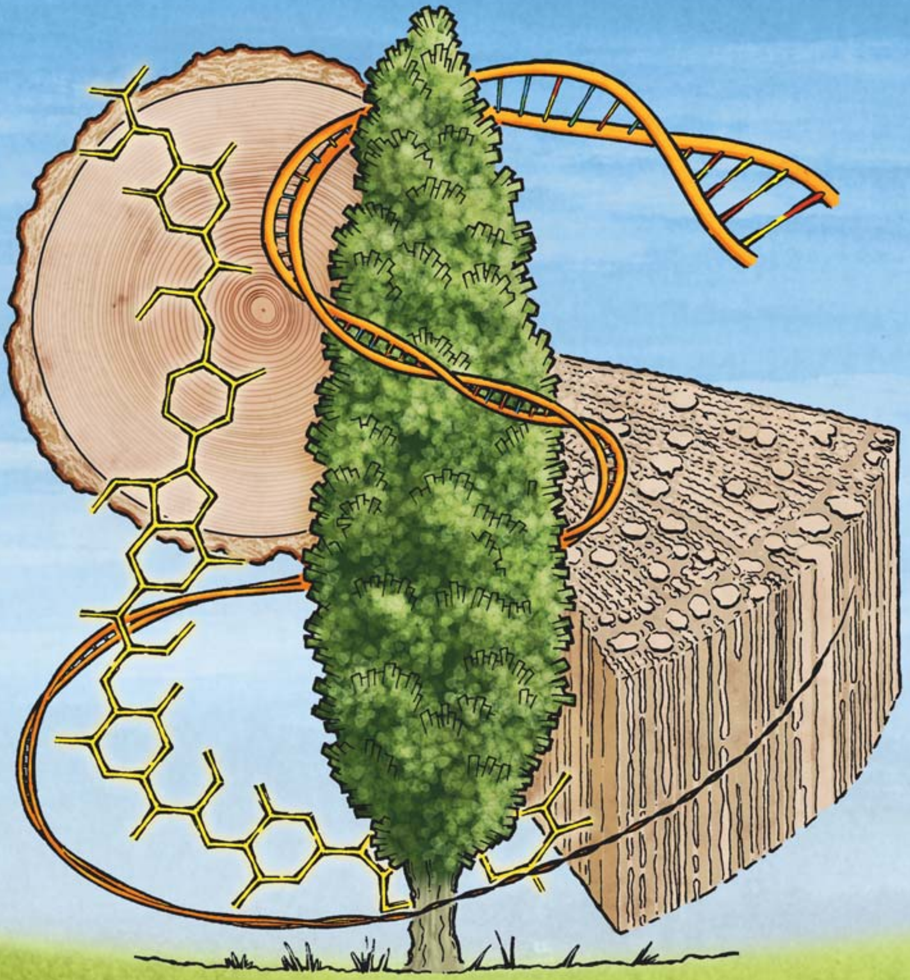
42nd New Phytologist Symposium

The biology of wood: from cell to trees



10–12 July 2018
Lake Tahoe, CA, USA

New Phytologist

Programme, abstracts and participants

42nd New Phytologist Symposium

The biology of wood: from cell to trees

Granlibakken, Lake Tahoe
CA, USA
10–12 July 2018

Scientific Organizing Committee

Andrew Groover
(USDA Forest Service and University of California, Davis, USA)
Shawn Mansfield
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New Phytologist Organization

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Acknowledgements

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The New Phytologist Trust is a non-profit-making organization dedicated to the promotion of plant science. It owns and produces the international journals *New Phytologist* and *Plants, People, Planet*. The Trust receives income through its publication activities and any excess revenue from publication of the journals 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
'The biology of wood' logo by
A.P.P.S., Lancaster, UK

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

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Information for Delegates

Symposium location

The 42nd New Phytologist Symposium will be held at Granlibakken, Lake Tahoe, CA 96145, USA http://granlibakken.com/ in the Mountain room.

Map

A map showing the location of the main lodge and cabins can be found at the back of this book.

Catering

Breakfast will be served from 07:30–09:00 daily in Granhall.
Coffee breaks will be served in the centrally located area outside the main meeting room.
Lunch and Dinner will be served either in Granhall or outside on the Garden Deck weather depending.

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

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 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 a dedicated poster session from 16:30–19:00 on Wednesday evening. 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 09:00 Thursday 12 July.

Internet access

Free wifi will be provided throughout the venue. Log in to ‘Granlibakken’. No password is required.

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 #42NPS in all of your tweets.

Excursion
On Thursday afternoon there will be an excursion to Fallen Leaf Lake. Vans will depart at 13:00 and return to Granlibakken around 18:00.

Altitude
The altitude of Lake Tahoe is 6,225 feet (1897 metres). This elevation does not usually cause altitude sickness, but to avoid problems such as dry sinuses and shortness of breath, particularly if you plan on hiking any of the surrounding peaks, drink plenty of water and cut down on your exercise for the first few days of your stay. You should also note that alcohol consumption can exacerbate the effects of altitude.

Photography
Photography will take place at the 42nd 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 42nd 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; tel: +44 7966 450 389.

Meeting Programme

Tuesday 10 July

13:00–14:30	Registration and arrivals
14:30–14:45	Welcome, Introductions and Information
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Session 1: Anatomy, Physiology and Biochemistry
Chair: Shawn Mansfield
16:00–19:00

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16:40–17:20	**S1.2 Wout Boerjan** Discovering and engineering phenolic metabolism in poplar
17:20–17:40	**Selected talk – Claire Depardieu: P7** A local adaptation signal for drought detected in white spruce woody traits
17:40–18:00	**Coffee/Tea break**
18:00–18:20	**Selected talk – Melis Kucukoglu: P14** Cytokinin regulation of cambium activity and wood formation in hybrid aspen
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Chair: Andrew Groover
09:00–12:45

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09:40–10:20	**S2.2 Georgia Drakakaki** Endomembrane dynamics and polysaccharide deposition
10:20–10:40	**Selected talk – Aleksandra Słupianek: P28** The role of the vessel-associated cells in the intercellular communication in secondary xylem
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Chair: Georgia Drakakaki
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	Genetic architecture of transcriptome variation shaping wood biology in *Eucalyptus*
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Thursday 12 July	
08:55–09:00	Announcements
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Speaker Abstracts

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

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Speaker abstracts

Model systems and wood formation: from cells to trees

Keynote

14:45–15:30

LACEY SAMUELS, M.J. MEENTS, Y. WATANABE, M. PERKINS, M. SCHUETZ

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Department of Botany, University of British Columbia, Vancouver, BC, Canada

Wood formation occurs within the inner bark of trees, where the production of new cells by vascular cambium and differentiation of xylem mother cells results in radial growth. These cells have key features that make them fascinating and challenging to study, such as the large vacuole occupying most of the cell volume, their extreme length, and rapid cytoplasmic streaming. Using cryo-fixation and transmission electron microscopy, we can describe many of the cellular events associated with cellulose, hemicellulose, and lignin deposition. However, getting mechanistic insights into these processes has required model systems such as the Vascular-Related NAC Domain7 (VND7) transcription factor induction system in Arabidopsis. The objective of this work is to discover cellular mechanisms of cell wall polysaccharide biosynthesis and lignification. Using mutant analysis, live cell imaging of fluorescently-tagged proteins, and electron microscopy, we have gained new insights into the dense and dynamic cellulose synthase complexes at the plasma membrane and the hemicellulose factory of the Golgi, as the polysaccharide matrix is deposited to form the thick secondary cell wall. Into this matrix, oxidative enzymes such as laccases and peroxidases are embedded, ready to control lignin deposition as monolignols flood the developing xylem cells. Applying the information gained from such model systems to wood formation *in situ* allows us to generate new hypotheses about secondary xylem production.

Session 1: Anatomy, Physiology and Biochemistry

Chair: Shawn Mansfield

Atypical lignification in Eastern Leatherwood (*Dirca palustris* L.)

S1.1

16:00–16:40

YASEEN MOTTIAR[1], NOTBURGA GIERLINGER[2], DRAGICA JEREMIC[3], EMMA R. MASTER[4], SHAWN D. MANSFIELD[1]

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The classical model of lignification depicts lignin polymerisation beginning at discrete nucleation sites in the cell corners and middle lamellae before spreading to the layers of the cell wall. In contrast to this, the cell corners and middle lamellae in the xylem of Eastern Leatherwood (*Dirca palustris* L.) remain unlignified. Eastern leatherwood is a slow-growing woody shrub native to eastern North America which is known for its flexible stems and branches. Our discovery of lignin-deficient middle lamellae explains this flexibility and also calls into question the classical model of lignification. In addition, we found that leatherwood xylem has low levels of lignin, a high syringyl lignin content, high levels of wall-bound acetyl, low cellulose crystallinity and a high microfibril angle – all important traits for biomass feedstock improvement efforts. Examination of xylem cross-sections revealed a peculiar lignification pattern wherein the oblique strands of vessels and vasicentric tracheids are highly lignified while the intervening libriform fibres are only weakly lignified. This atypical lignification regime vividly illustrates the diversity in plant secondary cell wall formation that abounds in nature and casts leatherwood as a new model for the study of lignin formation and deposition.

