40th New Phytologist Symposium

Plant epigenetics: from mechanisms to ecological relevance

12 – 15 September 2017
Vienna, Austria

New Phytologist

Programme, abstracts and participants

40th New Phytologist Symposium

Plant epigenetics: from mechanisms to ecological relevance

Department of Botany and Biodiversity Research, University of Vienna, Vienna, Austria
12–15 September 2017

Scientific Organizing Committee

Conchita Alonso *(Spanish National Research Council, Spain)*
Oliver Bossdorf *(University of Tübingen, Germany)*
Katrin Heer *(Philipps-Universität Marburg, Germany)*
Marie Mirouze *(Institute of Research for Development, France)*
Lars Opgenoorth *(Philipps-Universität Marburg, Germany)*
Ovidiu Paun *(University of Vienna, Austria*)
Christina Richards *(University of South Florida, USA)*
Koen Verhoeven *(Netherlands Institute of Ecology, The Netherlands)*

New Phytologist Organization

Helen Pinfield-Wells *(Deputy Managing Editor)*
Mike Whitfield *(Development Co-ordinator)*
Sarah Lennon *(Managing Editor)*
Jill Brooke *(Journal & Finance Administrator)*

Acknowledgements

The 40th New Phytologist Symposium is funded by the New Phytologist Trust
www.newphytologist.org

The New Phytologist Trust is a non-profit-making organization dedicated to the promotion of plant science. It owns and produces the international journal *New Phytologist*. The Trust receives income through subscriptions to *New Phytologist* and any excess revenue from the publication of the journal is put straight back into supporting plant science. This is achieved by funding a wide range of activities: the organisation and sponsorship of symposia, workshops and meetings; numerous grant schemes; sponsorship of various awards for early-stage career scientists including the Tansley Medal; and ensuring that research published in the journal is as widely and openly available as possible, as such, all of our Tansley series reviews and Forum articles are immediately available to access free of charge upon publication. All of these actions have a common goal to promote emerging areas of plant science and to encourage continued progress and innovation in the field.

Programme, abstracts and participant list compiled by Jill Brooke
'Plant epigenetics: from mechanisms to ecological relevance' logo by
A.P.P.S., Lancaster, UK

Contact email: *np-symposia@lancaster.ac.uk*

Table of contents

Information for Delegates

Symposium location
The 40th New Phytologist Symposium will be held at the Department of Botany and Biodiversity Research, Rennweg 14, A-1030 Vienna.
http://www.botanik.univie.ac.at/botanik/index.php?nav=h72

All presentations will be given in the Lecture Hall on the ground floor and posters will be displayed in the Cold Glasshouse of the Vienna Botanical Garden.

Catering
Breaks: coffee/tea will be served from several points during the breaks; outside the Lecture Hall and towards the Botanical Garden.
Lunches on Tuesday, Wednesday and Thursday will be in the Cold Glasshouse of the Vienna Botanical Garden. On Friday a packed lunch will be provided to all delegates.

If you have special dietary requirements please do make yourself known to the catering staff or ask Helen/Mike from *New Phytologist*. All our requirements have been provided to the catering team and they will have meals prepared accordingly.

Tram Tour and Symposium dinner
The Symposium dinner will be held on Thursday evening at Heuriger Schübel – Auer
http://www.schuebel-auer.at/
All delegates will be transported to Heuriger Schübel – Auer on a historic tram tour http://www.rentabim.at/english/english.html. Trams will depart at 19:00 from Rennweg, Line 71 direction Börse. There will be two trams (1 with 2 cars; 1 with 3 cars); please fill all seats in the cars of the first tram. Our tour will end at Nussdorf and we will walk to Heuriger Schübel – Auer arriving approx. 20:00 for dinner.

Buses will leave Heuriger Schübel – Auer at 23:00 and return to the Department of Botany and Biodiversity Research, Rennweg 14 (approx. 20 minutes by bus).

The symposium dinner and tram tour is included for all delegates attending the 40th NPS. If you are not able to attend the tram tour or dinner please do let us know.

Botanical Garden Tour

There is a tour on Thursday evening 17:00–18:30 of the Botanical Garden. There is no additional charge for this but we have asked you to sign up by email so we know how many guides we require for this. If you wish to attend, but have not signed up, please speak to Helen or Mike from the *New Phytologist* (np-symposia@lancaster.ac.uk). Visit http://www.botanik.univie.ac.at/hbv/index.php?nav=71 for more information about the Botanical Garden.

Posters

Posters should be prepared so that they are no larger than A0 size, portrait orientation (118 cm high x 84 cm wide). Posters should be put up during registration (Tuesday 12 September) and will be displayed for the duration of the meeting. Delegates are welcome to view posters during coffee and lunch breaks; there will also be dedicated poster sessions from 18:00–20:00 on Tuesday 12 September and 17:25–19:30 on Wednesday 13 September. Please stand by your poster for part of these sessions (we appreciate as poster presenters you will also want to view and discuss the other posters). Please note there will be prizes for the best posters. Drinks and snacks will be served throughout the poster session.

Prizes Posters will be assessed by your peers (the other delegates) and the posters that gain the most votes will receive prizes. A scoring sheet is included in your delegate pack. Please fill out and return this sheet to the registration desk by 14:00 on Thursday 14 September.

Internet access

The Eduroam wireless service is available across campus and will allow delegates from participating organisations to access the wireless using their home institution's credentials. For more information please visit https://zid.univie.ac.at/en/wi-fi/

Social media

We encourage all attendees to join in discussions on social media sites. Follow @NewPhyt on Twitter and fb.com/NewPhytologist on Facebook for updates before, during and after the meeting. Please use the hashtag #40NPS in all of your tweets.

Getting around

Vienna is a well connected city with an extensive public transport network and can be easily explored by foot or bicycle. The Conference Venue is located in the heart of Vienna and is reachable by tram and metro, making it a convenient location to get to.

Click here to plan your journey:
https://www.wienerlinien.at/eportal3/ep/channelView.do/channelId/-46649?routeFrom=&routeTo=&routeDatetime=now&immediate=false&deparr=Abfahrt

Touristic activities
For more information on sightseeing, shopping, wining & dining, practical tips, including opening times (e.g. museums, shops, banks, post offices) emergency phone numbers, WiFi spots etc. click here: https://www.wien.info/en/travel-info/tourist-info

Photography
Photography will take place at the 40th NPS.
The resulting photographs will be used by the New Phytologist Trust for the purpose of promoting its activities, and may be published on the New Phytologist Trust's website and social media channels.
If you do not wish to appear in the photographs, please speak to one of the organisers.

Code of conduct
The New Phytologist Trust celebrates diversity and we expect participants in our meetings to be respectful, considerate and supportive of each other, to offer constructive critiques and embrace the variety of opinions on offer. The 40th NPS is an opportunity to share, develop and broaden our viewpoints within a safe and inclusive setting, and we hope that you will enjoy the meeting. If you have any concerns or suggestions, please speak to one of the organisers.

Contact
For further information, and in case of any emergencies, please contact Helen Pinfield-Wells. Email: h.pinfield-wells@lancaster.ac.uk, np-symposia@lancaster.ac.uk;

Meeting Programme

Tuesday 12 September

11:00–13:00	Registration and lunch
13:00–13:10	Welcome, Introductions and Information
13:10–14:00	**Keynote: Frank Johannes** Epimutational processes shape the *A. thaliana* methylome

Session 1: Population epigenomics in natural systems
Chair: Ovidiu Paun
14:00–18:00

14:00–14:35	**S1.1 Conchita Alonso** Interspecific variation of DNA methylation: a trans-continental plant community approach
14:35–15:10	**S1.2 Victoria Sork** Climate associations and transgenerational inheritance of DNA methylated variants in oak populations.
15:10–15:25	**Selected talk - Sofia van Moorsel: P44** Evidence for rapid evolution in a grassland biodiversity experiment
15:25–16:00	**Coffee/Tea break**
16:00–16:35	**S1.3 Emiliano Trucchi** DNA methylation differentiation and heritability in alpine and montane ecotypes of *Heliosperma pusillum* (Caryophylaceae)
16:35–17:10	**S1.4 Danelle Seymour** The evolutionary dynamics of DNA methylation in gene bodies

17:10–17:45	**S1.5 Walter Durka** Genetic and epigenetic variation across larger scales: the case of an ephemeral plant with highly fragmented distribution range
17:45–18:00	**Selected talk - Jeanine Mounger: P39** Effects of genetic diversity and epigenetic change on trait variation in the foundation plant *Spartina alterniflora*
18:00–20:00	**Poster reception**

Wednesday 13 September

Session 2: Epigenetic regulation of plant phenotypes
Chair: Katrin Heer
09:00–12:25

09:00–09:35	**S2.1 Jo Hepworth (replacement speaker)** Releasing FLC back into the wild
09:35–10:10	**S2.2 Etienne Bucher** Mobilized transposable elements as a tool for crop improvement
10:10–10:25	**Selected talk - Thierry Halter: P23** Transcriptional control of immune-responsive genes by active DNA demethylation and its relevance in plant adaptation
10:25–11:00	**Coffee/Tea break**
11:00–11:35	**S2.3 Eriko Sasaki** Global natural variation of DNA methylation and its genetic architecture in *Arabidopsis thaliana*
11:35–12:10	**S2.4 Igor Yakovlev**

	The epigenetic memory and climatic adaptation in Norway spruce
12:10–12:25	**Selected talk - Abdelhak El Amrani: P7** Small RNAs and plant tolerance to xenobiotics: Responses under allopolyploidization in *Spartina* (Poaceae)
12:25–14:00	**Lunch**
Session 3: Transgenerational inheritance of epigenetic patterns **Chair: Oliver Bossdorf** **14:00–18:00**	
14:00–14:35	**S3.1 Bob Schmitz** Encyclopedia of plant methylomes - from patterns to mechanisms and functions
14:35–15:10	**S3.2 Koen Verhoeven** Epigenetic inheritance in asexual plants: transgenerational experiments and natural evolution within asexual dandelion genotypes
15:10–15:25	**Selected talk - Philippine Vergeer: P46** Adaptation to climate change: Evidence for epigenetically based transgenerational phenotypic effects
15:25–16:00	**Coffee/Tea break**
16:00–16:35	**S3.3 Claudia Köhler** Paternal easiRNAs regulate parental genome dosage in Arabidopsis
16:35–17:10	**S3.4 Jose Gutierrez-Marcos** Heritable phenotypic changes induced by stress and developmental reprogramming in plants
17:10–17:25	**Selected talk - Jack Colicchio: P10** Differential methylation in response to parental wounding explains transgenerational plasticity

17:25–19:30	**Poster reception**

Thursday 14 September

Session 4: Epigenetic regulation as a response to changes in environmental conditions
Chair: Lars Opgenoorth
09:00–12:25

09:00–09:35	**S4.1 Christina Richards** Understanding mechanisms of response to complex environmental conditions using model and non-model plants
09:35–10:10	**S4.2 Steven Eichten** Adaptation and response via the 'Extended Genotype' – causes and consequences of DNA methylome and transposon variation among various plant species
10:10–10:25	**Selected talk - Stéphane Maury: P38** DNA methylation and tree phenotypic plasticity in a context of global changes
10:25–11:00	**Coffee/Tea break**
11:00–11:35	**S4.3 Penny Tricker** The relevance of epigenetic response to abiotic stress in complex crop genomes
11:35–12:10	**S4.4 Yupeng Geng** Epigenetic modification may contribute to the progressive acclimatization of an asexual invasive plant to new environment
12:10–12:25	**Selected talk - Nader Aryamanesh: P2** Transcriptomics and methylation in locally adapted Arabidopsis *lyrata* populations
12:25–14:00	**Lunch**

Session 5: Epigenetic regulation of biotic interactions
Chair: Christina Richards
14:00–16:50

14:00–14:35	**S5.1 Claude Becker** An (epi)genomics approach to understanding plant–plant interaction
14:35–15:10	**S5.2 Vitek Latzel** Heritable epigenetic variation in ecology and evolution of (some) plants
15:10–15:25	**Selected talk - Jacob Herman: P25** Epigenetic effects of single- and multi-species inoculation of *Arabidopsis thaliana* with two natural pathogens, *Pseudomonas syringae* and *P. viridiflava*
15:25–16:00	**Coffee/Tea break**
16:00–16:35	**S5.3 Isabelle Fudal** Chromatin-based control of plant–fungi interactions
16:35–16:50	**Selected talk - Rebecca Kartzinel: P30** Ants and plants: specific host epigenetic responses in a multispecies mutualism
17:00–18:30	**Tour of Botanical Garden (optional)**
19:00	**Tram departs to Schübel-Auer**
20:00	**Symposium dinner at Schübel-Auer** Buses return to Dept. of Botany and Biodiversity at 23:00

Friday 15 September

Session 6: Bioinformatic analysis for plant epigenetics

Chair: Marie Mirouze
09:00–13:00

09:00–09:35	**S6.1 Maria Colomé-Tatché** Constructing chromatin state maps from whole genome epigenomic data
09:35–10:10	**S6.2 Peter Stadler** Computational tools for EpiGenomics
10:10–10:25	**Selected talk - Manu Dubin: P16** DNA methylation variation in barley is driven by both climate of origin and breeding efforts
10:25–11:00	**Coffee/Tea break**
11:00–11:15	**Selected talk - Ritushree Jain: P27** Epigenetic regulation of heritable immune priming in Arabidopsis
11:15–11:50	**S6.3 Christoph Bock** Epigenome mapping technology and bioinformatic analysis methods
11:50–12:25	**S6.4 Jörg Hagmann** Current advances in statistical calling of differential methylation in plant populations
12:25–13:00	**Concluding comments and close**
	Packed lunches available for all delegates

Speaker Abstracts

S=speaker abstract; P=poster abstract; Bold indicates presenting author

Speaker abstracts

Epimutational processes shape the *A. thaliana* methylome

Keynote

FRANK JOHANNES[1,2]

13:10–14:00

frank@johanneslab.org

[1]*Department of Plant Sciences, Technical University of Munich, Germany;* [2]*Institute for Advanced Study, Technical University of Munich, Germany*

Cytosine methylation is a pervasive feature of most plant genomes. Despite progress in dissecting the molecular pathways that establish and maintain DNA methylation patterns, little is known about the mechanisms that shape plant methylomes over time. Here I present Whole Genome Bisulphite Sequencing data from *A. thaliana* mutation accumulation lines and from natural accessions to argue that the accumulation dynamics of spontaneous epimutations is a major driver of methylome diversity over time-scales that are of agricultural and evolutionary relevance.

Session 1: Population epigenomics in natural systems
Chair: Ovidiu Paun

Interspecific variation of DNA methylation: A trans-continental plant community approach

S1.1

CONCHITA ALONSO[1], MÓNICA MEDRANO[1], RICARDO PÉREZ[2], AZUCENA CANTO[3], VÍCTOR PARRA-TABLA[4], CARLOS M HERRERA[1]

14:00–14:35

conalo@ebd.csic.es

[1]Estación Biológica de Doñana, CSIC, Spain; [2]Instituto de Investigaciones Químicas, CSIC-US, Spain; [3]Centro de Investigación Científica de Yucatán, CONACYT, México; [4]Universidad Autónoma de Yucatán, México

Understanding the ecological significance of epigenetic regulation in wild plant populations requires linking phenotypic variation in ecologically relevant traits with some 'epiphenotypic' trait possessing the capacity to summarize epigenetic variation across individuals, populations, and species. Global DNA cytosine methylation is a proxy for epigenetic variation that is related to genetic and ecological factors at the intraspecific level. However, little is known on the degree to which differences between species are related to evolutionary history, ecological settings and biogeography.
In this study, we adopted a plant community approach to analyze global methylation in species from two distant biogeographic regions, Mediterranean (Spain) and Tropical (Mexico), and four different communities per region. At each region, communities were distributed along an ecological gradient from the Atlantic coast to inland, which ran parallel to increasing rainfall. Global cytosine methylation varied widely between angiosperms (range 4.8–42.1%; N = 279 spp). Interspecific differences were related to their evolutionary trajectories, as denoted by a strong underlying phylogenetic signal. Genomes of tropical species were on average less methylated than those of Mediterranean ones and such difference remained for either woody or non-woody plants, supporting a significant biogeographic effect within growth form. Further, genomes of woody plants were less methylated than

those of herbaceous ones, and genomes of deciduous species less methylated than those of evergreen ones. However, the eight communities did not differ in average DNA methylation and exhibited similar interspecific variances in this ‘epiphenotypic’ trait. Altogether, taxonomic affiliation and intrinsic functional plant traits were much more important than community as predictors of origin the cytosine methylation level of individual species.

Climate associations and transgenerational inheritance of DNA methylated variants in oak populations

S1.2

VICTORIA L. SORK[1], SOREL FITZ-GIBBON[2], DYLAN BURGE[1], PAUL F. GUGGER[3], JESSICA WRIGHT[4], MATTEO PELLEGRINI [2]

14:35–15:10

vsork@college.ucla.edu

[1]Department of Ecology and Evolutionary Biology, University of California, Los Angeles, CA USA; [2]Department of Molecular, Cell, and Developmental Biology, University of California, Los Angeles, USA; [3]University of Maryland Center for Environmental Science, Frostburg, MD USA; [4]Pacific Southwest Experimental Station, US Forest Service, Davis, CA USA

DNA-methylation has the potential to shape plant phenotypes and improve plant response to environmental conditions. Variation in methylation across a population could be due to genetic regulation of methylation, which may occur in CG-methylation, or it could be due to environmentally induced methylation, which might occur in CHH-methylation (where H= A, T, or G). Here, we address three goals by utilizing a sample of ~67,000 single methylated variants (SMVs) generated by Reduced Representation Bisulfite Sequencing of leaf tissue from 48 naturally occurring *Quercus lobata* adults sampled throughout the species' range and 196 offspring grown in two environmentally-different common gardens. First, we assess environmental associations of adult SMVs, of all sequence contexts, with climate variables to identify significant loci as evidence of locally adapted methylation. Second, we test the hypothesis that CG methylation is transgenerationally inherited by using parent-offspring correlations as an estimate of broad-scale heritability. Third, we test the hypothesis that CHH methylation is more likely to reflect environmental effects by comparing DNA methylation levels of paired progenies grown in two gardens. Our findings show that (i) there are more significant climate-associated SMVs in the CG context but some CHG- and CHH-SMVs are also highly correlated with climate gradients; (ii) CG-SMVs show higher heritability levels than CHH-SMVs; and (iii) CHH-SMVs show variation in methylation levels between common gardens providing evidence that CHH-methylation may play a role in

phenotypic plasticity. Using our valley oak genome assembly, we identify genes with significant environmental associations, levels of methylation heritability, and methylation differences across common gardens. These findings illustrate that methylation can influence plant response to the local environment through both genetically-based local adapted methylation and epigenetically-mediated phenotypic plasticity.

