunoprecipitation (ChIP) was performed in leaf tissues of young rice seedlings using specific histone modification marks namely, H3K9ac, H4K5ac and H4K20me3. These assays allowed deciphering the pattern of specific histone modification and unvealed a differential enrichment of euchromatin marks at distinct chromatin domains along OsRMC promoter. These results indicated the involvement of nucleosome repositioning in the activation of OsRMC under salt stress.

P19.79 DEFENSIN EXPRESSION IN SUNFLOWER UNDERCOMBINED BROOMRAPE – DOWNY MILDEW ATTACK

MONDAY 9TH DECEMBER

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- Through the evolution cultivated sunflower (Helianthus annuusL.) has developed a wide range of pathogen defense mechanisms. In the last few decades, the pests that coexist with this crop went through fast evolution as a result of its massive commercial exploitation and use of different protection methods. Great efforts are being made to produce new genotypes that are resistant, or at least tolerant to new, aggressive pathogen races.
- Previous research that has been done on H. annuus was mainly focused on studying the model of separate diseases effect on this crop and rarely to the dualistic effect of different biotic factors. The aim of our study was to explore the possibility of sunflower immune response caused by one disease to defend the plant from another.
- In our experiment we were focused on two widespread sunflower pestsPlasmopara halstediiandOrobanchecumana.P. halstedii-resistant genotype was inoculated with the pathogen and planted into O. cumana-inoculated soil. Defensin expression was analyzed using RT-PCR in leaf samples taken at different stages from plants with combined broomrape-downy mildew infection, plants only infected with broomrape, and control plants.

P19.80 FASCIATA1 IS REQUIRED FOR DE-NOVO SHOOT REGENERATION IN ARABIDOPSIS THALIANA

TUESDAY 10 DECEMBER

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Tissue culture is not only important for agriculture to enable vegetative propagation of explants but also is widely used by plant breeders as a method of transgene introduction. Plants can regenerate in vitro via indirect organogenesis, which is a two-step process that consists of incubating a small piece of plant tissue (explant) on callus induction medium (CIM), where the explant will generate a mass of competent cell called callus. This step can be followed by incubation on shoot induction medium (SIM). Here the callus will acquire the competency needed to respond to shoot induction signals, and thus regenerate de-novo shoots.

However, de-novo shoot regeneration pathway is far away from being fully understood. With the aim to identify key players in this process, we performed a reverse genetic screen in Arabidopsis thaliana. From this screen, we identified FASCIATA1 (FAS1), a subunit of the histone chaperone Chromatin Assembly Factor 1 (CAF-1) complex. fas1 mutants display an extremely impaired shoot regeneration phenotype in tissue culture of Arabidopsis. This phenotype is accompanied by downregulation of several genes important for pluripotency acquisition and strong upregulation of genes related to DNA methylation. We will discuss FAS1-mediated gene regulation mechanism required for de-novo shoot regeneration.

P19.81 APPROACHING A DWARFING PHENOTYPE IN WHEAT BY EXPLORING THE EPIGENOME

MONDAY 9TH DECEMBER

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Genetic manipulation of Gibberellin (GA) metabolic pathway is a successfully used method to improve agricultural features of crops. Previously generated semidwarf durum wheat cultivar Icaro has been characterized as a GA-insensitive mutant with increased degradation of GA precursor caused by significant overexpression of GA-2 oxidase. As the nucleotide sequences of the predicted ORF of GA 2-oxidase were identical in the mutant Icaro and the original tall durum wheat variety Anhinga, we hypothesize that increased gene expression of this enzyme could result from disrupted transcriptional regulation involving non-coding DNA and/or DNA methylation.

Common methods to assess the regulatory landscape of a locus involve single chromosome pool methylome and local histone modification profiling as well as revealing long-range interactions between promoters and cis-regulatory DNA by 4C. Comparison of the epigenomic profiles of the GA-2 oxidase loci in the mutant semidwarf Icaro and tall Anhinga wheat varieties could help to decipher the causative variation of chromatin organization.

