

CSPB • SCBV 2025

Annual General Meeting

August 9th-12th, 2025
Dalhousie University, Halifax Nova Scotia



SCIENTIFIC PROGRAM



PRESIDENT'S MESSAGE

It is my great pleasure to welcome you all to the Canadian Society of Plant Biologists • la Société Canadienne de Biologie Végétale ([CSPB • SCBV](#)) 2025 Annual General Meeting. The organizers have curated an exceptional event that truly embodies our society's commitment to making the society diverse, inclusive and welcoming. This year's scientific program is designed to be highly impactful, featuring plenary sessions that delve into cutting-edge topics. These include *plant resilience - interactions with individual, and combinations of abiotic and/or biotic stresses; coastal eco-physiology and the changing climate; and artificial intelligence in advancing plant biology and sustainable agriculture*. In a first for CSPB-SCBV, and demonstrating our continued commitment to teaching and learning, we are proud to launch a dedicated plenary session on "Education - Teaching and Learning about Plants." This session, organized in collaboration with the CSPB-SCBV Education Committee, highlights our dedication to fostering Plant Biology knowledge at all levels. We remain steadfast in our commitment to training the next generation of plant scientists. This conference will proudly showcase the remarkable contributions of our early career scientists, graduate, and undergraduate students presented as oral talks in over 17 concurrent sessions and as poster displays in the exhibit hall. I extend my sincerest gratitude to the organizing committee for their tireless efforts in creating such a high-quality and enriching event. I eagerly anticipate meeting each of you in person.



Marcus Samuel

University of Calgary

Land Acknowledgement

Dalhousie University operates in the unceded territories of the Mi'kmaw, Wolastoqey, and Peskotomuhkati Peoples. These sovereign nations hold inherent rights as the original peoples of these lands, and we each carry collective obligations under the Peace and Friendship Treaties. Section 35 of the Constitution Act, 1982 recognizes and affirms Aboriginal and Treaty rights in Canada.

African Nova Scotian Acknowledgement

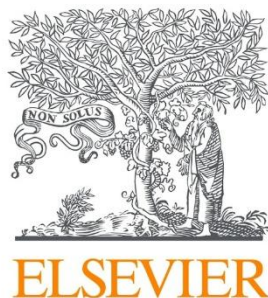
We recognize that African Nova Scotians are a distinct people whose histories, legacies and contributions have enriched that part of Mi'kma'ki known as Nova Scotia for over 400 years.

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Canadian Science Publishing Open Access Waiver Giveaway

Canadian Science Publishing and the journal *Botany* are proud to partner with the Canadian Society of Plant Biologists • la Société Canadienne de Biologie Végétale.

To be entered in the draw to win an open access waiver, which provides an **100% discount to publish a manuscript open access in the journal *Botany***, sign up for the Canadian Science Publishing newsletter below.

This giveaway is open to anyone attending the CSPB 2025 Annual General Meeting. The giveaway will close on Thursday August 14th, 2025, at midnight. [Please click this link](#) or scan the QR code to enter. If you have any questions, please contact Leanna Kalinowski at leanna.kalinowski@cdnsiencepub.com. *Good luck!*

SCAN ME





Flow Flow Genomics presents Canada's first historic award for SynBio: The Summer of Synthetic Biology (SoS) Prize

Presenting a poster at the CSPB-SCBV 2025 Annual General Meeting?

Submit it to us to be considered for an SoS prize! Each award is \$500 in cash and \$500 in sequencing credits with Flow Genomics. For the summer months of June, July, and August, we will be granting one SoS prize to recognize awesome scientific posters and overall achievements of students at Canadian universities.

For more info on eligibility, deadlines and submission portal, go to our information page at <https://www.synbiocanada.org/summer-of-synbio>



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Conference Giveaway: Win 2 BioCoupler[™] Glass Sets (3 winners will be selected). Scan the QR code for information on entering the giveaway.

Attendee Discount: 15% off your first order with code CSPB2025-0809 (valid on any order size, Aug 9 – Oct 31, 2025).

Scan QR code for more giveaway & discount information



ACKNOWLEDGEMENTS

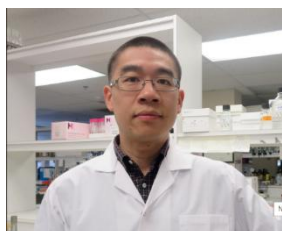
Local Organizing Committee



Sophia Stone, Co-chair
Department of Biology
Dalhousie University



David Chiasson, Co-chair
Department of Biology
St. Mary's University



Zhenyu Cheng
Department of Microbiology and Immunology
Dalhousie University



Ellie Goud
Department of Biology
St. Mary's University



Balakrishnan Prithiviraj
Department of Plant, Food, and Environmental Sciences
Dalhousie University

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Robin Cameron, Past President
Hugo Zheng, Vice President
David Bird, Secretary
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Gopal Subramaniam, Science Policy Director

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Paige Burns *Dalhousie University*
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Summer Carroll *St. Mary's University*
Elle Crilly *Dalhousie University*
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Grace Tadema *St. Mary's University*
Raja Vairavan *Dalhousie University*
Kapeeran Venkatesan *Dalhousie University*

A sincere thank you to all the volunteer judges! The time and effort you put into assessing the oral and poster presentations are greatly appreciated.

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Biology Education, Made in Canada:

A Community Workshop on Growing Our Own Teaching Resources

Join us to share ideas and resources for incorporating more Canadian content into biology teaching!

Come build your toolkit, connect with others, and help grow something rooted in the places we teach and live.

September 2nd, 2025

10:00 AM Pacific / 2:00 PM Atlantic



Scan for Registration





Dr. Miranda Meents
Lecturer
Simon Fraser University
Education Director



Dr. Solmaz Irani
Assistant Teaching Professor
Thompson Rivers University
Communications Committee