DNA methylation differentiation and heritability in alpine and montane ecotypes of *Heliosperma pusillum* (Caryophyllaceae)

S1.3

16:00–16:35

EMILIANO TRUCCHI[12], RUTH FLATSCHER[1], JULIANE BAAR[1], BOZO FRAJMAN[3], PETER SCHÖNSWETTER[3], OVIDIU PAUN[1]

emi.trucchi@gmail.com

[1]Department of Botany and Biodiversity Research, University of Vienna, Vienna, Austria; [2]Department of Life Sciences and Biotechnology, University of Ferrara, Ferrara, Italy; [3]Institute of Botany, University of Innsbruck, Innsbruck, Austria

Epigenetic modifications are expected to occur at a faster rate than genetic mutations, and to be a flexible way to respond to altered ecological conditions and to rapidly generate variation that can be visible to natural selection, even in the absence of genetic mutations. Epigenetics is thus expected to be particularly important in the early phases of ecological divergence. Alpine and low-mountain populations of *Heliosperma pusillum* constitute a perfect model system to test this hypothesis. They recently and recurrently diverged from one another in distinct ecological conditions with divergent morphologies, traits that are stable and heritable in common garden settings. Employing a reduced-representation approach (bisulfite-converted RAD sequencing – bsRADseq), we screened DNA methylation patterns of more than 200,000 nucleotide positions across six population pairs of the two ecotypes. Comparing DNA methylation state and context in 120 individuals, we generally found a conserved pattern of methylation independent of the native habitat type. This may be related to the over-representation of gene body regions within our dataset. Few outlier positions, some co-occurring within the same RAD loci, showed a signal of differential methylation between the two ecotypes. However, no patterns of differentiation between ecotypes have been consistently identified in more than two ecotype pairs indicating that most epigenetic divergence is driven either by drift or by adaptation to local conditions. In addition, across three generations of selfed plants of the two

ecotypes, we found a relatively high heritability rate, that is however context dependent. Nonmethylation was faithfully inherited in CHH and CHG context, followed by methylation at CpG positions, and non-methylation at CpG sites. Methylation at CHH and CHG contexts was instead highly instable in the subsample of the genome represented in our data. Altogether, our data provides insights on the epigenetic landscape during early phases of repeated ecological divergence, which may be a common process along the speciation continuum.

The evolutionary dynamics of DNA methylation in gene bodies

S1.4

DANELLE SEYMOUR[1], SHOHEI TAKUNO[2], BRANDON GAUT[1]

16:35–17:10

dkseym@uci.edu

[1]Department of Ecology and Evolutionary Biology, University of California, Irvine, CA, USA; [2]Department of Evolutionary Studies of Biosystems, SOKENDAI (The Graduate University for Advanced Studies), Hayama, Kanagawa, Japan

DNA methylation in plant genomes is predominantly localized to transposable elements (TEs) where it acts to repress transcription and inhibit transposition. While this TE-driven signal dominates genomic DNA methylation patterns in plants, a minority of genes are also heavily methylated (gene-body methylation, gbM). This type of DNA methylation is poorly understood. We do know that body-methylated genes are typically slowly evolving, broadly expressed, and that levels of gbM are positively associated with gene expression. This last observation is in stark contrast to the repressive effect of TE-linked methylation on gene expression. Most intriguingly, gbM levels are conserved over millions of years of evolution – from ferns to angiosperms. We sought to take advantage of this high degree of conservation to improve our understanding of gbM. To do so, we have surveyed gbM levels in multiple plant species to determine 1) the frequency at which gbM shifts in closely related species, 2) if shifts in body-methylation status are associated with any molecular consequences, and 3) whether changes in gbM status are preserved (i.e. beneficial). I will discuss how the dynamics of body-methylated genes can provide a glimpse into the evolutionary processes shaping gbM in plants.

Genetic and epigenetic variation across larger scales: the case of an ephemeral plant with highly fragmented distribution range

S1.5

WALTER DURKA[1,2], ANNETTE HÜBNER[3], ELKE RICHERT[3], HERMANN HEILMEIER[3], MARIA B. KRYUKOVA[4], GEORGIO TARAN[5]

17:10–17:45

walter.durka@ufz.de

[1]Helmholtz Centre for Environmental Research-UFZ, Dept. Community Ecology, Halle, Germany; [2]German Centre for Integrative Biodiversity Research (iDiv) Halle-Jena-Leipzig, Leipzig, Germany; [3]Institute of Biosciences, TU Bergakademie Freiberg, Freiberg, Germany; [4]Institute for Water and Ecological Problems, Russian Academy of Sciences, Khabarovsk, Russia; [5]Russian Academy of Sciences, Novosibirsk, Russia

The mossgrass *Coleanthus subtilis* (Poaceae) is a small, annual, ephemeral grass species growing on muddy soils of temporarily drained water bodies that display a high degree of water level dynamics, such as natural rivers, fishponds or water reservoirs. Its distribution is highly fragmented with regions of higher abundance ranging from Western France, Eastern Germany, Poland, Czech Republic, central Siberia, Russian Far East to Western United States. In Europe the species is mostly confined to anthropogenic ponds and water reservoirs. While it is declining in some regions due to changes in management and river dynamics, it was newly observed recently in other regions (Poland, Lower Lusatia(Germany)) suggesting a dynamic range due to seed dispersal both by birds and man. We wanted to identify dispersal routes through genetic markers and potential local adaptation through markers for methylation polymorphisms.

We used AFLP markers to unravel the genetic population structure across the species´ Eurasian range (214 individuals, 55 populations). Expecting that genetic variation will be low due to a highly selfing breeding system, we in addition wanted to assess the level and structure of epigenetic methylation variation using MSAP markers. We hypothesized that methylation variation will be related to local habitat conditions as a result of long term adaptation or short term response.

Genetic variation was extremely low (overall polymorphic loci: 20%; 3.8% at 5% level) with most variation partitioned among regions and populations (F_{ST}=0.61). Epigenetic variation was higher than genetic variation but still comparatively low (polymorphic H/M MSAP bands: 37%; 22% at 5% level; polymorphic MSAP epiloci: N=251, 65 (26%) at 5% level), with most variation residing within populations (F_{ST}=0.36). Epigenetic distance between individuals strongly exceeded, but was correlated to, genetic distance.
Results are discussed with respect to large scale range dynamics, recent range changes and local adaptation. We suggest that the highly specialized ecological and temporal niche limited both the level of genetic and epigenetic diversity due to genetic drift and selfing.

Session 2: Epigenetic regulation of plant phenotypes
Chair: Katrin Heer

Mobilome sequencing reveals real time transposable element activity in plants *(this speaker was unable to attend)*

S2.1

09:00–09:35

SOPHIE LANCIANO[1,2], MARIE-CHRISTINE CARPENTIER[2], CHRISTEL LLAURO[2], ALAIN GHESQUIERE[1], OLIVIER PANAUD[2], MARIE MIROUZE[1,2]

marie.mirouze@ird.fr

[1]Institut de Recherche pour le Développement (IRD), UMR232 DIADE, 911 Avenue Agropolis, 34394 Montpellier, France;
[2]University of Perpignan, Laboratory of Plant Genome and Development, 58 Avenue Alduy, 66680 Perpignan, France

Transposable elements (TEs) represent a main source of genomic diversity and an evolutionary force in both plants and animals. Host genomes have developed epigenetic mechanisms to control and prevent their proliferation. However, under specific stress conditions or at precise developmental stages, some TEs can be remobilized and proliferate in plant genomes. In plants, only a few active TEs have been identified and the mobile part of the genome or mobilome, comprising these elements, is unknown. Notably the extent of TE mobility in plant populations in the wild is not known.
To establish an unbiased repertoire of mobile TEs during plant development and in ecological conditions, we have developed a simple strategy based on high throughput sequencing to detect TEs extrachromosomal forms. Our method successfully identified known and novel active TEs in Arabidopsis and rice samples with destabilised epigenomes. I will also present how the method could be used in field experiments to analyze TE mobility on plants species with or without a reference genome. The mobilome-seq represents a novel approach to understand and evaluate the extent and impact of real time TE mobility on eukaryotic genomes.
This work is funded by an ANR grant (Extrachrom, 2014-2018).

Mobilized transposable elements as a tool for crop improvement

S2.2

MICHAEL THIEME[1], SOPHIE LANCIANO[2,3], SANDRINE BALZERGUE[4], NICOLAS DACCORD[4], MARIE MIROUZE[2,3], ETIENNE BUCHER[4]

09:35–10:10

etienne.bucher@inra.fr

[1]Botanical Institute, Zürich-Basel Plant Science Center, University of Basel, Hebelstrasse 1, 4056 Basel, Switzerland; [2]Institut de Recherche pour le Développement, UMR232 DIADE Diversité Adaptation et Développement des Plantes, Université Montpellier 2, Montpellier, France; [3]University of Perpignan, Laboratory of Plant Genome and Development, 58 Avenue Paul Alduy, 66860 Perpignan, France; [4]IRHS, Université d'Angers, INRA, AGROCAMPUS-Ouest, SFR4207 QUASAV, Université Bretagne Loire, 49045 Angers, France

The rapidly changing climate puts commonly used crop plants under strong pressure. It is therefore essential to develop novel breeding technologies to rapidly enhance crops to better withstand newly emerging stresses.
Interestingly, a clear link between transposable elements (TEs), crop improvement and varietal diversification exists. Furthermore, in recent years the importance of (TEs) in evolution and adaptation to stresses has been recognized. However, the use of TEs in crop breeding is currently very limited because it is not possible to trigger TE mobility in a controlled manner. We have now identified RNA polymerase II as a component of a novel highly conserved epigenetic silencing mechanism that represses the activity of TEs in Arabidopsis. Based on these findings we developed drug treatments capable of inhibiting this mechanism thus allowing us to generate controlled TE bursts in Arabidopsis. Because these drugs target highly conserved enzymes we were able to also mobilize TEs in rice using the same treatments. We are therefore now able to produce TE bursts in a controlled manner in virtually any plant. We can thus, for the first time, generate and study TE bursts in crop plants in real time. The described approach thus unlocks the use of TEs for plant breeding in virtually any crop irrespective of the availability of its genome sequence.

Global natural variation of DNA methylation and its genetic architecture in *Arabidopsis thaliana*

S2.3

ERIKO SASAKI[1], TAIJI KAWAKATSU[2], JOSEPH ECKER[2], MAGNUS NORDBORG[1]

11:00–11:35

eriko.sasaki@gmi.oeaw.ac.at

[1]Gregor Mendel Institute of Molecular Plant Biology, Austrian Academy of Sciences, Vienna Biocenter, 1030 Vienna, Austria, [2]The Salk Institute for Biological Studies, La Jolla, CA 92037, USA

Cytosine-methylation is an epigenetic mark that is involved in various molecular mechanisms, such as silencing of transposable elements (TEs) and chromatin modifications. Although natural variation of DNA methylation is largely regulated by genetic factors, the genetic architecture shaping this variation remains unclear. To address this, we carried out genome-wide association studies for DNA methylation levels at TEs using a large population of *Arabidopsis thaliana* from the 1001 methylome project (Kawakatsu et al., 2016). We identified seven regions that are significantly associated with the CHH methylation levels (H = C, A or T), and observed strong enrichment of *a priori* DNA methylation regulators in our candidate gene list including *ARGONAUTE 1*, *9*, *NRPE1 and CHROMOMETHYLASE2 (CMT2)*. Indeed, a polymorphism of *NRPE1*, a key component in RNA-directed DNA methylation (RdDM) pathways, had a large negative effect on the RdDM-targeted methylation levels, as well as the known polymorphism of *CMT2* (Dubin et al., 2015). These *NRPE1* and *CMT2* non-reference alleles showed essentially non-overlapping geographical distributions, and DNA methylation levels of these targeted-sites were correlated with different climate variables. Interestingly, individuals having both *NRPE1* and *CMT2* non-reference alleles were restricted to a narrow contact zone. All our findings suggest that natural selection could have shaped global variation of DNA methylation levels in *A. thaliana*.

The epigenetic memory and climatic adaptation in Norway spruce

S2.4

IGOR A. YAKOVLEV[1], THOMAS SOLVIN[1], HARALD KVAALINEN[1], ARNE STEFFENREM[1], MARCOS VIEJO[2], CARL GUNNAR FOSSDAL[1]

yai@nibio.no

[1]Norwegian Institute for Bioeconomy research, Ås, Norway; [2]Department of Plant Sciences, Norwegian University of Life Sciences, Ås, Norway

11:00–11:35

Adaptation to the changing environments is vitally important for long-lived plant species like forest trees. Epigenetic modifications and specifically epigenetic memory could be important mechanisms for diversifying environmental responses and widening the total plasticity of populations. The epigenetic memory of a plant is defined by the reproducible set of modifications of DNA and chromatin (without alteration of the DNA sequence) induced by external stimuli, which alter gene expression and therefore the properties and behavior of the plant. Memorizing of specific responses, laid down by epigenetic mechanisms, could provide significant strategic benefits to those plants, since the most successful response could be tuned or reenacted in response to a modified environmental condition and this would be retained in future cell lineages, and potentially inherited and altered by selection in future generations.

Such an important adaptive mechanism has been identified In Norway spruce and called epigenetic memory. The temperature and (probably) photoperiod during post-meiotic megagametogenesis and seed maturation epigenetically shifts the growth cycle program of the embryos. This results in significant and long lasting phenotypic change in the progeny such as advance or delay of several vital phenological processes of high adaptive values like bud break and bud set, allowing them to adapt rapidly to new and/or changing environments.

Here we summarize the information related to epigenetic memory regulation in Norway spruce with special emphasis on both phenotypic (quantitative) and molecular mechanism underlying this process. We will discuss how this phenomenon has changed our interpretation of clinal variation pattern in adaptive traits, and how we currently work to understand the molecular basis and regulation of this epigenetic memory.

Session 3: Transgenerational inheritance of epigenetic patterns
Chair: Oliver Bossdorf

Encyclopedia of plant methylomes - from patterns to mechanisms and functions

S3.1

BOB SCHMITZ[1], ADAM BEWICK, CHAD NIEDERHUTH, LEXIANG JI[2], XIULING SHI, ZEFU LU, BRIGITTE HOFMEISTER[2], NICK ROHR[2], YINWEN ZHANG[2]

14:00–14:35

schmitz@uga.edu

[1]Department of Genetics, University of Georgia, USA; [2]Institute of Bioinformatics, University of Georgia, USA

Advances in DNA sequencing technologies have led to the first single-base resolution maps of cytosine DNA methylation. These maps are revealing a diverse set of mechanisms that genomes utilize to control genome stability, transposon silencing, and gene expression. Our previous studies in natural and experimental *Arabidopsis thaliana* populations have uncovered errors in maintenance of DNA methylation that lead to the existence of spontaneous epigenetic alleles. The rates of formation of single methylation polymorphisms are about five orders of magnitude greater than the spontaneous mutation rate, whereas the formation of differentially methylated regions that affect gene expression occurs at a frequency that is similar to the spontaneous DNA mutation rate. To better understand the mechanisms governing maintenance of DNA methylation over longer evolutionary timescales we are sequencing methylomes from a range of plant species. Additionally, we have developed a novel high-throughput analysis method for profiling DNA methylation levels for any species regardless of the availability of a reference genome. We are unraveling the mechanistic underpinnings that evolved to establish and maintain DNA methylation by using this base modification as a molecular phenotype in combination with phylogenetic and molecular analysis of enzymes involved in DNA methylation. Lastly, we are developing methods of epimutagenesis to uncover cryptic variation which is a pervasive feature of plant genomes.

Epigenetic inheritance in asexual plants: transgenerational experiments and natural evolution within asexual dandelion genotypes

S3.2

KOEN VERHOEVEN[1]

14:35–15:10

k.verhoeven@nioo.knaw.nl

[1]*Netherlands Institute of Ecology (NIOO-KNAW), Department Terrestrial Ecology, Droevendaalsesteeg 10, 6708 PB Wageningen, Netherlands*

Because DNA methylation can be stably inherited in plants, stochastic and environment-induced DNA methylation modifications have the potential to affect heritable traits. Transgenerational stability of environment-induced epigenetic modifications is transient and seems restricted to the direct offspring of stress-exposed plants. However, transgenerational stability of epigenetic modifications may be different in asexual plants. Depending on the exact type of asexuality, reproduction takes place while bypassing either male germlines (apomixis) or both male and female germlines (vegetative propagation), thus also bypassing the epigenetic resetting mechanisms that operate in germlines. We use triploid apomictic dandelion as a model to explore the epigenetic contribution to heritable variation and adaptation. Asexual reproduction such as apomixis facilitates the study of epigenetic variants that are not confounded with genetic variants. I will show examples of heritable variation within individual apomictic lineages, both in gene expression and in phenotypes, that is associated with epigenetic mechanisms. This variation arises as a result of environmental experiences in previous generations and also as naturally evolved differences. To explore an underlying DNA methylation-based mechanism of epigenetic inheritance we developed a bisulfite sequencing extension to genotyping-by-sequencing to screen DNA methylation in a reduced subset of the genome. This revealed inherited methylation modifications mostly in CHG context, rather than CG context, after experimental exposure to a jasmonic acid treatment. Epigenetic variation between apomictic clone members in nature is in part associated with variable transposable element silencing; recent results indicate that specific low-abundant transposable element families show variable methylation and recent proliferation within the apomictic genome, representing a source of novel genetic and epigenetic variation within lineages.