Discovering and engineering phenolic metabolism in poplar

S1.2

16:40–17:20

MARINA DE LYRA SORIANO SALEME[1,2], LISANNE DE VRIES[1,2], SANDRIEN DESMET[1,2], BARBARA DE MEESTER[1,2], REBECCA VAN ACKER[1,2], IGOR CESARINO[1,2,3], LIVIA VARGAS[1,2], ANDREAS PALLIDIS[1,2], GEERT GOEMINNE[1,2], ALEXANDRA CHANOCA[1,2], JAN VAN DOORSSELAERE[4], HOON KIM[5], JOHN RALPH[5], KRIS MORREEL[1,2], RUBEN VANHOLME[1,2], WOUT BOERJAN[1,2]

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Wood is a promising feedstock for the biorefinery. The cell wall polysaccharides can be converted to fermentable sugars. However, for the production of fermentable sugars from the biomass, lignin needs to be extracted by costly pretreatments, hampering the transition from a fossil-based to a bio-based economy. Engineering lower amounts of lignin or altered lignin composition in biomass crops is therefore an attractive strategy to improve biomass quality. From a co-expression analyses with known genes involved in lignin biosynthesis, we recently discovered a gene encoding a new enzyme in the biosynthesis pathway to the monolignols, CAFFEOYL SHIKIMATE ESTERASE (CSE). The corresponding Arabidopsis mutant had a reduced lignin amount and a 4-fold increase in cellulose-to-glucose conversion without biomass pretreatment. Given these promising results, the *CSE* gene was down-regulated in poplar through a hairpin approach, and knocked out by a CRISPR/Cas9 approach. Wood of these trees was then investigated for improvements in saccharification efficiency. Furthermore, the effect of the pathway perturbation on phenolic metabolism was studied by comparative metabolomics.

In addition to lignin and cell wall polysaccharides, wood also contains numerous small molecules of which the identity has remained unknown. This hampers their valorization. We have developed an algorithm, called Candidate Substrate Product Pairs (CSPP) that allows prediction of the structures of unknown metabolites in plant tissues. Knowledge on the identities of metabolites can provide deeper insight into the metabolic pathways active during wood development and helps understanding the plants’ responses to pathway engineering.

Xylem structure and function: what have we learned and what do we still not know?

S1.3

N. MICHELE HOLBROOK

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Department of Organismic and Evolutionary Biology, Harvard University, Cambridge MA, USA

18:20–19:00

The xylem wears many hats: it transports water under tension, reversibly stores non-structural carbohydrates and water, and provides mechanical support. Here I focus on water transport and ask what are the structural and architectural features that allow plants to pull water from the soil. Because water is drawn through the xylem in a metastable state, the continuity of water needed for water transport under tension is at risk due to the potential for embolisation. Pit membranes serve as check valves that block the invasion of air, but also impede flow. The hydraulic resilience of wood depends on both the physicochemistry of the pit membranes and the ways in which they are deployed. I will discuss the nature of air movement across pit membranes, as well as the architecture of vessel networks. I will also consider the strategies plants use to limit the development of damaging tensions and thus to protect the xylem.

Session 2: Cell Biology

Chair: Andrew Groover

Emerging understanding on the mechanisms and significance of autolysis during cell death of Arabidopsis tracheary elements

S2.1

SACHA ESCAMEZ[1], BENJAMIN BOLLHÖNER[1], SIMON STAEL[2], JULIA VAINONEN[3], MICHAEL WRZACZEK[3], FRANK VAN BREUSEGEM[2], HANNELE TUOMINEN[1]

09:00–09:40

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Xylem differentiation culminates in cell death followed by complete autolysis of the cells. I will first of all summarize our current knowledge on cell morphology, cell wall chemistry and autolytic processes occurring during tracheary element (TE) and fiber cell death. Thereafter, I will focus on the *Arabidopsis thaliana* cysteine protease METACASPASE9 (AtMC9) that is crucial for the rapid autolysis of the tracheary elements. Suppression of *AtMC9* expression in Arabidopsis tracheary element (TE) cell cultures resulted in altered cysteine protease activities during TE differentiation, increased autophagic fluxes and cell death of not only the TEs but also ectopically in the non-TEs that normally rarely die. Supression of autophagic fluxes in these cultures by transgenic means reduced the ectopic cell death in the non-TEs. These results suggest that cell death inducing signals can be derived from maturing xylem elements in a non-cell autonomous manner and that appropriate activation of the autolytic machinery as well as suppression of autophagy by AtMC9 is critical to guarantee rapid and complete degradation of the TE contents and to prevent ectopic cell death during xylem maturation. Evidence is provided that this control mechanism involves a cascade of cysteine proteases in an analogous manner to the animal caspase cascades. Further understanding to the underlying molecular mechanism is provided by identification of a peptide that is present in the culture medium of the *AtMC9* supressed TE cell cultures, and that is able to induce cell death when applied to the wild type TE cell cultures or to whole seedlings. It seems therefore plausible that this pro-death peptide needs to be maintained in the TEs by the action of AtMC9 to prevent spreading of the cell death to the neighbouring cells.

Endomembrane dynamics and polysaccharide deposition

S2.2

09:40–10:20

<u>GEORGIA DRAKAKAKI</u>[1], DESTINY DAVIS[1], NATASHA WORDEN[1], WENLONG BAO[1], MICHEL RUIZ ROSQUETE[1], SIVAKUMAR PATTATHIL[2], ANGELO PERALTA[2], DAVID DOMOZYCH [3], MICHAEL HAHN[2], THOMAS WILKOP[1,4]

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The plant endomembrane system controls many aspects of plant development including cell wall deposition. Combining chemical genomics and proteomics we studied various components of the endomembrane system present in the *trans*-Golgi Network (TGN), a site of polysaccharide trafficking and recycling of endosomal components.

We have identified novel pharmacological inhibitors, targeting cell wall deposition during cell division and/or during cell elongation, among them the small molecule endosidin 7 (ES7). It targets specifically callose synthase activity during late cytokinesis and arrests cell plate maturation. Towards a better understanding of vesicle dynamics and polysaccharide deposition in cytokinesis, we established a methodology allowing in vivo, quantitative 4D microscopy of the cell plate formation. The temporal interplay between cell-plate-specific post-Golgi vesicle populations and callose accumulation reveals the unique contributions of secretory and endosomal vesicles providing detailed cues for the understanding of cell plate formation.

In a complementary proteomic and glycomic approach, we separated TGN vesicles identified by the syntaxin SYP61, and analyzed their proteome and glycome. Interestingly, the characterization of the SYP61 vesicle proteome revealed the

presence of proteins involved in cell wall biosynthesis and modification and in stress responses. The function of novel proteins identified in SYP61 vesicles was investigated. In parallel, using an automated high throughput ELISA assay employing a comprehensive suite of cell wall glycan directed monoclonal antibodies (mAbs), the polysaccharide composition of the isolated vesicles was determined. Based on the cumulative data, we propose that the SYP61 pathway mediates trafficking of cell wall and stress response components in the cell.

Session 3: Genomics and bioinformatics

Chair: Missy Holbrook

A novel Ca^{2+}-dependent Nuclease regulates secondary vascular tissue differentiation in *Populus*

S3.1

<u>JUAN DU</u>[1,2], CHENG JIANG[1], JUN-GANG WANG[1],SHU-TANG ZHAO[1], XUE-QIN SONG[1], YING-LI LIU[1], YING-HUA GUO[1], HEVEN SZE[2], SUZANNE GERTTULA[3], ANDREW T. GROOVER[3], MENG-ZHU LU[1]

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14:00–14:40

Vascular tissues that conduct water and nutrients in plants are formed from division of cambium meristem cells followed by cell differentiation. Xylem and phloem mother cells give rise to cells that can undergo programmed cell death as a normal part of their development. However, the molecular events that trigger vascular development and programmed cell death in woody tree remain poorly understood. Here we present evidences that a Ca^{2+}-dependent Nuclease is a trigger in vascular tissue differentiation. Initial experiments found that DNA fragmentation and Ca^{2+}-dependent Nuclease activity occurred in the cambium zone at the onset of secondary growth in poplar. A gene encoding a Ca^{2+}-dependent Nuclease was cloned and named *PtoCAN*. Transgenic plants over-expressing *CAN* showed increased girth

and length in the newly developed internodes, where the early appearance of differentiated secondary xylem and phloem was accompanied by the early and enhanced nuclei DNA fragmentation. CRISPR-mediated knock-out lines of *CAN* had reduced internodes girth and length in the early secondary growth stage, delayed secondary xylem and phloem formation and reduced DNA fragmentation. Moreover, genes encoding proteins involved in vascular cambium maintenance, cell division and expansion, master regulators of secondary cell wall deposition, programmed cell death and final autolysis were up-regulated in *CAN* over expressing plants, and down-regulated in *CAN* knock-out plants. Together our results suggest that *CAN* play a role in early vascular differentiation.

