Welcome to the 2024 ASPS & NZSPB Conference

28th November 2024
University of Western Australia (Crawley campus) and online

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AUSTRALIAN SOCIETY OF PLANT SCIENTISTS

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ASPS2024 WA Meeting Program
Thursday 28 November 2024

8.00 am	Registration
8.30 am	Welcome Address and Acknowledgement of Country Judith Lichtenzveig (ASPS WA Conference Chair)
8.45 am	**Session 1 (Wilsmore Lecture Theatre, UWA)**
	Chair: Judith Lichtenzveig
8.45 am	**Plenary talk 1: Shahal Abbo (Hebrew University of Jerusalem)** What can we learn from the study of Near Eastern plant domestication?
9.15 am	**FL1 Virginia Wainaina (CSIRO)** Cracking the Code: transcriptional insights into lupin seed development.
9.30 am	**FL2 Lina Isaza Lopez (UWA Albany)** Interactions fire and demographic attributes of *Banksia* media in the Fitz-Stirling region, Southwestern Australia.
Speed talks (3 minutes)	
9.45 am	**SP1 Yan Ai (UWA)** Atrazine application on triazine-resistant canola suppresses white leaf spot (*Neopseudocercosporella capsellae*) development.
9:48 am	**SP2 Agyeya Pratap (UWA)** Unravelling heat tolerance in bread wheat (*Triticum aestivum* L.) using physiology and proteomics approaches.
9:51 am	**SP3 Andrew Tuckey (UWA)** Negative regulation of KAI2 signalling in angiosperms.
9.54 am	**SP4 Madhoolika Karunakaram (UWA)** Examining changes in the spatial expression of key photorespiratory enzymes during the evolution of C4 photosynthesis in the genus *Flaveria*.
9:57 am	**SP5 Benjamin Hearn-Thomas (UWA)** Investigating the functionality of LYR domain proteins in *Arabidopsis thaliana*
10.00 am	**Morning Tea & Poster session (Bayliss Building)**
	Note: SP1- 5 will have corresponding abstracts under the 3-minute talks section in the booklet, SP6-SP12 abstracts are found in the "poster-only" section.

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10.30 am	**ASPS & NZSPB Joint Awards session (Wilsmore Lecture Theatre)**
	Chairs: Martha Ludwig & Mark Waters (ASPS)
via Zoom	**NZSPB Roger Slack Award: David Chagné (Plant & Food Research, Papaioea; Genomics Aotearoa)** Genomics for restoring a critically threatened tree species in the rohe of Rangitāne o Manawatū
via Zoom	**ASPS Goldacre Award: Peter Crisp (UQ)** The DNA hypomethylome: unlocking crop epigenomics to uncover and engineer hidden diversity
via Zoom	**ASPS Education and Outreach Award:** **Ashley Jones and Benjamin Schwessinger (ANU)** Genomics for all: a cross-sector effort to make cutting-edge genomics a cornerstone of education, industry, government, and public health.
via Zoom	**NZSPB Elected Fellows to the NZ Royal Society:** **Andy Allan (University of Auckland, New Zealand)** Fast flowering as a tool for gene discovery in woody perennials
	Kevin Davies (Plant & Food Research, New Zealand) The evolution of flavonoid biosynthesis
via Zoom	**ASPS Jan Anderson Award: Jenny Mortimer (UoA)** Sweet green tales: efforts to unravel the complexities of plant polysaccharides
Live!	**ASPS J.G Woods Award: Sergey Shabala (UWA)** Cell-based phenotyping for breeding crops for future climates.
1:15 pm	**Lunch and Poster session (Bayliss Building)**
	Let's vote! Note: SP1- 5 will have corresponding abstracts under the 3-minute talks section in the booklet, SP6-SP12 abstracts are found in the "poster-only" section.
2.15 pm	**Session 2 (Wilsmore Lecture Theatre, UWA)**
	Chair: Razlin Azman (CSIRO)
2.15 pm	**Plenary talk 2: Megan Ryan (UWA)** Arbuscular mycorrhizal fungi: ideology vs strong science rooted in a farming systems context
2.45 pm	**FL3 Fatima Naim (Curtin University)** Spatiotemporal phenotyping and multi-omic analysis of net blotch disease in barley

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3.00 pm	**FL4 Alistair Hockey (UWA)**
	Natural factors affecting genome stability and interspecific gene flow – the case of *Cicer echinospermum,* a close relative of chickpea
3.15 pm	**FL5 Samantha Norman (UWA)**
	Caught in the middle: the role of the parenchymatous sheath in C4 *Neurachne* species.
3.30 pm	**FL6 Ping Yun (UWA)**
	Using MIFE technique to determine cellular ion flux: a case study on leaf salinity tissue tolerance in wild rice *Oryza coarctata*.
3.45 pm	**Afternoon Break**
4.00 pm	**Session 3 (Wilsmore Lecture Theatre, UWA)**
	Chair: Alistair Hockey (UWA)
4.00 pm	**FL7 Ruby Wiese (UWA)**
	Investigation of changes in seed yield and quality during maturation of subterranean clover to evaluate a novel seed harvesting method.
4.15 pm	**FL8 Clément Gille (UWA)**
	Accumulation of manganese in leaves of carboxylate-releasing *Hakea* species (Proteaceae) depends on cluster-root physiology.
4.30 pm	**FL9 Saurabh Saha (UWA)**
	The LYR domain of the Complex I Subunit B22 is crucial for an interaction with the mitochondrial Acyl Carrier Protein ACP2.
4.45 pm	**FL10 Eleonora Davide (UWA)**
	Cyclic mononucleotide-dependent ion fluxes in *Arabidopsis thaliana* - a tale of two isomers
5.00 pm	**Closing address, prize presentation, group photo**
	Wilsmore Lecture Theatre, UWA
5.30 pm	**Social gathering & conference dinner**
	Sunken Gardens, UWA

Welcome

Welcome to the ASPS WA conference 2024!

The WA organising committee acknowledges we are meeting in Whadjuk Bodja, lands and water of the Noongar people at the University of Western Australia (UWA). The university campus is by Goordandalup (Crawley Bay), which continues to be a place to gather and learn for tens of thousands of years. UWA has the ultimate privilege of sitting on this sacred soil where Western Australian kaartdijin (knowledge) began[1]. We pay our respect to the past and present elders of the Noongar and other First Australian nations.

The Australian Society of Plant Scientists (ASPS) exists to promote the interests of the plant science profession in Australia. This year's conference is a hybrid event, where in-person sessions combine with an online session coordinated across Australia and, for the first time, colleagues from the New Zealand Society of Plant Biologists.

This conference is an excellent opportunity for Western Australia's young plant scientists to highlight and share their exciting work. The meeting represents one of the few opportunities for junior scientists to share the podium with more established researchers and will have a relaxed and informal atmosphere that allows all those attending to exchange views and develop useful contacts within academia and industry.

We are very pleased to be joined by Dr Shahal Abdo (Hebrew University of Jerusalem) and Prof Megan Ryan (School of Agriculture and Environment, UWA) as our plenary speakers this year. In addition, we welcome talks from the 2024 ASPS award winners Peter Crisp (UQ, Peter Goldacre Award), Jenny Mortimer (UoA, Jan Anderson Award), Ashley Jones and Benjamin Schwessinger (ANU, Education and Outreach Award) and Sergey Shabala (UWA, JG Wood Lecture) at the online session.

The organising committee would like to thank all the sponsors whose generous support made this year's event possible: BioPlatforms Australia; ARC Centre of Excellence for Plant Success in Nature and Agriculture; ThermoFisher Scientific, Functional Plant Biology; Agrisera Antibodies; and Annals of Botany Company. In addition, we also thank our local WA sponsors, namely the Australian Plant Phenomics Network (APPN), AGRF, Qiagen, and John Morris Group.

Finally, we would like to thank all delegates for your participation in the 2024 ASPS conference, and we hope you will enjoy the opportunity to get out of the lab/office and talk shop with fellow researchers.