Paternal easiRNAs regulate parental genome dosage in Arabidopsis

S3.3

16:00–16:35

GERMAN MARTINEZ[1], PHILIP WOLFF[1], ZHENXING WANG[1], JORDI MORENO-ROMERO[1], JUAN SANTOS-GONZÁLEZ[1], LEI LIU CONZE[1], CHRISTOPHER DEFRAIA[2], KEITH SLOTKIN[2], CLAUDIA KÖHLER[1]

claudia.kohler@slu.se

[1]Department of Plant Biology, Uppsala BioCenter, Swedish University of Agricultural Sciences and Linnean Center of Plant Biology, Uppsala 75007, Sweden; [2]Department of Molecular Genetics and Center for RNA Biology, The Ohio State University, 500 Aronoff Laboratory, 318 West 12th Avenue, Columbus, Ohio 43210, USA

Polyploidy, the presence of more than two sets of chromosomes within the nucleus, is a common phenomenon among plants that has shaped genome organization and is thought to be a major driver of speciation. The triploid block acts as a reproductive barrier that prevents successful backcrosses of newly formed polyploids with their progenitors. This barrier is established in the endosperm, an ephemeral tissue that nurtures the developing embryo and induces the abortion of triploid seeds through a yet unknown mechanism. Interploidy hybridizations involving diploid (2x) maternal parents and tetraploid (4x) pollen donors cause failure in endosperm cellularization, leading to embryo arrest. Our work revealed that paternal epigenetically activated small interfering RNAs (easiRNAs) are responsible for the establishment of the triploid block-associated seed abortion in *Arabidopsis thaliana*. Paternal loss of the plant-specific RNA polymerase IV suppressed easiRNA formation and rescued triploid seeds by restoring small RNA-directed DNA methylation at transposable elements, correlating with reduced expression of paternally expressed imprinted genes. We propose that excess of paternally derived easiRNAs in diploid pollen prevents establishment of DNA methylation, leading to triploid seed abortion. Thus, easiRNAs form a quantitative signal for chromosome number and their balanced dosage is required for seed viability. Our data highlight the importance of transgenerational epigenetic regulation for post-fertilization genome stability.

Heritable phenotypic changes induced by stress and developmental reprogramming in plants

S3.4

ANJAR WIBOWO[1,2], CLAUDE BECKER[2,5], JULIUS DURR[1], JONATHAN PRICE[1], STIJN SPAEPEN[3], HADI PUTRA[1], SALLY HILTON[1], QUENTIN SAINTAIN[1], SARAH HARVEY[1,4], GARY BENDING[1], PAUL SCHULZE-LEFERT[3], DETLEF WEIGEL[2], JOSE GUTIERREZ-MARCOS[1]

16:35–17:10

j.f.gutierrez-marcos@warwick.ac.uk

[1]School of Life Sciences, University of Warwick, Coventry CV4 7AL, UK; [2]Department of Molecular Biology, Max Planck Institute for Developmental Biology, 72076 Tübingen, Germany; [3]Department of Plant Microbe Interactions, Max Planck Institute for Plant Breeding Research, Carl-von-Linné-Weg 10, 50829 Köln, Germany; [4]Department of Biology, University of York, York YO10 5DD, UK; [5]Gregor Mendel Institute of Molecular Plant Biology, Austrian Academy of Sciences, Vienna Biocenter (VBC), 1030 Vienna, Austria

Eukaryotes are able to effectively adapt to both long term and short term adverse environmental conditions through either genetic or epigenetic changes/response mechanisms. While the latter response is thought to be heritable, the precise mechanism(s) by which this occurs remains unknown. We have found that in Arabidopsis, recurrent exposure to stress directs epigenetic changes to newly identified epigenetically labile genomic regions. These acquired epigenetic marks are associated with conditional heritable adaptive phenotypic stress responses, which gradually reset in the absence of stress. We also found that these stress responses are not transmitted equally through parental sexual lineages due to the extensive epigenetic reprogramming affecting the male germline. We hypothesized that this epigenetic resetting could be bypassed in somatic embryos and thus assessed the epigenomic changes taking place during somatic embryogenesis. We found that regeneration from terminally differentiated somatic cells induce epigenetic changes that are heritable over multiple generations leading to novel phenotypes.

Session 4: Epigenetic regulation as a response to changes in environmental conditions

Chair: Lars Opgenoorth

Understanding mechanisms of response to complex environmental conditions using model and non-model plants

S4.1

CHRISTINA L. RICHARDS[1], MARIANO ALVAREZ[1,2], MARTA ROBERTSON[1], JULIE FERREIRA DE CARVALHO[3,4], ARMEL SALMON[3], MALIKA L. AINOUCHE[3], ARMAND CAVÉ-RADET[3], ABDELHAK EL AMRANI[3], THOMAS P. VAN GURP[5], NIELS C.A.M. WAGEMAKER[6], KOEN J.F. VERHOEVEN[5]

clr@usf.edu

[1]Department of Integrative Biology, University of South Florida, 4202 East Fowler Avenue, Tampa FL 33620 USA; [2]Present address: Department of Biology, Duke University, 130 Science Drive, Box 90388, Durham, NC 27708 USA; [3]Université de Rennes I, UMR CNRS 6553 EcoBio, Campus Beaulieu, Rennes, France; [4]Present address: UMR1349 IGEPP, INRA BP35327, 35653 Le Rheu cedex, France; [5]Netherlands Institute of Ecology, (NIOO-KNAW), Dept. Terrestrial Ecology, Droevendaalsesteeg 10, 6708 PB Wageningen, Netherlands; [6]Department of Experimental Plant Ecology, Radboud University Nijmegen, Nijmegen, Netherlands

09:00–09:35

Understanding how organisms are able to respond at different time scales is an essential component of deciphering the impact and long-term consequences of changing environment. Rapidly developing genomic tools for model plants grown in controlled conditions can now be used to examine the mechanisms of phenotypic response in a broad array of wild organisms and biologically relevant conditions. Studies in wild settings allow for exploring how phenotypic variation is modulated by variation in gene expression resulting from sequence polymorphisms and

regulatory mechanisms. While studies interested in adaptation have largely assumed that trait variation is based on sequence variation, we now know that epigenetic effects can result in heritable, novel phenotypes even without variation in DNA sequence and could therefore provide an unappreciated source of response. My lab group uses reduced representation bisulphite sequencing and transcriptomic approaches to explore the potential role of genetic and epigenetic processes in natural and controlled studies of native and invasive salt marsh species like *Spartina alterniflora* and Japanese knotweed. We also leverage the power of the eudicot *Arabidopsis* and monocot *Brachypodium distychum* model plant species to confirm our findings in these non-model plants. Combined these studies will enhance our understanding of how genetic and epigenetic variation interact in response to environment on different time scales, and ultimately contribute to adaptation.

Adaptation and response via the 'Extended Genotype' – causes and consequences of DNA methylome and transposon variation among various plant species

S4.2

S.R. EICHTEN[1], D. GANGULY[1], P. CRISP[1], T. STUART[2], J STREICH[1], K.D. MURRAY[1], B.J. POGSON[1], R. LISTER[2], J.O. BOREVITZ[1]

09:35–10:10

steven.eichten@anu.edu.au

[1]ARC Centre of Excellence in Plant Energy Biology, Australian National University, Canberra, Australia; [2]ARC Centre of Excellence in Plant Energy Biology, University of Western Australia, Perth, Australia

Phenotypic variation and environmental response is often due to allelic diversity found among populations. However, populations of largely clonal individuals are observed in nature and may display phenotypic variation in the absence of observed genetic diversity. DNA methylome variation, a possible epigenetic factor not associated with DNA sequence changes, may act as a source of novel variation within these populations. Although a possible epigenetic factor, DNA methylation is also a chromatin mark that is often associated, and controlled by genetic variation. As older SNP based genetic diversity may be limited in clonal populations, there are opportunities for rapid variation to arise through the activity of transposable elements within the genome leading to rapid, and possibly reversible, natural variation in these populations. These 'extended genotype' factors, and their interactions, are being studied across Australian and global populations of the model cereal *Brachypodium distachyon* to determine their role in adaptive life history strategies. These results, combined with additional evidence from *Zea mays* and *Arabidopsis thaliana*, highlight how extended genotype factors may act as a source for novel and rapid phenotypic variation. However, minimal evidence is present regarding heritable chromatin-based responses to environmental stresses.

The relevance of epigenetic response to abiotic stress in complex crop genomes

S4.3

PENNY TRICKER[1], JANNATUL FERDOUS[1], MOUMOUNI KONATE[1], ARON CORY[2], STEPHEN ROBINSON[3], EVERARD EDWARDS[4], GEORGE GIBBINGS[5], CARLOS RODRIGUEZ-LOPEZ[1]

11:00–11:35

penny.tricker@adelaide.edu.au

[1]School of Agriculture, Food and Wine, University of Adelaide, Adelaide, SA., Australia; [2]Crop Development Centre, University of Saskatchewan, Saskatoon, Saskatchewan, Canada; [3]Agriculture and Agri-Food Canada, Saskatoon, Saskatchewan, Canada; [4]CSIRO, Adelaide, SA, Australia; [5]University of Reading, Reading, Berks., UK

Although initial discoveries in epigenetics were made in crops, many of the advances in molecular and mechanistic understanding have been made in model organisms under laboratory conditions. Crop improvement through conventional breeding depends on intra-species diversity, or hybridization between closely related species, and has advanced through the identification and selection of genetic markers, making use of forward genetic approaches. Genetic association studies are complicated by large genotype x environment interactions and lack of heritability. Forward epigenetic populations, however, in both crop and non-crop species, have identified heritable phenotypes for adaptive traits, expression patterns, associated epi-alleles and differentially methylated regions, with high heritability. This suggests that our knowledge of crop responses to environment will be significantly augmented with epigenomic information, opening new possibilities for improvement.

Data from different, complex epigenomes and populations are valuable additions to our fundamental understanding of the impact of epigenetic regulation on physiology under abiotic stress, phenotypic plasticity and evolvability and the mediation of intragenomic conflicts. As we attempt to increase diversity for crop improvement by branching out into different genepools, our experiments suggest that a focus on epigenetics will not only enable novel breeding techniques in crops, but also broaden our knowledge of the ecological and evolutionary relevance of epigenetic mechanisms in changing and challenging climates.

Epigenetic modification may contribute to the progressive acclimatization of an asexual invasive plant to new environment

S4.4

WEN SHI [1], LEXUAN GAO[2], OLIVER BOSSDORF[3], XIAOJIE CHEN[1], CHENG-YUAN XU[4], JI YANG[2], YUPENG GENG[1]

11:35–12:10

ypgeng@ynu.edu.cn

[1]School of Ecology and Environmental Sciences, Yunnan University, China; [2]Ministry of Education Key Laboratory for Biodiversity Science and Ecological Engineering, Fudan University, China; [3]Plant Evolutionary Ecology group, University of Tübingen, Germany; [4]School of Medical and Applied Sciences, Central Queensland University, Australia

Epigenetic variation may play an important role for plants to cope with heterogeneous environments. Significant epigenetic differentiation among natural plant populations has been frequently observed, but its stability and adaptive significance remain largely unknown. Here, we use field monitoring and mutli-generation common garden experiment to investigate the stability and plasticity of epigenetic variation in natural populations of *Alternanthera philoxeroides*, a clonal perennial weed. We found little genetic variation across a broad geographic distribution, which is consistent with its unique life history of asexual reproduction. In contrast, we found high levels of epigenetic variation within and among natural populations. Repeated measurements indicated stable epigenetic differentiations among natural populations in field, but a progressive convergence of epigenetic differentiation in common garden. Loci-based analyses suggested that nearly half of epiloci were stable across years and populations in both field and common garden environment. Notably, some epiloci showed typically plasticity in response to the environmental switch from field to common garden. Environmental association analyses indicate that the global epigenetic differentiation and certain candidate epiloci were significantly related with a few climate variables of temperature and precipitation. Our results suggested that both stability and plasticity have contributed to the observed epigenetic variation in natural populations of *Alternanthera philoxeroides*, which help these asexual populations to acclimate to changing environments across a broad geographic distribution.

Session 5: Epigentic regulation of biotic interactions
Chair: Christina Richards

An (epi)genomics approach to understanding plant–plant interaction

S5.1

PATRICK HUTHER[1], JUDIT GAZDAG-KOVACS[1], DANIELA RAMOS CRUZ[1], NIKLAS SCHANDRY[1], CLAUDE BECKER[1]

14:00–14:35

claude.becker@gmi.oeaw.ac.at

[1]Gregor Mendel Institute of Molecular Plant Biology (GmbH), Austrian Academy of Sciences, Vienna Biocenter (VBC), 1030 Vienna, Austria

In natural and agricultural habitats, plants grow in organismal communities in which they compete for limited resources. Plants have evolved various strategies to outcompete their neighbours and to secure their survival and reproduction. One of them is to release into the rhizosphere phytotoxic substances that inhibit the germination and growth of plants in close proximity, a process known as allelopathy. Despite the importance of allelopathy in shaping natural plant communities and for agricultural production, the underlying molecular mechanisms are largely unknown. We have found that allelochemicals derived from the common class of cyclic hydroxamic acid root exudates directly affect the chromatin-modifying machinery in *Arabidopsis thaliana*. These allelochemicals inhibit histone deacetylases and exert their activity through locus-specific alterations of histone acetylation and associated gene expression. Our multilevel analysis collectively shows how biochemical plant–plant interference affects a fundamental cellular process, histone acetylation, by targeting an evolutionarily highly conserved class of enzymes. Our findings lead to a mechanistically informed model for the molecular mode of action of allelopathic compounds in the target plant and provide insights into the potent competition strategy of allelochemical-producing plants. Here, I will highlight some of our current research efforts to determine the specificity of the allelochemicals in their target interaction, and to disentangle the genetic basis underlying resistance towards allelopathic competitors.

Heritable epigenetic variation in ecology and evolution of (some) plants

S5.2

VÍT LATZEL

14:35–15:10

vit.latzel@ibot.cas.cz

Institute of Botany of the Czech Academy of Sciences, Průhonice, Czech Republic

Epigenetic variation can cause heritable variation in plant phenotypes. It is therefore intuitive to expect that epigenetic variation can be significant player in ecological and evolutionary processes in plants. However, evidence for this expectation is still scarce. By using of epigenetic recombinant inbred lines (epiRILs; lines that greatly differ in their heritable patterns of DNA methylation but not in DNA sequence) of *Arabidopsis thaliana* I will demonstrate that heritable epigenetic variation can provide the raw material for rapid evolution of plant phenotypes in response to changing environments. I will also show that epigenetic diversity, just as species and genetic diversity, alters the productivity and stability of populations and ecosystems. In addition, by using of a novel method in demethylation of mature plants, based on periodical spraying of 5-azacytidine aqueous solution on established plants, I will show that the growth and behaviour of clonal plant *Trifolium repens* can be affected by environments that are no longer present at the time of their growth due to environmentally induced epigenetic memory. Such epigenetic memory can be important component of the success of clonal plants in some environments.

Chromatin-based control of plant–fungi interactions

S5.3

ISABELLE FUDAL

Isabelle.Fudal@inra.fr

BIOGER, INRA, AgroParisTech, Université Paris-Saclay, Avenue Lucien Brétignières. 78850 Thiverval-Grignon, France

16:00–16:35

Plant pathogens, including plant-associated fungi, secrete during plant infection an arsenal of small secreted proteins acting as effectors that modulate host immunity to facilitate infection. Genome-wide transcriptomic studies have shown waves of concerted expression of effector genes that correspond to different stages of plant tissue infection and colonization. In parallel, effector genes were often found to be located in 'plastic' genomic regions, enriched in transposable elements. The location of effector genes in regions enriched in transposable elements has been shown to have an impact on adaptability of fungi but could also provide for tight control of effector gene expression through chromatin-based regulation. Recently, chromatin structure was shown to be an important regulatory layer of effector gene expression in several plant-associated fungi with different lifestyles. Chromatin-based control of effector gene expression is likely to provide an evolutionary advantage by preventing the expression of genes not needed during vegetative growth and allow for a massive concerted expression at particular time-points of plant infection.