GENERAL INFORMATION

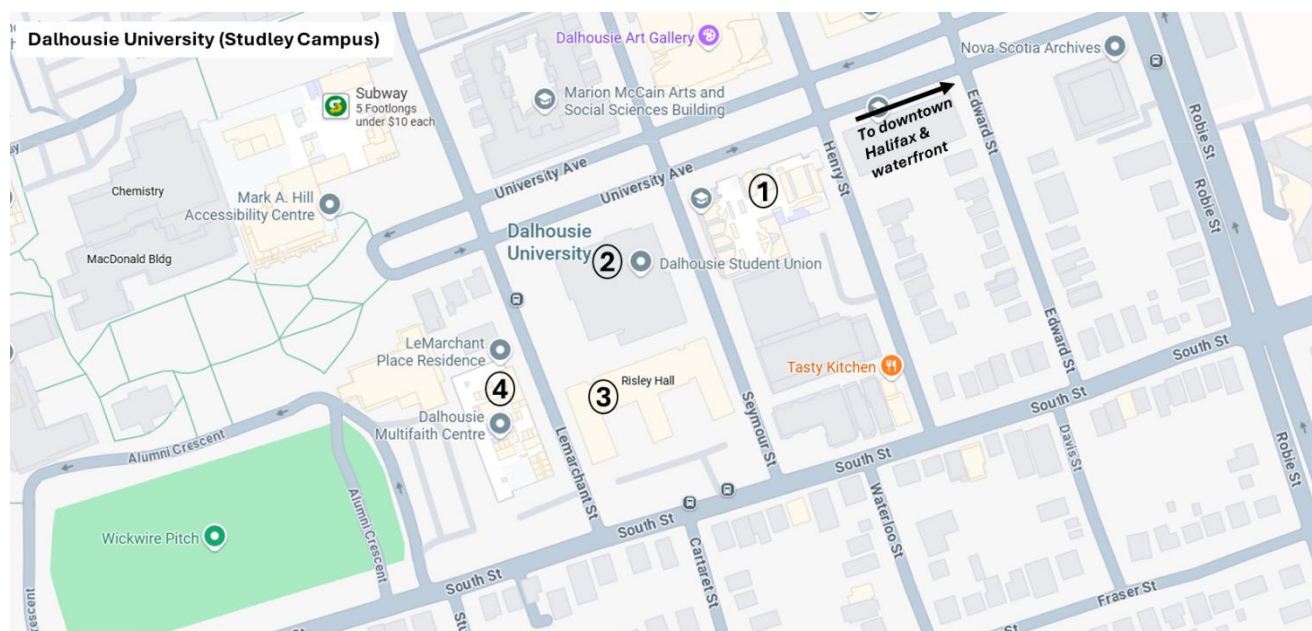
Meeting Venues

The Canadian Society of Plant Biologists • la Société Canadienne de la Biologie Végétale (CSPB • SCBV) 2025 Annual General Meeting will be held at Dalhousie University (Studley Campus; see map below) on August 9th to 12th, 2025, in Halifax, Nova Scotia, Canada.

Seminars (oral presentations), annual business meeting (ABM) and coffee breaks will take place in the **Kenneth C. Rowe Management Building (ROWE bldg.** [see 1 on map]; 6100 University Ave, Halifax, NS B3H 4R2). All plenary and award presentations will be given in the Potter Auditorium, room 1028 of the ROWE bldg. Concurrent Sessions will take place in rooms 1007/9/11/14/16 on the first floor of the ROWE bldg.

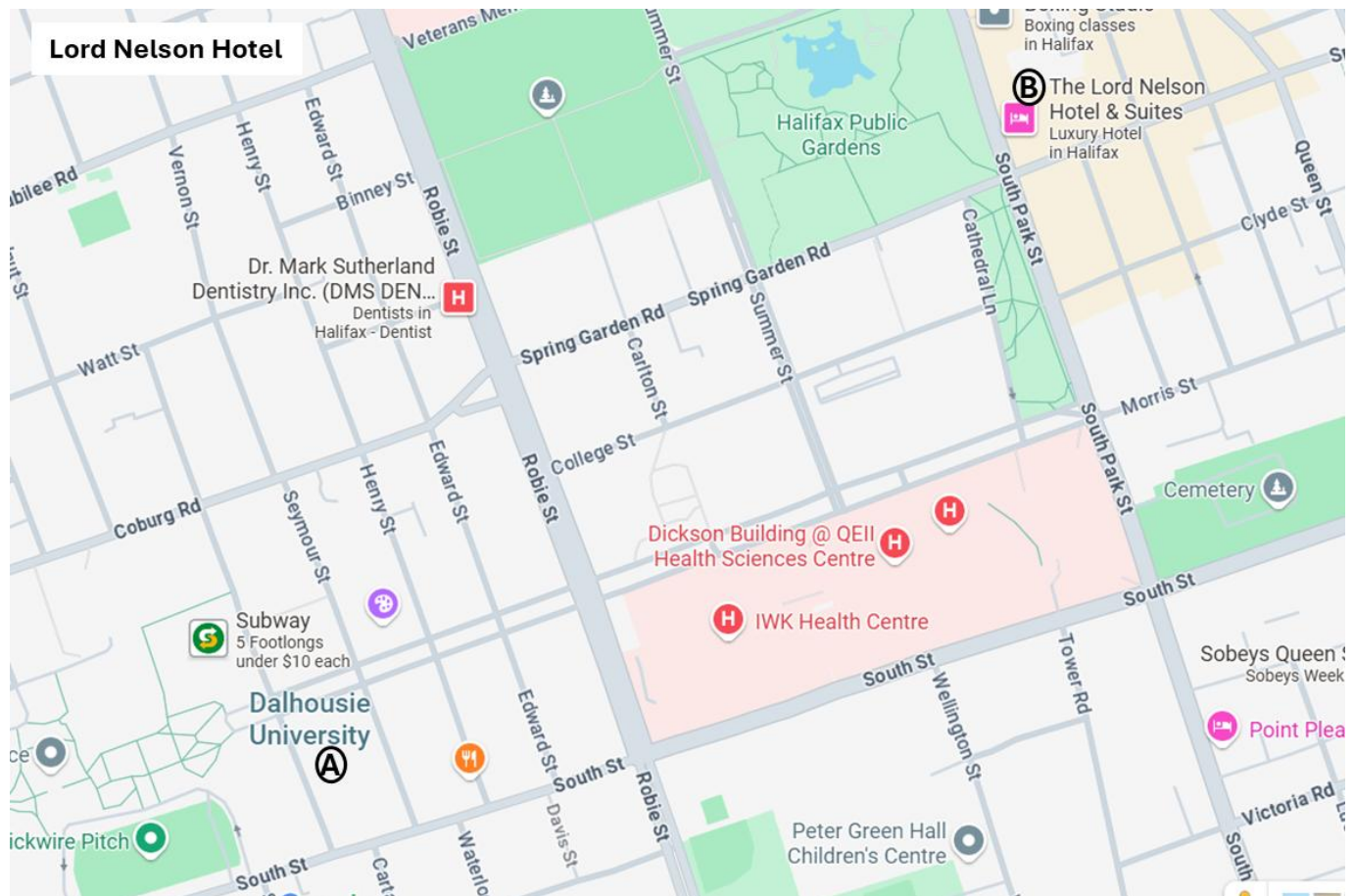
Poster Sessions, lunch, Welcome Reception and Social/Mixer will be held in the **Student Union Building (SUB** [see 2 on map]; 6136 University Ave, Halifax, NS B3H 4R2). For the nature walk, the bus will pick-up participants at the main entrance of the SUB on University Ave.

On campus accommodations are available in Risley Hall (see 3 on map; 1233 LeMarchant Street, Halifax, NS B3H 4) and LeMarchant Place (see 4 on map; 1246 LeMarchant Street, Halifax, NS B3H 4R2).