The organising committee

The organising committee

The following people have generously donated their time and talent to make the WA ASPS 2024 hybrid conference possible:

Judith Lichtenzveig (UWA)
Mark Waters (UWA)
Razlin Azman (CSIRO, Phytogen editor)
Alistair Hockey (UWA)

In addition, the following people have been working to make the ASPS 2024 conference become reality nationwide:

Martha Ludwig (UWA)
Kim Johnson (La Trobe University)
Crystal Sweetman (Flinders University)
Dugald Reid (La Trobe University)
Lim Chee Liew (La Trobe University)
Lucas Auroux (La Trobe University, Phytogen editor)
Frances Sussmilch (UTAS)
Mark Waters (Treasurer, UWA)
Janet Wheeler (ASPS webmaster)
Nipuni Thanthrige (UQ)
Brett Williams (QUT)
Brent Kaiser (Sydney University)
Kristine Crous (Western Sydney University)
Rob Sharwood (Western Sydney University)
Nijat Imin (Western Sydney University)

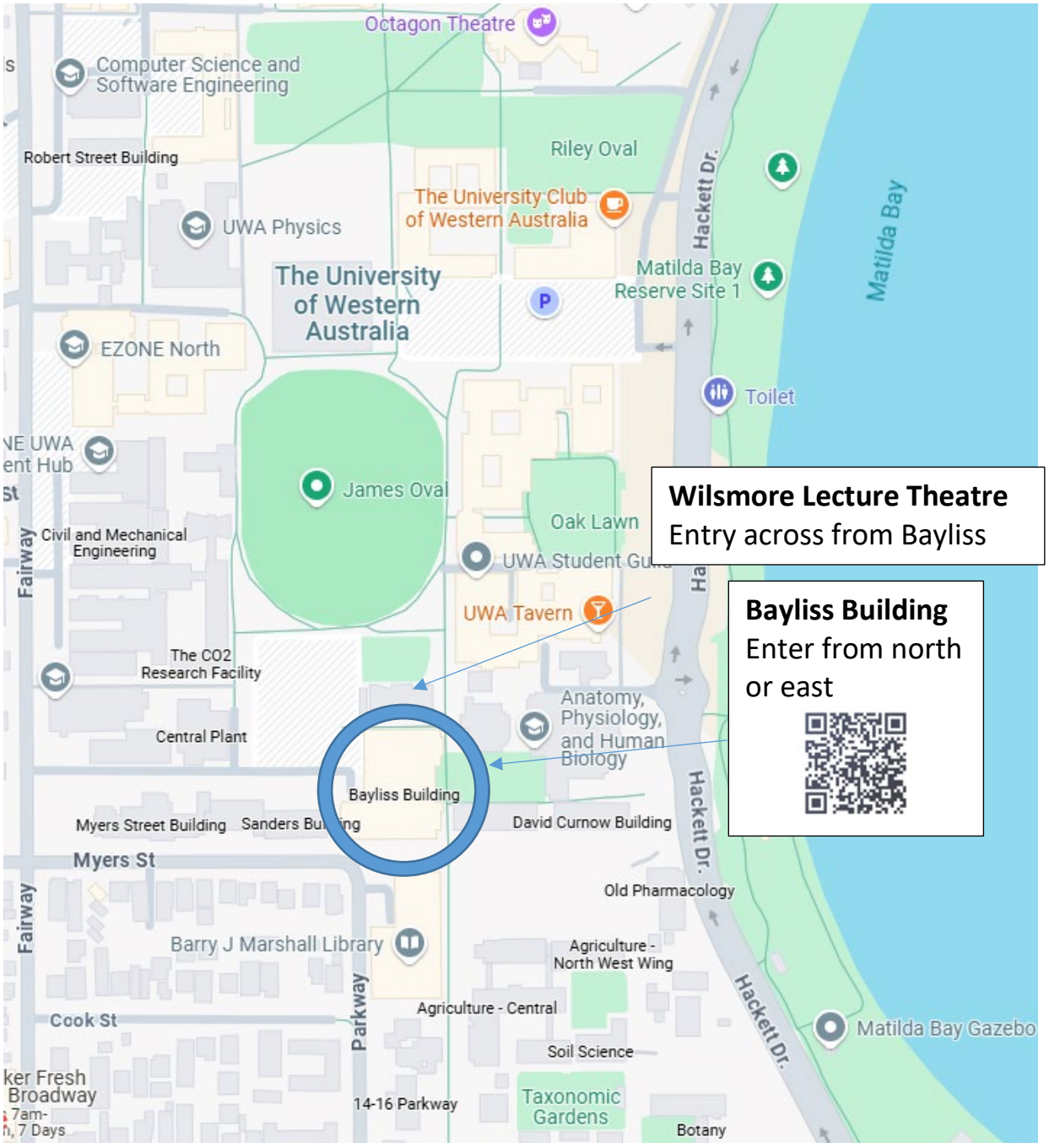
We would like to thank all our session chairs and talk and poster judges for their time and assistance.

General information

Registration desk

Registration is at the Baylis building foyer of UWA Crawley campus and will open from 8 am. If you are exhibiting a poster, please try to put it up in the Bayliss building foyer before registering, so it is displayed in time for the morning tea session.

Use the QR codes for directions.



Poster presentations

The poster session will be held at morning tea 10:00-10:30am in the Bayliss foyer. Please put up your poster (according to your poster number) as instructed by the organizing committee. Velcro for hanging on the poster boards will be available on the boards themselves. Please be prepared to give a summary to your audience between 10:10 and 10:30am

Oral presentations

All sessions will be held in the Wilsmore Lecture Theatre. Speakers will have access to a PC desktop. Unless prior arrangements have been made, speakers should bring their presentation on a USB stick to the registration desk at least 20 minutes before their session starts, or at the end of the prior session. Speakers are allocated 3- 15- or 30-minute slots according to the program. This will include 13 or 18 minutes for the presentation, with an additional two minutes for questions. The session chairs have been asked to enforce these timings. 3-minute presentations (SP1-5) will have no questions, as these speakers will also have a poster to complement their talk.

ASPS Joint Awards presentations

We will be joining up with the other ASPS nodes for live presentations. All will be screened in the Alexander Lecture Theatre; we will have the benefit of Prof Sergey Shabala, presenting locally in person. Each presentation will be chaired in turn by the local host node, but we encourage audience questions from all nodes.

Sustainability

To reduce the environmental impact of this meeting, we encourage you to minimise waste. Please use your own water bottles, print posters on non-laminated paper, return unwanted lanyards and recycle this programme book. We also encourage you to bring your own food container to keep any leftovers from lunch.

Conference Dinner

The conference dinner will be held at The Sunken Gardens, UWA. Your dinner ticket includes one alcoholic or soft drink – after that, drinks are on your own dime!

Plenary talks

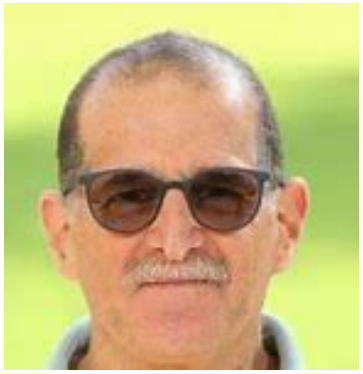
What can we learn from the study of Near Eastern plant domestication?

Shahal Abbo

The Hebrew University of Jerusalem, Israel

In recent years many researchers describe plant domestication as a long (protracted), unconscious (circumstantial), geographically diffused (culturally independent) process. This reconstruction is endorsed by geographers, archaeologists, archaeobotanists, population geneticists and plant scientists who are interested in plant domestication. An alternative model of plant domestication as a punctuated, knowledge-based human initiative that was rapid, geographically localized, fully conscious, mostly monophyletic, that gave rise to a nutritionally agronomically balanced crop package will be presented. The conceptual distinction between 'plant domestication' and 'crop evolution' will be presented as a potential guiding principle to overcome some of the inherent problems associated with the popular (protracted) model. Possible future breeding and research implications of this distinction will be highlighted.

About the speaker



Shahal Abbo is an agronomist and plant geneticist at the Hebrew University of Jerusalem, Israel. Through comparative study of grain legumes and cereals, both domesticated and wild, across Mediterranean agro-eco-systems, he developed several new practical and conceptual tools pertaining to plant domestication and crop evolution.

Arbuscular mycorrhizal fungi: ideology vs strong science rooted in a farming systems context

Megan Ryan

School of Agriculture and Environment, University of Western Australia, Australia

Arbuscular mycorrhizal fungi (AMF) are commonly portrayed in the popular and scientific press, as well as by product sales-people, as “saviours” of the soil and critical for global food security. This narrative highlights AMF as pivotal to the functioning of farming systems and suggests they should be prominent in the minds of farmers as they make management decisions. Yet, the vast majority of research has been undertaken under controlled conditions in glasshouse and for southern Australia cropping systems there are no data from field trials that suggest farmers need to actively manage AMF. In this talk I will explore why there is a mismatch between the AMF-focussed literature and the agronomy literature and why these matters (a lot!).