Session 6: Bioinformatic analysis for plant epigenetics
Chair: Marie Mirouze

Constructing chromatin state maps from whole genome epigenomic data

S6.1

A. TAUDT, D. ROQUIS, A. VIDALIS, R. WARDENAAR, M. A. NGUYEN, M. HEINIG, F. JOHANNES, MARIA COLOMÉ-TATCHÉ

09:00–09:35

maria.colome@helmholtz-muenchen.de

Advances in Next Generation Sequencing (NGS) technologies have enabled high-resolution measurements of various epigenetic marks at a genome-wide scale. Among them, post-translational modifications of histone residue tails are an important component of genome regulation. It is becoming increasingly clear that the combinatorial presence and absence of various modifications define discrete chromatin states which determine the functional properties of a locus. The de-facto standard procedure for genome wide mapping of histone modifications is chromatin immunoprecipitation followed by next generation sequencing (ChIP-seq). However, the statistical analysis and integration of data from several histone modifications remains challenging. Here I will present chromstaR, an algorithm for the computational inference of combinatorial chromatin states across an arbitrary number of conditions, such as different tissues or individuals. ChromstaR uses a multivariate Hidden Markov Model to assign every genomic region to a chromatin state based on the presence/absence of each modification in every condition. chromstaR is a versatile computational tool, written in C++ and available as an R package [1], that facilitates a deeper biological understanding of chromatin organization and dynamics.
Another widely studied epigenetic mark is DNA methylation. Cytosine methylation (5mC) is a widely conserved epigenetic mark with important roles in the regulation of gene expression and the silencing of transposable elements and repeats. Whole-genome Bisulfite sequencing (WGBS-seq) has become the standard method for interrogating plant methylomes at base pair resolution. However, deep WGBS-seq measurements remain cost prohibitive for large, complex genomes and for

population level studies. I will present METHimpute, a Hidden Markov Model based imputation algorithm for the analysis of shallow or deep WGBS-seq data. Unlike existing methods, METHimpute enables the construction of complete methylomes by inferring the methylation level of all cytosines in the genome regardless of coverage. Like that, METHimpute yields fully saturated methylomes even with low-coverage WGBS datasets, making it a valuable tool for increasing statistical power in genome-wide methylation QTL (meQTL) mapping studies, or in ecological studies that aim to correlate site-specific methylation states with environmental/climatic variables. METHimpute is written in C++ and available as an R package [2].

[1] www.chromstar.org
[2] github.com/ataudt/methimpute

Computational tools for EpiGenomics

S6.2

09:35–10:10

HELENE KRETZMER[1,2], RAINER MACHNÉ[3], DANIEL GERIGHAUS[4], CHRISTIAN OTTO[5], JAN ENGELHARDT[6], ANNE HOFFMANN[6], FABIAN AMMAN[7,8], LYDIA MÜLLER[9], DIRK ZECKZER[10], STEVE HOFFMANN[1,3], PETER F. STADLER[6,4,8,11,12,13]

studla@bioinf.uni-leipzig.de

[1]Transcriptome Bioinformatics, LIFE Research Center, Univ. Leipzig, Leipzig, Germany; [2]Interdisciplinary Center for Bioinformatics, Univ. Leipzig, Leipzig Germany; [3]Heinrich-Heine- Univ., Düsseldorf, Germany; [4]Competence Center for Scalable Data Services and Solutions Dresden/Leipzig, Univ. Leipzig, Germany; [5]ecSeq Bioinformatics GmbH, Leipzig Germany; [6]Bioinformatics, Institute for Computer Science, Univ. Leipzig, Leipzig, Germany; [7]Department of Chromosome Biology, Univ. Vienna, Vienna, Austria; [8]Institute for Theoretical Chemistry, Univ. Vienna, Vienna, Austria; [9]Natural Language Processing, Institute for Computer Science, Univ. Leipzig, Leipzig, Germany; [10]Image and Signal Processing, Institute for Computer Science, Univ. Leipzig, Leipzig, Germany; [11]Max-Planck-Institute for Mathematics in the Science, Leipzig, Germany; [12]Fraunhofer Institute for Cell Therapy and Immunology, Leipzig, Germany; [13]The Santa Fe Institute, Santa Fe, NM, USA

The analysis of epigenomic data requires a series of specialized tools. This ranges from specialized read-mappers that efficiently operate on reduced alphabets to efficient methods for calling differentially methylated regions, peak calling for ChIP-seq data, and the solution of data segmentation problems. A particularly difficult challenge is the incorporation of repetitive or near repetitive sequence elements in the analysis. These are often ignored, but recent case studies emphasize the importance of such genomic features in particular in the context of gene regulation. In this presentation I will discuss recent advances as well as open problems for the future development of computational tools for the efficient processing of epigenomic and epitranscriptomic data.

Epigenome mapping technology and bioinformatic analysis methods

S6.3

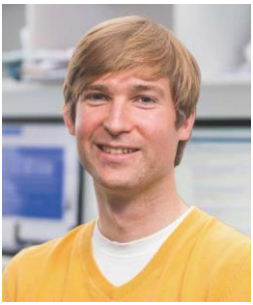
CHRISTOPH BOCK[1,2,3]

11:15–11:50

cbock@cemm.oeaw.ac.at

[1]CeMM Research Center for Molecular Medicine of the Austrian Academy of Sciences, 1090 Vienna, Austria; [2]Department of Laboratory Medicine, Medical University of Vienna, 1090 Vienna, Austria; [3]Max Planck Institute for Informatics, 66123 Saarbrücken, Germany

In my presentation, I will summarize our ongoing work developing experimental and computational methods for epigenome analysis. Main results include ChIPmentation for high-throughput chromatin analysis (Schmidl, Rendeiro *et al*. 2015 *Nature Methods*), RefFreeDMA for DNA methylation analysis without a reference genome (Klughammer *et al*. 2015 *Cell Reports*), and single-cell whole genome bisulfite sequencing (Farlik, Sheffield *et al*. 2015 *Cell Reports*). I will also discuss our research in the context of the International Human Epigenome Consortium, aimed at reconstructing complex epigenome landscapes using bioinformatic methods.



Current advances in statistical calling of differential methylation in plant populations

S6.4

JÖRG HAGMANN[1], PATRICK HÜTHER[2], CLAUDE BECKER[2], SEBASTIAN J SCHULTHEISS[1]

11:50–12:25

joerg.hagmann@computomics.com

[1]Computomics GmbH, Tübingen, Germany; [2]Gregor Mendel Institute, Vienna, Austria

The accurate identification of DNA methylation variation between individuals or conditions is crucial for understanding its role in shaping phenotypic diversity and in coping with environmental influences. Most epialleles in natural populations consist of methylation differences in regions rather than at single sites, which highlights the importance to statistically identify differentially methylated regions (DMRs). The vast majority of previous efforts to call DMRs in plant studies relied either on identifying variable single sites first, or testing genome-wide sliding windows, which both limits statistical power due to the high number of performed tests. More importantly, population-scale epigenome studies become increasingly common, and their typical approach to perform all pairwise comparisons between samples or conditions is difficult to interpret and computationally inefficient.
In this talk, I will address challenges and present current advances in analyzing bisulfite sequencing data for calling DMRs, including a new algorithm developed by us, MethylScore. This approach highly reduces the number of statistical tests by only focussing on the methylated genome space and by avoiding all-against-all sample comparisons. Methylated regions are identified unsupervised based on a Hidden Markov Model, and on this informed selection of regions, samples are clustered based on their methylation levels so that only clusters of samples are compared against each other.
I discuss the challenge in determining DMRs in plant populations to balance the tradeoff between sensitivity and performing a small number of statistical tests to retain detection power and computational efficiency.

Poster abstracts

P=poster abstract. S=speaker abstract. Bold=presenting author

Poster abstracts

Poster abstracts are ordered alphabetically by presenting author (underlined).

P1 Cold induces large and specific methylome changes in maize

Z. ACHOUR[1], H. SELLIER[3], J.P. PICHON[4], H. DUBORJAL[4], M. LEVEUGLE[4], H. CORTI[5], A. ROUSSELET[5], C. GIAUFFRET[2], C. VITTE[6]

[1]Université Paris–Sud, [5]INRA, [6]CNRS / UMR GQE - Le Moulon, Gif-sur-Yvette, France, [2]INRA UR GAP, [3]INRA UE GCIE / Estrées-Mons, Péronne, France, [4]Biogemma, Chappes, France

In Northern Europe, maize development leads to high financial and environmental drying costs at harvest. Early sowing has been proposed as a strategy to overcome this problem, but this cycle shift leads plants to be subjected to cold in the first phase of their development. Such an early and prolonged cold affects maize metabolism, leading to yield reduction. Origin of this physiological modification is not well understood, and detection of candidate genes using genetic-based studies is challenging. Whether DNA methylation changes are involved in this phenotype remains to be elucidated.

Our work aims at analyzing the impact of cold on the maize methylome, and compare it among three genotypes with contrasted phenotypic response. We use whole genome bisulfite-sequencing to detect Differentially Methylated Regions (DMRs) between 'cold' and 'control' plants in each genotype. Prolonged cold induces both hypermethylated and hypomethylated regions in CG, CHG and CHH contexts. Most of these changes are conserved among 'cold' plants, suggesting targeting of specific regions. As expected, CG DMRs are enriched in genes while CHG DMRs are enriched in genes and transposons. The two cold-sensitive lines show more DMRs than the tolerant one, suggesting a possible role of DNA methylation in maize response to cold.

P2 Transcriptomics and methylation in locally adapted *Arabidopsis lyrata* populations

NADER ARYAMANESH, WEIXUAN NING, TUOMAS HÄMÄLÄ, HELMI KUITTINEN, OUTI SAVOLAINEN

Department of Ecology and Genetics, Biocenter Oulu, 90014 University of Oulu, FIN-90014, Finland

There is a growing evidence that genome-wide methylation plays an important role in local adaptation. In this research, we studied the transcriptomics and epigenetics in locally adapted *Arabidopsis lyrata.* Populations from Norway (Spiterstulen and Lom) and Germany (Plech) were grown and phenotyped in a common garden experiment in two locations (high altitude Spiterstulen and low altitude Lom) in Norway. Reciprocal transplant experiment showed that the Norwegian populations were locally adapted, with native populations having higher fitness than non-native ones at each site. RNA sequencing and whole genome bisulfite sequencing was performed to investigate the gene expression and methylation signatures in local adaptation. Methylation levels in *Arabidopsis lyrata* were higher at the high altitude than the low altitude site for all different CpG, CHG and CHH contexts. Overall, Norwegian populations were more highly methylated than the German population. Results showed that environment plays a major role in the number of differentially expressed genes and differentially methylated regions between populations. The common garden experiment in the model perennial plant, *Arabidopsis lyrata*, made it feasible to dissect the roles of environmental effects and genetic differences in expression and methylation responses in the different environments.

P3 Environmental regulation of transposon activity in tomato

M. BENOIT[1], J. PASZKOWSKI[1]

[1]The Sainsbury Laboratory, University of Cambridge, Cambridge, UK

We are investigating the role of transposons in the regulation of plant development, by monitoring transposon activity during critical developmental processes in tomato. Long terminal repeat (LTR) retrotransposons represent a large proportion of plant genomes [1]. The annotation of the LTR retrotransposons in tomato revealed more than 5800 intact elements, some of them suggesting recent transposition events as revealed by sequence identity between LTRs [2]. Therefore, tomato represents a powerful model to study transposon activities and their links to associated environmental responses.
The phytohormone ABA (abscisic acid) plays an important role in mediating plant adaptation to stress, including water, light and temperature-related stresses. We present here evidences of a LTR retrotransposon family taking advantage of increased ABA levels upon drought stress to substantially increase its transcription level and activity. Moreover, we provide here evidences that perturbations of DNA methylation, here the RdDM pathway, enhance activity of this transposon family.
In summary, our work identified a family of LTR retrotransposons subjected to environmental regulation through ABA-specific transcriptional activation. Our results reveal an important plasticity in the environmental and epigenetic control of retrotransposons in tomato.

P4 BRY"O"MICS: Application of high-sensitive and high-throughput molecular tools to disentangle the mechanisms of heavy metals accumulation and tolerance in mosses: epigenetic and transcriptomic approaches

M. TERESA BOQUETE[1,2], CONCHITA ALONSO[1], CHRISTINA L. RICHARDS[2], CARLOS M. HERRERA[1]

[1]Estación Biológica de Doñana, CSIC, Avenida Américo Vespucio 25, Isla de la Cartuja, Sevilla 41092, Spain; [2]Department of Integrative Biology, University of South Florida, 4202 E Fowler Ave, Tampa, FL 33620, USA

Current knowledge on the mechanisms used by plants to deal with environmental stress is mostly derived from tracheophytes. However, bryophytes due to their phylogenetic position between green algae and tracheophytes, are especially interesting organisms to unravel the complexities of the plant-environment interactions from an evolutionary perspective. Current evidence suggests that epigenetic changes allow angiosperms to respond to environmental stress. The role of epigenetics in the phenotypic variation of ecologically important traits in bryophytes is not as well established.

Exposure to heavy metals (HM) imposes a strong environmental pressure to plants and yet bryophytes are able to thrive in highly polluted environments and even specialize to live in them. Thus, BRY"O"MICS aims to provide a deep understanding of the mechanisms underlying the existence of phenotypic variability for HM tolerance and hyperaccumulation in bryophytes. Two ecologically different moss species, *Scopelophila cataractae* (a 'copper-moss' restricted to HM enriched habitats) and *Ceratodon purpureus* (a cosmopolitan species occurring in a variety of substrates) were collected in the field and cultured in the laboratory under different HM treatments. Reduced representation bisulfite sequencing (RRBS-seq) and RNA sequencing (RNA-seq) will be used to build the methylome and transcriptome profiles of these two species and gain insight in their capacity to accumulate and deal with these pollutants.

Preliminary results show that gametophytes of *S. cataractae* collected in the most polluted sites were significantly smaller that the collected in less contaminated sites. Besides, we observed a significant sex-specific inhibition

of growth (males' growth significantly more inhibited that females') of *C. purpureus* plants under Cd and Cu treatments.
Also, we expect to find interspecific differences in the HM accumulation capacity as well as differences in the molecular mechanisms used to cope with HMs.

P5 RADseq and bsRADseq as genome-wide approaches to understand allopolyploid evolution in *Dactylorhiza* (Orchidaceae)

M. K. BRANDRUD[1], E. TRUCCHI[1], J. BAAR[1], M. T. LORENZO[1], M. HEDÉN[2], O. PAUN[1]

[1]Department of Systematic Botany and Biodiversity Research, University of Vienna, Rennweg 14, A-1030 Vienna, Austria, [2]Department of Biology, Biodiversity, University of Lund, Sölvegatan 37, SE-223 62 Lund, Sweden

Genomic inferences based on RADseq (i.e. restriction-site associated DNA sequencing) are currently widely applied to answer evolutionary questions in diploid organisms, but their use in polyploids is hindered by their complicated genomes and inheritance patterns. We focus here on *Dactylorhiza* allopolyploids that have originated independently from hybridization of two diploid lineages; *D. fuchsii* and *D. incarnata*. Based on thousands of RAD loci, we investigate the patterns of genetic divergence, gene flow dynamics and polyploid origins of 53 populations sampled across Europe, over the distribution range of the allopolyploid complex. We complement this with results obtained in selected individuals with bisulfite-converted RADseq, an approach to quantify the level of DNA methylation differentiation across multiple individuals. Our genetic results show a phylogeographic signal, but document a genome-wide absence of genetic differentiation between allopolyploids, giving evidence for frequent and extensive gene flow between the sympatric sibling allopolyploids in Central Europe. In contrast, significant methylation differences are uncovered that are fully methylated in one polyploid and not methylated in the other. We conclude that, in the face of gene flow, the observed phenotypic divergence between the sibling *Dactylorhiza* polyploids is maintained by a strong divergent selection, potentially related to their ecological specialization.

P6 Epistatic interactions mediated by DNA methylation control tocochromanol metabolism in tomato plants

ESTANISLAO BURGOS[1], BELEN M. DE LUCA[1], LEANDRO QUADRANA[2], LUISA BERMUDEZ[1], FERNANDO CARRARI[1*]

[1] *Instituto de Biotecnología, CICVyA INTA Castelar, Argentina;* [2]*Institut de Biologie de l'Ecole Normale Supérieure, (IBENS), Paris, France*

Plants are sessile organisms that had developed a plethora of response mechanisms to environmental constrains. Tocochromanol biosynthesis is triggered to cope with these conditions. Regulation of this pathway can be explained at different organization levels; from changes in leaf anatomy to epigenomics. One of the last steps in tocopherol biosynthesis is catalyzed by the 2-methyl-6-phytylquinol methyltransferase, which in tomato is encoded by two paralog genes, *VTE3(1)* and *VTE3(2)*. We have pointed that differential expression of *VTE3(1)* underlie a major QTL for tocopherol contents in fruits. Moreover, we demonstrated that variable expression of this gene is associated with differential DNA methylation of a SINE retrotransposon inserted in the promoter region of *VTE3(1)*. Here, using a combination of wild and domesticated alleles for both paralogs, we explored the existence of potential epistatic interaction(s) between paralogs as well as (epi)genetic X environmental (GxE). Our results show that the mRNA accumulation of the different alleles follows a coordinated fashion with the methylation levels of their promoters. Furthermore, tocopherol contents in fruits show both, epistatic as well as GxE interactions. These results contribute to understand the epistatic interactions of low heritability traits and might result in novel tools to be applied in breeding programs.

P7

Small RNAs and plant tolerance to xenobiotics: Responses under allopolyploidization in *Spartina* (Poaceae)

A. CAVÉ-RADET, A. EL AMRANI, D. GIRAUD, M. L. AÏNOUCHE, A. SALMON

UMR CNRS-OSUR 6553, ECOBIO (Ecosystèmes, Biodiversité, Evolution)
Université de Rennes 1, Campus de Beaulieu, 35042 Rennes, France

Allopolyploidization is known to induce a myriad of genetic and epigenetic changes resulting from both genome merger (hybridization) and redundancy (polyploidy), which ultimately lead to genes expression reprogramming and/or transposable element activation. This dynamics if of particular interest under stress conditions, as it may promote the formation and persistence of new adaptive phenotypes.
In the present work we examined the effect of hybridization and polyploidy on small RNA expression under phenanthrene (a Polycyclic Aromatic Hydrocarbon common in polluted environments) stress, in the *Spartina* system (Poaceae). We compared two parental species (*S. alterniflora* and *S. maritima*), their F1 hybrid, and the allopolyploid *S. anglica* which exhibit higher tolerance to phenanthrene.
Small RNA sequencing from control and phenanthrene stressed plants was performed. Annotations of miRNA and their targets were conducted. A total of 113 conserved and 481 novels miRNA were annotated. Differential expression analysis highlights changes between species, with specific sRNA expression patterns under control and treatment conditions next to hybridization and genome doubling. Additionally, we detected 103 miRNA differentially expressed (DE) under PAH stress that may represent potential key regulators of stress tolerance. Functional validation of DE miRNA on homologous *MIR* genes and targets in *A. thaliana* are performed.