P19.82 INHIBITION OF JASMONIC ACID BIOSYNTHESIS INDUCED POWDERY MILDEW RESISTANCE IN WHEAT

TUESDAY 10 DECEMBER

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Jasmonic acid (JA) is an important plant hormone associated with plant pathogen defense. To study the role of JA in plant-fungal interactions, we applied an inhibitor of JA biosynthesis, sodium diethyldithiocarbamate (DIECA), on wheat leaves. Our results indicated that resistance to wheat powdery mildew was induced when DIECA treatment was applied prior to powdery mildew infection. Transcriptome analysis identified 364 up-regulated and 68 down-regulated differentially expressed genes (DEGs) in DIECA-treated leaves. Gene ontology)GO) enrichment analysis of those DEGs revealed important GO terms and pathways, such as response to growth hormones, activity of glutathione metabolism (e.g., glutathione transferase activity), oxalate oxidase, and chitinase activity. Furthermore,pathogenesis-related(PR) genes, such asPR1.1,PR1,PR10,PR4a,Chitinase 8,beta-1,3-glucanase,RPM1,RGA2, andHSP70were included among the up-regulated DEGs by DIECA. Most of these transcriptional changes have been previously reported in associations with plant-fungi interaction and in ROS response. Quantification of plant hormones revealed that in addition to the inhibition of JA biosynthesis, the level of Auxin (IAA) was decreased and Brassinosteroid (BR) was increased. Moreover, glutathioneamount was elevated and reactive oxygen species (ROS) lesions were observed. Our results indicate that inhibition of JA biosynthesis triggered hormonal crosstalk and transcriptional reprogramming, leading to an induced resistance to powdery mildew. This approach can be applied to increase plant immunity and reduce the severity of powdery mildew disease in wheat fields.

P19.83 DEVELOPMENT AND COMPARISON OF SEGMENTATION METHODS FOR THE ANALYSIS OF THE PLANT'S 3D NUCLEUS

MONDAY 9TH DECEMBER

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The nucleus is a compartmentalized organelle containing distinct nuclear domains. To link nuclear structure and function, we are developing an ImageJ plugin to quantify in 3D the nuclear morphology as well as positioning and organization of nuclear domains. This plugin named NucleusJ (Desset et al., 2018; Poulet et al., 2015), applies for a batch of images a modified Otsu thresholding method to segment the nucleus and a 3D watershed algorithm to delimit chromatin domains by partitioning the nucleus.

However, a first limitation of the current version is the time needed to delimit each bounding volume including the considered nucleus. To overcome this manual step and automatically capture large numbers of nuclei at various depths in the original wide field image, we implemented an automatic detection of the nucleus (autocrop) as a starting point of our workflow. Spatial positions of the cropped nuclei are then recorded in order to estimate distance maps as a new estimator of spatial distribution of the nuclei in a whole tissue context. A second limitation is the segmentation of the nucleus from the background which in few cases escaped from our Otsu segmentation procedure. To this aim we developed a 3D gift-wrapping method to define the final boundary. Finally, these new functionalities were applied to generate annotated datasets subsequently used to train a neural network based on the U-net architecture.

We will provide a comparative analysis of these new tools and their potential to decipher the function of nuclear envelope-associated proteins in a big data context.

P19.84 EPIGENETIC RESPONSES TO DROUGHT STRESS AND RECOVERY PERIOD IN OLEA EUROPAEAL.

TUESDAY 10 DECEMBER

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Epigenetic regulation is a significant mechanism that is implicated in a wide range of biological processes, such as gene expression, genome stability, developmental regulation, and diseases In plants, the alteration of histone modification and DNA methylation are mediated with changes in the stress-responsive gene expression to adapt to environmental changes. Under abiotic stress conditions, several chromatin regulators have been reported to be involved in the regulation of stress-responsive gene networks. However, the entire correlation network between abiotic stress responses and epigenetic information, such as the targeted stress-responsive genes, the stress-responsive epigenetic modifiers, and the specific histone modification sites remain unclear.

Drought stress can severally damage plant growth and the most important factor in the reduction of yield. The slow-growing character of olive trees make field trials costly and time-consuming. Therefore, it is beneficial to take advantage of those morphological and physiological traits relevant to drought tolerance to help the breeding and selection process.

In order to understand the epigenetic mechanism to drought stress and recovery period, RNA-Seq and ChIP-seq analysis will be performed in this research. For the evaluation of the agronomic traits of different olive genotypes under drought- stressed and control conditions, some of physiological and biochemical responses (lipid peroxidation, H2 O2 content and antioxidant enzyme activity) were examined. Selected genotypes showing different stress responses (two drought-tolerant and one susceptible) were used for RNA-seq experiments. This work will identfy differential gene expression and altered plant phenotypes upon exposure to drought stress and recovery period to illuminate the role of nuclear domains and hence epigenetic regulation and epigenetic memory of the stress in strategic plant olive.