About the speaker

Professor Megan Ryan is a teaching and research academic at the University of Western Australia. Megan’s research interests encompass crops and pastures, with a focus on annual pasture legumes, as well as organic amendments and agronomy. A strong research theme has been roots and plant nutrition, especially in regard to phosphorus. For over 30 years, Megan has studied arbuscular mycorrhizal fungi with a focus on their role in farming systems: her team recently discovered that these fungi are more diverse than previously suspected, being spread across two phyla.

2024 ASPS and NZSPB Award Winner Talks

NZSPB Roger Slack Award

Genomics for restoring a critically threatened tree species in the rohe of Rangitāne o Manawatū

Colan Balkwill[1], Keith Funnell[2], Emily Koot[2], Julie Deslippe[1], Alana Nuku[3], Paul Horton[3], Wayne Blissett[3], David Chagné[2,4]

[1] Victoria University of Wellington, Wellington; [2] Plant & Food Research, Papaioea; [3] Rangitāne o Manawatu, Papaioea; [4] Genomics Aotearoa

Swamp maire (*Syzygium maire*; maire tawake) is an endemic tree species of Aotearoa's swamp forests that is currently listed as nationally critical due to habitat loss and, most recently, infection by myrtle rust. With fewer than twenty mature trees of swamp maire remaining within the Rangitāne o Manawatū rohe, including a remnant population under threat from the construction of Te Ahu a Turanga Manawatū Tararua, a Mana Whenua-led project was set up for conserving the species in the rohe, in accordance with Rangitānenuiarawa (Rangitāne o Manawatū tikanga). Genome sequencing of naturally occurring trees and seedlings from within the rohe was performed to generate knowledge of genetic diversity. A high-quality reference genome was assembled for the species, becoming the first genome sequence to be named by an indigenous group (Ngā Hua o te Ia Whenua). This genomics-based mahi focused on understanding the past and current population structure, how much inbreeding has occurred and how related trees are to each other and to other populations in Aotearoa. This research contributed to developing a restoration plan integrating Mātauranga Māori, genetic diversity, and habitat suitability for replanting.

ASPS Peter Goldacre Award Seminar

The DNA hypomethylome: unlocking crop epigenomics to uncover and engineer hidden diversity.

Peter Crisp

School of Agriculture and Food Sustainability, The University of Queensland, Brisbane, Qld, 4072, Australia

Decoding the information stored in nucleic acids has been transformative to our understanding of life and inheritance. However, beyond the sequence of genes, it has been more challenging to understand the rules of the DNA regulatory code in the non-coding portion of plant genomes, particularly in the vast genomes of many crop species. In addition to the DNA bases A, T, G and C, heritable information can also be stored using modified bases, such as 5-methylcytosine, commonly known as DNA methylation. Profiling the patterns of DNA methylation now enables us to rapidly distil a genome down to the relatively small fraction of regions that are functionally most valuable for trait variation. Genomic regions that lack DNA methylation, named Unmethylated Regions (UMRs), provide very useful information for decoding a plant genome because they can predict loci enriched for cis-regulatory elements. We have developed approaches that use DNA methylation profiling of a single tissue (e.g. a leaf) to discover and characterise plant UMRs, which collectively comprise the 'hypomethylome' of a species. We are using this approach in multiple plant species, in particular cereals such as sorghum, wheat, barley and maize to annotate the regulatory portion of these genomes and to investigate natural epigenetic variation. Using the new hypomethylome annotations we have also selected novel gene regulatory regions as targets for gene editing to engineer gene expression variation for trait improvement.

About the speaker

Dr Peter Crisp is a Group Leader and Senior Lecturer in the School of Agriculture and Food Sustainability at The University of Queensland. Peter's research program is focused on crop functional genomics, epigenetics and biotechnology, and has significantly advanced our understanding of the contribution of epigenetics to heritable phenotypic variation in plants. His group has invented groundbreaking technologies for harnessing (epi)genetic variation and their discoveries have led to exciting new avenues for decoding genomes and for the rational engineering of gene regulation for trait improvement in plants. Having benefited immensely from brilliant mentors, Peter is passionate about training. He leads a budding group of talented students and researchers and is a Chief Investigator in the ARC Training Centre in Predictive Breeding and the International Research Training Group for Accelerating Crop Genetic Gain. Peter is also an affiliate of the Queensland Alliance for Agriculture and Food Innovation and the ARC Centre of Excellence for Plant Success in Nature and Agriculture.

Peter completed his PhD in functional genomics and intracellular signalling at the Australian National University under the supervision of Prof Barry Pogson; then from 2017 was a Postdoctoral Fellow at the University of Minnesota, USA under the mentorship of Prof Nathan Springer working on epigenomics in maize. Peter joined UQ in 2020 as a Group Leader and Lecturer Crop Biotechnology and genomics in the School of Agriculture and Food Science in the Plant Science cluster. At UQ Peter also held an ARC DECRA Fellowship followed by a UQ Amplify Fellowship.

ASPS Education and Outreach Award Seminar

Genomics for all: a cross-sector effort to make cutting-edge genomics a cornerstone of education, industry, government, and public health.

Ashley Jones and Benjamin Schwessinger

Australian National University, Canberra, Australia

Rapid advancements in long-read DNA sequencing technologies offer unprecedented potential to transform genomic research. However, the complexity of these technologies can hinder widespread adoption. Over the past seven years, we have been at the forefront of integrating long-read sequencing into various sectors, including academia, industry, government, and public health. Through hands-on workshops, open-source resources, and collaborative networks, we have facilitated knowledge sharing, skill development, and the application of genomic technologies across diverse fields. Our efforts have led to significant advancements in student education, empowering them with practical skills, leading to improved engagement and learning outcomes. In the community, we have applied genomics to develop valuable genome resources for Australia's native flora, enhance biodiversity conservation and improve agricultural genotyping. Our collaboration with government agencies has enhanced biosecurity practices to rapidly detect invasive pathogens, while partnerships with public health organisations have contributed to addressing pressing issues like the COVID-19 pandemic. We believe our collaborative approach to education and outreach fosters a scientifically skilled workforce that empowers individuals to utilise the full potential of genomics for innovative advancements.

About the speakers

L: Ashley Jones; R:Benjamin Schwessinger

Over the past seven years, we have continuously innovated our teaching of the latest genomic analysis approaches in plant sciences, spearheading the establishment of Oxford Nanopore sequencing across Australia and in the class room. Our impact in plant science teaching and beyond has been significant, influencing multiple sectors including universities, industries, government, and communities. Key to these achievements, is that we pursue a highly integrative approach to teaching from wet lab protocol development to dry lab analysis pipeline sharing. In addition, we focus on development of wider scientific communities, as we share teaching materials and protocols online via Protocols.io.

ASPS Jan Anderson Award Seminar

Sweet green tales: efforts to unravel the complexities of plant polysaccharides

Jenny C. Mortimer[1,2,3,4]

[1]School of Agriculture, Food and Wine, & Waite Research Institute, University of Adelaide, Australia; [2]ARC Centre of Excellence in Plants for Space, University of Adelaide, Australia; [3]ARC Training Centre in Future Crops Development, University of Adelaide, Australia; [4]Joint BioEnergy Institute & Lawrence Berkeley National Laboratory, Berkeley, CA 94608, USA

Plant glycosylation is a highly complex and essential biological process, yet it is still poorly understood. From glycan modifications of proteins and metabolites to storage polysaccharides such as starch, to the structural complexities of cell wall polysaccharides, these shape how plants grow and respond to the environment. My research has focused on understanding how these complex glycans are formed, how their structure relates to their function, and how we can harness them to support a transition away from fossil-fuel based technologies.

Here, I will highlight our work in identifying key enzymes involved in cell wall and sphingolipid biosynthesis, and our progress in linking glycan structure to function. I will also explore how we are engineering these glycans to enhance biomass traits for biorefinery applications, extending their utility beyond serving as a carbon source for microbial conversion to a broader range of sustainable uses. Finally, I will discuss how understanding the regulation of plant cell wall biosynthesis is important when growing plants in a controlled environment, whether in a vertical farm on earth, or on the lunar surface.