The CSPB Dinner will take place at the Lord Nelson (see B on map below; 1515 South Park St, Halifax, NS B3J 1G9). The dinner will be held in the Georgian Lounge, Regency and Imperial Ballrooms of the h

otel. Dalhousie University (see A on map below) is a 15-minute walk from the Lord Nelson Hotel.



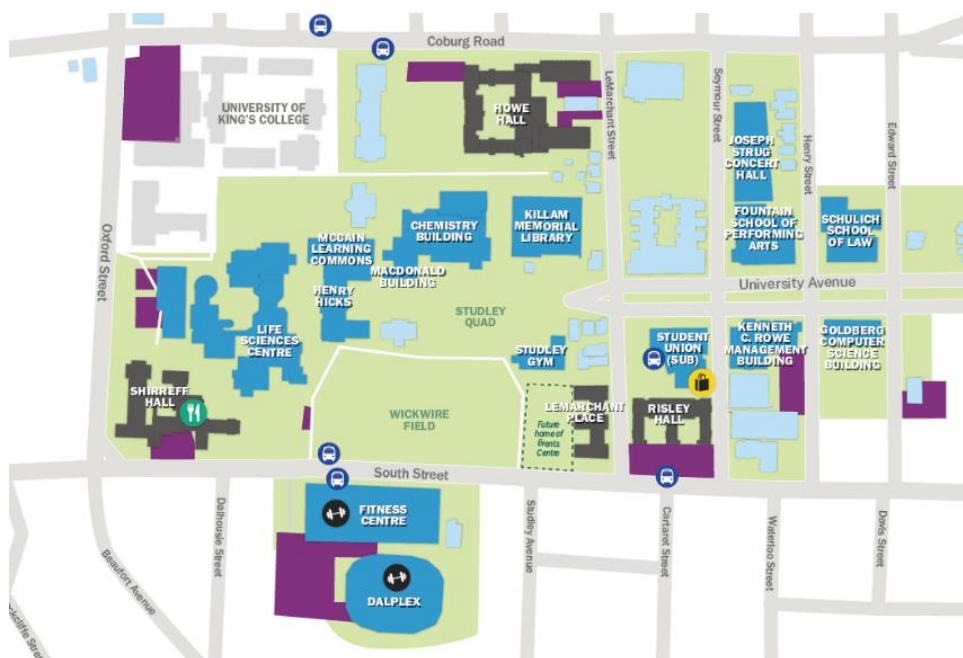
Internet Access

Each attendee will be given a guest account to access Dalhousie's Eduroam wireless network at registration. The credentials (NetID and password) to access the wireless network are unique for each registrant, so *please do not share*. For internet access, connect to the Eduroam network and login with the supplied NetID and password to authenticate. Access will be available on 2025-08-08 and expire on 2025-08-12 at 11:59pm.

If you are visiting from another [eduroam](#)-affiliated institution, you can also use your own home institution's wireless credentials to access Dalhousie's Eduroam wireless network. [Set up eduroam](#) at your home institution first to confirm that it works.

Parking

Parking is available at lots close to the Kenneth C. Rowe Management Building (ROWE bldg) and the Student Union Building (SUB). Parking lots are highlighted purple on the map below. The campus 'Parking Map' can also be found at <https://www.dal.ca/campus-maps/parking/parking-maps.html>. Parking is included with your stay in residence buildings, Risely Hall and LeMarchant Place. If you are not staying in residence, parking passes are available for purchase (cost \$12/day) at the front desk of Risely Hall and LeMarchant Place. Hourly parking is also available at the lot adjacent to Shirreff Hall.



Food

Coffee, tea and other beverages will be available in the lobby of the ROWE bldg during Coffee Breaks. Muffins and other pastries will also be served at the morning Coffee Breaks. Boxed lunches, including sandwiches or wraps, fruit, cookie (or other baked items) and drink, will be provided on Sunday August 10th and Monday August 11th in the SUB, 2nd floor lobby. Food (small bites/ hor d'oeuvres) will also be served at the Welcome Reception and Social/Mixer. Food served at Coffee Breaks, Lunches, Welcome Reception and Social/Mixer are included in the registration fee. At the Welcome Reception non-alcoholic drinks will be available (free) and alcoholic drinks may be purchased at the bar. Social/Mixer attendees will be given a drink ticket for one alcoholic/non-alcoholic drink, and additional drinks may be purchased from the bar. Alcoholic and non-alcoholic drinks will be available on the table during the CSPB dinner, and additional drinks may be purchased at the bar during the reception and dinner.

See <https://dal.campusdish.com/LocationsAndMenus> for on-campus dining locations and hours of operation (note: most dining location are operating on summer hours). A coffee shop is also located on the 1st floor of the SUB.

MEETING SCHEDULE

Saturday, 9th August 2025		
14:00 - 17:00	Registration <i>Lobby, SUB</i>	Poster Set-up <i>McInnes Room, Student Union Building (SUB)</i>
15:00 - 17:00	Welcome Reception <i>Council Chambers, Room 270, SUB</i>	

Sunday, 10th August 2025					
08:00 - 13:30	Poster Set-up <i>McInnes Room, SUB</i>				
08:00 - 17:00	Registration <i>Lobby, Kenneth C. Rowe Management Bldg (ROWE)</i>				
08:50 - 09:00	Opening Remarks <i>Potter Auditorium, Room 1028, ROWE</i>				
09:00 - 10:30	Plenary Session I <i>Potter Auditorium, Room 1028, ROWE</i>				
10:30 - 11:00	Coffee Break <i>Lobby, ROWE</i>				
11:00 - 11:45	Plenary Session I cont.				
11:45 - 12:30	Award Session I <i>Potter Auditorium, Room 1028, ROWE</i>				
12:30 - 13:30	Lunch <i>Lobby, SUB</i>				
13:30 - 15:00	Concurrent Session A1 <i>Room 1009, ROWE</i>	Concurrent Session A2 <i>Room 1011, ROWE</i>	Concurrent Session A3 <i>Room 1014, ROWE</i>	Concurrent Session A4 <i>Room 1007, ROWE</i>	Student and Post-doc led Workshop <i>Room 1016, ROWE</i>
15:00 - 15:30	Coffee Break <i>Lobby, ROWE</i>				
15:30 - 17:00	Concurrent Session B1 <i>Room 1009, ROWE</i>	Concurrent Session B2 <i>Room 1011, ROWE</i>	Concurrent Session B3 <i>Room 1014, ROWE</i>	Concurrent Session B4 <i>Room 1007, ROWE</i>	
17:00 - 18:30	Poster Session I <i>McInnes Room, SUB</i>				
18:30 - 21:30	CSPB Dinner <i>Lord Nelson Hotel, 1515 South Park Street, Halifax</i>				