P8 DNA methylation profile of grafted eggplants

E. CERRUTI[1], A. ACQUADRO[1], S. LANTERI[1], E. PORTIS[1], C. COMINO[1], P. GRAMAZIO[2], J. PROHENS[2], C. GISBERT[2]

[1]*DISAFA Department of Plant Genetics and Breeding, University of Turin, Largo Paolo Braccini 2, 10095 Grugliasco (TO), ITALY;* [2]*Instituto de Conservación y Mejora de la Agrodiversidad Valenciana, Universitat Politècnica de València, Valencia, Spain*

The molecular mechanisms behind grafting are still poorly understood, despite its extensive use for enhancing the performance of horticultural crops.
We performed a DNA methylation profiling study to assess the alterations in DNA methylation of grafted eggplant (*Solanum melongena*) plants. The Black Beauty (BB) eggplant commercial cultivar was selected as scion for self-grafting as well as for hetero-grafting onto two different commercial rootstocks: (i) *Solanum torvum,* the most commonly used rootstock in eggplant and (ii) the tomato hybrid 'Emperador RZ', which is also resistant to *Verticillium.* Non-grafted BB plants were used as controls.
Based on methylation sensitive restriction enzymes approach we generated three reduced complexity genomic libraries. Each library is specific for CG, CHH or CHG methylation context and is composed of samples coming from two developmental stages of both leaves and fruits. A set of 400 M Illumina raw reads were bioinformatically processed. Loci, target of methylation, were analysed with edgeR, revealing a trend of variation of DNA methylation in both leaves and fruits of grafted plants compared to the controls. Functional annotation and target bisulfite validation of the differentially methylated *loci* is ongoing.

P9 Investigating the involvement of the chromatin state in the control of effector gene expression in *Leptospharia maculans*

COLIN CLAIRET, NICOLAS LAPALU, ADELINE SIMON, FRANÇOISE BLAISE, JULIE GERVAIS, JESSICA L. SOYER, ISABELLE FUDAL

UMR BIOGER, INRA, AgroParisTech, Université Paris-Saclay, 78850 Thiverval-Grignon, France

Leptosphaeria maculans, a hemibiotrophic fungus responsible of stem canker, colonises oilseed rape in two stages: an early stage of cotyledon or leaf colonisation, and a late colonisation stage during which the fungus colonises systemically without visible symptom the plant before stem canker appears. *L. maculans* presents a bipartite genome structure alternating gene-rich and transposable element (TE)-rich regions. TE-rich regions, which encompass one third of the genome, are enriched in putative effector-encoding genes that are expressed specifically in an early stage of infection ('early effectors'). In contrast, gene-rich regions were recently reported to contain putative effector-encoding genes specifically expressed during the late stages of stem infection ('late' effectors). We have previously investigated the involvement of the chromatin structure of repeat-rich regions on the expression of 'early' effector genes: RNAi silencing of two genes encoding key players in heterochromatin assembly through histone modification H3K9me3, HP1 and KMT1, induced an over-expression of genes located in AT-isochores, particularly 'early' effector genes but no modification of 'late' effector genes expression. Here, we performed analysis of nucleosome positioning, chromatin structure and gene expression at the genome scale combining MAINE-seq, ChIP-seq and RNA-seq data during *in vitro* growth of *L. maculans*.

P10 Differential methylation in response to parental wounding explains transgenerational plasticity

JACK COLICCHIO, BENJAMIN BLACKMAN, JOHN KELLY, LENA HILEMAN

University of Kansas, Ecology and Evolutionary Biology, 1200 Sunnyside Avenue, Lawrence, KS, USA

Structured landscapes, climatic oscillations, and many other patterns in nature lead to substantial similarities in the conditions faced by individuals living nearby and shortly following the previous generation. Often the conditions faced by successive generations are autocorrelated, meaning they are more similar than expected by white-noise; this leads to fitness benefits associated with transgenerational phenotypic plasticity. One example of this transgenerational plasticity can be found in *Mimulus guttatus,* where the offspring of wounded plants differentially express hundreds of genes and produce leaves with more trichomes. However, in this system and many others, the mechanism through which gene expression and phenotype are altered has remained elusive. Here, utilizing whole genome methylome sequencing in the progeny of control and wounded individuals, we identify thousands of differentially methylated regions. An overabundance of these regions are near differentially expressed genes involved in hormone synthesis, making these regions prime candidates as key epigenetically inherited loci. Finally, along with targeted changes in DNA methylation, there are increases in the variability of methylation in the progeny of wounded plants. The ability for parental environmental stress to cause targeted epigenetic shifts and increase epigenetic variation has implications on the trajectory of evolution in variable landscapes.

P11 DNA methylation profile of micropropagated globe artichoke

C. COMINO[1], E. CERRUTI[1], A. ACQUADRO[1], G. MARCONI[2], A. REPETTO[3], A. PISANU[3], R. PILIA[3], E. ALBERTINI[2], E. PORTIS[1]

[1]DISAFA, Plant Genetics and Breeding, University of Torino, Grugliasco, Italy; [2]Department of Agricultural, Food, and Environmental Sciences, University of Perugia, Italy; [3]Agris Sardegna - Agenzia Regionale per la ricerca in agricoltura - Servizio Ricerca sui Sistemi Colturali Erbacei, Cagliari, Italy

We report our study on the methylation profile in micropropagated globe artichoke (*Cynara cardunculus* L, Asteraceae family, 2n=2x=34) plants. 'Spinoso sardo' is the most economically relevant globe artichoke ecotype in Sardinia (Italy). Its *in vitro* propagation has promoted an increased frequency of 'out-types' plants, which show two different phenotypes: the 'selvatico' characterised by highly pinnate-parted leaves and the late budding of the inflorescence, in contrast to the normal phenotype, here called 'gentile'. This unpredictable phenomenon may occur both *in vitro* and in open fields and it is reversible through globe artichokes generations, suggesting that it might be driven by epigenetics mechanisms. To test this hypothesis, a methylation profile study has been conducted by sequencing reduced complexity genomic libraries of leaves sampled from plants presenting both 'gentile' and 'selvatico' phenotypes. Bioinformatics analyses showed significant differences in CG, CHG and CHH methylation contexts. In addition, functional annotation of the differentially methylated loci enabled the identification of a set of candidate genes involved in: (i) flower development and its regulation, (ii) maintenance of epigenetic modifications and (iii) vegetative development, which will be validated by target bisulfite sequencing.

P12 Characterization of the wild emmer wheat methylome using whole genome bisulfite sequencing

ARON T CORY[1], ANDREW G. SHARPE[2], STEPHEN J. ROBINSON[3], CURTIS J. POZNIAK[1]

[1]Crop Development Centre, University of Saskatchewan, College of Agriculture and Bioresources, 51 Campus Drive, Saskatoon, SK, S7N 5A8, Canada; [2]Global Institute of Food Security, University of Saskatchewan, 110 Gymnasium Place, Saskatoon, SK, S7N 4J8, Canada; [3]Saskatoon Research and Development Centre, Agriculture and Agri-Food Canada, 107 Science Place, Saskatoon, SK, S7N OX2, Canada

The allotetraploid wild emmer wheat (WEW; *T. turgidum ssp. dicoccoides* (Körn.) Thell.; genome BBAA) is the progenitor of modern durum wheat (*Triticum turgidum subsp. durum*) which represents 10% wheat produced world wide[1]. Characterization of the methylome and transcriptome of the newly constructed cv. 'Zavitan' reference sequence has been initiated for 14 day old (Zadok stage 13) leaf tissue. This study represents the first comprehensive analysis of a tetraploid wheat using Whole Genome Bisulfite Sequencing (WGBS) and RNAseq. While analysis is ongoing, initial results confirm WEW to be highly methylated in CpG (92.4%) and CHG (64.7%) context but CHH methylation remains relatively low (3.3%). Methylation profiles of genes and their flanking regions show distinct patterns between active and inactive genes, constitutively expressed genes and genes with tissue dependent expression levels. Methylation of homeologous genes show some genome specific methylation. However, methylation of homeologous genes seems evenly distributed, favouring neither the A nor B genome and RNAseq data indicate no implicit genome dominance in the tissue studied. The large, repetitive and polyploid genome of WEW highlights novel bioinformatic challenges and opportunities when working with crop species.

P13 Epigenetic diversity and adaptive potential of *Fragaria vesca* populations

H. DE KORT[1], B. PANIS[2], O. HONNAY[1]

[1]Laboratory for Plant Conservation and Population Biology, University of Leuven, Kasteelpark Arenberg 31, 3001 Leuven, Belgium; [2]Department of Biosystems (Division Crop Biotechnics), W. de Croylaan 42, box 2427, 3001 Leuven, Belgium

Natural communities continue to collapse as a consequence of global environmental changes, urging for a better understanding of the mechanisms underlying the potential of species to adapt to these widespread threats. Although genetic variation underlying quantitative trait variation is considered to provide most of the adaptive potential needed to withstand local environmental disturbances, epigenetic variation is increasingly recognized as a potentially important component of adaptive potential.
We have propagated *Fragaria vesca* (Woodland strawberry) genotypes *in vitro*, to obtain genetically identical copies of plants originating from distinct natural environments. These plants (n = 1700) are randomly distributed across three different soil moisture regimes, allowing disentangling within and among-genotype responses to these environmental stressors. The resulting phenotypic, genetic and epigenetic (DNA methylation) data will offer a variety of opportunities to study the eco-evolutionary role of epigenetic change in conservation biology.
This research will provide novel insights into the molecular processes underlying the potential of natural populations to cope with unusual drought and flooding as a consequence of climate change. Considering the availability of a large cohort of clones with distinct environmental histories and drought stress conditions, I look forward to discuss the many opportunities that could arise from this epigenetics project.

P14 Natural variation of epigenetic mechanisms regulating nutritional quality of tomato fruits

BELEN M. DE LUCA, ESTANISLAO BURGOS, LUISA BERMUDEZ, FERNANDO CARRARI

Institute of Biotechnology, CICVyA, INTA-Castelar, Argentina

Under limiting environmental conditions, landraces are an important source of well-adapted biodiversity. Tocopherol biosynthetic pathway is regulated by the methylation levels of the promoter region of a dimethyl-phytylquinol-methyltransferase encoding gene *VTE3(1).* This is true for tomato fruits of the bred cultivar M82 harvested from contrasting environments, probably involving RdDM mechanisms. Moreover, the existence of *VTE3(1)* epialleles in natural population of tomato Andean landraces correlated with γ-tocopherol accumulation in fruits. In order to evaluate how conserved is this mechanism we further analyzed the co-variation between the gene expression-DNA methylation for related *tocopherol-pathway-genes* and through different organs cultivated under contrasting environmental conditions. Our results show that similar epigenetic mechanisms operate for the *VTE3(2)* paralog gene and that natural variation exists for this mechanism in our collection of landraces. Furthermore, genes operating upstream in the pathway, such as *DXR*(1-deoxy-D-xylulose-5-phosphate reductoisomerase encoding), seem to be regulated by different mechanism(s). These evidence points to the importance of the phenotypic and genetic variation preserved in tomato landraces, as a key biodiversity source to understand the epigenetic and genetic basis of characters related to health and organoleptic traits in plant crop species.

P15 Methylation map of young developing leaves grown under optimal and drought stress conditions in *Brachypodium distachyon*

C. DE QUATTRO[1], N. DACCORD[2], E. BUCHER[2], E. MICA[3], M.E. PÈ[1], E. BERTOLINI [1,4]

[1]*Institute of Life Sciences, Scuola Superiore Sant'Anna, Piazza Martiri della Libertà 33, 56127 Pisa, Italy;* [2]*Institut de Recherche en Horticulture et Semences (IRHS), INRA, AGROCAMPUS-Ouest, 42 rue Georges Morel – BP 60057, 49071 Beaucouzé cedex, France;* [3]*Genomics Research Centre, Consiglio per la Ricerca in agricoltura e l'analisi dell'economia agraria, Via S.Protaso 302, 29017 Fiorenzuola d'Arda, Italy;* [4]*Donald Danforth Plant Science Center, 975 North Warson Road, Saint Louis, MO 63132 USA*

Epigenetic modifications play a crucial role in plant development and abiotic stress responses.
We performed a whole-genome bisulfite sequencing of proliferating and expanding cells collected from the third developing leaf of *Brachypodium distachyon,* an extremely drought tolerant species, to generate a DNA methylation map of the entire genome. We grew *Brachypodium* plants under a severe drought stress condition that does not negatively affect cell proliferation rate in young leaves (1). BS-Seq libraries of proliferating and expanding leaf cells were generated and sequenced using a 2 X 125 bp strategy with an expected coverage of 20X, with three biological replicates. On average 20% of cytosine residues were found methylated as 5-methylcytosine and strong differences in the three sequence contexts were observed between proliferating and expanding cells both in optimal and drought conditions. We identified 41,532 differentially methylated regions (DMRs) among the developmental gradient and 13,454 DMRs in response to drought. These data are in agreement with what was observed from the transcriptomic profiles of both coding and noncoding genes previously reported (1; 2). The data here presented will contribute to the better understanding of the complex regulatory network underlying leaf cells differentiation and its response to environmental perturbations.

1. Verelst W et al., 2013. Molecular Plant 6: 311–322
2. Bertolini E et al., 2013. Molecular Plant. 6:423–443

P16 DNA methylation variation in barley is driven by both climate of origin and breeding efforts

M. J. DUBIN[1], B. J. HOFINGER[2], B. J. TILL[2,3]

[1]Oxford Software Ltd., 7A Whero Ave, 8971 Diamond Harbour, New Zealand; [2]Plant Breeding and Genetics Laboratory, Joint FAO/IAEA Division, International Atomic Energy Agency, Vienna/International Centre, PO Box 100, 1400 Vienna, Austria; [3]Department of Chromosome Biology, University of Vienna, Dr. Bohrgasse 9, 1030 Vienna, Austria

While strongly influenced by underlying genetic variation, DNA methylation patterns are also sensitive to environment, suggesting that they may have role in environmental adaptation or in the mediation of genome-environment interactions. Supporting this, we previously found that DNA methylation varies with the local environment in wild *Arabidopsis thaliana* populations where it is correlated with changes in gene expression and appears to be under selection (Dubin *et al*., 2015).
To better understand this phenomenon, here we investigate DNA methylation in a worldwide panel of landrace and elite Barley cultivars using whole genome bisulfite sequencing.
No correlation between DNA methylation and habitat (e.g. winter vs spring Barley) or other agronomic traits were apparent, but an inverse correlation between DNA methylation over gene bodies and latitude was observed in both the northern and southern hemispheres. These latitudinal correlations appear to be driven by climate variables and, in particular, winter temperatures. These correlations were more pronounced in elite lines (compared to landraces) and elite lines also had higher overall levels of DNA methylation.

P17 Improved demethylation in ecological epigenetic experiments: testing a simple and harmless foliar demethylation

JAVIER PUY, HANA DVOŘÁKOVÁ, CARLOS P. CARMONA, FRANCESCO DE BELLO, INGA HIIESALU, VÍT LATZEL

Department of Botany, Faculty of Science, University of South Bohemia, Na Zlaté Stoce 1, 370 05, České Budějovice, Czech Republic; Institute of Botany, Czech Academy of Science, Dukelská 135, 379 82, Třeboň, Czech Republic; Institute of Ecology and Earth Sciences, Department of Botany, University of Tartu, Lai 40, 51005, Tartu, Estonia; Institute of Botany, Czech Academy of Sciences, Zámek 1, CZ-252 43 Průhonice, Czech Republic

Experimental demethylation of plant DNA enables testing for epigenetic effects in a simple and straightforward way without the use of DNA sequencing. Plants are commonly demethylated during their germination by the application of agents such as 5-azacytidine (5-azaC). However, this approach causes developmental constrains, especially in the root system, hindering a full comparison with untreated plants. Here we tested a simple alternative method of plant demethylation, designed to overcome the shortcomings of the germinating method. We compared a novel method of demethylating plants by periodical spraying of 5-azaC solution on established seedlings, with the traditional method where seeds were germinated directly in 5-azaC solution. We quantified the amount of methylated DNA and measured various aspects of plant performance. We found that the demethylating efficiency of both methods was comparable but spraying method avoided unwanted impacts on plant growth traits. In conclusion, the regular spraying of 5-azaC solution onto established seedlings surpassed the germination-in-solution method in terms of vigor and fitness of treated plants. This novel method could thus be better suited for experimental studies seeking valuable insights into ecological epigenetics. Furthermore, the spray method can be suitable for clonal species and it opens the possibility of community-level experimental demethylation of plants.

P18 Tomato landraces reveal new natural epigenetic variants

F. FEDI, G. B. SEYMOUR

School of Biosciences, University of Nottingham, Sutton Bonington Campus, Loughborough, UK

Fruits are an essential part of a healthy and balanced human diet which provides mineral and nutrients to prevent human diseases. Tomato is a well-studied model system for fruit development and ripening process and it has a global export value of more than $88 billion. Tomato domestication has contributed to a substantial genetic erosion leading to a limited gene pool to develop new varieties. Using an untapped genetic resource composed of more than 300 tomato landraces we aimed to identify natural epigenetic variations. We selected 8 lines with contrasting fruit shelf life and we identified DNA sequences with altered cytosine methylation using pyrosequencing. The first natural stable epi-variant shows an increase in fruit firmness and increased DNA methylation in the promoter of a gene involved in fruit softening. RIN binding domains are localised in the differentially methylated area and preliminary results show the interaction of the DNA sequence with the transcription factor RIN. We are currently developing transgenic lines with increased and targeted DNA methylation to increase tomato shelf life. The results shows the likely presence of an epimutation which delays fruit softening. This suggests that the phenotypically different fruits could be a consequence of epigenetic mechanism and not of SNPs.