Reverse engineering networks of secondary growth and their evolution in plants

S3.2

15:50–16:30

<u>ESHCHAR MIZRACHI</u>[1], DESRE PINARD[1], DANIELLE ROODT[1], RIAAN SWANEPOEL[1], ANDREW BEHRENS[1], MARK MAISTRY[1], VICTORIA MALONY[1], YVES VAN DE PEER[2,3], KATHLEEN MARCHAL[3], ALEXANDER MYBURG[1]

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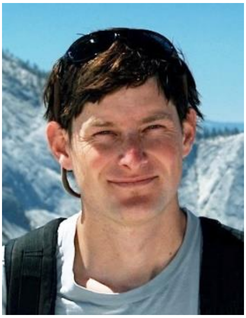
Our understanding of the biological networks regulating secondary growth is especially extensive for the biosynthesis of various cell wall biopolymers, but major gaps remain in understanding other aspects of secondary xylem formation. Two points of major interest are the evolution and regulation of cell patterning across the various lineages in vascular plants, and how carbon in the form of simple sugars is partitioned at a subcellular level between homeostasis and the synthesis of cellulose, hemicellulose and lignin in xylem sink tissues. We investigate networks involved in xylogenesis by comparative genomics and transcriptomics from lineages representing key transitions in secondary growth and/or secondary cell wall evolution, including under-represented monocots and non-coniferous gymnosperms. Additionally, we have constructed secondary xylem gene co-expression networks from *Eucalyptus* populations to investigate the genetic architecture of regulation, focusing on the specific role of organelles (plastids and mitochondria) during xylogenesis. We demonstrate evidence of distinctly regulated (nuclear-encoded) plastid *targeted* genes with progressive phases of xylogenesis, as

well as differential upregulation of plastid *encoded* genes during this process. This upregulation of organellar encoded genes is primarily governed by nuclear-encoded factors, as well as regulation by RNA-binding proteins as evidenced by small RNA footprints. Functionally, we are testing (in both *Arabidopsis* and *Populus*) perturbations to the pentose-phosphate pathway, to further clarify individual enzymes roles in carbon partitioning towards aromatic amino acid biosynthesis during wood formation.



Geology of the Sierra Nevada and the Tahoe Basin

Invited Lecture

GREG STOCK

20:30–21:15

greg_stock@nps.gov

National Park Service - Yosemite National Park, 5083 Foresta Road Box 700, El Portal, CA 95389, United States

From volcanic flows to glacial moraines, earthquake faults to giant lakes, the eastern Sierra Nevada displays some of the most unique and spectacular geology anywhere in the world. This talk will provide an overview of the geology of the Sierra Nevada, describe the effects of millions of years of climate variability, and discuss how the underlying geology influences the distribution of plant species.

Session 4: Ecology and Evolution

Chair: Missy Holbrook

Mountain forests respond to climate change; Adaptation, biogeography, and…wood

S4.1

CONSTANCE MILLAR

cmillar@fs.fed.us

USDA Forest Service, Pacific Southwest Research Station, Albany, CA, USA

09:00–09:40

In the spirit of trans-disciplinary integration in wood biology research, I offer perspectives of a climate ecologist working in Great Basin mountain forests. In the face of climate challenges, forests respond successfully through changes in community composition, structure, and function; distribution shifts marked by colonization events; and genetic adaptation rendered by changes in natural selection, gene flow, genetic drift, and breeding structure. Unsuccessful responses to climate drivers include extirpations and species extinction. At the heart of survival and reproduction are healthy, functioning trees supported by the critical hydraulic, mechanical, metabolic, and defense functions of wood. Thus our interests in how forests will cope with ongoing climate change rest on understanding how wood functions are able to respond adaptively – or how limits to their capacities will dictate population and species losses.

I share basic knowledge about the behaviour of Earth's climate system, including processes of natural climate variability and their historic ecological and genetic consequences on mountain forests. Knowledge of historic processes provides valuable context informing us how modern forests might respond to climate change. I address the novel anthropogenic climate forces and summarize projections for changes anticipated in mountainous western North America. Of particular importance (I hope) to wood biologists are challenges that confront basic wood functions to maintain healthy forests from: accelerated warming; increasing extreme weather and inter-annual climate variability, hotter droughts and extreme winter storms; changes in snowpack, recharge, and runoff; increasing atmospheric CO_2; and changes in fire regimes, insects and pathogens. Beyond individual drivers,

climate change is catalysing interaction of these agents in increasing megadisturbance events. My hope is that wood biologists will gain a better understanding of the nature of population genetic, evolutionary, and ecological forcings that are challenging the fundamental wood framework of our forests and, through their expertise, forge innovative paths in wood research.

Genes and mechanisms underlying the evolution and development of wood in angiosperm trees

S4.2

<u>ANDREW GROOVER</u>[1,2], SUZANNE GERTTULA[1], MATT ZINKGRAF[3], HELOISE BASTIAANSE[1], VLADIMIR FILKOV[4], SHUTANG ZHOU[5], MENGZHU LU[5]

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09:40–10:20

Secondary growth and wood formation are ancestral traits of angiosperms. Currently little is known regarding the ancestral mechanisms regulating wood formation or how they have been modified to produce the amazing array of wood anatomy in extant angiosperms. In this presentation, we present three examples of how genomic and computational approaches can provide new insights into the evolution and development of wood formation. First, we describe how gravitropic responses and tension wood formation of woody poplar stems serves as a model for dissecting wood formation based on gene co-expression analyses. Second, we show how gravitropic response and co-expression analyses in a broad survey of angiosperm trees can be used to identify groups of co-expressed genes that represent putative ancestral mechanisms. Third, we describe the use of a unique population of poplars carrying genomically mapped insertions and deletions for the dissection of wood anatomical, biochemical and physiology traits.

Evolution of genetic mechanisms regulating tracheary element differentiation

S4.3

TAKU DEMURA, MISATO OHTANI

demura@bs.naist.jp

Division of Biological Sciences, Graduate School of Science and Technology, Nara Institute of Science and Technology, Ikoma, Nara, Japan

11:30–12:10

Land plants developed water transport system during plant evolution, which influences the viability of plants on dry land. Tracheary elements, namely tracheids and vessel elements, found in xylem of vascular plants as the major constituents of the water transport system, are elongated dead cells with lignified and patterned secondary cell walls (SCWs). Recent molecular genetic studies, mainly in *Arabidopsis thaliana*, revealed the gene regulatory network (GRN) governing the differentiation of tracheary elements: NAC transcription factors VASCULAR-RELATED NAC-DOMAIN (VND) and their homologous proteins VNS (VND, NST/SND, SMB-related) are the master regulators that can induce the genes involved in programmed cell death and cell wall biosynthesis/modification. It was found that this GRN is widely conserved in vascular plants, including angiosperms and gymnosperm such as loblolly pine. In addition, we showed that the VNS-based GRN for programmed cell death and cell wall biosynthesis/modification is conserved in the differentiation of hydroids, dead water-transport cells, and stereids, supporting cells with thickened cell walls, of the moss *Physcomitrella patens*. Interestingly, a phylogenetic analysis of NAC transcription factors indicated that the VNS group occurs not only in some other land plants (*Marchantia polymorpha*, *Selaginella moellendorffi*, and *Amborella trichopoda*) but also in the charophyte green algae *Coleochaete orbicularis*, suggesting the emergence of the VNS-based GRN probably occurred before, or almost simultaneously with, the evolution of early land plant species.

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Poster abstracts

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P1

Tree rings reveal that climatic and competitive growth constraints trade off at large scales but rarely explain local tree range boundaries

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We must understand the drivers of species ranges to anticipate and manage climate change-induced range shifts. Long-standing ecological theory predicts a tradeoff between climatic constraints in harsh environments and biotic constraints in benign environments, but this theory has rarely been tested. Using a large network of tree cores, we measured competitive and climatic constraints on tree growth in multiple species across their elevation ranges in three climatic regions to test whether a climate-competition tradeoff can explain altitudinal distributions. We show that tree growth at environmentally harsh range boundaries was climatically constrained for most species. However, tree growth at environmentally benign range boundaries was rarely constrained by competition. As a result, a climate-competition tradeoff explained few elevational ranges. Additionally, it was difficult to predict a-priori which range boundaries (low or high) were climatically vs. competitively constrained. However, climatic growth constraints increased and competitive constraints decreased in harsh environments across all species and sites, consistent with a broad-scale climate-competition tradeoff. Our findings highlight multiple processes that complicate local range dynamics, but suggest that the constraints on tree

performance at a large-scale (e.g. latitudinal) may be predicted from ecological theory.