MEETING SCHEDULE *continued*

Monday, 11th August 2025					
08:50 - 10:20	Plenary Session II <i>Potter Auditorium, Room 1028, ROWE</i>				
10:20 - 10:45	Coffee Break <i>Lobby, ROWE</i>				
10:45 - 12:00	Plenary Session III <i>Potter Auditorium, Room 1028, ROWE</i>				
12:00 - 14:00	Lunch <i>Lobby, SUB</i>	Annual Business Meeting (ABM) <i>Potter Auditorium, Room 1028, ROWE</i>			
14:00 - 15:30	Concurrent Session C1 <i>Room 1009, ROWE</i>	Concurrent Session C2 <i>Room 1011, ROWE</i>	Concurrent Session C3 <i>Room 1014, ROWE</i>	Concurrent Session C4 <i>Room 1016, ROWE</i>	Concurrent Session C5 <i>Room 1007, ROWE</i>
15:30 - 16:00	Coffee Break <i>Lobby, ROWE</i>				
16:00 - 17:00	Concurrent Session D1 <i>Room 1009, ROWE</i>	Concurrent Session D2 <i>Room 1011, ROWE</i>	Concurrent Session D3 <i>Room 1014, ROWE</i>	Concurrent Session D4 <i>Room 1016, ROWE</i>	
17:00 - 18:30	Poster Session II <i>McInnes Room, SUB</i>				
18:30 - 20:30	CSPB Social/Mixer <i>Council Chambers, Room 270, SUB</i>				

Tuesday, 12th August 2025	
08:00 - 13:00	Poster Take-down <i>McInnes Room, SUB</i>
09:00 - 10:30	Plenary Session IV <i>Potter Auditorium, Room 1028, ROWE</i>
10:30 - 11:00	Coffee Break <i>Lobby, ROWE</i>
11:00 - 12:30	Award Session II <i>Potter Auditorium, Room 1028, ROWE</i>
12:30 - 12:50	Award Announcements and Closing Remarks <i>Potter Auditorium, Room 1028, ROWE</i>
13:30 - 16:00	Nature Walk at Duncans Cove <i>SUB Main Entrance</i>

SCIENTIFIC PROGRAM

Saturday, 9th August 2025	
14:00 - 17:00	Registration <i>Lobby, SUB</i>
14:00 - 17:00	Poster Set-up <i>McInnes Room, Student Union Building (SUB)</i>
15:00 - 17:00	Welcome Reception <i>Council Chambers, Room 270, SUB</i>

Sunday, 10th August 2025	
08:00 - 13:30	Poster Set-up <i>McInnes Room, SUB</i>
08:00 - 17:00	Registration <i>Lobby, Kenneth C. Rowe Management Bldg (ROWE Bldg)</i>
08:50 - 09:00	Opening Remarks <i>Potter Auditorium, ROWE Bldg, Room 1028</i>
09:00 - 10:30	Plenary Session I: Plant Resilience - Interactions with Individual, and Combinations of Abiotic and/or Biotic Stresses <i>Chair: Robin Cameron (McMaster University)</i> <i>Potter Auditorium, ROWE Bldg, Room 1028</i>
09:00 - 09:45	A forward genetic screen in <i>Sclerotinia sclerotiorum</i> : finding the most unexpected, Xin Li , University of British Columbia
09:45 - 10:30	Building the toolbox for cannabis improvement, David Joly , Université de Moncton
10:30 - 11:00	Coffee Break <i>Lobby, ROWE Bldg</i>
11:00 - 11:45	Plenary Session I cont.
11:00 - 11:45	Absciscic acid, calcium signaling, and plant responses to high humidity, Eiji Nambara , University of Toronto
11:45 - 12:30	Award Session I <i>Chair: Marcus Samuel (University of Calgary), CSPB President</i> <i>Potter Auditorium, ROWE Bldg, Room 1028</i>
11:45 - 12:30	Mary E. Spencer Award - Plant defense under water: apoplast hydration in plant-microbe interactions, Peter Moffett , Université de Sherbrooke
12:30 - 13:30	Lunch <i>Lobby, Student Union Building (SUB)</i>
13:30 - 15:00	Concurrent Session A1: Biotic Stress I - Cellular Signalling <i>Chair: Zhenyu Cheng (Dalhousie University)</i> <i>Room 1009, ROWE Bldg</i>
13:30 - 13:45	From Arabidopsis to Canola: CPK28 Homologs in Pathogen Resistance, Virginia Miguel , Queen's University
13:45 - 14:00	Investigating the role of CPK4-mediated phosphorylation of the Arabidopsis E3 ubiquitin ligase ATL6 in immune signaling, Faranak Soleimani , Queen's University
14:00 - 14:15	AIL7 transcription factor contributes to defense against clubroot disease, Kethmi Jayawardhane , University of Alberta
14:15 - 14:30	Identification of cyclic nucleotide-gated channels in chitin-triggered immune responses in <i>Arabidopsis thaliana</i> , Hyunsuh Lee , University of Toronto
14:30 - 14:45	Does Overexpressed BnNAC19 in <i>Brassica napus</i> Enhance Plant Resistance Against <i>Verticillium longisporum</i> , Nick Archibald , University of Wilfrid Laurier
14:45 - 15:00	Uncovering new players in Arabidopsis kinase-mediated signaling during abiotic stress, Nathan Doner , Western University