P19.85 CORK OAK YOUNG AND TRAUMATIC PERIDERMS SHOW PCD TYPICAL CHROMATIN PATTERNS BUT DIFFERENT CHROMATIN-MODIFYING GENES EXPRESSION

MONDAY 9TH DECEMBER

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Tree stems are enveloped by a periderm made of cork cells, resulting from the activity of the phellogen, fundamental to their survival. DNA methylation and posttranslational histone modifications have important roles in the regulation of plant cell differentiation. However, studies on its involvement in cork differentiation are scarce despite periderm importance. Cork oak periderm was used as a model to study the formation of secondary protective tissues. Nuclei structural changes, dynamics of DNA methylation, and histone modifications were assessed in young and traumatic periderms formed after cork harvesting. Lenticular phellogen producing atypical non-suberized cells was also studied, due to high impact for cork industrial uses. Immunolocalization of active and repressive marks, transcription analysis of the corresponding genes, and correlations between gene expression and cork porosity were investigated. Cork cells differentiation was accompanied by drastic chromatin remodelling, evidenced by chromatin condensation and accumulation at the nuclear periphery. Alongside, progressive increase in DNA methylation and constant levels of H3K4me3 and H3K18ac gene activation associated marks were observed. Lenticular cells nuclei were highly fragmented with faint 5-mC labelling. Distinct gene expression patterns in young and traumatic periderms suggest that cork differentiation might be under specific silencing regulatory pathways. Significant correlations were found between QsMET1, QsMET2, and QsSUVH4 gene expression and cork porosity. This work provides

the first insights into chromatin dynamics during cork and lenticular cells differentiation pointing to a distinct type of remodelling associated with cell death, and evidences that DNA methylation and histone modifications play a role in periderm formation.

P19.86 POPULATION GENETIC AND EPIGENETIC VARIABILITY AND DISTRIBUTION OF THE ENDANGERED GREEK ENDEMICCICER GRAECUMUNDER CLIMATE CHANGE SCENARIOS

TUESDAY 10 DECEMBER

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Crop wild relatives (CWR) are an incredible resource for crop improvement due to their high level of genetic diversity. The Mediterranean hot-spot includes numerous endemic plant species, seriously threatened by climate change and habitat loss. On the other hand, genetic and epigenetic variation in natural populations with different habitats might be an important component, towards plant adaptation to environmental stress. In this study, the genetic and epigenetic diversity of five populations ofCicer graecum, an endangered endemic species from Northern Peloponnese, Greece, wild relative of the cultivatedC. arietinum, was investigated using ISSR, AFLP and methylation-sensitive AFLP (MSAP) markers. Nei's gene diversity (GD) for ISSR markers ranged from 0.191 to 0.228 and for AFLP markers from 0.120-0.148, indicating medium to high genetic diversity at the population level despite of their small size. The ecological adaptation ofC. graecumpopulations was also investigated by correlation of their genetic diversity with certain environmental variables. Aridity arose as the dominant factor positively affecting the genetic diversity ofC. graecumpopulations. Furthermore, we used species distribution modeling in order to predict present habitat suitability for the species, using distribution data and several environmental variables. The possible effects of climate change in habitat suitability were calculated according to different climate change scenarios. Our results forecast that the long term survival of this species in its natural habitat is especially uncertain.These findings were evaluated towards developing an effective management and conservation scheme forC. graecumand exploiting the most promising populations in future breeding programs.