P2

Bibliometrics evaluation of research status and hotspots in *Picea* species: China relative to the rest countries of world

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Picea A. Dietrich is an important component of boreal vegetation and subalpine coniferous forests and has a wide geographical range that covers the Northern Hemisphere and extends from the Eurasian continent to North America. This paper digs up and filters the trace left by the researches on the Web of Science and the China National Knowledge Infrastructure in 2002-2016: literature derivation distribution, high quoted literature and nuclear researchers. The result could offer insights concerning the best way to face future research gaps, and guide researchers through the concept of *Picea* because it shows the literatures that has to be included in further analyses. Furthermore, this study compared the differences between China and the world in the study of *Picea*, to help serve as a point of reference and a preliminary approach for new researchers seeking to familiarize themselves with the *Picea* literature and trends, in order that research level of China scholars about *Picea* can keep up with the pace of the world as soon as possible.

P3 *EVE* regulates vessel dimensions in flowering plants

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The transition of plants to grow on land required the creation of a vascular system, leading them to acquired water-conducting tracheary elements. The physical dimensions of these specialized cells have played a critical role in plant growth and evolution. However, the genetic control of the physical dimension of tracheary elements has remained largely unknown. Here we show that a previously uncharacterized gene, *ENLARGED VESSEL ELEMENT 1 (EVE1)* containing the domain of unknown function 3339, regulates tracheary element formation, or vessels, in flowering plants. To determine the developmental function of *EVE1*, we generated *EVE 1* overexpressing and knockout lines. Xylem vessel mean diameter and count were significantly higher in transgenic plants overexpressing *EVE1*. On the other hand, *EVE1* knockout lines showed a significant reduction in vessel mean diameter and count. Plants overexpressing *EVE1* have higher hydraulic conductivity and an increase in the rate of photosynthesis and growth. In plants, *EVE* first emerged in streptophyte algae, but expanded dramatically among vessel-containing angiosperms. Other than in streptophytes, *EVE* is only found in the genomes of algae-infecting prasinoviruses. *EVE* plant homologues resemble the structure of viral genes, suggesting it originated in plants from an ancient viral lateral gene transfer event.

P4

Ectopic callose accumulation in dicots secondary cell walls and its potential as a substrate for biofuel and biomaterials applications

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We previously identified several gain-of-function mutations in a locus coding for a callose synthase isoform (CALS3) in Arabidopsis, and combined them to produce an enzyme highly active in callose polymerization (cals3m). When compared to cellulose, callose shows appealing biochemical properties providing significant potential to lower biomass recalcitrance to deconstruction. Here, we took advantage of our cals3m tool to specifically increase callose content in xylem cell types of Arabidopsis and hybrid Aspen. We aim to demonstrate that callose enriched wood biomass is sustainably easier to process than its non-enriched counterparts for subsequent bioethanol. We also want to address the biochemical and biophysical properties associated with callose enrichment for potential biomaterial applications. Preliminary results show a stable accumulation of callose in secondary cell walls, along with a tight association with cellulose microfibrils. Interestingly, callose enriched primary cell walls also show increased stiffness, a promising feature when addressing biomaterial applications.

P5 New facts about content of antioxidants in yacon leaves

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Yacon (*Smallanthus sonchifolius*) belongs to the Asteraceae (Compositae) family, Asteraceae subfamily (Astrodeae), Sunflower tribe (Heliantheae). Yacon can be described as a delicious and nutritious vegetable that can be eaten raw or cooked, it also can help to lose weight and it is also a unique vegetable with various health benefits. This root crop of South American origin, is considerably rich in health-related substances, such as antioxidants and carbohydrates.

The study examined the total content of antioxidants in the yacon leaves during cultivation under various types of illumination and under the influence of growth stimulant drug, such as Albit and Lebame.

The study of hydrophilic low molecular antioxidants content in yacon leaves during cultivation under various types of illumination shows that the spectrum radiation irradiator, equipped with a white light lamp, should be optimize with LED emitters. Thus, biosynthesis activation reactions of biologically active substances and antioxidants, and plants species, variety and development stage must be considered.

Irradiation of yacon plants with phytolamp light significantly increases the content of photosynthetic pigments and hydrophilic low-molecular antioxidants in leaves as compared to fluorescent lamps control (daylight).

P6 Quantifying vulnerability to xylem embolism in *Aesculus californica* in a hotter, drier California

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California is predicted to undergo substantial future climate change with most models predicting prolonged and hotter droughts. The most recent drought resulted in widespread tree mortality, highlighting the need to understand species-level traits and physiological mechanisms that control plant response to drought. Recent findings indicate that xylem embolism is a critical process that plants experience during dehydration, suggesting that we can extract critical thresholds from xylem vulnerability curves. However, much controversy surrounds the capacity of plants to resist embolism and many species remain uninvestigated. We investigated the physiological responses and vulnerability to embolism of a California native tree species, *Aesculus californica*, to drought via a greenhouse dry-down experiment. *Aesculus californica* (Buckeye) is unique in that it emerges rapidly across California in the spring but quickly loses its leaves in summer. Using the optical technique, we quantified the leaf and stem xylem vulnerability to embolism of nine individuals of Buckeye to examine whether this is a component of a drought avoidance strategy. Leaf level gas exchange, sapflow and chlorophyll fluorescence were also quantified. Our results show that Buckeye has several mechanisms to avoid low water potentials, including stomatal closure and leaf shedding. Further, Buckeye is not vulnerable to embolism, and avoids critical threshold by reducing water loss and persisting through dry periods with very low sapflow. We conclude that Buckeye has a drought avoidance strategy, supporting the idea that embolism is a critical point that plants aim to avoid during desiccation. These data will ultimately aid in predicting and managing the fate of Buckeye as extreme climate events continue to occur in California.

P7 A local adaptation signal for drought detected in white spruce woody traits

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Worldwide, an increase in temperature and in the frequency of extreme drought events is projected due to ongoing climate change. For spruce species that largely dominate the boreal forest ecosystem, their physiological responses to drought throughout their lifespan remain largely unknown. Dendroecology approaches using common garden experiments have recently proven to be useful in determining species sensitivity to climate variability. We have taken advantage of an existing set of tree ring data obtained from a *Picea glauca* provenance/family test. We aimed to (1) determine the main climatic factors influencing growth and woody traits (cell wall thickness, lumen diameters) and (2) compare growth resilience among provenances. Data were obtained by SilviScan technology for 1595 trees representing 43 provenances. Dendro-climatic relationships were examined between residual chronologies of woody traits and monthly climatic variables. At planting site, soil water availability was the major limiting factor for radial growth. Interestingly, the provenances originating from drier locations compared to the test site were relatively more resilient to drought than those originating from rainier locations. This can be interpreted as a signal of local genetic adaptation to drought. Wood trait-climate relationships revealed differences in climate sensitivity between provenances, which might reflect different drought-coping strategies in white spruce.

P8

Surviving but not expanding – a large fraction of trees for hyperdominant species do not exhibit annual increase in diameter in a central Amazon forest

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The low growth rates of adult trees in the Central Amazon forests suggest a high competition for resources. This work is showing that adult trees of hyperdominant species spend a long time without expressing xylem production in the DBH (diameter at breast height). A high percentage of trees with zero increments were detected in permanent plots with two precisions level, 1mm and 0.01mm. Zero increments can occur sporadically or for several consecutive years. In years influenced by gaps in the canopy (post-logging), are smaller numbers of trees with zero increment ($p=7.6x10^{-12}$). The number of days without rain throughout the year also affects the occurrence of zero or negative increment in hyperdominant trees ($R^2= 0.70$ e $p= 0.0004$) and by trees community represented by 109 species ($R^2= 0.59$ e $p=0.002$). Ours results demonstrated a cessation of xylem production for many years in the adult trees of the Central Amazon 'Terra Firme' Forests. During these periods, trees probably do not form growth ring, increasing the intensity of partial or missing rings throughout the lifetime of trees. Moreover, these results require more attention in the data treatment of the forest inventory and from dendrocronologists to estimate biomass or cut cycle to forest management.