13:30 - 15:00	Concurrent Session A2: Abiotic Stress I - Nutrient Uptake and Signalling <i>Chair: Yunfei Jiang (Dalhousie University)</i> <i>Room 1011, ROWE Bldg</i>
13:30 - 13:45	Ammonium-responsive repressor NRI1 orchestrates nitrogen use strategies in <i>Chlamydomonas reinhardtii</i> , Jae-Hyeok Lee , University of Manitoba
13:45 - 14:00	Nitrogen form substitution identifies nitrogen use efficiency management pathways in maize, Joseph Amoah , University of Sydney
14:00 - 14:15	Examining the role of a RING-type E3 in jasmonic acid-mediated regulation of iron deficiency response in <i>Arabidopsis thaliana</i> , Hannah Iwuoha , Dalhousie University
14:15 - 14:30	How cells know when nitrogen is scarce: A divergent glutamine synthetase essential for nitrogen sensing in algae, Hasni Nimalka Dharmasiri , University of Manitoba
14:30 - 14:45	Role and control of the γ -aminobutyrate (GABA) shunt during acclimation of <i>Arabidopsis thaliana</i> to nutritional phosphorus starvation, Kirsten Benidickson , Queen's University
13:30 - 15:00	Concurrent Session A3: Hormones <i>Chair: Sophia Stone (Dalhousie University)</i> <i>Room 1014, ROWE Bldg</i>
13:30 - 13:45	Investigating the dynamics of strigolactone receptors across the rhizosphere, George Ly , University of Toronto
13:45 - 14:00	CRY2-SMAX1 Crosstalk Illuminates a New Layer of Germination Control in Arabidopsis, Andrea Taylor , University of Toronto
14:00 - 14:15	Identifying a novel plant hormone for the orphan receptor, AtHTL/KAI2, Jenna Hountalas , University of Toronto
14:15 - 14:30	Silver nitrate inhibits ABA2 xanthoxin dehydrogenase, an ABA biosynthetic enzyme, Benjamin P. Brookbank , University of Toronto
14:30 - 14:45	Small Molecule Uptake Through Fungal Transporters: New Perspectives on Symbiosis and Antifungal Action, Freddie Montague , University of Toronto
14:45 - 15:00	Regulation of jasmonate biosynthesis in pepper: Temporal expression of lipoxygenases in response to wounding, Juliette Keith , McGill University
13:30 - 15:00	Concurrent Session A4: Genome Engineering <i>Chair: Lewis Lukens (University of Guelph)</i> <i>Room 1007, ROWE Bldg</i>
13:30 - 13:45	A simple method to efficiently generate structural variation in plants, Satyaki Rajavasireddy , University of Toronto Scarborough
13:45 - 14:00	Optimization of Solid Silica Nanoparticles for Transformation of Protoplasts and Microspores in Canola (<i>Brassica napus</i>), Zhila Osmani , University of Alberta
14:00 - 14:15	CRISPR Makes Its Oat Debut: A breakthrough in oat genomics, Mehtab Singh , McGill University
14:15 - 14:30	Functional Characterization of Thaumatin-like Protein 8 (TLP8) Using the CRISPR-Cas9 System, Cali Kaye , McGill University
14:30 - 14:45	CRISPR-Cas9 editing of de novo DNA methyltransferase alters development and cold responses in <i>Brachypodium distachyon</i> , Luc Ouellette , McGill University
13:30 - 15:00	Student and Post-doc led Workshop: Navigating scholarly publishing - Insights from the editors <i>Room 1016, ROWE Bldg</i>
15:00 - 15:30	Coffee Break <i>Lobby, ROWE Bldg</i>
15:30 - 17:00	Concurrent Session B1: Biotic Stress II - Pathogenicity and Response Mechanisms <i>Chair: Darrell Desveaux (University of Toronto)</i> <i>Room 1009, ROWE Bldg</i>
15:30 - 15:45	Harnessing Plant Evolutionary Diversity to Decode <i>Pseudomonas</i> Virulence, Philip Carella , John Innes Centre
15:45 - 16:00	Swiss Needle Cast and the Foliar Mycobiome of Coastal Douglas-fir (<i>Pseudotsuga menziesii</i>), Jürgen Ehrling , University of Victoria
16:00 - 16:15	Shooting the Messenger: The Importance of Small RNAs in Plant Immunity, Rylan Vincent , University of Toronto - Scarborough

16:15 - 16:30	A clubroot pathogen pseudokinase induces tolerance to the disease in Arabidopsis and canola, Jiaxu Wu , Université Laval
16:30 - 16:45	Pinpointing candidate genes in quantitative trait loci (QTL) associated with resistance to bacterial brown spot (BBS) by exploiting the molecular interactions between adzuki bean (<i>Vigna angularis</i>) and <i>Pseudomonas syringae</i> pv. <i>syringae</i> , Ujomonigho Omoregie , University of Guelph
16:45 - 17:00	Regulation and virulence function of <i>Fusarium graminearum</i> secondary metabolites during Arabidopsis seedling infection, Marielle Zouein , University of Ottawa
15:30 - 17:00	Concurrent Session B2: Abiotic Stress II - Ecophysiology and Climate Change Chair: Sheila Macfie (University of Western Ontario) Room 1011, ROWE Bldg
15:30 - 15:45	Shrub ecophysiological responses to metal stress in degraded peatlands, Ellie Goud , Saint Mary's University
15:45 - 16:00	Plant-derived methane: What do we know about it? Mirwais Qaderi , Mount Saint Vincent University
16:00 - 16:15	Warmer Autumns Delay the Development of Photoprotective Sustained Non-Photochemical Quenching (NPQ) in White Spruce, Anchalya Balasubramaniam , University of Toronto Mississauga
16:15 - 16:30	Intraspecific Variation in Populus balsamifera Indicates Divergent Drought and Elevated [CO ₂] Responses Among Ecotypes, Sahari Inoue , Hokkaido University
16:30 - 16:45	Improving heat stress tolerance in canola (<i>Brassica napus</i> L.) by regulating diacylglycerol acyltransferase activity, Xiaoyu Wang , University of Alberta
16:45 - 17:00	Microtopography and soil moisture gradients drive variation in turgor loss point, xylem conductivity, and growth in white spruce seedlings, Emilie Isasa , University of Toronto Mississauga
15:30 - 17:00	Concurrent Session B3: Biochemistry I - Stress Response Chair: Malinda Thilakarathna (University of Alberta) Room 1014, ROWE Bldg
15:30 - 15:45	Phenotypic Variation in a Batch Culture of <i>Chlamydomonas reinhardtii</i> with Different Light Tolerances, Dion Durnford , UNB
15:45 - 16:00	Do Ginsenosides Alter the Pathogenicity of <i>Ilyonectria</i> ? Anka Colo , Western University
16:00 - 16:15	Role of Reactive Oxygen Species in the Acetate-dependent aging of <i>Chlamydomonas reinhardtii</i> , Navpreet Kaur , University of New Brunswick
16:15 - 16:30	Linking Metabolomic Variation to Larval Performance and Breeding Potential in Conifer Resistance, Mingming Cui , Laval University
16:30 - 16:45	Antioxidant Enzyme Responses and Gene Regulation in Early Interaction between <i>Brassica napus</i> - <i>Verticillium longisporum</i> , Ayomi Thilakarathne , Wilfrid Laurier University
16:45 - 17:00	Molecular Insights into Herbicide Resistance in Wild Oat: Toward Early Detection and Better Management, Jacob Weedmark , Carleton University
15:30 - 17:00	Concurrent Session B4: Reproduction and Reproductive Development Chair: Satyaki Rajavasireddy (University of Toronto, Scarborough) Room 1007, ROWE Bldg
15:30 - 15:45	Single-cell genomics highlights the molecular impacts of <i>Indeterminate1</i> on <i>Zea mays</i> vasculature development and the floral transition, Mark Allan Alexander Minow , University of Georgia
15:45 - 16:00	Effects of Ascorbic Acid Seed Priming on Germination Performance of Seven Broccoli Cultivars, Vijaya ragavan Mohan , Dalhousie University
16:00 - 16:15	The Yellow Submarine: Discovering Suberization in the Silique and Its Functional Role Enabling Endocarp a Degradation, Justin Nichol , University of Calgary
16:15 - 16:30	Free fatty acid biosynthesis precursors are involved in pollen-stigma interactions in Brassica, Kumar Abhinandan , University of Calgary
16:30 - 16:45	Investigating the Receptor-Like Kinase, RKF1, and its Interactors in <i>Arabidopsis thaliana</i> Pollen-Pistil Interactions, Stephen Bordeleau , University of Toronto
16:45 - 17:00	Investigating Arabidopsis stigmatic factors in the regulation of pollen hydration, Raymond Liu , University of Toronto
17:00 - 18:30	Poster Session I McInnes Room, Student Union Building (SUB)
18:30 - 21:30	CSPB Dinner Lord Nelson Hotel, 1515 South Park Street, Halifax