About the speaker

Jenny Mortimer is Associate Professor of Plant Synthetic Biology in the School of Agriculture, Food and Wine at the University of Adelaide (UoA), Australia, where she is also Deputy Director (Interim) of the Waite Research Institute. She is Chief Investigator (CI) and UoA node leader of the ARC Centre of Excellence Plants for Space (P4S). She is also an Affiliate Staff Scientist at Lawrence Berkeley National Laboratory, USA, and a Director of Plant Systems Biology at the Joint BioEnergy Institute, USA. After completing her PhD at Cambridge University, UK, she began exploring how engineering the plant cell wall could deliver sustainable and economically viable biofuels: first as a postdoc in Cambridge, then as a research fellow at RIKEN Japan, before joining Berkeley Lab in 2014, and Adelaide in 2021. Her team's research focuses on understanding and manipulating plant cell metabolism, with a focus on complex glycosylation. The goal is to develop knowledge and crops which contribute to a sustainable and renewable bioeconomy.

At Adelaide, her group is using synthetic biology to develop new crops for food and materials production in controlled growth environments – including for Space settlement (P4S), applying new agricultural biotechnologies to develop resilient field crops as a CI in the ARC Training Centre for Future Crops Development, and developing Australian feedstocks for sustainable jet fuel as a CI in the ARC Research Hub for Engineering Plants to Replace Fossil Carbon. She collaborates extensively internationally, and projects include a UK Space Agency funded project to develop a plant growth facility for Axiom Station, and a NASA funded project to develop a payload for Artemis III, the mission that will return humans to the surface of the moon. She was selected as a World Economic Forum Young Scientist (2016/17), where she contributed to the WEF Code of Ethics for Researchers, and she is an editor for the society journals Plant Cell Physiology and Plant Journal. You can find out more information on the lab here: mortimerlab.org

ASPS JG Wood Lecture

Cell-based phenotyping for breeding crops for future climates

Sergey Shabala

School of Biological Sciences, University of Western Australia, Crawley WA6009, Australia

Agriculture is vulnerable to climate change, and sustainable agricultural food production will be not achievable by the current agronomical and breeding practices, due to impact of climate changes and associated abiotic stresses on crop performance. At the same time, tolerance to key abiotic stresses (such as drought; heat; salinity; flooding) is conferred by multiple mechanisms. Each of them operates in a specific tissue/cell type and is regulated by multiple genes. In this context, the (empirical) whole-plant phenotyping (regardless of whether it is hyperspectral imaging, or ionomics, or any other whole-plant based trait) will be always critical for a final validation of genetic material (e.g., in field trails) but is unlikely reveal the role of a specific mechanism/gene, amongst others. To be more effective, breeding targets can be directed towards specific mechanisms. In this talk, I will argue for a need for a paradigm shift from whole-plant to cell-based phenotyping approach and discuss its current prospects and limitations. Using salinity stress as an example I will show the pitfalls of the whole-plant phenotyping approach for crop breeding, and then illustrate how using cell-based phenotyping platforms allow to overcome this problem. I will then demonstrate how combining novel electrophysiological and imaging techniques can be used for discovery of the candidate genes and/or QTLs conferring not only salinity but also tolerance to other abiotic and biotic stresses. I will also argue for a need for a broader use of wild relatives, to regain abiotic stress tolerance that was lost during domestication process.

About the speaker

I was trained as an electrical engineer (B Eng Hon 1984) but then became fascinated by living systems and moved into biology, receiving a PhD in Plant Physiology in 1989 from the Institute of Experimental Botany in Minsk (former USSR). In 1995 I came to University of Tasmania as a post-doc in biophysics. In 1998 I got my tenure in the School of Agricultural Science where I have been working until 2023. During this time, I have built a highly productive laboratory focusing on stress physiology and membrane transport in plants, exploring mechanisms of plant sensing and adaptation to harsh environmental conditions such as drought, salinity, waterlogging, oxidative stress, and nutritional disorders. After working at UTAS for 28 years, I have moved to Western Australia to become a UWA Chair in Plant Physiology in June 2023. Over my research career, I have published over 460 peer-reviewed papers and supervised to completion 58 PhD students.

NZSPB Elected Fellows to the NZ Royal Society

The Evolution Of Flavonoid Biosynthesis

Kevin M. Davies[1], Nick W. Albert[1], Yanfei Zhou[1], Samarth Kulshrestha[1], Rubina Jibran[2], John W. van Klink[3], David Chagné[1], Marco Landi[4], Peter Schafran[5], Fay-Wei Li[5], Stefan J. Hill[6], John L. Bowman[7]

*[1] Plant & Food Research, Private Bag 11600, Palmerston North 4442, NZ; [2] Plant & Food Research, Private Bag 92169, Auckland Mail Centre, Auckland 1142, NZ; [3] Plant & Food Research, Department of Chemistry, Otago University, Dunedin 9054, NZ; [4] Department of Agriculture, Food and Environment, University of Pisa, Italy; [5] Boyce Thompson Institute, Ithaca, NY 14853, USA; [6] Scion, Private Bag 3020, Rotorua 3046, NZ; [7] School of Biological Sciences, Monash University, Melbourne, VIC 3800, Australia * kevin.davies@plantandfood.co.nz*

The flavonoid pathway is characteristic of land plants and a central biosynthetic component enabling life in a terrestrial environment. It is one of the most studied plant characters, and the subject of >15,000 journal articles each year. However, it is only with the recent advent of model systems for non-seed plants, that we have started to understand how and why the pathway may have evolved. The phylogenetic and functional data on non-seed plants challenge the idea of a canonical flavonoid pathway inherited from the Last Common Ancestor (LCA) of all land plants. Rather, it suggests extensive gene losses and gains within each lineage.

Some flavonoid pathway branches are well conserved and may have been present in the LCA. In particular, the UVR8/HY5-mediated induction of colourless flavonoids for tolerance of UVB-light is strongly conserved between Arabidopsis and the liverwort *Marchantia polymorpha*. In contrast, the stress-related red pigments have striking biosynthetic and functional diversity. Notably, the red pigments of liverworts are a previously unreported flavonoid type 'auronidins' that are cell-wall located polymers that provide protection against abiotic and biotic stresses. One lineage, the hornworts, has lost flavonoid biosynthesis entirely. Yet genome sequencing for eight hornwort genera found a single 'canonical' flavonoid biosynthetic gene in the phylogenetic outlier hornwort species. Thus, the hornwort ancestor may have inherited the flavonoid pathway, but the biosynthetic and regulatory genes were lost during lineage-specific evolution. The results illustrate the importance of extending studies out from the usual suspects of plant models and across the embryophyte diversity.

Full Length Talk Abstracts

ASPS24 WA – TALK FL1

Cracking the Code: Transcriptional Insights into Lupin Seed Development

Virginia Wainaina[1], Tina Rathjen[2], Annelie Marquardt[3], Natalie Fletcher[1], Hayley Cassaroto[1], Meredith McNeil[3], Kerensa McElroy[2], Lingling Gao[1]

[1]CSIRO, Floreat, WA, Australia; [2]CSIRO Black Mountain, ACT, Australia; [3]CSIRO St Lucia, QLD, Australia

Lupin (Lupinus spp.) seeds are valued for their high protein content (35-40%) for both human and animal consumption. Seed development in crop plants is a critical factor influencing seed fate and yield, hence, understanding the molecular mechanisms of seed development is essential. This study conducted a transcriptome analysis of Narrow Leaf Lupin (NLL) seed development at 3, 6, 9, 12, 15, 18, and 21 days after flowering (DAF) to investigate transcriptional dynamics and identify key candidate genes that control seed development. Key findings include the upregulation of cytochrome P450 genes in later stages, implicating their role in secondary metabolism and defense during seed maturation. At 3 DAF, genes like annexins and lipoxygenases were active, supporting initial development and stress responses. GO term enrichment analysis showed parallels with other dicot seeds, highlighting cell division, expansion, and maturation phases, with enriched functions in catalytic and metabolic processes within cellular structures. Stage-specific expression patterns revealed a reduction in transcriptome activity as seeds matured, with genes encoding glutathione transferase and thioredoxin-like proteins supporting oxidative stress protection and cellular metabolism in early stages. The conglutin protein families (ALPHA, BETA, and DELTA) were upregulated in late stages, pointing to their role in nutrient storage. Additionally, limited expression of the lysine/ornithine decarboxylase (LCD) gene, essential for alkaloid biosynthesis, suggests potential antinutritional regulation. These findings provide insights into lupin seed biology and suggest target genes for future breeding to enhance nutritional qualities and stress resilience in lupin.