P19 Natural epigenetic diversity in the common grassland plant *Plantago lanceolata*

B. GASPAR[1,2], W. DURKA[2], O. BOSSDORF[1]

[1]Plant Evolutionary Ecology, University of Tübingen, Auf der Morgenstelle 5, 72076 Tübingen, Germany; [2]Department of Community Ecology, Helmholtz Centre for Environmental Research UFZ, Theodor-Lieser-Strasse 4, 06121 Halle, Germany

Within-species diversity is an important driver of population and ecosystem processes such as productivity, stability or invasion resistance. Some evidence from model systems suggests that, even in the absence of genetic variation, within-species diversity in ecologically important traits can also be created by underlying epigenetic variation. However, it is still unknown whether this is relevant in natural populations. Here, we present work based on 60 natural grassland populations of *Plantago lanceolata*, distributed across three regions in Germany, and covering various environmental gradients and different land-use types and intensities. We analysed genetic and epigenetic diversity using amplified fragment length polymorphisms (AFLP) and methylation-sensitive amplification polymorphisms (MSAP). To assess the heritability of epigenetic differences, we used leaf tissue collected directly from the field as well as from the F1 generation grown in a common greenhouse environment. Our first results show that there was only weak but nevertheless significant epigenetic population structure, and that epigenetic variation was generally lower in the greenhouse than in the field, indicating a significant environmentally-induced component of epigenetic variation. However, the observed epigenetic diversity of or epigenetic distances between the studied populations were unrelated to their geographic, genetic or environmental differences.

P20 Water primrose, an invasive aquatic plant: a model for the study of epigenetic mechanisms to the adaptation of terrestrial environment?

J. GENITONI[1], D. RENAULT[2], D. VASSAUX[3], S. MAURY[4] D. BARLOY[1]

[1]Agrocampus Ouest, UMR Ecology and Ecosystem Health, 64 rue de Saint Brieuc, 35 042 Rennes, France; [2]UMR CNRS 6553 EcoBio, 263 Avenue du Gal Leclerc, CS 74205, 35042 Rennes Cedex, France; [3]INRA, UMR Ecology and Ecosystem Health, 64 rue de Saint Brieuc, 35 042 Rennes, France; [4]University of Orléans, LBLGC EA1207 INRA USC 1328, rue de Chartres, 45067 Orléans cedex 2, France

In a context of expansion of invasive species, their ability to adapt to a new environment is crucial. In France, the aquatic invasive plant, *Ludwigia grandiflora,* is recognized as harmful in rivers and its recent dispersion in wet meadow results in depreciation of their fodder values and losses of financial aids for farmers.
Invasive species represent an opportunity to study short-term adapting mechanisms. Epigenetic is a possible source of flexibility that could participate to short-term adaptation and constitutes actually a broad field to explore. In addition, water primrose exhibiting clonal propagation and high phenotypic plasticity between terrestrial and aquatic environments could represent a relevant model to explore the role of epigenetic in short-term adaptation of invasive species. In this context, two morphotypes, respectively living in terrestrial and aquatic environments, were grown with or without a hypomethylating agent: Zebularine. The adaptive response of both morphotypes in emerged and submerged environments through the observations of different morphological traits and their metabolomic responses were carried out. This preliminary step will be followed by a genomic approach involving RNA-seq and MeDIP-seq to identify genes differentially expressed and methylated that could be candidates for adaptive potential.

P21 Climatically induced variation of cytosine methylation patterns across the range of European beech (*Fagus sylvatica* L.)

MATÚŠ HRIVNÁK[1], DIANA KRAJMEROVÁ[1], JOSEF FRÝDL[2], DUŠAN GÖMÖRY[1]

[1]Technical University in Zvolen, Faculty of Forestry, TG Masaryka 24, 96053 Zvolen, Slovakia; [2]Forestry and Game Management Research Institute, Strnady 136. 252 02 Jíloviště, Czech Republic

Methylation-sensitive amplification polymorphism (MSAP) markers were investigated in the international provenance experiment on European beech (*Fagus sylvatica* L.). Two climatically contrasting trial plots (Tale, Slovakia and Zbraslav, Czech republic) were studied represented by twenty and ten provenances, respectively, covering a major part of the range of European beech. Correlations of cytosine-methylation patterns at several loci and overall DNA methylation with climatic conditions of the sites of population origin and budburst phenology were detected. As plants were grown under uniform environments since sowing, this suggests that methylation at particular loci was influenced by the weather or photoperiod at the site of origin, during the embryogenesis or even earlier. Frequencies of methylation patterns at three loci differed between the two trial locations, indicating that the change of methylation induced by the climate of the planting site appeared during the ontogeny. The results suggest that the rules for collection, transfer, and use of forest reproductive materials should also consider epigenetic effects.

P22 Epigenetic modifications in response to environmental constraints: Study of shade avoidance response in *Antirrhinum majus*

D. GOURCILLEAU[1], M. MOUSSET[1], M. LATUTRIE[1], S. MARIN[1], S. MAURY[2], B. PUJOL[1]

[1]CNRS, University of Toulouse 3 Paul Sabatier, Ecole Nationale supérieur de la Formation de l'enseignement Agricole, UMR5174 Laboratoire Evolution et Diversité Biologique, 31062 Toulouse, France; [2]LBLGC EA1207 INRA USC1328 University of Orléans, rue de Chartres, CoST, 45067 Orléans, France

Shade avoidance is a common example of phenotypic plasticity in plants that can be advantageous when there is competition for light and pollination. Its phenotypic syndrome includes elongation of internodes, apical dominance (reduced branching) and early flowering. Our aim is to test whether this shade avoidance response is epigenetically controlled. Natural populations of *Antirrhinum majus* have shown strong stem elongation in response to shade. We conducted greenhouse experiments and analyzed DNA methylation percentages by using HPLC on several inbred lines under different artificial shade treatments in order to answer the following questions: what is the shade avoidance syndrome in *A. majus*? What are its phenotypic components (height, node number, stem diameter, leaf number, SLA and flowering)? Is it associated with changes in methylation patterns? Are these epigenetic changes heritable?

P23 Transcriptional control of immune-responsive genes by active DNA demethylation and its relevance in plant adaptation

T. HALTER, J. WANG, E. LASTRUCCI, D. AMESEFE, A. PEREZ-QUINTERO, A. YU, G. LEPERE, L. BAPAUME, L. NAVARRO

Institut de Biologie de l'Ecole Normale Supérieure, 46 rue d'Ulm, 75005 Paris, France

DNA methylation is an epigenetic mark that silences transposable elements as well as some genes carrying repeats in their vicinity. In Arabidopsis, CG and CHG methylation marks are maintained by MET1 and CMT3, respectively, while CHH methylation is maintained by the RNA-directed DNA methylation (RdDM) machinery. Furthermore, the demethylase ROS1 actively erases DNA methylation in all cytosine sequence contexts. We have previously shown that MET1 and RdDM factors negatively regulate resistance against a virulent strain of *Pseudomonas syringae*, and accordingly, siRNAs and DNA methylation were found to repress the expression of defense genes. By contrast, we demonstrated that *ROS1* positively regulates plant defense against this bacterium and that it actively demethylates a canonical resistance (*R*) gene to ensure its rapid and pervasive induction upon pathogen detection. Here, I will report the dynamics and biological significance of these regulatory processes in the context of antibacterial defense and the extent to which the active demethylase REPRESSOR OF SILENCING 1 (ROS1) controls transcriptional reprogramming during plant defense. In addition, I will provide evidences indicating that modulation of active demethylation activity can fine-tune defense gene expression in nature and has likely evolved to promote plant adaptation to specific environmental conditions. Therefore, this work not only describes the possible mechanism by which active demethylation could control transcriptional reprogramming during plant immunity, but also highlights the ecological relevance of this regulatory process.

P24 Releasing *FLC* back into the wild

JO HEPWORTH[1], REA ANTONIOU KOUROUNIOTI[1], KRISTINA BERGGREN[2], CATJA SELGA[3], REBECCA BLOOMER[1], DEBORAH COX[1*], TORBJÖRN SÄLL[3], SVANTE HOLM[2], MARTIN HOWARD[1], CAROLINE DEAN[1]

[1]John Innes Centre, Norwich, UK; [2]Mid Sweden University, Sundsvall, Sweden; [3]Lund University, Lund, Sweden; [*]Now at Queen's University, Belfast, UK

The integration of information from internal and/or external cues at the epigenetic level is a key process in the control of developmental transitions. In the model plant *Arabidopsis thaliana*, the switch from vegetative to reproductive development is entrained to the seasons by temperature and day length. Photoperiods long enough to induce flowering occur in both autumn and spring and therefore the memory of winter is necessary to distinguish between the two. This process is known as vernalisation and the floral repressor and Polycomb target *FLOWERING LOCUS C* (*FLC)* is its major determinant. Detailed genetic study in standard conditions has shown that constant low temperature results in the transcriptional shutdown and stable epigenetic repression of *FLC*, allowing flowering upon return to inductive photoperiod and temperature conditions. These studies have been very informative in understanding the mechanics of *FLC* regulation, but the relevance of these mechanisms in the field, where Arabidopsis can experience daily temperature fluctuations spanning twenty degrees, remains unknown. Here we exploit this well-characterised genetic mechanism to examine the sensing, integration and memory of a very noisy environmental cue at a single locus. We combine measurement of behaviour in natural environments, genetic analysis and mathematical modelling to provide mechanistic insight into the signal integration behaviour of a Polycomb target. In so doing, we find that instead of averaging the entire temperature profile, different thermosensory mechanisms respond to diverse aspects of the daily temperature cycle. While presence of cool temperatures promotes the shutting down of transcription at *FLC*, the absence of higher temperatures permits the action of Polycomb Repressive Complex in accumulating histone-based memory, separating these two processes, and that they vary among natural populations.

P25 Epigenetic effects of single- and multi-species inoculation of *Arabidopsis thaliana* with two natural pathogens, *Pseudomonas syringae* and *P. viridiflava*

JACOB J. HERMAN, MAXIME POLICARPO, JOY BERGELSON

Department of Ecology and Evolution, University of Chicago, 1101 E. 57th St., Chicago, IL, 60637, USA

Mounting evidence suggests that DNA methylation plays a key role in defense against plant pathogens. When infected with a single pathogen, various plants have been shown to undergo DNA methylation changes that correlate with changes in defense gene expression. However, plants in nature are typically subject to simultaneous infection, potentially involving pathogens that induce counteracting defenses. The effects of such co-infection on genome-wide DNA methylation patterns remain unexplored, as does the extent of host genetic variation for these epigenetic responses. We inoculated 20 *Arabidopsis thaliana* accessions with natural strains of the biotroph *Pseudomonas syringae* and the necrotroph *P. viridiflava*, each isolated from *A. thaliana* in the Midwestern United States. On average, single-inoculation with *P. viridiflava* elicited the most severe disease symptoms, followed by co-inoculation and then single-inoculation with *P. syringae. Arabidopsis* accessions differed significantly in disease symptoms in both single and co-inoculation treatments. I will present whole-genome bisulfite sequencing data from two accessions with contrasting disease symptoms across treatments. These data will reveal the extent to which co-infection alters DNA methylation responses as compared to single inoculations, and will provide first insights into the nature of genetic variation in epigenetic responses to pathogen infection in *A. thaliana*.

P26 Modelling the invasion and maintenance of epialleles in plant populations

L. HUYGHE, D. J. SCHOEN

Department of Biology, McGill University, 1205 avenue du Docteur Penfield, Montreal H3A 1B1, Canada

Epigenetic variation may provide both heritable and non-heritable variation that is triggered by environmental cues. But it is unclear whether such variation contributes to the adaptive potential of natural populations. In plants, there have been a number of reports of what appear to be transgenerational inheritance of environmentally-induced variation. Using a forward simulation approach, we developed a model that captures several key characteristics of plant life-cycle such as the sedentary life-style coupled with limited gamete dispersal by pollen and seed. As proposed in non-plant models, we adopt a two-locus system to depict how the environment acts on epialleles (alleles that can be epigenetically modified). We investigated under which set of parameters such epialleles could be maintained in a typical plant population with spatial heterogeneity and selection. We find that the balance between migration rate (pollen and seed), strength of selection, and cost of switching between epialleles is important for the maintenance of epialleles.

P27 Epigenetic regulation of heritable immune priming in Arabidopsis

R. JAIN, L. FURCI, D. ROQUIS, O. BERKOWITZ, V. COLOT, F. JOHANNES, J. TON

Plant Environmental Signalling Group, Department of Animal and Plant Sciences, University of Sheffield, Western Bank, Sheffield, S10 2TN, UK

Diseased Arabidopsis plants are capable of epigenetically priming defence responses enhancing resistance against biotrophic pathogens, such as *Hyaloperonospora arabidopsidis* (*Hpa*). This immune priming can be mimicked by mutations affecting DNA methylation. DNA (de)methylation is known to regulate nearly half of the defence-related transcriptome in Arabidopsis. Here, we analysed a population of 123 epigenetic recombinant inbred lines (epiRILs) for basal resistance to *Hpa*. These lines are isogenic, but vary in DNA hypo-methylation inherited from the *ddm1-2* mutant, which results in intense demethylation of transposable elements (TEs) in heterochromatic regions. These epiRILs show a spectrum of resistance, ranging from no resistance to near complete levels of resistance to *Hpa* without plant growth defects, unlike the parent *ddm1-2* mutant. Correlation analysis between *Hpa* resistance and epigenetic markers identified four statistically significant epiQTLs. Genome wide RNA-seq analysis of selected resistant epiRILs after *Hpa* infection confirmed that their resistance is associated with primed gene clusters that are strongly enriched for defence-related gene ontology terms. Surprisingly, however, almost all hypo-methylated genes with a primed expression profile within the epiQTL intervals were genes annotated as TEs. Based on these results, we propose that DDM1-targetted TEs in the resistance epiQTLs *trans*-regulate transcriptional responsiveness of distant defence genes.

P28 Epigenetic variation in seagrass clones

A. JUETERBOCK[1], C. BOSTRÖM[2], T.B.H. REUSCH[3], J.L. OLSEN[4], A. DHANASIRI[1], I. SMOLINA[1], S. ARNAUD-HAOND[5], G. HOARAU[1]

[1]Marine Molecular Ecology Group, Faculty of Biosciences and Aquaculture, Nord University, Universitetsalleen 11, 8026 Bodø, *Norway; [2]Department of Biosciences, Environmental and Marine Biology, Åbo Akademi University, Artillerigatan 6 FI-20520 Åbo, Finland; [3]GEOMAR Helmholtz-Centre for Ocean Research Kiel, Evolutionary Ecology of Marine Fishes, Düsternbrooker Weg 20, D-24105 Kiel, Germany; [4]Ecological Genetics-Genomics Group, Groningen Institute of Evolutionary Life Sciences, University of Groningen, 9747 AG Groningen, The Netherlands ; [5]Ifremer, Station de Sète, Avenue Jean Monnet, CS 30171, 34203 Sète Cedex, France*

Evolutionary theory predicts that low genetic variation reduces a population's ability to cope with environmental variability and to adapt to changing environments. However, the evolutionary success of >1000-year old clones of the seagrass *Zostera marina* challenges the direct relationship between genetic diversity and adaptation potential. We aim to test the hypothesis that epigenetic variation, a hitherto overlooked layer of evolutionary relevant variation, is the key to this paradox. Our main objective is to describe the spatial pattern of epigenetic variation in a ~1000-year old seagrass meadow of low genetic diversity from the Åland Islands, northern Baltic Sea. In 2015, we sampled 100 shoots along a transect of 250 meters. We screened within-clone variation of cytosine methylation using MethylRAD sequencing. Results on the first eight sequenced samples indicate that genes are differentially methylated between shoots and that epigenetic variation is not correlated with spatial distance in this predominantly clonal meadow. Each shoot (ramet) was assigned to its clonal origin (genet) based on its multi-locus microsatellite genotype, allowing to characterize the interplay between genetic and epigenetic structure. This work is a first step to uncover whether epigenetic variation may compensate for the absence of genetic variation in seagrass clones.

P29 Genome-wide analysis of DNA methylation and transcriptional trends in fertile hybrid and inbred lines of Pigeonpea

ALIM JUNAID, KISHOR GAIKWAD

[1]National Research Centre on Plant Biotechnology, LBS Centre, Pusa campus, New Delhi, 110012, India

DNA methylation is an important heritable landmark that confers epigenetic changes in hybrids and has fascinated biologists and plant-breeders over the years. Although occurrence of epigenetic changes have been documented in rice and maize hybrids, such investigations have not been reported in Pigeonpea. Here we present genome-wide CG, CHG and CHH methylation profiles of pigeonpea-inbred lines (11A and 303R) and their F1 hybrid at single base resolution. We found that methylation levels in sterile line (11A) were higher than fertile hybrid and fertile parent (303R). Furthermore, identification of differentially methylated regions (DMRs) and analysis of their methylation levels revealed remarkable differences between methylation patterns in sterile parent and fertile hybrid. By comparing whole transcriptome data with complete genome bisulfite sequencing data, we identified several DMRs associated with differentially expressed genes that are known to play a role in fertility restoration. Further, inbred parents and F1 hybrid displayed a negative correlation between gene expression and CG methylation. Lastly, since lncRNAs are known to regulate fertility, we also identified lncRNAs in Pigeonpea and analysed their methylation patterns. Over all, our results provide an understanding of epigenetic changes and their role in regulating gene expression in Pigeonpea hybrids from genome-wide point of view.