Monday, 11th August 2025	
08:50 - 10:20	Plenary Session II: Using Computational Tools and AI to Advance Plant Biology and Sustainable Agriculture <i>Chair: Jaswinder Singh (McGill University)</i> <i>Potter Auditorium, ROWE Bldg, Room 1028</i>
08:50 - 09:35	Data to Decisions: AI Applications in Crop Systems and Agricultural Decision-Making, Daniela Jones , North Carolina State University
09:35 - 10:20	Quantifying apple diversity for a future of sustainable agriculture, Zoë Migicovsky , Acadia University
10:20 - 10:45	Coffee Break <i>Lobby, ROWE Bldg</i>
10:45 - 12:00	Plenary Session III: Education - Teaching and Learning about Plants <i>Chair: Abel Rosado (University of British Columbia)</i> <i>Potter Auditorium, ROWE Bldg, Room 1028</i>
10:45 - 11:15	Creative Assessment: students can write an exam and have fun, too! Jennifer McDonald , Saint Mary's University
11:15 - 11:45	The terminological thicket: helping students untangle the language of plant biology, Gillian Gass, Lara Gibson, Jennifer Van Dommelen , Dalhousie University
11:45 - 12:00	Destressing Exams, Barbara Ehling , University of Victoria
12:00 - 14:00	Lunch <i>Lobby, SUB</i>
12:00 - 14:00	Annual Business Meeting (ABM) <i>Potter Auditorium, ROWE Bldg, Room 1028</i>
14:00 - 15:30	Concurrent Session C1: Biotic and Abiotic Stress Interactions <i>Chair: Jacquie Bede (McGill University)</i> <i>Room 1009, ROWE Bldg</i>
14:00 - 14:15	Exploring the Potential of <i>Cannabis sativa</i> and <i>Chelidonium majus</i> Endophytes in Promoting Growth and Abiotic Stress Tolerance in Yellow Beans, Laura Amaya Quiroz , McGill University
14:15 - 14:30	Harnessing bacterial synthetic communities to alleviate drought stress in alfalfa and red clover, Maniruzzaman Sikder , Dalhousie University
14:30 - 14:45	Double Trouble in Canola: Understanding the interaction between <i>Verticillium longisporum</i> and <i>Leptosphaeria maculans</i> , Vidushika Madduma Bandara , Wilfrid Laurier University
14:45 - 15:00	The Role Ubiquitin Ligase, XBAT35, in Plant Response to Combination of Abiotic and Biotic Stresses, Rajaswaminathan Vairavan , Dalhousie University
15:00 - 15:15	Effect of abiotic stress on Verticillium stripe disease in Canola (<i>Brassica napus</i> L.), Shruti Kashy , University of Manitoba
15:15 - 15:30	<i>Bacillus toyonensis</i> and <i>Pseudomonas koreensis</i> Cell-Free Supernatants: A Novel Approach to Enhance Soybean Growth under Saline Conditions, Rania Alrasheed , McGill University
14:00 - 15:30	Concurrent Session C2: Abiotic Stress III - Hormones and Signalling <i>Chair: Belay Ayele (University of Manitoba)</i> <i>Room 1011, ROWE Bldg</i>
14:00 - 14:15	Aldehyde dehydrogenases control the carbonylation of photosynthesis-related proteins in <i>Arabidopsis thaliana</i> , Tagnon Missihoun , University of Quebec Trois-Rivieres
14:15 - 14:30	Teasing out the mechanism behind the elevated CO ₂ attenuation of the jasmonate burst in C ₃ plants, Amelia Redl , McGill University
14:30 - 14:45	A Thaumatin-like Protein Bridges Photosystem II and Brassinosteroid Pathways for Guard Cell-Mediated Drought Adaptation, Paul Gamueda , University of Toronto
14:45 - 15:00	Investigation into the roles of calmodulin-binding transcription activators (CAMTAs) in the Arabidopsis heat stress response, Liam Duff , Queen's University
15:00 - 15:15	Overexpression of Canola PLA1ly3 Altered Chloroplast Lipid Homeostasis and Reduced Thermotolerance in Canola, Xinjie Liu , University of Alberta
15:15 - 15:30	Developing drought-tolerant and compact canopy architecture in wheat by manipulating the brassinosteroid pathway, Manpartik Gill , University of Calgary