Fire and demographic attributes of Banksia media in the Fitz-Stirling region, Southwestern Australia.

Lina Marcela Isaza Lopez
The University of Western Australia, Australia

Banksia media commonly known as the golden stalk Banksia, is one of 170 recognized species within the genus, which are keystones for biodiversity in southwestern Australia. Found on nutrient-poor soils along the southern coast, these plants have yellow, orange, or brown flowers that develop into woody cones with seeds enclosed in follicles. *Banksia media* is well-adapted to fire, as heat triggers seed release, a vital regeneration mechanism for these plants. This adaptation, however, faces challenges due to shifts in climate, fire regimes, and land use changes, affecting their ability to thrive and maintain population density and fertility across different fire-interval sites.

This study examined *B. media* across three sites with varying fire intervals, analyzing population density, plant size, and reproductive traits. Findings showed that older sites had lower plant densities and greater canopy sizes, while younger sites displayed higher density with smaller canopy areas. Cones from different age groups (young, medium, and old) were also evaluated, revealing significant differences in productivity and cone age distribution between sites. The oldest site produced the most cones and showed higher fertility rates compared to younger sites, indicating that time since fire (TSF) strongly influences B. media's growth habits, density, and reproductive output. This research highlights the importance of fire intervals and site age in sustaining *Banksia media* populations and the broader ecosystem they support.

Spatiotemporal phenotyping and multi-omic analysis of Net blotch disease in barley.

Yutathkarn Coles[1], Sabrina Cuellar[1], Ciara Gifford[1], Jade Davis[1], Robabeh Ghavamabad[1], Ayalsew Zerihun[1], Mark Gibberd[1], Fatima Naim[1]

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Net blotch is a damaging barley disease caused by the necrotrophic fungal pathogen *Pyrenophora teres*. It occurs in two phenotypically distinct forms: net form net blotch caused by *Pyrenophora teres f. teres* (Ptt), and spot form net blotch caused by *Pyrenophora teres f. maculata* (Ptm). The recent emergence of fungicide-resistant and increasingly virulent pathotypes and the lack of resistant barley germplasm pose a serious threat to our barley production. To understand the mechanisms of pathogenicity, differences in virulence, and differential phenotypes, we have applied spatially resolved high-resolution microscopy and multi-omic techniques. This included a combination of leaf images to capture disease development, confocal microscopy, long-read genome assemblies of the pathogen, and gene expression profiling using long-read sequencing from targeted tissue sampling. I will present our recent findings in disentangling this complex interaction paving the way to better management of the disease.

Natural factors affecting genome stability and interspecific gene flow – the case of *Cicer echinospermum*, a close relative of chickpea.

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The genesis, persistence, and effect of post-zygotic barriers to gene flow is consequential in the genomic stability and genetic isolation of diploid species; near-complete homologous chromosome pairing at meiosis is required to produce viable gametes and genetic incompatibilities are less likely to be buffered by variable expression of gene copies. Here, we discuss how genome instability intrinsic to *Cicer echinospermum* P.H. Davis shapes gene flow within the species and with its close relative species, chickpea (*C. arietinum* L.). The two autogamous diploid species (2n = 16) occur in natural sympatric populations and differ in genome structure by at least one reciprocal translocation. Lower than expected pollen viability and lack of seed set in interspecific hybrids suggest additional barriers to gene flow in *C. echinospermum*. We evaluated the microspore development, pollen viability, seed fertility and made cytological observations for a diverse panel of *C. echinospermum* accessions and C. arietinum × *C. echinospermum* F 1 hybrids (10 parental combinations) generated in this study. Abnormal microspore development and aberrant chromosome pairing in *C. echinospermum* accessions and interspecific hybrids indicate there are multiple factors that impede complete homologous pairing. There is evidence of large genome structure variants (i.e., reciprocal translocations, inversions) and a recalcitrant genetic factor with non-Mendelian inheritance within *C. echinospermum* that impede viable gamete formation. The nature and persistence of such post-zygotic barriers have significant implications for the species coherence of *C. echinospermum*, its viability in nature, and its utility as a genetic resource for crop improvement.

Caught in the middle: the role of the parenchymatous sheath in C4 *Neurachne* species.

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The carbon concentrating mechanism (CCM) of C4 plants reduces photorespiration, providing them with an advantage over their C3 counterparts in hot, dry climates. A key feature of the C4 CCM is the physical separation of photosynthetic carbon assimilation (PCA) and photosynthetic carbon reduction (PCR) reactions, typically occurring in mesophyll and bundle-sheath cells, respectively. This specialized leaf anatomy, known as "Kranz anatomy", involves mesophyll cells arranged around enlarged bundle-sheath cells that surround the vascular tissue. *Neurachne* is an Australian native grass genus which contains distinct, closely related C3, C3 -C4 intermediate, and C4 species. Although the two C4 species of this genus, *N. muelleri* and *N. munroi* show independent evolutionary origins, they both display a form of Kranz anatomy known as Neurachneoid-type Kranz anatomy. In this anatomy, two sheath layers surround vascular bundles: an inner mestome sheath, and an outer parenchymatous sheath. This outer parenchymatous sheath, which contains very few organelles, separates mesophyll cells – that house PCA – from mestome sheath cells in which PCR occurs. While the spatial separation of PCA and PCR is crucial for the C4 CCM, the movement of metabolites between the cells in which these reactions occur is essential. This raises questions about the role of the parenchymatous sheath in C4 *Neurachne* species. To investigate this, the localisation and distribution of key C4 -associated enzymes in C3, C3 -C4 intermediate, and C4 *Neurachne* species was examined using immunolocalisation techniques at both light and electron microscopy levels. The results of these experiments have provided insights into how the C4 pathway functions across the three cell types and the evolution of a C4 syndrome in *Neurachne.*

Using MIFE technique to determine cellular ion flux: a case study on leaf salinity tissue tolerance in wild rice *Oryza coarctata*

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Known as a key trait that confers the adaptive potential of halophyte species, tissue tolerance represents a valuable resource in improving overall salinity stress tolerance in traditional staple crops. The wild rice *Oryza coarctata* is the only halophyte in *Oryza* species with outstanding salinity tolerance, while cultivated rice (*Oryza sativa*) is much more vulnerable. Considering this, this study employed the MIFE technique to determine ion fluxes from mesophyll cells of *O. coarctata* and dissect its contribution to leaf salinity tissue tolerance in this wild rice. Salinity induced ca 80% decline in the mesophyll cell viability in cultivated rice, whereas only 15% reduction was observed in the wild rice. In response to NaCl treatments, mesophyll cells of *O. coarctata* possessed less Na^+ uptake and better K^+ retention than cultivated rice. This difference in plant ionic relations was partially attributed to the difference in transcriptional changes in AKT1, SOS1, and HKT1 transporters. It is concluded that better K^+ retention and Na^+ exclusion confer mesophyll salt tolerance in *O. coarctata*. The above traits should be considered as potential targets in the rice breeding program for salt tolerance enhancement.

Investigation of changes in seed yield and quality during maturation of subterranean clover to evaluate a novel seed harvesting method

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Subterranean clover (*Trifolium subterraneum* L.*; subclover*) is a winter-active annual pasture legume originating in the Mediterranean Basin and parts of Western Asia and Europe which remains the most widely sown annual pasture legume in southern Australia. Subclover's popularity is attributed largely to its unique trait of seed burial which promotes high levels of persistence but makes seed harvesting inherently difficult. Current harvesting methods involve tillage preparation followed by a suction-based harvester: while effective this process is inefficient, causes soil disturbance and erosion, and relies on ageing, outdated harvesters. Swathing offers an alternative harvesting solution for subclover in which plant roots are cut, plants lifted and windrowed, then dried plants harvested with a combine. Trials using technology adapted from peanut production have demonstrated that swathing subclover seeds shows promise, however performance is highly dependent on plant senescence stage. Swathing must occur while the plants are still green with sap in stems and peduncles to ensure seed-containing burrs remain attached to plants and are lifted during swathing, yet seed development must also be sufficiently advanced to ensure adequate quality and yield. Further trials have shown that swathing can achieve higher seed retention with sufficient seed quality compared to conventional subclover seed harvesting methods, but that both seed retention and quality change rapidly with time. Identification of the optimal swathing window is hence crucial for the success of swathing subclover. In my PhD I am investigating changes in burr retention, seed quality and yield as seed development advances and plants senesce, to develop a method that farmers can apply to identify the ideal swathing window.