P9

Seasonal variation in redwood branch hydraulic conductivity: the critical link between climate and tree growth

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Redwoods grow in a bimodal climate and tend to experience substantial water stress (low xylem water potential, Ψ) during the hot, dry summers. This stress can cause xylem dysfunction and decreased hydraulic conductivity (K_s), but magnitude and phenology of this effect have not been studied. We investigate the question: *How does redwood K_s respond to seasonal variation in Ψ?*

The Ψ threshold at which redwood K_s decreases in the lab often occurs naturally in summer. We hypothesize, therefore, that: *Redwood K_s decreases seasonally during stressful conditions and episodes of low Ψ.* Summer is also the redwood growing season. We further hypothesize: *Xylem growth compensates for decreases in K_s by functionally replacing damaged cells.*

This study combines time-series measurements of Ψ, K_s, and xylem growth in stand-dominating redwood tree tops at Big Creek Reserve. We also calculate the potential K_s of whole branch segments, embolized cells, and newly grown cells.

Despite measuring record-breaking low Ψ, we have observed no significant change in K_s. New growth may mask losses of K_s, but no growth has occurred. This apparent resilience contradicts the results of lab studies of xylem vulnerability, suggesting that the lab-determined Ψ threshold may not be applicable in the field.

P10

Implications of xylem network organization on the spread of xylem-dwelling bacteria

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Xylella fastidiosa (Xf) is a xylem-dwelling bacterium and is the causal agent of wide range woody plant diseases, including grapevine, olive, and oak. Once the Xf are introduced into the xylem, they proliferate spreading systemically. This spread has been associated with water transport dysfunction resulting in significant losses in crop productivity and plant death. This is because the xylem tissue is a network of pipe-like cells transporting water in a range of different anatomical pathways: water moves axially within conduits and radially through connection between these conduits. The radial movement is a critical factor in maintaining water transport by creating multiples pathways, however greater xylem lateral connectivity also increases the vulnerability to the spread of air embolism and pathogens. Our objective is to understand the relationship between the xylem connections and the Xf spread. We applied microCT imaging to identify and quantify vessels relays in grapevines species.

Vitis arizonica (Xf-resistance) presented less vessels relays than the vinifera varieties, but the pit membranes air-seeding pressure were not significant different between *V. arizonica* 1.46MPa (±0.87), Chardonnay 1.82MPa (±0.95), and Lenoir 1.44MPa (±0.53). These data could support future research in wood anatomy by providing detailed cellular ultrastructure organization of grapevine with different levels of Xf-tolerance.

P11

Nutrient resorption of ten mineral elements in the assimilation organ of desert shrubs and its relation to aridity

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Nutrient resorption from senescent assimilation organs to the woody parts, such as stems, trunks and roots in autumn or dry season, is a key strategy for nutrient conservation for deciduous woody plants. Drought that often occurs in arid regions was supposed to lower nutrient resorption efficiencies in woody plants. We here test this hypothesis via determination of ten mineral nutrients [*i.e.*, N, P, K, Mg, Ca, Mn, Zn, Al, Fe, and Cu] in green and senescent leaves or assimilation branches sampled from the dominant desert shrubs in northern Xinjiang of China. Resorption happened for six elements (N, P, K, Mg, Cu, and Mn) during assimilation-organ senescence, while the other four nutrients tended to accumulate in senesced-organs, showing negative resorption efficiencies. Aridity showed strikingly different effects on the resorption of the ten nutrients. Resorption efficiencies of N, P, K, Cu, and Zn generally decreased with aridity, suggesting that drought stress had negative effects on the resorption of these elements. However, the others, most tending to accumulate during assimilation-organs senescing, showed a reverse trend with aridity. The contrasting resorptive responses of different elements to aridity in typical desert shrubs suggest potential decoupling of nutrient cycles in arid ecosystems under climatic change.

Keywords: aridity, desert shrubs, mineral elements, nutrient resorption, woody plants

P12

DNA barcoding authentication for the wood of eight endangered *Dalbergia* species using machine learning approaches

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Illegal logging is becoming a serious problem, threatening not individual wood species, but even entire ecosystem, which necessitates the reliable wood identification methods at the species level. Reliable analytical methods and reference library are essential to the application of DNA barcoding, a promising tool for forensic wood identification. We collected xylarium wood specimens of eight *Dalbergia* species to investigate the efficacy of DNA barcodes for species resolution using machine learning approaches (BLOG and WEKA) in comparison with distance-based and tree-based methods, and verify the reliability of the machine learning approaches for wood species identification based on the reference dataset of four barcodes (ITS2, *matK*, *trnH-psbA*, and *trnL*) and their combinations. The results demonstrated that the machine learning classifier SMO performed the best (98%~100%) in correct identification rates among the three analytical methods. Moreover, the two-locus combination ITS2+*trnH-psbA* showed the highest success rate for discriminating the eight *Dalbergia* species based on the three methods, and the non-vouchered specimens were entirely identified by ITS2+*trnH-psbA* with 100% of identification rate using SMO classifier and NJ tree. The present results demonstrate the feasibility of the machine learning approaches and DNA barcodes libraries for identification and conservation of endangered *Dalbergia* species.

P13

Morphological, physiological and transcriptomic analyses of stem growth in a tropical timber tree species, *Shorea leprosula*

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Stem growth is an important trait for trees as it affects its architecture, biomass and timber value. However, its regulation in trees still remains unclear partly because of complicated regulations due to their intermittent growth and difficulties in the observations of young leaves and stems enclosed in buds in many tree species. To overcome these obstacles, we focused on a tropical timber tree species, *Shorea leprosula*, which shows intermittent growth and whose buds are not tightly packed. Observation of elongating stems showed that well-growing regions exist around developing leaves. Excision of the developing leaves significantly reduced stem elongation, but exogenous application of bioactive gibberellin (GA) restored stem elongation. Whole-genome transcriptome analysis suggested GA responses in both growing stems and leaves during leaf flushing. However, up-regulation of GA biosynthesis genes was observed only in the developing leaves, suggesting that GA produced by developing leaves promoted stem elongation. Furthermore, we found that elongation rate of stem decreased before leaf development completed. GA biosynthesis genes were still expressed in these leaves, suggesting that GA-independent mechanism may exist in the termination of stem elongation. Further analysis comparing with publicly available transcriptome data of leaf flushing from temperate tree species will be discussed.

P14 Cytokinin regulation of cambium activity and wood formation in hybrid aspen

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Radial expansion of the stems and roots in plants derives from the activity of the vascular cambium - a meristematic tissue that contains the vascular stem cells and generates xylem (wood) on the inside and phloem on the outside. Proliferation and differentiation of the vascular stem cells in the cambium is tightly regulated to achieve an organized vascular development. A recent study from our group displayed that cambial cell division rate and biomass production can be stimulated dramatically in hybrid aspen trees through overexpression of the cytokinin biosynthesis gene, *ISOPENTENYLTRANSFERASE 7 (IPT7)*. To understand how cytokinin orchestrates the cambium activity and wood formation, we collected genome-wide profiling data from the wood-forming regions of wild-type (WT) and mutant trees with enhanced cytokinin production, and from the stem of WT trees treated with cytokinin. As a result, several new regulators of cambium development in hybrid aspen was identified. Currently we are studying the functions of these candidate genes in trees through transgenic approach. Moreover, we are developing a DNA-free CRISPR/Cas9 genome editing method for silver birch (*Betula pendula*) using protoplasts to facilitate tree genomic studies. Our recent results in this research avenue will also be presented.

P15

Variation in xylem anatomy between living and drought-killed *Pinus ponderosa* and *P. jeffreyi* in the Sierra Nevada

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The California drought of 2012–2016 was more severe than any observed in the previous 1200 years and left an estimated 130 million standing dead trees in the Sierra Nevada. We sampled living and dead *Pinus ponderosa* and *P. jeffreyi* in drought-stricken stands and compared annual rings with measurements of xylem cell (tracheid) dimensions to ask how climatic variables influenced ring traits in living and dead trees. Annual growth in living trees increased slightly during and following the drought, and decreased in dead trees prior to mortality. Fall precipitation explained growth patterns in drought survivors, but not dead trees. Average tracheid diameter was positively correlated with previous November temperatures and negatively correlated with January temperatures. The ratio of tracheid wall thickness to diameter ('thickness-to-span', T-S) increased over the length of the drought, and was higher in living trees than drought-killed trees. Mixed linear models showed that cold winters induce large rings with a high density of small tracheids. These relationships show that small, thick-walled tracheids increase drought resilience, and that pines need cold, wet winters to maintain increased growth of safe xylem.