14:00 - 15:30	Concurrent Session C3: Biochemistry II - Enzymes and Metabolism <i>Chair: Jeffrey Waller (Mount Allison University)</i> <i>Room 1014, ROWE Bldg</i>
14:00 - 14:15	Unlocking the enzyme network driving Amaryllidaceae alkaloids biosynthesis, Isabel Desgagne-Penix , Université du Québec à Trois-Rivières
14:15 - 14:30	Dissecting the function of arogenate dehydratases from <i>Arabidopsis thaliana</i> , Susanne Kohalmi , University of Western Ontario
14:30 - 14:45	Discovery and Characterization of Coenzyme A Ligases Involved in Phytocannabinoid and Bibenzyl Biosynthesis in <i>Radula complanata</i> , Jacob Perley , University of New Brunswick
14:45 - 15:00	Flavonol peroxidation in senescing arugula leaves, Yifan Yan , University of Guelph
15:00 - 15:15	Screening of novel genes associated with seed oil production, Tharangani Somarathna , University of Alberta
15:15 - 15:30	Mechanistic insights into the toxicity of naphthenic acid fraction compounds: Toward plant-based solutions for oil sands process water remediation, Kyle Symonds , University of Calgary
14:00 - 15:30	Concurrent Session C4: Cell Structure and Function <i>Chair: Yang Xu (University of Guelph)</i> <i>Room 1016, ROWE Bldg</i>
14:00 - 14:15	Developing new tools to quantify plant endomembrane dynamics, Emily Larson , University of Montreal
14:15 - 14:30	Characterizing the targeting mechanism of OEP6, a novel chloroplast outer membrane protein, Holly Ferguson , Wilfrid Laurier University
14:30 - 14:45	Protein interaction analyses for plant molecular chaperone HSP90C via Turbo, Morvenley Mamenta , University of Toronto
14:45 - 15:00	What goes up must come down: modulation of the microtubule protein clasp is required for control of cell proliferation, Sean Ritter , University of British Columbia
15:00 - 15:15	Unexpectedly Diverse - Anatomical Variations in the Rosette Leaves of <i>Arabidopsis thaliana</i> , Lamha Kumar , McGill University
15:15 - 15:30	Formation of a stable tubular ER network requires localized phosphatidylcholine synthesis in Arabidopsis, Weina Wang , McGill University
14:00 - 15:30	Concurrent Session C5: Omics' Based Approaches <i>Chair: Rhea Amor Lumactud (Dalhousie University)</i> <i>Room 1007, ROWE Bldg</i>
14:00 - 14:15	Comparative transcriptomics of divergent wheat cultivars reveals epigenetic regulation in microspore embryogenesis, Hai Ying Yuan , National Research Council Canada
14:15 - 14:30	Proteomic analysis of compound leaves of two pea (<i>Pisum sativum</i> L.) cultivars reveals accumulation of regulatory clues for differential reproductive development in two phenotypes, Ravinder Goyal , Agriculture and Agri Food Canada
14:30 - 14:45	Multi-Omics-Driven Genetic Improvement of Pea (<i>Pisum sativum</i> L.) Seed Quality, Bianyun Yu , National Research Council Canada
14:45 - 15:00	Identification of genetic loci for culm breaking strength in spring wheat (<i>Triticum aestivum</i> L.), Ginelle Grenier , University of Manitoba
15:00 - 15:15	From Infection to Insight: De Novo Assembly of the Sunflower Transcriptome Under Fungal Stress, Tien Nguyen , University of British Columbia
15:15 - 15:30	Radicle proteomics of soybean and green pea, in response to lipo-chitooligosaccharide and Thuricin 17, Sowmyalakshmi Subramanian , McGill University
15:30 - 16:00	Coffee Break <i>Lobby, ROWE Bldg</i>
16:00 - 17:00	Concurrent Session D1: Biotic Stress III - Improving Resilience <i>Chair: Yuelin Zhang (University of British Columbia)</i> <i>Room 1009, ROWE Bldg</i>
16:00 - 16:15	Enhancing disease resistance in potato through heterologous expression of multiple host defense peptides, Dmytro Yevtushenko , University of Lethbridge

16:15 - 16:30	Interference of fungal nucleases protects crop plants from disease using next generation RNA fungicides, Brigo Castillo , University of Manitoba
16:30 - 16:45	Exploring the Phenotypic Impacts of the Soybean Endophytic Microbiome Using Shotgun Proteomics, Haleema Tariq , McGill University
16:45 - 17:00	Use of a rapid screen to identify formulations that enhance plant resistance to viral and bacterial infections, Fathy El-Gebaly , McMaster University
16:00 - 17:00	Concurrent Session D2: Applying Computational Strategies <i>Chair: Bianyun Yu (National Research Council Canada)</i> <i>Room 1011, ROWE Bldg</i>
16:00 - 16:15	Selection has acted upon regulatory alleles to improve hexaploid bread wheat, Lewis Lukens , University of Guelph
16:15 - 16:30	Genomic Insights into Aroma Using Whole Genome Sequencing in Apple (<i>Malus domestica</i>), Clay Elzinga , Acadia University
16:30 - 16:45	Weeding Out the Weeds: Genomic tools for improving identification of noxious plant species, Saanchi Singh , Carleton University
16:45 - 17:00	Conservation Challenges and Opportunities for Native Apples (<i>Malus</i> spp.) in Canada, Terrell Roulston , Acadia University
16:00 - 17:00	Concurrent Session D3: Biochemistry II - Enzymes and Metabolism cont <i>Chair: Shawn Mansfield (University of British Columbia)</i> <i>Room 1014, ROWE Bldg</i>
16:00 - 16:15	Metabolic Engineering of Terpene Metabolism in Lavender Glandular Trichomes, Soheil Mahmoud , UBC Okanagan
16:15 - 16:30	Functions and Phosphorylation-Dependent Regulation of the CCT1 Enzyme in Phosphatidylcholine Biosynthesis and Root Development in Arabidopsis, Guanqun (Gavin) Chen , University of Alberta
16:30 - 16:45	Deciphering the role of CYP96T enzymes in the biosynthesis of galanthamine-type Amaryllidaceae Alkaloids from <i>Hippeastrum papilio</i> , Sajjad Sobhanverdi , Université du Québec à Trois-Rivières
16:45 - 17:00	A Novel Model of the PAL-Mediated Salicylic Acid Biosynthesis Pathway: A Functional Rediscovery of Two Hypersensitivity-Related Genes, Dawei Ma , University of Victoria
16:00 - 17:00	Concurrent Session D4: Cell Structure and Function cont <i>Chair: Emily Larson (University of Montreal)</i> <i>Room 1016, ROWE Bldg</i>
16:00 - 16:15	Cell-Tissue Specific Expression Does Not Encode RLCK Signalling Specificity, Eleanor Khochaba , Western University
16:15 - 16:30	Biostimulant Potential of Endophytic Bacteria from <i>Cannabis sativa</i> and Chelidonium majus as Sustainable Alternatives to Agrochemicals, Mohammad Jamil Kaddoura , McGill University
16:30 - 16:45	Genetic Screening with Markers can Assist Backcrossing to Introduce a Novel Trait into Cranberry Dry Bean (<i>Phaseolus vulgaris</i>) Varieties, Sajida Noor , University of Guelph
16:45 - 17:00	Ethylene-mediated sexual plasticity as an example of plant developmental plasticity in <i>Cannabis sativa</i> , Adrian S Monthony , Université Laval
17:00 - 18:30	Poster Session II <i>McInnes Room, Student Union Building (SUB)</i>
18:30 - 20:30	CSPB Social/Mixer <i>Council Chambers Room 270, SUB</i>

Tuesday, 12th August 2025	
08:00 - 13:00	Poster Take-down <i>McInnes Room, Student Union Building (SUB)</i>
09:00 - 10:30	Plenary Session IV: Coastal Eco-physiology and the Changing Climate <i>Chair: Dion Durnford (University of New Brunswick, Fredericton)</i> <i>Potter Auditorium, ROWE Bldg, Room 1028</i>
09:00 - 09:45	Latitude, Light & O2: Phytoplankton from Coast to Coast to Coast, Douglas Campbell , Mount Allison University
09:45 - 10:30	Canadian kelp forests in a changing ocean: consequences of loss and our narrative for the future, Karen Filbee-Dexter , University of Western Australia
10:30 - 11:00	Coffee Break <i>Lobby, ROWE Bldg</i>
11:00 - 12:30	Award Session II <i>Chair: Marcus Samuel (University of Calgary), CSPB President</i> <i>Potter Auditorium, ROWE Bldg, Room 1028</i>
11:00 - 11:30	Carl Douglas Award - Investigating the role of Rosaceae subgroup IV CDPKs in plant immunity, Márcia Gonçalves Dias , Queen's University
11:30 - 12:30	The Gold Medal Award - Pollen acceptance or rejection? Intersecting signaling pathways in the stigma regulate Brassicaceae reproduction, Daphne Goring , University of Toronto
12:30 - 12:50	Award Announcements and Closing Remarks <i>Potter Auditorium, ROWE Bldg, Room 1028</i>
13:30 - 16:00	Nature Walk at Duncans Cove <i>SUB Main Entrance</i>