Accumulation of manganese in leaves of carboxylate-releasing Hakea species (Proteaceae) depends on cluster-root physiology

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In extremely phosphorus (P)-impoverished environments, adaptations have evolved in plant species to acquire P efficiently. Most Proteaceae, a prominent family in the Southwest Australian Biodiversity Hotspot, produce cluster roots, which are ephemeral non-mycorrhizal specialised root structures comprising hundreds to thousands of hairy rootlets. Cluster roots release carboxylates and protons that mobilise poorly available P sorbed onto soil particles. In addition to P, carboxylates and protons mobilise a range of micronutrients, including manganese (Mn). Assessing carboxylate exudation in roots is laborious and often requires growing plants in controlled environments with easy access to the root system. However, Mn uptake in roots is poorly regulated, and Mn accumulates in mature leaves after being mobilised by root-exuded carboxylates and protons. As a result, Lambers et al. (2015, 2021) conceptualised leaf Mn concentration as a proxy for rhizosphere carboxylate concentrations. However, some Hakea species (Proteaceae) that produce cluster roots and release carboxylates do not accumulate Mn in their leaves, challenging this conceptual model. Using *Hakea prostrata* (Mn accumulating) and *H. flabellifolia* (non-Mn accumulating), we explored the physiology of carboxylate release in non-cluster and cluster roots and the determinants of Mn accumulation in leaves. Our results show that Mn accumulation in leaves does not depend on carboxylate release, but on protons concomitantly released to drive the release of carboxylates. Species using the proton gradient to release other cations, such as $^{K+}$ or Mg^{2+}, do not acidify the rhizosphere and therefore do not increase the rhizosphere Mn availability and do not accumulate Mn. We highlight the limitations of leaf Mn concentration as a proxy for rhizosphere carboxylate concentration and propose a revised model. We further discuss how different physiologies of carboxylate exudation contribute to the ecological success of carboxylate-releasing species, regarding modulating rhizosphere pH to enhance nutrient uptake and alleviate element toxicity.

The LYR Domain of the Complex I Subunit B22 is Crucial for an Interaction with the Mitochondrial Acyl Carrier Protein ACP2

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Mitochondrial Complex I (CI), a large multi-subunit respiratory complex contains two LYR (leucine/tyrosine/arginine) domain-containing subunits, B14 (NDUA6/LYRM6) and B22 (NDUB9/LYRM3). Mitochondrial LYR (LYRM) proteins are soluble matrix-located proteins that have been implicated in diverse functions such as iron-sulphur cluster insertion, OXPHOS complex assembly and mitoribosome biogenesis. B14 and B22 are unique to other LYRM proteins in that they are integral components of CI. To explore the function of B22, we examined T-DNA insertional knockdown lines, which displayed a mild growth defect linked to reduced CI activity and abundance. Notably, this defect could not be rescued by complementation with a B22 variant that contained a mutated LYR domain, indicating the domain's critical role in B22's function. Protein interaction assays further revealed that the LYR domain is crucial for B22's interaction with neighbouring CI subunit, mitochondrial acyl carrier protein ACP2. Similarly, T-DNA insertional knockdown lines of SDAP1 showed a comparable CI defect, suggesting that the interaction between B22 and ACP2, mediated by the LYR domain, is essential for the function and assembly of CI.

Cyclic mononucleotide-dependent ion fluxes in Arabidopsis thaliana - a tale of two isomers

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Cyclic mononucleotides, including 3',5'-cyclic adenosine monophosphate (3',5'-cAMP) and its positional isomer 2',3'-cyclic adenosine monophosphate (2',3'-cAMP) are increasingly recognised as key signalling molecules in *Arabidopsis thaliana*. 3',5'-cAMP is synthesized from ATP by adenylate cyclases (ACs) and metabolised by cyclic mononucleotide specific phosphodiesterases (PDEs). In contrast, 2',3'-cAMP is an RNA degradation product, accumulates under stress and may also have a role in stress signalling, although its specific mechanisms of action remain somewhat unclear. A recent proteomic pilot study has suggested that, despite a small overlap in commonly regulated proteins, the two isomers affect distinct sets of proteins. This is consistent with distinct roles for each isomer in cellular processes. Here we have set out to further delineate the specific actions of these two isomers and their effects on ion fluxes. As a target, the *Arabidopsis thaliana* CNGC2 channel (AtCNGC2) was selected because of its annotated role in abiotic stress response. Experiments using Xenopus laevis oocytes expressing AtCNGC2 revealed that while 3',5'-cAMP induced K^+ currents, 2',3'-cAMP failed to elicit any currents, indicating no specific interaction with the channel. This supports the hypothesis that AC generated 3',5'-cAMP but not the RNA degradation product 2',3'-cAMP participates in in cAMP-dependent modulation of ion homeostasis. Our research further explores the effects of the cAMP isomers on ion fluxes under stress conditions in *A. thaliana*. Transgenic lines overexpressing the "cAMP sponge" (cAS) that buffers 3',5'-cAMP at low cytosolic concentrations will be used to compare stress-induced signatures of 3',5'-cAMP and 2',3'-cAMP. The aim is to establish if 2',3'-cAMP can restore ion flux signatures in 3',5'-cAMP depleted cells. Exogenous applications of both isomers will shed light on the roles of these messengers in plant stress responses and adaptation.

Short Talk Abstract

*Note: Abstracts SP1-SP5 will be presented in 3-minute session, followed by poster presentation at morning tea

ASPS24 WA – SP1

Atrazine application on triazine-resistant canola suppresses white leaf spot (*Neopseudocercosporella capsellae*) development

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White leaf spot, caused by *Neopseudocercosporella capsellae* is an economically important disease of many Brassicaceae genera and species in Australia, Europe, and north America. It is concerning that its pathogenicity is increasing in Australia and in the United Kingdom. In Western Australia triazine-tolerant canola varieties have at times constituted up to 80% of the area sown. Atrazine is routinely applied within the first two months of crop development, often corresponding with the period of most severe white leaf spot disease. As atrazine application is known to affect white leaf spot incidence and severity, studies are being undertaken to define exactly how atrazine application at various timings before and after *N. capsellae* infection affects white leaf spot development on canola varieties of different resistance to the disease. We found that suppression of WLS by atrazine application varied with cultivar susceptibility and with application timing, and that there was a strong interaction of cultivar x application timing. That suppression of white leaf spot from application of atrazine occurred both before and after *N. capsellae* infection is perhaps surprising as herbicide application can stress and weaken plants, more generally encouraging rapid fungal colonisation. The outcome of these studies is important as they will highlight unique opportunities for farmers to exploit better cultivar choices in conjunction with manipulating the timing of atrazine application to maximize white leaf spot suppression and consequent canola yield.

Unravelling heat tolerance in bread wheat (*Triticum aestivum L.*) using physiology and proteomics approaches.

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Heat stress significantly impacts global bread wheat productivity. Our study aimed to identify underlying mechanisms of heat tolerance in wheat flag leaves and spike tissues. We compared physiology, yield, and protein abundance changes of wheat genotypes with contrasting heat tolerance (two tolerant [RAJ3765 and HD2932] and two susceptible [HD2329 and HD2733]) under short- and long-term heat stress (32°C) at ear peep. This experiment revealed that heat tolerant genotypes maintained grain yield under short-term heat exposure by maintaining photosynthesis, membrane stability, chlorophyll content, pollen viability, and redox homeostasis. Heat stress during ear peep reduced grain number, above-ground biomass, harvest index less in heat-tolerant than -susceptible genotypes while increased thousand grain weight and grain protein content, with significant genotype x treatment interactions. Notably, long-term heat stress reduced thousand grain weight more in heat-susceptible than -tolerant genotypes. We identified 31 and 60 changes in protein abundances in flag leaves and spike tissues, respectively. Key pathways in flag leaves included photosynthesis, RNA processing, heat shock proteins, redox homeostasis, carbohydrate metabolism, chromatin organisation, and protein breakdown, translation, and translocation. In spikes, prominent pathways included carbohydrate, lipid, and secondary metabolism, cell wall and chromatin organisation, redox homeostasis, membrane transport, methylation, protein folding, breakdown and translocation, RNA processing, lipid transfer, cell morphogenesis, heat shock proteins, and reproduction. Co-expression analysis revealed proteins correlated with important agronomic traits. These proteins provided insights into mechanisms of heat tolerance associated wheat physiology and yield.