P16 Peeking beneath the hood of the wood economics spectrum

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The wood economics spectrum characterizes a set of inter-relationships between wood traits and functional trade-offs such as lower carbon investment (low wood density) and increased growth and mortality. Furthermore, wood is a vital component of nutrient dynamics in forests given the high nutrient stocks stored in the trunks of trees, although showing lower concentrations than leaves. Despite of the suggested general patterns of trade-offs and associations between life history and wood traits, a comprehensive understanding of the underlying causes of trait variation and their consequences for tree biology are still missing. We explored the physiological and structural implications of the trait correlations and partitioned their variances across different scales (environmental and biological). We collected wood samples and analyzed their density and nutrient concentrations across tropical forests in three continents to tackle the questions underlying the causes of wood trait variation and how they are inter-associated. Wood density showed a large proportion of variation due to differences between species, however intra-specific variation had a substantial contribution. Nutrients varied widely across scales, thereby showing that different drivers control the variation of each woody trait. Here we disentangle the main drivers of wood trait variations and discuss the ecological implications of their associations.

P17

Branching of conifer xylan and its interaction with cellulose microfibril in spruce wood

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Relatively little is known about the biosynthesis and molecular architecture of conifer wood. Softwood is mainly composed of cellulose, galactoglucomannan, xylan and lignin. We have recently demonstrated that in hardwoods, xylan interacts with the cellulose fibril as a two-fold screw, and that GUX (GlucUronic acid substitution of Xylan)-dependent xylan branching is critical for biomass recalcitrance. Here we investigated the decoration of softwood xylan by GUX enzymes and studied the interaction of xylan with cellulose in spruce wood. Using *in vitro* and *in planta* assays we demonstrated that two distinct conifer GUX enzymes are active glucuronosyltransferases. Interestingly, these enzymes have different specific activities with one adding evenly spaced GlcA branches and the other one adding consecutive GlcA decorations. Two-dimensional solid state NMR analysis of never-dried ^{13}C-enriched spruce wood revealed that the majority of conifer xylan is cellulose-bound in a two-fold screw conformation. We speculate that the evenly spaced GlcA branches allow xylan-cellulose interaction whereas the consecutive decorations added by the second enzyme inhibit it. This work therefore elucidates the molecular architecture of native spruce wood using 2D ^{13}C-^{13}C solid state NMR and to characterise conifer enzymes likely to determine the recalcitrance of softwood to processing.

P18

Water deficiency and potassium supply trigger interconnected signals to modulate wood formation in *Eucalyptus*

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Fast growing *Eucalyptus* tree is well adapted to various soils and climate environment, but its growth varies strongly according to these factors. Thanks to potassium fertilization, productivity in the south of Brazil is one of the highest in the world. The development of more sustainable cultural practices requires an improved understanding of mineral nutrition, especially in interaction with water stress, which is more and more threatening plant culture. We aimed to characterize the effect of water availability and nutrition supply on wood formation and quality. An experimental design was set up on field with a highly productive *Eucalyptus grandis* clone planted in a split-plot design, with two factors tested in interaction: water availability set up with rainfall exclusion system, and K+ fertilization. We analyzed wood properties and performed large-scale analysis of transcriptome (RNAseq) and metabolome in developing xylem. These data were integrated using multivariate statistical analyses and co-regulation networks. We identified promising transcription factors potentially involved in the regulation of wood formation. The functional characterization of one candidate in *E. grandis* transgenic roots demonstrated its implication in secondary cell wall biosynthesis, confirming the potential of our system biology approach to identify new key regulators of wood formation in woody plants.

P19

Genetic architecture of transcriptome variation shaping wood biology in *Eucalyptus*

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Biotechnology approaches such as transcriptional rewiring and metabolic engineering have been proposed as avenues to modify wood biology and develop lignocellulosic crops tailored for various bioproducts. Such approaches depend on detailed understanding of the regulation of wood formation and the degree to which genes and pathways can be perturbed without compromising tree growth and defence. We exploited natural genetic variation affecting xylem gene expression in *Eucalyptus* interspecific hybrids to dissect the genetic architecture of wood development in these trees. Towards this, we performed transcriptome-wide co-expression and expression QTL (eQTL) analysis in 283 *E. grandis* x *E. urophylla* F2 backcross progeny. We identified 26 loci (eQTL hotspots) that perturb and shape xylem gene co-expression modules, many of which are enriched for pathways and processes key to cell wall biology. Query-based analysis of genes associated with glucuronoxylan modification, a key recalcitrance factor in industrial wood processing, identified genetic loci and expression modules linking xylan acetylation and (methyl) glucuronic acid substitution to interdependent metabolic precursor pathways. Besides identifying genetic drivers of wood property traits, our results suggest tight genetic coordination of wood cell wall formation, a strong carbon sink, with primary metabolism to ensure cellular homeostasis during the biosynthesis and deposition of wood biopolymers.

P20

Genome sequence of an Asian dipterocarp suggest paleopolyploidization with enrichment of drought response genes

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Dipterocarpaceae, which consists of more than 500 species, dominate the Asian tropical rainforest by its subfamily Dipterocarpoideae also known as the Asian dipterocarps. We present the genome sequence of an ecologically and economically important Asian dipterocarp, *Shorea leprosula* a tall emergent tree species. Our assembled genome contains 60,563 predicted protein-coding genes. Many of these protein-coding genes had similar paralogous genes and the Ks distribution for the paralogous gene pairs

suggested a whole-genome duplication event. Transcriptome data from seven different genera of the Asian dipterocarps independently supported that the whole-genome duplication (WGD) occurred in the ancestor of these dipterocarps around the period close to the Cretaceous-Paleogene extinction event ~65 mya when several other plant species also underwent a WGD. The gene ontology enrichment test revealed that a large number of drought response genes retained their paralogous pairs after the duplication event. Differential expression analysis from drought experiment further confirmed the function of the drought responsive genes. The retention of duplicated drought response genes in Asian dipterocarps after WGD may explain their current distributions in the seasonal and aseasonal tropics. Furthermore, our hypothesis that links the enriched drought response genes to the evolution of tall emergent Asian dipterocarps will be discussed.

P21

ABA – a new player in wood formation?

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Trees have to cope for decades or even centuries with ever-changing environmental conditions. A particular threat to growth and survival under climate change is low water availability. The plant hormone abscisic acid (ABA) plays a pivotal role in plant acclimation to drought. In perennials ABA has also a central function in seasonal regulation of growth when trees enter dormancy. Here, we investigated the molecular events during adjustment of the hydraulic system to water limitation. Under moderate drought which still affords low growth, the production of wood switches from few large to many small vessels with thicker cell walls. We found a strong induction of ABA levels and ABA-induced transcription factors in the developing xylem of drought-stressed trees, suggesting a function of ABA in the hydraulic acclimation of wood. To elucidate whether ABA signaling is crucial for the anatomical structure of wood we conducted ABA feeding experiments and studied transgenic poplars with modifications in ABA perception and signal processing. The results of these studies will be shown.

P22

The environmental and ecological determinants of xylem and phloem cell structural traits within and across species

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Variability in the structural traits of xylem and phloem cells lies at the basis of the differences in water and carbon economics of trees. In this study, we researched the relationships between xylem and phloem cell traits and quantified cell structural variability along gradients in water availability for 90 populations of six dominant tree species in Catalonia, NE Spain. The selected tree species included three Fagaceae (*Fagus sylvatica*, *Quercus ilex* and *Quercus pubescens*) and three Pinaceae (*Pinus sylvestris*, *Pinus halepensis* and *Pinus nigra*) for which five individuals per population (15 per species) were sampled. Xylem and phloem cell characteristics (lumen area of transport cells, transport cell density and spatial organisation and cell wall thickness) were measured on manually made micro-sections by use of Roxas and ImageJ software. For all 450 individuals (five individuals x 15 populations x six species) a comprehensive set of leaf and wood structural and hydraulic traits was also measured at individual branch and tree level to help understand and interpret the observed cell trait patterns. Population-specific climate, soil and forest structure data allowed to identify the environmental and ecological determinants of trait variation. We will present the results of the study during the conference.

P23

Characterizing the genetic basis of xylem response to drought stress in hybrid poplar

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Wood anatomy can help determine how trees respond to drought. Xylem that is efficient at transporting water tends to have larger and more interconnected vessels than less efficient xylem. Greater hydraulic efficiency, however, has been associated with greater vulnerability to drought stress and hydraulic failure. Vessel diameter decreases while vessel frequency increases in response to water stress. It is currently unknown, however, what genes are responsible for vessel size variation and drought-induced anatomical changes in xylem. A unique collection of poplar irradiation hybrid genotypes will be used during the summer of 2018 to perform dosage-dependent quantitative trait loci analyses (dQTL). These genotypes have large-scale insertions and deletions that have been mapped using whole-genome sequencing. We will measure vessel element diameter, frequency, circularity and grouping, and compare between genotypes (220) and treatments (chronic drought and control). We conducted a series of preliminary studies on 23 genotypes from this population. Our results indicated that there is significant variation in wood anatomical properties between genotypes.