P30 Ants and plants: specific host epigenetic responses in a multispecies mutualism

REBECCA Y. KARTZINEL[1,2], BRIDGETT M. VONHOLDT[1], CORINA E. TARNITA[1], ROBERT M. PRINGLE[1]

[1]Department of Ecology and Evolutionary Biology, Princeton University, Princeton, NJ 08544, USA; [2]Department of Ecology and Evolutionary Biology, Brown University, Providence, RI 02912, USA

Understanding the factors that stabilize mutualistic symbioses has long been a central goal in evolutionary ecology. In multispecies mutualisms, organisms must be able to rapidly adjust to different partner species that impose different costs and benefits - especially when mismatches in longevity mean that one side will necessarily experience partner turnover during its ontogeny. We investigated epigenetic changes in ant-plants (*Acacia drepanolobium*) associated with occupancy by each of its four protective ant mutualists. Using reduced-representation bisulfite sequencing, we show that rather than having a universal epigenetic signature of ant occupancy, each ant species induces unique DNA methylation patterns in host trees. These methylation signatures vary both in nature (genes affected) and extent (total number of sites affected). Results of functional analyses are consistent with our ecological knowledge of the mutualism, e.g. ants requiring high energy inputs from host trees disproportionally affect genes related to photosynthesis and metabolism. We also identify several genes, strongly affected by ant occupancy, related to pathogen defense and abiotic stress that provide novel insights into the ant-plant interaction. This work complements our extensive ecological knowledge of mutualism by providing a first look at the molecular basis of host response to complex ecological interactions in natural settings.

P31 Epigenetic gene-regulation under Al stress in cotton plant (*Gossypium hirsutum* L.) growth and development

ATREYEE KUNDU[1], MARKKANDAN GANESAN[1]

[1]Department of Life Sciences, Presidency University, Kolkata 700073, West Bengal, India

Worldwide aluminium (Al^{3+}) toxicity is one of the major factors that limit plant growth and yield in acid soils. Al^{3+} induced exudation of organic acids (citrate and malate) into the rhizosphere from the root has been identified as a major Al^{3+}-tolerance mechanism in many plant species. Based on our preliminary experiments, we observed that the cotton plants displayed very sensitive phenotypes under high Al-$^{3+}$ and low pH stress conditions. With reference to Arabidopsis system, we cloned the key genes responsible for Al^{3+} tolerance in cotton. Based on the homology, we successfully cloned cotton *MATE*, *ALS3*, *STOP1* and *STAR*. The genes were isolated using polymerase chain reaction-based cloning techniques. The expression profile studies will be discussed in the presentation. Recently epigenetic gene regulation has been well studied and this wide ranging system is related to adaptation for various environmental stresses in plants via methylations of DNA, RNA and histones as well as other modifications of histones. Studies suggested the existence of *AvSAMS1*-related epigenetic gene-regulation under Al stress. Based on the above reasons, we are aimed to introduce *AvSAMS1*, into *Gossypium hirsutum* to investigate its biological functions by *Agrobacterium* mediated gene transfer method. The transgenic plants are expected to show existence of *AvSAMS1*-related epigenetic gene-regulation under Al stress and leading to increased tolerance to Al^{3+} stress.

P32 Epigenome role in the pathogenesis of *Alternaria brassicicola*

ANTHONY KWASIBORSKI, FRANCK BASTIDE, JUSTINE COLOU, PASCAL POUPARD, PHILIPPE SIMONEAU, THOMAS GUILLEMETTE

IRHS-Fungisem, UMR1345 University of Angers-INRA-Agrocampus Ouest, 42 rue Georges Morel 49071 Beaucouzé cedex, France

Alternaria brassicicola is the causing agent of black spot disease in the *Brassicaceae*. This pathogenic fungus is able to transmit itself on and by seeds. Then it has developed resistance mechanisms against hydric, osmotic and osmolar stresses.
Epigenetic mechanisms and miRNAs are hereditary modifications of gene activities without mutations in the DNA sequence and show an implication in the virulence and plant defense molecules response of pathogen fungi. In *A. brassicicola*, 2 methyltransferases (abDIM-2, abDIM-5) and 2 endoribonucleases (abDICER-1, abDICER2) have been identified.
In this study, we investigated the implication of epigenetic mechanisms on the virulence and the seed transmission of *A. brassicicola*.
Phenotyping of abDICERs and abDIMs deleted mutants didn't show involvement of epigenetic mechanisms in their growth, sporulation, virulence or susceptibility against plant defense molecules. However, DIM mutants are able to transmit better to seeds due to an increased resistance to hydric and osmotic stresses. In parallel, small RNAs sequencing from wild-type and DICER mutant identified 17 miRNAs in *A. brassicicola*. The synthesis of these miRNAs is not influenced by DICER proteins suggesting the possible existence of an alternative DICER-independent miRNA synthesis pathway. Moreover, *A. brassicicola* miRNAs expression is not modified by the presence of *Brassicaceae* defense molecules.

P33 Parental effects that cause home-site advantage under natural conditions are associated with 20-fold change in GC-methylation

CHRISTIAN LAMPEI[1], BIANCA DÖßELMANN[1], STEFAN SCHOLTEN[2], KARL J. SCHMID[1]

[1]University of Hohenheim, Institute 350b, Fruwirthstrasse 21, 70599 Stuttgart, Germany; [2]Biozentrum Klein Flottbek, Entwicklungsbiologie und Biotechnologie, Ohnhorststr. 18, 22609 Hamburg, Germany

Transgenerational plasticity is increasingly recognized as an important source of phenotypic variation, affecting potentially adaptation to environmental change. It was postulated that epigenetic mechanisms are driving this plastic response. We cultivated 16 diverse genotypes in two sites at different altitudes (400 m and 700 m) for two years. In the third year, the offspring were reciprocally transplanted revealing strong differences in fitness (biomass as proxy) due to the parental environment. On average, we discovered the classical home-site advantage suggesting adaptation to the parental environment caused by the parental experience of the site – not by selection. However, these parental effects were strong in some genotypes, accompanied with up to 30 days flowering time differences, but absent in others. We used this difference and sequenced the methylome of two genotypes with the same parental effect and two genotypes without parental effects. We found that differences in GC-methylation between offspring of plants from 400 m and 700 m respectively, were 20 fold higher in genotypes with parental effects. In these genotypes, genes with differentially methylated regions (DMRs) were enriched for more than 100 Gene-Ontology classes with 'Gene silencing by RNA', 'histone modification', 'macromolecule methylation', 'covalent chromatin modification', 'gene silencing' among the top 10.

P34 Unique and universal microRNAs mediating plastic internode elongation in the amphibious species *Alternanthera philoxeroides* in contrasting hydrological habitats

GENGYUN LI, YING DENG, YUPENG GENG, LEXUAN GAO, JI YANG

Ministry of Education Key Laboratory for Biodiversity Science and Ecological Engineering, Fudan University, Shanghai 200438, China

Amphibious plant can successfully withstand variable water regimes from wet land to dry land via eco-devo process that cause wide variety of anatomical, morphological and physiological responses to submergence. However, little is known about how microRNAs, which regulate gene expression at post-transcriptional level, participate in this process. In this study, a time course experiment was processed by using high-throughput sequencing technology to characterize the dynamic expression pattern of flooding responsive microRNAs in amphibious plant *Alternanthera philoxeroides*. In total, 128 known and 18 novel microRNAs differentially expressed under contrasting hydrological habitats, most of them present a temporal regulation pattern and response quickly to sudden flooding. The plasticity of *A. philoxeroides* was detected at microRNAome level, several microRNAs presented a diverse expression pattern in contrasting hydrological habitats. In addition, functional analysis of target gene indicate microRNAs involved in plastic adaptive phenotypic plasticity by regulate adaptive trait adjustment related genes. This result provide knowledge to the role microRNAs played in plastic eco-devo process and highlight the importance of time course experiment in stress responsive microRNAs researches.

P35 Epimutations control *Arabidopsis* response to *Plasmodiophora brassicae*

B. LIEGARD[1], A. GRAVOT[1], V. BAILLET[2], J. BENEJAM[1], C. LARIAGON[1], J. LEMOINE[1], V.COLOT[2], M.J. MANZANARES-DAULEUX[1], M. JUBAULT[1]

[1]AGROCAMPUS OUEST, INRA, Université Rennes 1, UMR 1349 IGEPP, F-35653 Le Rheu, France; [2]Institut de Biologie de l'Ecole Normale Supérieure, Centre National de la Recherche Scientifique, Institut National de la Santé et de la Recherche Médicale, Ecole Normale Supérieure, Paris, France

Plant phenotypic variations are classically explained by the presence of genomic sequence variations in the causal locus. Currently very few cases of phenotypic variations due to the presence of epimutational variations were described in plant. Two regions involved in Arabidopsis response to clubroot, a major disease of Brassica crops caused by the obligate protist *Plasmodiophora brassicae*, were identified in our laboratory as potentially regulated by epimutations. The first region was detected in the *ddm1-2* x Col-0 epiRIL population as well as in natural accessions by linkage analysis and genome-wide association analysis. In this region, two potentials Differentially Methylated Regions were identified in epiRIL population as well as in natural accessions: the first one showed constitutive methylation variations according to clubroot phenotype whereas the second one showed methylation variations induced by infection. The second region, identified by QTL fine mapping, is reduced to two genes which displayed constitutive differential methylation state and gene expression, correlated with clubroot response. These results revealed thus the implications of natural epimutations in the variations of plant response to clubroot. Combination of favorable epiallelic variations to allelic ones could therefore constitute new opportunities in plant breeding.

P36 The epigenetic basis of plant immunity

A. LÓPEZ SÁNCHEZ, R. JAIN, J. STASSEN, L. FURCI, J. TON

Animal and Plant Sciences Dpt, The University of Sheffield, Western Bank, Sheffield S102TN, UK

Plants can prime their immune system after perception of certain environmental signals, leading to a more efficient response to future stresses. Our laboratory previously demonstrated that progenies of disease-exposed Arabidopsis display a primed defence maintained over 2 stress-free generations. Interestingly, DNA methylation is the predominant mechanism responsible for inheritance of epigenetic traits, infections by *P. syringae* induce genome-wide DNA hypomethylation and mutants defective in RNA-directed DNA methylation (RdDM) show constitutive priming, suggesting a role of DNA methylation in the inheritance of immune responsiveness. Therefore, our overarching objective is to investigate how plants epigenetically adjust their immune system to adapt to long-term hostile environments. To address this question we:
1) Genetically dissected the RdDM-dependent DNA methylation and ROS1-dependent de-methylation pathways to assess their interplay in plant immunity (recently published).
2) Analysed the methylome of progenies from stress-exposed Arabidopsis to elucidate the role of DNA methylation in transgenerational priming (unpublished).
3) Studied transgenerational priming against different biotic and abiotic stresses to assess the ecological and evolutionary relevance of transgenerational traits (unpublished).

P37 Prefoldin complex could be a new player in chromatin remodeling in Arabidopsis

C. MARÍ-CARMONA, D. ESTEVE-BRUNA, N. BLANCO-TOURIÑÁN, M.A. BLÁZQUEZ, D. ALABADÍ

Instituto de Biología Molecular y Celular de Plantas (IBMCP), Ingeniero Fausto Elio, s/n 46022 Valencia, Spain

Prefoldin (PFD) is an evolutionarily conserved heterohexameric chaperonin that delivers unfolded tubulin to the main cytosolic chaperone CCT (Siegert *et al*., 2000). This function takes place in the cytosol, but it has been shown that when DELLA proteins are accumulated, PFD moves to the nucleus, impairing tubulin folding and microtubules arrangement (Locascio *et al*., 2013). This DELLA-dependent localization also opens the possibility that PFD may also perform a role in the nucleus.
An in silico analysis of the public interactomes from yeast and flies has allowed us to identify numerous putative nuclear interactors for PFD subunits in Arabidopsis. We have demonstrated that some of these interactions are conserved, suggesting new roles for the PFD complex in plants. In particular, the interaction with different subunits of SWR1 complex suggests that PFD might control gene expression by modulating the deposition of the histone variant H2A.Z, a hypothesis that is being challenged by genomic approaches.

P38 DNA methylation and tree phenotypic plasticity in a context of global changes

S. MAURY[1], A.-L. LE GAC[1], M.D. SOW[1], A. DELAUNAY[1], R. FICHOT[1], F. BRIGNOLAS[1], D. CONDE[2], I ALLONA[2], S. LANCIANO[3], M. MIROUZE[3], C. GRUNAU[3], V. SEGURA[4], M.-A. LELU-WALTER[4], V. COURDAVAULT[5], J. TOST[6], S. STRAUSS[7]

[1]*LBLGC EA1207 INRA USC1328 University Orléans, rue de Chartres, CoST, 45067 Orléans, France;* [2]*Centro de Biotecnologia y Genomica de Plantas, Universidad Politecnica de Madrid Campus de Montegancedo UPM, 28223 Madrid, Spain;* [3]*IHPE, University Perpignan, IRD 58 Avenue Paul Alduy, 66860 Perpignan, France;* [4]*AGPF, INRA, 45160, Ardon, France;* [5]*BBV, University Tours, Parc de Grandmont, 37200 Tours, France;* [6]*LEE, CEA, 2 rue Gaston Crémieux, 91057 Evry, France ;* [7]*Forest Ecosystem and Society, University Oregon, 338 Richardson Hall, Oregon, USA*

Global climate changes in progress will impact forest productivity notably through reduced water availability and heat periods. One possibility to adapt is phenotypic plasticity for which epigenetic mechanisms are proposed to be a main source of flexibility. Our objective is to evaluate the potential of DNA methylation to significantly participate to phenotypic plasticity in trees, fixed and perennials organisms with major ecological roles. Over the 10 last years, using an integrative approach with ecophysiological, biochemical, transcriptomics, epigenomics (MeDIP, WGBS, Mobilome) and reverse genetics (RNAi lines) tools, we were able to dissect in the shoot apical meristem (center of the shoot morphogenesis) the response of trees to environmental variations. This work was assessed in distinct experimental set-ups from greenhouse to field plantations as well as during the stress or months post-stress. Our data (recently published and unpublished) showed that Differentially Methylated Regions (DMRs) are associated to active TE and differentially expressed genes with biological functions related to stress response and phytohormone signaling. Altogether, our data proposed that DNA methylation is a source of flexibility associated to phenotypic plasticity in trees opening perspectives for tree breeding.

P39 Effects of genetic diversity and epigenetic change on trait variation in the foundation plant *Spartina alterniflora*

J. M. MOUNGER, C. L. RICHARDS, A. R. HUGHES, C. A. GEHRING, M. H. ROBERTSON, S. VOORS

Department of Integrative Biology, 4202 East Fowler Ave, SCA110 Tampa, FL 33620, USA

While ecological genetics approaches have informed us about the structure of genetic diversity in natural populations, we still know surprisingly little about the mechanisms that permit organisms to adapt to variable environmental conditions. Using MS-AFLP, our previous work showed a weak, but significant correlation of epigenetic variation with habitat in the salt marsh foundation plant *Spartina alterniflora*. In this study, we used the more powerful genomics approach of epigenotyping-by-sequencing (epiGBS) to examine differential methylation polymorphisms (DMPs) and single-nucleotide polymorphisms (SNPs) to investigate genetic and epigenetic diversity in natural populations across salinity gradients, and from a reciprocal transplant study of *S. alterniflora*. In addition, we used sequencing of the ribosomal ITS region to identify variation in fungal endophyte communities, which may be particularly beneficial under stressful environmental conditions. We will present DMPs and SNPs, and associated candidate genes that are correlated with trait response, environmental gradients and symbiotic relationships in this critical coastal species.

P40 Transgenerational effects in plant interactions: does parental competition improves offspring adaptability?

JAVIER PUY[1], HANA DVOŘÁKOVÁ[1], INGA HIIESALU[2], FRANCESCO DE BELLO[1,3], VÍT LATZEL[4], CARLOS P. CARMONA[1,2]

[1]Department of Botany, Faculty of Science, University of South Bohemia, Na Zlaté Stoce 1, 370 05, České Budějovice, Czech Republic; [2]Institute of Ecology and Earth Sciences, Department of Botany, University of Tartu, Lai 40, 51005, Tartu, Estonia; [3]Institute of Botany, Czech Academy of Science, Dukelská 135, 379 82, Třeboň, Czech Republic; [4]Institute of Botany, Czech Academy of Sciences, Zámek 1, CZ-252 43 Průhonice, Czech Republic

Heritable epigenetic mechanisms enable plants to remember their former environment, adjust their phenotype, and transmit the acquired knowledge to their offspring, promoting local adaptation. While such transgenerational effects have been demonstrated in response to different abiotic, the role of biotic interactions mediating transgenerational effects is still largely unknown. Here we test whether the effect of parental plant–plant competition could influence offspring performance and adaptability. We conducted an experiment where genetically identical individuals of *Taraxacum brevicorniculatum* were grown, for one season, alone or in competition with different species and itself. Subsequently, we conducted another experiment where the offspring from the first experiment were grown experiencing the same or different competition than the mother. We analyzed the relationship between the intensity of competition on parental generation and the offspring performance. Additionally, we analyzed if the intensity of competition suffered changed between generations depending on the parental condition. The results show that parental competition affected offspring performance beside the prevailing effect of the present competition. More interestingly, offspring suffered less than parents with higher level of competition experienced by the parental plant. Our experiment suggests that competition can trigger transgenerational epigenetic adjustments that could influence plant–plant interactions and local adaptation.