P19.87 QUANTIFICATION OF MORPHOLOGICAL FEATURES IN INTERPHASE NUCLEI OF ARABIDOPSIS THALIANA

MONDAY 9TH DECEMBER

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Arabidopsis is a powerful system to study chromatin domains and chromatin dynamics inside the nucleus. With standard fluorescence microscopy and a DNA stain it is already possible to analyze morphological parameters, to quantify nuclear phenotypes, to determine cell-specific states and species-specific dynamics of heterochromatin and to identify genetic factors of nuclear morphology. We performed a detailed morphometric analysis of nuclei from different cell types, tissues and organs in several accessions. Epidermal nuclei are significantly different from parenchyma and vascular nuclei. They have the highest DNA density and the highest heterochromatin fraction (RHF = 0.19). In epidermal cells a significant percentage (~5.4%) of gene regions resides in heterochromatic domains (chromocenters), whereas in other cell types this fraction is practically zero. Arabidopsis accessions differ significantly in heterochromatin fraction, chromatin dynamics and chromocenter association. The accession Cvi has many (~93.7%) transposons outside chromocenters. Significantly more association of NOR chromocenters was found in Cvi (45%) and Ws (43%) compared to Col (27%), Ler (17%) or C24 (23%). This feature corresponds to the low rDNA copy number and low rDNA methylation in Cvi and Ws. The results further suggest that Ws and C24 have more parenchyma

cells in G2 phase than the other accessions. The Ler nuclear phenotype is more robust, whereas in Cvi the organization is more variable, possibly due to light stress. QTL analysis indicates the photoreceptor PhyB as an important factor in nuclear morphology. Hence, basic quantitative microscopic analysis can reveal detailed information of nuclear organization.

P19.88 PHYLOGENETIC ANALYSIS OF THE TANDEM KINASE-PSEUDOKINASE (TKP) PROTEIN FAMILY INVOLVED IN PLANT IMMUNITY

TUESDAY 10 DECEMBER

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Yellow rust, caused by Puccinia striiformis f. sp. tritici (Pst), is a devastating fungal disease threatening much of global wheat production. Yr15 is a broad-spectrum resistance gene derived from wild emmer wheat (WEW), Triticum dicoccoides, which encodes a putative kinase-pseudokinase protein, designated as Wheat Tandem Kinase 1 (WTK1), comprising a unique R-gene structure in wheat (Klymiuk et al. Nature Communications 2018). WTK1 orthologs and paralogs are found in all group 1 and 6 wheat chromosomes. The exon-intron structure of WTK1 orthologues copies is similar to that of WEW functional copy, but differ in numerous SNPs and indels that cause changes in reading frames. The unique protein architecture, similar to WTK1, was found also in 92 putative proteins across the plant kingdom, including the barley RPG1 and a candidate forUg8, suggesting that they are members of a distinct family of plant proteins, termed here tandem kinase-pseudokinases (TKPs). We found that 175 out of 184 kinase/pseudokinase domains of these TKPs are associated with receptor-like kinases (RLKs), indicating that TKPs are involved in plant defense mechanisms. Further phylogenetic analysis indicated that TKP family members originated from either gene duplication or gene fusion events, implying a polyphyletic origin of the TKPs. The decoy role of pseudokinase domain can be proposed as one of the potential mechanisms of function of the TKP family members in immune response. However, further studies are required in various plant species in order to elucidate the mechanism of resistance conferred by this unique protein family.

P19.89 SPATIOTEMPORAL DISTRIBUTION OF PLOIDY LEVELS AND PLOIDY SPECIFIC TRANSCRIPTOME DURING TOMATO FRUIT DEVELOPMENT

MONDAY 9TH DECEMBER

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Endoreduplication, a process during which nuclear DNA content is increased through successive genome duplication without cell division, plays pivotal functions throughout the plant life cycle and in response to environmental stresses. A potential effect of endoreduplication is that, by increasing gene copy number, transcription could be increased. In Tomato, the fruit pericarp tissue (fleshy part) is composed of a heterogeneous population of cells displaying a large variety of ploidy levels reaching up to 256C. These high ploidy levels are generally correlated with large cells. However, little is known about the onset and progression of endoreduplication during tomato fruit growth and its consequences on the regulation of cell size and gene expression. We, therefore, aim to determine the in situ distribution of gene expression based on the ploidy levels in the pericarp during fruit development. For that, ploidy distribution in the pericarp is first quantified in situ by Fluorescent in situ Hybridization. In parallel, cell size is measured to study the potential link between ploidy and cell growth. Second, RNA extracted from nuclei sorted based on their ploidy level is used for sequencing. From this transcriptome data, a search for potential markers of ploidy and/or genes having a ploidy specific expression will be done. These ploidy distributions and transcriptomics experiments are done by harvesting fruits at five stages from 3 to 17 days post-anthesis during fruit growth. The ultimate step will be to generate a virtual map showing the distribution of ploidy, cell size and gene expression in the pericarp.

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