P24

Torus-margo pit structure underpins the growth-longevity tradeoff in ponderosa pine

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Consistent with a ubiquitous life history tradeoff, trees exhibit a negative relationship between growth and survival both among and within species. Yet the proximate physiological mechanisms underlying this life history tradeoff are not well understood. Our hypothesis is that a morphological trait with multiple but conflicting physiological functions, such as hydraulic efficiency versus safety imposed by torus-margo pit structure in the conifer xylem, provides a mechanistic basis for the apparent life history tradeoff. We tested the extent to which torus-margo pit structure, along with other xylem morphological traits (wood density and tracheid diameters), relate to growth rates and longevity (as a proxy for survival) in two mixed-age ponderosa pine stands. Our results indicate that trees with larger torus overlap have slower growth rates and live longer. In contrast, neither wood density nor tracheid diameters explained the growth-survival tradeoff. Our results highlight a critical role of torus-margo pit structure in tree life history strategies is due to its conflicting effects on embolism resistance and water transport efficiency. Thus, we show that a measurable morphological trait, torus-margo pit structure, provides a proximate mechanism for the growth-survival tradeoff in a long-lived conifer species.

P25

Immunolocalization of matrix polysaccharides during selective delignification and simultaneous decay by white rot fungus provide evidence for specific interaction between hemicellulose and lignin in the primary and secondary cell wall of wood fibre of *Dalbergia sisso* Roxb.

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Alterations in the structure and chemistry of cell wall polymers during simultaneous decay and preferential delignification by white rot fungi could be one of the most important experimental systems to study the association of different cell wall polymers and biology of plant microbe interaction. Therefore, in the present study, we investigated the spatial and temporal changes in the distribution pattern of lignin, β-1-3 galactans (LM-5), xyloglucans (CCRCM1), less substituted xylans (LM10) and highly substituted xylans (LM 11) in the fibre cell walls of *Dalbergia sissoo* wood degraded by *Lenzites betulina* and *Daedaleopsis flavida* using immuno-electron microscopic (IEM) methods. TEM analysis of fibre walls infected with *Lenzites betulina* showed degradation of lignin from the S_1 layer of secondary walls resulting cell separation followed by selective removal of lignin from the S_2 and S_3 layers while lignin removal from cell corners and middle lamellae occurred at advanced stages of decay. The secondary walls of fibres inoculated with *Daedaleopsis flavida* did not show any gradation in the staining contrast, indicating that all wall polymers degrade simultaneously. IEM analysis revealed that LM 5 labelling is more in the compound middle lamellae while weak labelling was evident in secondary walls of degraded fibres underwent selective delignification. Less substituted xylans (LM10) distribution was more in the S_1 layer while relatively weak labelling was evident in S_2 and S_3 layers of selectively delignified fibre walls. Highly substituted xylans labelled with LM11 also showed more distribution in outer layers of secondary wall even at late stages of degradation. In contrast, the secondary wall region adjacent to cell corners was characterized by more distribution of highly substituted xylans. CCRCM1 labelling revealed the degradation of xyloglucans from the CML region undergoing selective delignification. In conclusion, our study suggests a close association between the lignin-xyloglucan in the primary wall and lignin-xylans in the secondary wall as they removed simultaneously during selective delignification in the xylem fibres.

Key words: Matrix polysaccharides, wood decay, *Dalbergia sissoo,* selective delignification, Immuno-electron microscopy.

P26 Low vulnerability to xylem embolism in leaves and stems of North American oaks

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Although recent findings suggest that xylem embolism represents a significant, drought-induced damaging process in land plants, substantial debate surrounds the capacity of long-vesselled, ring-porous species to resist embolism. We investigated whether recent methodological developments could help resolve this controversy within Quercus, a long-vesselled, ring-porous temperate angiosperm genus, and shed further light on the importance of xylem vulnerability to embolism as an indicator of drought tolerance. We used the optical technique to quantify leaf and stem xylem vulnerability to embolism of eight Quercus species from the Mediterranean-type climate region of California, USA, to examine absolute measures of resistance to embolism as well as any potential hydraulic segmentation between tissue types. We demonstrated that our optical assessment reflected flow impairment for a subset of our sample species by quantifying changes in leaf hydraulic conductance in dehydrating branches. Air-entry water potential varied twofold in leaves, ranging from −1.70 ± 0.25 MPa to −3.74 ± 0.23 MPa, and fourfold in stems, ranging from −1.17 ± 0.04 MPa to −4.91 ± 0.30 MPa. Embolism occurred earlier in leaves than in stems in only one out of eight sample species, and plants always lost turgor before experiencing stem embolism. Our results show that long-vesselled North American Quercus species are more resistant to embolism than previously thought, and support the hypothesis that avoiding stem embolism is a critical component of drought tolerance in woody trees. Accurately quantifying xylem vulnerability to embolism is essential for understanding species distributions along aridity gradients and predicting plant mortality during drought.

P27

Involvement of S1/P1, CAN1/CAN2 nucleases in PCD and plants development

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Programmed cell death (PCD) is an important cellular process that plays crucial role in morphogenesis, defense responses to pathogens and also in xylem development. Till now BFN1 was found as involved in nucleic acids degradation in xylem development. In previous studies have demonstrated that in plants *S1/P1 and CAN1/CAN2 nuclease families* are responsible for nucleic acids degradation. Ions and pH that establish nucleases activity were specified and suggest sequential action during PCD. GUS expression regulated by *nucleases promoters revealed expression in i.a. A. thaliana* transport tissues and trichomes. Moreover, in *ps1/ps2* line which exhibit accelerated chlorosis upon dark treatment, CAN2 was shown to have greater activity. Ps1/Ps2 and CAN1/CAN2 interacting proteins have been investigated to elucidate crosslinks between these nucleases and intramembrane proteases. Interestingly in both cases nucleus localized RNA-binding proteins were identified as Ps1/Ps2 and CAN1/CAN2 interacting proteins.

P28

The role of the vessel-associated cells in the intercellular communication in secondary xylem

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Wood (secondary xylem) is a heterogeneous tissue composed of both dead cells, like tracheary and strengthening elements and living cells, like xylem parenchyma cells. Thus, the long- and short-distance transporting pathways in wood are carried out via apoplasm and symplasm. The most interesting among living xylem parenchyma cells are vessel-associated cells (VACs), as they possess large and densely packed contact pits facing the tracheary elements and are an intriguing example of cells with possible communication at the apoplasm/symplasm interface.

To decipher the mechanisms of solute uptake from vessel elements to VACs we employed various approaches, like dye loading, co-localization studies or drug treatments. We experimentally proved that in the wood of three different tree species (maple, ash and poplar) VACs are involved in solute uptake at the apoplasm/symplasm interface, what will be discussed in the context of the mechanisms governing the processes of intercellular communication in wood.

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P29

Occurrence of tyloses in chestnut oak

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Chestnut oak (*Quercus montana,* a.k.a *Q. prinus)* is a member of the white oak group but its wood is excluded from use in wine and whiskey barrels because of inconsistent occurrence of tyloses. Wine and whiskey barrels are examples of ‘tight cooperage’ and the component boards (‘staves’) must not leak. The white oaks commonly used for staves, e.g. *Q. alba* and *Q. robur*, have pores that are consistently blocked by tyloses. As a preliminary study, I collected samples from chestnut oak logs at sawmills at different locations in the eastern United States and examined them for tyloses. The results showed great variation in the prevalence of tyloses across the radius, with no apparent association with tree growth rate, tree age, or the latitude of the source. There was some evidence for an increase in the occurrence of tyloses in trees grown at higher elevation.

P30

Functional analysis of the 5-hydroxyconiferaldehyde O-methylation in *Dendrocalamus latiflorus*

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Ma bamboo (*Dendrocalamus latiflorus*, Dla) is the most widely distributed bamboo in Taiwan. It grows extremely fast and can reach more than 20 meters within one growing season. During its growth, ma bamboo's cell walls accumulate S lignin. The 5-O-methylation of angiosperm's S lignin has long been believed to be catalyzed by the caffeic acid O-methyltransferases (COMTs). However, the information related to the 5-O-methylation of S lignin in this bamboo is limited. To gain insight into the mechanisms underlying the 5-O-methylation of S lignin in ma bamboo, we characterized the structure of ma bamboo lignin and the function of DlaCOMTs. The results showed that the S/G ratios of ma bamboo culms increase during its growth. The cloned DlaCOMTs are highly expressed in the lignified bamboo tissues. 5-O-methylation of S lignin in ma bamboo might be similar to that in dicotyledons which use 5-hydroxyconiferaldehyde (5-HCAld) as the preferred substrate. Overexpression of DlaCOMTs in tobacco plants increase the S/G ratio of tobacco plants. Thus, the cloned DlaCOMTs function as 5-HCAld OMTs in ma bamboo lignin biosynthesis.