P41 Measuring adaptive potential in British populations of Ash (*Fraxinus excelsior*): genetic differences and phenotypic plasticity in leaf spring phenology

C. ROSIQUE-ESPLUGAS[1,2,3], J. COTTRELL[2], S. CAVERS[3], R. ENNOS[1]

[1]Institute of Evolutionary Biology, Ashworth laboratories, Kings Buildings, Charlotte Auerbach Road, Edinburgh EH9 3FL, Scotland, UK; [2]Forest Research, Northern Research Station, Roslin, Midlothian EH25 9SY, Scotland, UK; [3]Centre for Ecology & Hydrology, Bush Estate, Penicuik, Midlothian EH26 0QB, Scotland, UK

Rapid climate change is a significant threat to the long-term persistence of native tree populations. Concern has been expressed that tree populations might fail to adapt due to rate of change, insufficient adaptive variation in tree populations and limits to dispersal. In contrast, others have contended that most tree species have high phenotypic plasticity, maintain high levels of within-population genetic variation and exhibit effective gene dispersal capability, all characteristics which should enable an adaptive response. To assess the potential for adaptive mitigation in forest tree populations we need to focus on quantifying patterns of adaptive genetic variation and the likely response of such variation to environmental change.
Assessments of variation in multiple adaptive traits have been conducted within comprehensive experimental provenance trials of common ash (*Fraxinus excelsior*) across Great Britain. We compared spring leaf phenology (bud flush) in 42 different ash populations in two contrasting experimental sites which find significant genetic differences among populations and high levels of phenotypic plasticity. The genetic variation including phenotypic plasticity is crucial to the survival of the forest species, together with epigenetics, which plays as well a very important role in long-term adaptation. These results contribute to the understanding of the adaptive capability of long-lived trees to the changing environment.

P42 Plasticity of hypomethylated poplar clones under drought stress

MAMADOU DIA SOW[1], ANNE-LAURE LE GAC[1], SOPHIE LANCIANO[2], ALAIN DELAUNAY[1], ISABELLE LE JAN[1], JORG TOST[3], VINCENT SEGURA[4], CRISTIAN CHAPARRO[5], CHRISTOPH GRUNAU[5], RÉGIS FICHOT[1], MARIE MIROUZE[2], FRANCK BRIGNOLAS[1], STEEVE STRAUSS[6], STÉPHANE MAURY[1]

[1]LBLGC EA1207, INRA, Université d'Orléans, USC 1328, 45067, Orléans, France; [2]IRD, UMR 232 DIADE, University of Perpignan, Laboratory of Plant Genome and Development, France; [3]Laboratory for Epigenetics and Environment, Centre National de Génotypage, CEA-Institut de Génomique, Evry, France; [4]AGPF, INRA, UMR 588, 45160, Ardon, France; [5]IHPE, Université de Perpignan, UMR 5244, 66100, Perpignan, France; [6]Department of Forest Ecosystems and Society, Oregon State University, Corvallis, OR 97331-5752, USA

As sessile organism, plants have constantly to evolve and develop mechanisms enabling them to adapt and to survive for the incoming climatic changes. Trees as perennials organisms are particularly relevant models to study the effects of repeated cycles of environmental constraints such as drought. Epigenetic mechanisms represent a valuable resource since they can be triggered by the environment conditions. Because of its stability, roles in gene expression, transposon silencing and more generally development and response to stress and facility to study at the genomic levels, DNA methylation is actually the most studied epigenetic marks. However, evidences for its role in tree phenotypic plasticity are still lacking (Bräutigam *et al*., 2013; Plomion *et al*., 2016). In this context, we develop a study on DNA methylation at the genomic level in poplar, a model tree with sequenced genome, submitted to drought and we particularly focused on the shoot apical meristem (SAM), which is the center of shoot morphogenesis (Gourcilleau *et al*., 2010; Lafon-Placette *et al*., 2013; Bastien *et al*., 2015; Le Gac, 2017; Lafon-Placette *et al*, submitted). In order to assess the role of DNA methylation variations previously observed in the poplar SAM, we used control and RNAi clones of *Populus tremula x alba* (*ddm1-15* [B] and *ddm1-23* [D]) obtained by downregulation of the two orthologous of the *Arabidopsis thaliana DDM1* genes (Zhu *et al*., 2013) grown under two watering regimes: Well-Watered (WW) and Water-Deficit followed by ReWatering (WD-RW). An

exhaustive phenotyping was realized on these plants showing that hypomethylated trees show a modify response to changes in water ability. To explain this phenomenon, Differentially Methylated Regions (DMRs) were identified using Whole Genome Bisulfite Sequencing (WGBS) while a mobilome-seq was performed in order to identify the extrachromosomal forms of TEs reflecting the activation of these repetitive elements. Altogether, our data proposed that DNA methylation play a role in the orchestration of the SAM response during variations of water availability to ensure developmental plasticity.

P43 Drought-induced histone modifications in barley

A. TEMEL, K. HUMBECK

Institute of Biology, Martin Luther University Halle-Wittenberg, Weinbergweg 10, 06120 Halle, Germany

Binding of several groups to histone proteins specifies expression of genes. The effects of histone modifications in cell cycle, centromere formation and senescence have been shown in different plant species including barley. However, it is not clear in barley how histone modifications affect gene expression under drought, a major type of stress. In this study, 5 day-old barley (*Hordeum vulgare* cv. Carina) plants were subjected to progressive drought by withholding water for 22 days. Expression of *hsp17* gene, a drought tolerance-associated gene in barley, increased after 12 days of drought. Levels of several histone modifications (H3, H3K4me3, H3K9ac, H3K9me2) in 4 regions of *hsp17* gene and house-keeping (*actin* and *pp2a*) and silent genes (*bm9* and *cereba*) were analyzed by Chromatin Immunoprecipitation followed by Real-time PCR. *hsp17* gene expression showed a gradual increase that was prominent after 12 days of drought. Drought affected H3, H3K4me3, H3K9me2 but not H3K9ac levels. Drought-induced changes in H3, H3K4me3, H3K9me2 levels depended (interaction) on the gene region. Transcription status of housekeeping (*actin* and *pp2a*) and silent genes (*bm9*, *cereba*) were not altered by epigenetic marks. H3K4me3 may be a critical mark for the induction of a gene.

P44 Evidence for rapid evolution in a grassland biodiversity experiment

S.J. VAN MOORSEL[1], M.W. SCHMID[1], C.A.M. WAGEMAKER[2], T. VAN GURP[2], B. SCHMID[1], P. VERGEER[2,3]

[1]Department of Evolutionary Biology and Environmental Studies, University of Zurich, Zurich, Switzerland; [2]Department of Experimental Plant Ecology, Radboud University Nijmegen, Nijmegen, Netherlands; [3]Plant Ecology and Nature Conservation Group, Wageningen University, Wageningen, Netherlands

In long-term grassland biodiversity experiments biodiversity effects commonly increase with time, but the underlying mechanisms are still unclear. Previously it was shown that within one species, differential selection in monoculture and mixed species grassland communities could lead to the rapid emergence of monoculture and mixture types (Zuppinger-Dingley *et al*. 2014). We hypothesize that in biodiversity experiments pre-adapted genotypes or epigenetic variants could be sorted out from the standing genetic or epigenetic variation (Bossdorf *et al*. 2008).
To test if biodiversity acted as a selective environment, we grew offspring from plants grown for twelve years in monocultures or mixtures of such a biodiversity experiment under controlled greenhouse conditions. Using epiGBS (van Gurp *et al.* 2016), a novel method assessing genetic and epigenetic diversity at once, we could show that populations originating from monocultures and mixtures were genetically distinct and that epigenetic variation mostly followed the underlying genetic variation. This pattern was consistently observed across different plant species. These results suggest that, in perennial grassland species, selection of genetic variation underlies the rapid emergence of monoculture and mixture types.

P45 Transgenerational arbuscular mycorrhizal fungal effects are mediated by changes in DNA methylation levels

S. VARGA, C. D. SOULSBURY

School of Life Sciences, University of Lincoln, LN6 7TS, Lincoln, United Kingdom

Most land plants grow in association with arbuscular mycorrhizal fungi (AMF) in their roots and these fungi can cause transgenerational effects (TE) on plants' offspring. Such impacts may be caused by changes in DNA methylation of the offspring. In this study, we compared the amount of global DNA methylation in seeds of the gynodioecious plant *Geranium sylvaticum* in relation to the gender and the AMF status of the parents producing the seeds. We quantified DNA methylation from seeds produced by females or hermaphrodites plants that were originally inoculated with *Claroideoglomus claroideum*, *Glomus hoi* or left non-inoculated, and fertilised by pollen from these AMF treatments. The amount of DNA methylated was positively related to seed mass. Additionally, we observed a sex-AMF specific interaction in the amount of global DNA methylated. Seeds produced by females had a similar proportion of methylated DNA regardless of the AMF status of the father siring the seed; whereas in seeds from hermaphrodites, differences in DNA methylation between AMF fathers occurred. We show for the first time that AMF can have paternal TE via DNA methylation and that these changes are further dependent on the gender of the mother producing the seeds.

P46 Adaptation to climate change: Evidence for epigenetically based transgenerational phenotypic effects

P. VERGEER[1], C.A.M. WAGEMAKER[2], T.P. VAN GURP[3], J.P. GRIME[4], J.D. FRIDLEY[5]

[1]Plant Ecology and Nature Conservation Group, Wageningen University, Wageningen, the Netherlands; [2]Experimental Plant Ecology, Radboud University Nijmegen, Nijmegen, the Netherlands; [3]Netherlands Institute of Ecology, Wageningen, the Netherlands; [4]Department of Animal and Plant Sciences, University of Sheffield, Sheffield, United Kingdom; [5]Department of Biology, Syracuse University, Syracuse, NY, USA

Transgenerational phenotypic effects are increasingly recognized as an important source of phenotypic variation. In addition, they are postulated to play a role in adaptation to environmental change. However, experimental evidence is scarce.
At Buxton Climate Change Impacts Laboratory (BCCIL), a semi-natural grassland has been exposed to almost 20 years of simulated climate change including drought, additional rainfall, and winter warming treatments. *Scabiosa columbaria*, a short-lived perennial, has proven to be highly resistant to these climatic manipulations.
Offspring from established plants that received long-term summer drought, additional summer rain and control treatment at BCCIL were used to set up a drought-stress experiment. Plant performance was measured as well as genetic and epigenetic variation using next-generation sequencing to test for genetic and/or epigenetic signatures of selection.
F2-generation plants from grandmothers that received summer drought treatments grew significantly larger in the drought treatment compared to plants from grandmothers that received control or additional summer rain treatments, illustrating transgenerational inheritance of adaptive responses to drought. No genetic differentiation, but patterns of epigenetic differentiation were observed in response to climatic manipulations. The heritability of these epigenetic changes may have contributed to the stability of *Scabiosa columbaria* through almost 20 years of climatic manipulations.

P47 Bud burst timing in clonal epitypes of *Picea abies* reflects a stable epigenetic memory set up during embryogenesis

MARCOS VIEJO[1], ELENA CARNEROS[2], HUGH CROSS[2], IGOR YAKOVLEV[2], CARL GUNNAR FOSSDAL[2], JORUNN E. OLSEN[1]

[1]Norwegian University of Life Sciences (NMBU), Department of Plant Science (IPV), Ås, Norway; [2]Norwegian Institute of Bioenconomy Research (NIBIO), Ås, Norway

Bud burst timing in *P. abies* is a trait affected by the temperature conditions during zygotic embryogenesis through the establishment of a long-lasting epigenetic memory. Similarly, during somatic embryo production different temperatures lay down an epigenetic memory generating warm (WE) and cold epitypes (CE) which display different timing of autumnal bud set in the regenerated seedlings.
In spite of the accumulated knowledge, little is known about the persistence of the epigenetic memory from a molecular perspective and its reflection in the later phenology of the epitype trees. To shed light on this, detailed spring bud burst stages were monitored in 15-year old clonal WE and CE individuals growing in field in Ås, Norway (69°N latitude). Furthermore, a targeted transcriptomic analysis of microdissected mother cells and leaf primordia in addition to whole bud sections from apical buds from the first whorl was conducted on materials harvested in early spring several weeks before bud burst.
The analysis of phenology not only showed differential timing of bud swelling and bud burst in the epitypes with the CE being advanced development relative to the WE, but also confirmed a long-lasting effect of the epigenetic memory. The transcriptomic data and Gene Ontology terms showed distinct cell-type upregulation of transcripts related to light and temperature signalling in the CE. Besides, integration of external and internal cues through modulation of chromatin and thus transcription is likely cell-type specific for genes involved in DNA methylation and histone modifications (H3K9me, H3K27me or H3S10p). Consistent with advanced preparation for bud burst, the CE showed upregulation of cell cycle-related genes whereas hormone signalling pathways showed cell type-specific upregulation in the WE, probably reflecting its delayed bud burst-preparation. These results

demonstrate that the epigenetic memory established during embryogenesis leads to differential bud burst timing in the epitypes through cell type-specific transcriptomes.

P48 Environmental maternal effects on the morphology, physiology and interactions of *Eucalyptus grandis*

M. VIVAS[1], M. KEMLER[1,2], V. ROLO[3], M. J. WINGFIELD[1], B. SLIPPERS[1]

[1]Department of Genetics, Forestry and Agricultural Biotechnology Institute (FABI), University of Pretoria, Cnr Lynnwood and University Road, 0083 Hatfield, Pretoria, South Africa; [2]Geobotany, Ruhr-University Bochum, Universitätsstrasse 150, 44780 Bochum, Germany; [3]Indehesa Institute, University of Extremadura, Av. de España 43, 10600 Plasencia, Spain

The maternal environmental 'experience' of plants can influence their offspring without changes to their DNA sequence. This study considers the environmental maternal effects on the morphology, physiology and resistance of *Eucalyptus grandis* seedlings, and their impact on fungal communities in the leaves of the seedlings. Seeds were collected from two *E. grandis* clonal seed orchards, with different environmental conditions, and grown in a common garden. Seedling morphology, physiology and response to a pest and a pathogen were measured. Fungal communities in the leaves of the seedlings were assessed using a metabarcoding approach. The contrasting maternal environments influenced seedling morphology (i.e. height, leaf mass ratio and specific leaf area) and physiology (leaf intercellular CO_2 and intrinsic water use). Seedling resistance to pathogens differed significantly between maternal environments. Fungal communities were also distinguishable between the offspring from the two maternal environments. The study demonstrates that differences in the environment in which *Eucalyptus* trees grow can affect the phenotype, physiology, and resistance of their progeny. Additionally, we suggest that the maternal environment can regulate the structure of fungal communities in the subsequent generation.

P49 Plant grafting: How genetic exchange promotes vascular reconnection

JING WANG[1], LIBO JIANG[1], RONGLING WU[1,2]

[1]Center for Computational Biology, College of Biological Sciences and Technology, Beijing Forestry University, Beijing 100083, China; [2]Center for Statistical Genetics, Pennsylvania State University, Hershey, PA 17033, USA

Grafting has been widely used to improve horticultural traits. It has also served increasingly as a tool to investigate the long-distance transport of molecules that is an essential part for key biological processes. Many studies have revealed the molecular mechanisms of graft-induced phenotypic variation in anatomy, morphology and production. Here, we review the phenomena and their underlying mechanisms by which macromolecules, including RNA, protein, and even DNA, are transported between scions and rootstocks via vascular tissues. We further propose a conceptual framework that characterizes and quantifies the driving mechanisms of scion-rootstock interactions toward vascular reconnection and regeneration.

P50 Identification of a protein family involved in gene silencing and maintenance of genome stability

K. WÖLTJE, D. MELLENTIN, Z. MAGYAR, M. WEINGARTNER

Department of Molecular Plant Physiology, University of Hamburg, Biozentrum Klein Flottbek, Ohnhorststrasse 18, 22609 Hamburg, Germany Institute of Plant Biology, Biological Research Centre, 6726 Szeged Temesvari krt 62, Hungary

In plants, the majority of TEs are transcriptionally silenced by DNA methylation, repressive histone modifications and short interfering RNA pathways. We have identified a gene family in Arabidopsis thaliana consisting of 4 members which share a highly conserved transposon related domain. Due to their mutant phenotype we named these genes *MAINTENANCE of MERISTEMS* (*MAIN*) and *MAINTENANCE of MERISTEMS LIKE 1, 2 and 3* (*MAIL1, MAIL2, MAIL3*). Loss of function of MAIN and MAIL1 leads to root growth arrest soon after germination associated with accumulation of DNA damage, cell death of stem cells and loss of stem cell niche activity. In addition, both mutants show release of silencing of transposable elements (TEs) in all tissues. The mechanism by which MAIL1 and MAIN control gene silencing is still unknown but seems to be independent of known TE silencing pathways, since DNA methylation, repressive histone modifications and short interfering RNA levels are unchanged. We performed Co-Immunoprecipitation experiments followed by mass spectrometry to identify the protein complex in which MAIL1 acts. Here, we present our first results on validated interaction partners for MAIL1. Possible mechanisms how MAIL1 and MAIN might act to control gene silencing and the maintenance of genome stability will be discussed.

P51 The transcriptomic drivers of ecological divergence after recurrent allopolyploidization in *Dactylorhiza* (Orchidaceae)

T. WOLFE, E. TRUCCHI, F. BALAO ROBLES, M. LORENZO, J. BAAR, O. PAUN

Early-generation allopolyploids need to quickly accommodate divergent genomes into one nucleus by adjusting organization and function, thereby altering ecological properties and adaptive success. With powerful transcriptomic approaches we are screening the genome-wide natural diversity
among ecologically-divergent, sibling allopolyploids in *Dactylorhiza* (Orchidaceae) in order to identify the genomic mechanisms that, following iterative genome doubling, drive adaptation to different environments leading to isolation. RNAseq results identify several genes with a putative ecological function, which are found to be expressed at significantly different rates between the polyploids. The differential post transcriptional regulation via smRNAs is less significant, and affects a different population of transcripts, processes and functions. The phenotypic divergence between the polyploids appears mediated by a general parental dominance in opposite directions in the sister polyploids, a pattern partly retained also at the level of transgressively expressed transcripts and at the level of post-transcriptional regulation by smRNAs. We conclude that the major transcriptomic divergence among the diploid parental species of these polyploids became reconciled in different ways in the sibling *Dactylorhiza* polyploids, most probably as a result of an interplay between stochastic genetic and epigenetic alterations and distinct selection pressures specific for divergent environments.