Negative regulation of KAI2 signalling in angiosperms

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Butenolide signalling is a conserved biological mechanism across multiple domains and kingdoms of life, enabling organisms to respond to specific chemical stimuli. In plants, this signalling is an integral part of both hormone-driven development and smoke-stimulated germination and is increasingly recognised for its significance in mediating agricultural outcomes, such as drought tolerance and grain yields. Butenolide phytohormone signalling and smoke response shares a common feature: the requirement of an α/β-hydrolase receptor-enzyme to perceive the butenolide molecules. This is accomplished by DWARF14 (D14), which perceives strigolactone hormones, and KARRIKIN INSENSITIVE 2 (KAI2), an ancestral homologue of D14 that is essential for the recognition of karrikin molecules found in smoke. Additionally, there is strong evidence that KAI2 perceives a distinct, yet undiscovered class of butenolide hormones, tentatively named 'KL'. Flowering plants harbour a third homologue of KAI2 and D14 known as DWARF14-LIKE 2 (DLK2), which remains poorly understood despite its strong conservation within angiosperms. Intriguingly, DLK2transcription is strongly upregulated by KAI2 activity. This project evaluates the role of DLK2 in the light of KAI2 homeostasis through protein assays and in planta genetic experiments. The findings point towards a mechanism in which DLK2 has lost receptor-like signalling function, but retained hydrolase activity, and catalyses the degradation of KL as KAI2 activity increases.

Examining changes in the spatial expression of key photorespiratory enzymes during the evolution of C4 photosynthesis in the genus Flaveria

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Carbon concentrating mechanisms (CCM), such as C2 and C4 photosynthesis, reduce the deleterious effects of photorespiration and improve the photosynthetic efficiency of plants using these pathways in arid environments. The ancestral C3 pathway is hypothesised to have given rise to the C2 CCM which, in turn, preconditioned the C4 CCM. The genus *Flaveria* is an exceptional model to study photosynthetic evolution, as it comprises distinct, closely related species that employ either C3, C2, or C4 photosynthesis. Investigations into this genus have shown that the evolution of CCMs is dependent upon changes to leaf anatomy, and the abundance and spatial patterning of enzymes. The C2 CCM relies on the recapture of photorespired CO2 released during the metabolism of glycine-by-glycine decarboxylase (GDC). This CCM is established via the compartmentation of GDC in bundle sheath cells (BSCs). However, GDC activity also liberates NH3, potentially creating a nitrogen imbalance between leaf mesophyll cells (MCs) and BSCs. In parallel with the movement of glycine into BSCs, movement of earlier photorespiratory metabolites such as glycolate and/or glyoxylate may mitigate nitrogen imbalances and precondition the evolution of an incipient C4 cycle. Examining the cell-specific expression pattern of glycolate oxidase (GOX), an enzyme involved in glycolate/glyoxylate metabolism, may provide insights into the evolution of nitrogen balancing mechanisms in C2 species. In this study, immunolocalisation of GOX and GDC in the leaves of *Flaveria* species that use either C3, C2, or C4 pathways was determined using confocal and electron microscopy. The BSC-specific localisation of GDC was apparent in C2 and C4 species. By contrast, GOX labelling was detected in both MCs and BSCs in C2 and C4 *Flaveria* species, suggesting that glycine, along with glycolate and/or glyoxylate, may move into the BSC. These findings provide further understanding of the molecular involved in the evolution of C4 photosynthesis.

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Investigating the functionality of LYR domain proteins in Arabidopsis thaliana

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Mitochondrial energy generation in plant systems plays a unique functional role when compared to that of other eukaryotic systems. While the mitochondrion and the respiration that occurs within it is responsible for most of the energy generation in common eukaryotic systems, plants utilise the mitochondrion in tandem with the chloroplast, after it has matured as an organelle. The functionality of the mitochondrion in plant systems is contingent on the cooperative activity of several multi-subunit oxidative phosphorylation (OXPHOS) protein complexes. Many subunits comprising these complexes must be transported from the cytosol or the mitochondrial matrix to the mitochondrion itself before their assembly into a larger complex. Additionally, other subunits must undergo maturation through the coordinated chaperoning and insertion of cofactors before electron transport, and thus energy generation, may take place. Proteins known as assembly factors are responsible for these transport and maturation processes, and these have been shown to exhibit intricate interactivity with many mitochondrial proteins. Previous work conducted in my Honours research project examined the functionality of a plant-specific assembly factor of the OXPHOS complex succinate dehydrogenase (SDH) that was suspected to fulfill a role analogous to mammalian SDH assembly factor 3 (SDHAF3), which facilitates iron-sulphur (Fe-S) cluster insertion into the subunit SDH2. This PhD project will aim to further examine both the functionality and genetic characteristics of this assembly factor in Arabidopsis thaliana , while expanding the scope of the Honours project by encompassing several other potential assembly factor proteins belonging to the same family as SDHAF3, known as the leucine, tyrosine, arginine (LYR) domain-containing proteins.

ASPS24 WA – SP6

Why does thiazole synthase turn over very rapidly and can this be changed by directed mutation to alter its catalytic properties and thiamin content in plants?

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Reducing enzyme turnover rates can potentially decrease cellular energy demands, benefiting plant efficiency and productivity. A notable example is thiazole synthase (THI4), a high-turnover enzyme central to thiamin (vitamin B1) biosynthesis, an essential cofactor for various enzymes in plants and humans. THI4, a 'suicide enzyme,' irreversibly inactivates itself as it donates a sulfur atom from a cysteine residue to form thiamin's thiazole ring, leaving a dehydroalanine (DHAla) residue in its active site. This self-inactivation leads to rapid THI4 turnover, requiring continuous resynthesis in plants, an energy-intensive process that impact crop yield. The degradation rates of functional versus inactivated THI4, and how these are influenced by environmental conditions, remain unexplored. Understanding THI4's stability and identifying strategies to reduce its turnover rate could be key to enhancing crop efficiency and reducing the energy costs associated with thiamin regeneration. I will present our findings comparing the developmental thiamine requirements of wild-type (WT), THI4 complementation line, and THI4 knockout (tz<-1) Arabidopsis plants. Additionally, I will showcase our current efforts to engineer THI4 variants replacing the cysteine residue in Arabidopsis, alongside our methodology to quantify thiamine content across various thiamine mutants. This approach aims to investigate the role of thiamine in plant development in greater detail and assess whether the turnover rate of THI4 can be altered by replacing this suicidal cysteine residue.

How does ENOD93 drive Mitochondrial ATP production to enhance Nitrogen Use Efficiency in plants?

Selin Altintas[1], Harvey Millar[1]

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Early Nodulin 93 (ENOD93) was first identified within a group of genes, known as ENODs, that are expressed during early nodule development. ENOD93 is the only ENOD protein predicted to be localized in plant mitochondria. In Arabidopsis, ENOD93 appears as a small 12kDa protein in the mitochondrial proteome. ENOD93 homologs have been found in other plants, generally consisting of 90-120 amino acids. In soybean, silencing ENOD93 reduces nodule formation and biological nitrogen fixation, while overexpressing OsENOD93-1 in rice enhances nitrogen use efficiency. Moreover, oil palm ENOD93 is essential for somatic embryogenesis, and overexpressing EgENOD93 in *Arabidopsis thaliana* enhances shoot regeneration. Loss of AtENOD93 function in Arabidopsis leads to reduced root growth, a decline in adenylate levels, and disrupted mitochondrial metabolism. Recent studies from the Millar lab show that ENOD93 regulates COX subunits in plants and resembles the N-terminus of yeast Respiratory Supercomplex Factor 2 (RCF2), which contains both an ENOD93-like N-terminus and a C-terminal Hypoxia-Inducible Domain (HIGD). These regions may be proteolytically cleaved into separate fragments under varying growth conditions and energy sources. It has been reported HIGD proteins are present in plants and animals. Both RCFs and HIGD proteins are involved in COX biogenesis, either as independent units or within respiratory supercomplexes, playing critical roles in regulating COX activity and cellular respiration, particularly in response to hypoxia and oxidative stress. In this project evolutionary relationship between plant ENOD93 and HIGD proteins and yeast RCF2, as well as the interoperability of ENOD93 and HIGD proteins in plant mitochondria, are being explore. Also, we will be characterizing the potential functions of ENOD93 gene families in different crops, to determine how the high demand for mitochondrial ATP during nitrogen-linked processes can enhance plant tolerance to hypoxia and improve nutrient utilization