P31

Elucidating transcriptional regulation using eQTL mapping in *Populus*

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Genetic control of transcription is highly complex in higher plants, especially in woody species. To reveal the genetic regulatory overview in woody model plant *Populus trichocarpa*, we performed an expression quantitative trait loci (eQTL) mapping enabled by the whole-genome resequencing analysis and RNA-seq analysis of gene expression. From 390 *P. trichocarpa* accessions leaves and 444 accessions xylems transcriptomic data, 16,030 leaf genes and 15,496 xylem genes with high variation in expression in the population were used in eQTL mapping. We found that *cis*-eQTL accounted for 10.9% and 13.7% of the top 1000 variable genes in leaf and xylem from the population, respectively. Enriched transcription factor binding sites (TFBS) including *cis*-eQTL showed tissue-specific divergence. *trans*-eQTL analysis identified 31 and 83 hotspots significant associated more than 50 genes' expression in leaf and xylem, respectively. In addition, the phenotypes including 13 photosynthesis parameters, 37 metabolites profiles and 20 ions contents were combined to explore their key transcriptional regulators. We identified novel transcriptional regulators modulating carbon flow between the shikimate and glycolysis pathways and the phenylpropanoid and tryptophan pathways. The eQTL study is being applied to identify transcriptional regulators underlying complex phenotypes of biomass quality and yield.

P32

Wood physical and mechanical traits of major gymnosperm *vs* angiosperm trees in China

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Wood is central to our basic understanding of the evolution and diversification of land plants. As the defining feature of trees, wood provides mechanical support and other functions for tree body. However, there is large variation across diverse tree species, but lack of a systematic assessment. Here we combined allometry, PCA and phylogeny analyses to explore the variations in 12 traits of the major gymnosperm and angiosperm trees in China. The results are summarized below: i) Wood traits were numerically lower in gymnosperms than angiosperms. ii) Air-dried density (ADD) and tangential shrinkage coefficient (TSC) are key traits that summarize the correlations among wood traits for gymnosperms, while ADD and radial shrinkage coefficient (RSC) represent those for angiosperms. The allometric analysis showed that traits were positively related with each other, except that TSC was negatively related with density-related (ADD, BD) and hardness-related traits (HES, HRS and HTS) in gymnosperms. iii) Phylogeny had significant effects on some wood traits of gymnosperms, but no effects on the traits of angiosperms. Our analyses demonstrated a systematic difference in wood traits between two major plant categories, which suggests the evolutionary divergence (TSC, RSC) and convergence (ADD) in key functional traits among woody plants.

Keywords: allometry, functional traits, phylogeny, shrinkage coefficient, wood density

P33 Field trial study of transgenic poplars expressing a monolignol 4-*O*-methyltransferase

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Producing lignocellulosic biofuels and bio-based chemicals from woody biomass is currently hindered by the presence of lignin in plant cell walls. Manipulating the monolignol biosynthetic pathway offers an approach to improve processability, but often impairs plant growth and development. Previously a set of novel monolignol 4-*O*-methyltransferases (MOMTs) was engineered in our lab. MOMT4, one of these OMT variants, effectively catalyzes the 4-*O*-methylation of monolignols, and shows a catalytic preference for methylating sinapyl alcohol. Previously we found that expressing the MOMT4 in poplar trees alters lignin structure and reduces lignin content in the transgenic plants and woody biomass from the transgenic poplars grown in a greenhouse exhibited significantly increased digestibility and ethanol yield when they were subjected to fermentation. Moreover, no biomass yield penalty was observed in the engineered trees grown in a greenhouse. Our purpose in this field trial study is to further examine the growth and cell wall biomass properties of the transgenic poplars grown in the natural environment. We harvested 75 transgenic plants each grown for a period of about 1 year and 2 years in the field and determined their woody biomass yield, wood density, saccharification efficiency and fermentative conversion to ethanol. The data indicate that transgenic poplars grown in the field keep the promising properties that were observed in the trees grown in greenhouse, suggesting the engineered trees could be the promising feedstock used in industry.

P34

The costs of tension wood formation in *Populus*

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Tension wood (TW) in *Populus* is characterized by gelatinous (G) fibers, reduced vessel frequency, and eccentric radial growth, the latter often ascribed to accelerated cambial activities. Presumably, therefore, TW formation incurs local growth costs against opposite wood (OW) development. Here we report that the overall growth of tilted trees was severely penalized, indicative of both local and systemic effects of TW formation on growth. Whole-plant and time-course monitoring revealed decreases early on in leaf expansion, leaf emergence, height, and diameter growth of tilted trees, followed by increases in diameter growth, but only in a relatively small zone. In contrast, G-fiber deposition and reduction in vessel number were observed along the entire inclined stem regardless of whether radial growth was increasing (classic) or decreasing (atypical TW). Fasciclin genes were upregulated differentially in classic vs. atypical TW, and most NAC transcription factors exhibited altered expression only in classic TW. Sharply increased bark hexose levels on the OW side did not correlate with radial growth or TW volume, and instead point toward metabolic adjustments related to water distribution. This is supported by widespread misregulation of aquaporins both across and along the tilted stem, and by decreased vessel numbers and impaired water uptake on the TW side. These findings suggest that TW formation depends on multiple, perhaps independent responses to tilting, and that molecular responses pertinent to cell wall modifications and cambial activities under TW-forming conditions could be decoupled for wood property engineering.

P35

Xylogenesis and seasonal radial growth patterns in *Pinus ponderosa* from a hyperarid montane site in the Mojave Desert indicate high drought resilience and phenological plasticity

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Vulnerability of tree species in the Southwestern United States under future warming scenarios depends upon their plasticity to adapt to more frequent and severe droughts. Cambial phenology of 12 ponderosa pines (*Pinus ponderosa*) from a montane stand in the Mojave Desert (Southern Nevada) was monitored for 2 consecutive years (2015–2016) characterized by opposite-sign anomalies between summer and winter precipitation. The onset of xylogenesis was driven by moisture availability and new xylem cells were observed more than two months earlier in 2016, after a wet cold-season, compared to 2015, when cellular division began only in mid-July together with monsoonal rainfalls. Abundant monsoonal precipitations in 2015 also affected the anatomical features of woody tracheids (i.e. lumen area, cell wall-thickness), triggering the formation of several Intra Annual Density Fluctuations (IADFs) which were absent in 2016. Sub-hourly stem size variations measured by point dendrometers confirmed the opportunistic growth strategy of ponderosa pine and its ability to utilize episodic precipitation events. These findings show the high resilience to drought of this species expressed as phenological avoidance of hyperarid periods. Given the uncertainties in future precipitation patterns simulated by global change models for the Southwestern United States, the dependency of ponderosa pine on seasonal moisture implies a greater conservation challenge than for species responding mostly to temperature.

P36

Wood water storage and its anatomical underpinnings in temperate angiosperms

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Water stored in wood creates a buffer, which presumably plays a significant role during drought. Most information about water storage strategies relates to tropical trees and we know very little about temperate trees. The structural drivers of water storage remain unclear too. We asked: how much water can be stored and released from wood into the transpiration stream, and which tissue drives this hydraulic behavior?

We examined water storage, capacitance (water released per change between predawn and midday water potential) and wood anatomy in twigs of 30 temperate, deciduous tree species grown in the Arnold Arboretum of Harvard University in Boston, USA. The species represented a broad phylogenetic range and diverse anatomies.

The difference in wood water content between predawn and midday ranged from ~0.5 to 5% and capacitance varied from near 0 to ~100 kg m^{-3} MPa^{-1}, with one outlier species reaching above 400 kg m^{-3} MPa^{-1}. Species with higher parenchyma lumen fraction tended to store water closer to their maximum storage capacity, but only in ring- and semi-ring-porous species, not in diffuse-porous. Species with higher fibre lumen fraction tended to store less water. Capacitance did not correlate with any tissue fraction suggesting tissue fractions do not constrain capacitance.

Late abstract

Plant height and hydraulic vulnerability to drought and cold

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Understanding how plants survive drought and cold is increasingly important as plants worldwide experience dieback with drought in moist places and grow taller with warming in cold ones. Crucial in plant climate adaptation are the diameters of water-transporting conduits. Sampling 537 species across climate zones dominated by angiosperms, we find that plant size is unambiguously the main driver of conduit diameter variation. And because taller plants have wider conduits, and wider conduits within-species are more vulnerable to conduction-blocking embolisms, then taller conspecifics should be more vulnerable than shorter ones, a prediction we confirm with a plantation experiment. As a result, maximum plant size should be short under drought and cold, which cause embolism, or increase if these pressures relax. That conduit diameter and embolism vulnerability are inseparably related to plant size helps explain why factors that interact with conduit diameter, such as drought or warming, are altering plant heights worldwide.

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