Genetic and evolutionary insights into the KAI2-dependent Pectate Lyase-Like 21 (PLL21) in *Arabidopsis thaliana*

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Early Nodulin 93 (ENOD93) was first identified within a group of genes, known as ENODs, that are expressed during early nodule development. ENOD93 is the only ENOD protein predicted to be localized in plant mitochondria. In Arabidopsis, ENOD93 appears as a small 12kDa protein in the mitochondrial proteome. ENOD93 homologs have been found in other plants, generally consisting of 90-120 amino acids. In soybean, silencing ENOD93 reduces nodule formation and biological nitrogen fixation, while overexpressing OsENOD93-1 in rice enhances nitrogen use efficiency. Moreover, oil palm ENOD93 is essential for somatic embryogenesis, and overexpressing EgENOD93 in *Arabidopsis thaliana* enhances shoot regeneration. Loss of AtENOD93 function in Arabidopsis leads to reduced root growth, a decline in adenylate levels, and disrupted mitochondrial metabolism. Recent studies from the Millar lab show that ENOD93 regulates COX subunits in plants and resembles the N-terminus of yeast Respiratory Supercomplex Factor 2 (RCF2), which contains both an ENOD93-like N-terminus and a C-terminal Hypoxia-Inducible Domain (HIGD). These regions may be proteolytically cleaved into separate fragments under varying growth conditions and energy sources. It has been reported HIGD proteins are present in plants and animals. Both RCFs and HIGD proteins are involved in COX biogenesis, either as independent units or within respiratory supercomplexes, playing critical roles in regulating COX activity and cellular respiration, particularly in response to hypoxia and oxidative stress. In this project evolutionary relationship between plant ENOD93 and HIGD proteins and yeast RCF2, as well as the interoperability of ENOD93 and HIGD proteins in plant mitochondria, are being explore. Also, we will be characterizing the potential functions of ENOD93 gene families in different crops, to determine how the high demand for mitochondrial ATP during nitrogen-linked processes can enhance plant tolerance to hypoxia and improve nutrient utilization.

Elucidating the role of butenolides in plant physiology and plant-fungal symbiosis

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When deficient in mineral nutrients, plant roots exude strigolactones (SLs) into the rhizosphere to encourage the symbiosis with arbuscular mycorrhizal (AM) fungi. Chemically classified as a butenolides with a four-carbon heterocyclic lactone moiety, SLs also serve as plant hormones to regulate shoot branching by inhibiting axillary bud outgrowth. Two related α/β-hydrolases encoded in plant genomes are implicated in butenolide perception: DWARF14 (D14) is the SL receptor, whereas KARRIKIN INSENSITIVE2 (KAI2) is required for response to butenolide compounds from smoke and is a likely receptor for a hypothetical butenolide ligand termed KL. The ligand(s) and function of a third α/β-hydrolase found in seed plants, D14-LIKE2 (DLK2), are unknown. KAI2 positively regulates the expression of DLK2 and is necessary for arbuscular mycorrhizal (AM) symbiosis (Gutjahr et al., 20151; Meng et al., 2015). Meanwhile, DLK2 negatively regulates AMF arbuscule branching and the expression of AM-responsive genes in tomato (Ho-Plágaro et al., 2021). Thus, we hypothesise that KAI2 is a positive regulator of AM symbiosis, whereas DLK2 limits AMF colonisation by regulating the availability of KAI2 substrates such as KL. Due to the genetic tractability and agricultural importance of barley (Hordeum vulgare), this project aims to to isolate loss-of-function mutations in DLK2 and other genes in barley via CRISPR/Cas9 mutagenesis, and to characterise their impacts on AM symbiosis and plant morphology. we also characterised the enzymatic properties and ligand preferences of two DLK2 homologues from barley. Our study will increase our knowledge about AM symbiosis, which may provide new approaches to reducing fertiliser use in agriculture.

Root response to environmental stress under changing climates

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Root system and root-soil interface has attracted interest among scientists to address future food security under climate change. They are multi-faceted, incorporating key variables that are individually & interactively affected by climatic factors (rainfall, radiation, temperature, eCO2, pollution). The world is becoming hotter, drier and more vulnerable. Such changes in climate exacerbates soil-related stresses for agriculture. Understanding root system and root-soil interactions is critical for identifying root traits for breeding cultivars with improved resource uptake and adaptation to stressful environments. Crop root systems are often poorly adapted to soils with the major limiting factors under changing climates. Root traits that overcome abiotic constraints are vital to maintaining structural and functional properties, and important target in breeding programs for rainfed agriculture under changing climates. This talk will present some examples on how root-soil interface responds to climate-related stresses, such as drought, salinity, Al toxicity in acid soil, and soil compaction.

Cyclic mononucleotide-dependent ion fluxes in Arabidopsis thaliana - a tale of two isomers

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Cyclic mononucleotides, including 3',5'-cyclic adenosine monophosphate (3',5'-cAMP) and its positional isomer 2',3'-cyclic adenosine monophosphate (2',3'-cAMP) are increasingly recognised as key signalling molecules in *Arabidopsis thaliana*. 3',5'-cAMP is synthesized from ATP by adenylate cyclases (ACs) and metabolised by cyclic mononucleotide specific phosphodiesterases (PDEs). In contrast, 2',3'-cAMP is an RNA degradation product, accumulates under stress and may also have a role in stress signalling, although its specific mechanisms of action remain somewhat unclear. A recent proteomic pilot study has suggested that, despite a small overlap in commonly regulated proteins, the two isomers affect distinct sets of proteins. This is consistent with distinct roles for each isomer in cellular processes. Here we have set out to further delineate the specific actions of these two isomers and their effects on ion fluxes. As a target, the *Arabidopsis thaliana* CNGC2 channel (AtCNGC2) was selected because of its annotated role in abiotic stress response. Experiments using Xenopus laevis oocytes expressing AtCNGC2 revealed that while 3',5'-cAMP induced K^+ currents, 2',3'-cAMP failed to elicit any currents, indicating no specific interaction with the channel. This supports the hypothesis that AC generated 3',5'-cAMP but not the RNA degradation product 2',3'-cAMP participates in in cAMP-dependent modulation of ion homeostasis. Our research further explores the effects of the cAMP isomers on ion fluxes under stress conditions in *A. thaliana*. Transgenic lines overexpressing the "cAMP sponge" (cAS) that buffers 3',5'-cAMP at low cytosolic concentrations will be used to compare stress-induced signatures of 3',5'-cAMP and 2',3'-cAMP. The aim is to establish if 2',3'-cAMP can restore ion flux signatures in 3',5'-cAMP depleted cells. Exogenous applications of both isomers will shed light on the roles of these messengers in plant stress responses and adaptation.

The Impact of Nitrogen Management on Wheat Protein Composition in Western Australians Soils

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Understanding the impact of nitrogen on wheat protein composition is crucial for enhancing the agronomic and commercial value of wheat. My research utilises a proteomic approach to analyse how different nitrogen management strategies effect the grain protein composition of wheat grown in the arid Western Australian soils. The significance of my research stems from the need to optimise nitrogen utilisation while improving wheat grain quality without compromising yield. To examine the influence of nitrogen on wheat protein composition, I performed a range of field and glasshouse trials and analysed plant development, yield and protein content in relation to the amount and timing of nitrogen applications. I identified specific changes in protein composition resulting from the different nitrogen application strategies. The findings of my research confirm that nitrogen application in terms of the amount and timing of nitrogen can significantly influence wheat grain protein composition, which could lead to downstream grain quality changes. The contribution of my research provides valuable insights into the proteomic alterations associated with nitrogen management and suggests that protein composition can be altered through targeted nitrogen application strategies. Additionally, these findings will help develop protein targets for bioassays of harvested grain to guide future nitrogen management practices. My research highlights the critical role of nitrogen management in shaping wheat protein composition and establishes a scientific foundation for advancing more sustainable and productive agricultural practices with potentially lower nitrogen inputs. I will present the agronomic and proteomic outcomes of our trials on nitrogen application strategies, highlighting their implications for future wheat production.

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