ABSTRACTS

PLENARY AND AWARD SESSIONS

Sunday, August 10th, 2025

PLENARY SESSION I

1 - A forward genetic screen in *Sclerotinia sclerotiorum*: finding the most unexpected

Yan Xu^{1,2}, Lei Tian^{1,3}, Tina Tan^{1,3}, Weijie Huang¹, Josh Li¹, Yuelin Zhang^{2,3}, Xin Li^{1,3}

¹Michael Smith Laboratories, University of British Columbia; Vancouver BC, V6T 1Z4, Canada, ²The College of Life Sciences, Sichuan University; Chengdu, Sichuan, 610064, China, ³Department of Botany, University of British Columbia Vancouver BC, V6T 1Z4, Canada

Sclerotinia sclerotiorum is one of the worst soilborne pathogens around the world. Despite its economic importance, it has been understudied due to its multinucleate cells, making molecular genetic analyses difficult. We recently developed a forward genetics pipeline in *S. sclerotiorum*, which combines mutant screening with high-throughput next-generation sequencing to enable quick gene discovery. From analyses of mutants that cannot form sclerotia and/or cannot cause diseases, we uncovered the importance of RAS signaling, cAMP, autophagy, and MAPK kinase cascades to the basic biology of *Sclerotinia sclerotiorum*, including sclerotia formation and virulence. We also serendipitously found that chromosomes in haploid cells of *Sclerotinia sclerotiorum* and *Botrytis cinerea* are separated into different nuclei, with each nucleus containing a fraction of the chromosomes. The partition of chromosome subsets into distinct nuclei brings new questions and opens fresh avenues for chromosome biology.

2 - Building the toolbox for cannabis improvement

David Joly¹

¹Université de Moncton

As the legal cannabis industry expands globally, the need for resilient cultivars is critical to ensure consistent yield and product quality. Disease pressure from pathogens such as *Botrytis cinerea*, *Fusarium* spp., and *Golovinomyces ambrosiae* (powdery mildew) poses a significant threat to production systems. However, breeding for disease resistance in *Cannabis sativa* is hindered by two major challenges: (1) limited access to genetically diverse germplasm, and (2) a lack of validated biotechnological tools such as functional transformation systems.

I will review our current screening efforts for resistance to powdery mildew. Using phenotype-based selection under high-inoculum pressure, we identified several promising cultivars exhibiting distinct forms of resistance, from complete immunity to partial resistance, or a form of resistance that appears to be lost after transitioning into flowering. Using structured crossing schemes and molecular markers targeting the previously characterized *CsMLO1* insertion, we unraveled resistance genes hidden behind the loss-of-susceptibility conferred by the *CsMLO1* insertion, but also cultivars where the *CsMLO1* insertion was not sufficient to confer loss-of-susceptibility.

In parallel, I will describe the potential utility (and risk!) of the Hop Latent Viroid (HLVd) as a minimal, stable, and highly mobile RNA backbone for gene silencing, offering a novel alternative to virus-induced gene silencing (VIGS) systems. Indeed, the unique properties of HLVd make it particularly attractive for functional genomics in cannabis, where traditional transformation methods are inefficient or unavailable. Using engineered HLVd constructs with sequence-specific silencing motifs, we demonstrated successful systemic spread and targeted downregulation of gene expression.

This work opens new frontiers with regards to cannabis improvement.

3 - Absciscic acid, calcium signaling, and plant responses to high humidity

Saad Hussain¹, Mohammad Ismu Daud¹, Dylan Bradizza¹, Christopher Antkowiak¹, Christine Nguyen¹, Hiraku Suda², Masatsugu Toyota², Keiko Yoshioka¹, Eiji Nambara¹

¹Department of Cell & Systems Biology, University of Toronto, ²Department of Biochemistry & Molecular Biology, Saitama University

Absciscic acid (ABA) is a plant hormone that regulates plant growth and physiology in response to environmental changes, particularly water status. CYP707A is an ABA 8'-hydroxylase that catalyzes the committed step in ABA catabolism, converting ABA to 8'-hydroxyABA, which non-enzymatically isomerizes to phaseic acid, a less bioactive metabolite. CYP707A is critical for decreasing ABA levels under various water-sufficient conditions. When *Arabidopsis* plants are exposed to high humidity (HH) conditions, the expression of CYP707A3 is induced within 10 minutes, followed by a decrease in ABA levels. We found that HH induces an increase in cytosolic calcium concentrations within a few minutes, activating calcium signaling to induce the transcription of CYP707A3. Calcium signaling also impacts the action of other hormones in response to environmental changes, triggering various downstream physiological responses. We will present our recent results depicting plant hormone- and calcium-mediated plant responses to changing environments.

AWARD SESSION I

MARY E. SPENCER AWARD

4 - Plant defense under water: apoplast hydration in plant-microbe interactions

Peter Moffett¹

¹Université de Sherbrooke

Many plant pathogens seek to colonize the plant extracellular space, known as the apoplast. However, the apoplast is typically unfavorable to microbial growth due to limited water and nutrient availability, as well as both constitutive and inducible antimicrobial defenses. Reflecting the strategic importance of this compartment, plants and microbes engage in a dynamic struggle to shape its composition. Using the *Arabidopsis thaliana*-*Pseudomonas syringae* pathosystem, we have investigated how pathogen effectors contribute to the establishment of an effector-driven extracellular niche (EDEN) by manipulating host physiological processes to enrich the apoplast in water and nutrients. Our findings show that a core mechanism of EDEN formation involves pathogen-induced activation of host absciscic acid signaling, which promotes stomatal closure and leads to water accumulation in the extracellular space, which manifests as water-soaked lesions. In opposition to this virulence strategy, plants mount defense responses, including the production of salicylic acid, which act to maintain stomatal opening, thereby depriving the bacteria of water. In addition, apoplastic water has complex and contrasting effects on plant immune responses. Consistent with the observation that water-soaking is induced by a large number of plant pathogens, extracellular water appears to induce defense signaling, similar to damage-associated molecular patterns. However, if pathogens are able to suppress defense signaling, or if water soaking is established rapidly, a hydrated apoplast results in strong pathogen growth. Our results highlight the complex effects that pathogen-induced microenvironments can have on their host immune responses and *vice versa*.

PLENARY SESSION II

5 - Data to Decisions: AI Applications in Crop Systems and Agricultural Decision-Making

Daniela Jones¹

¹North Carolina State University

Artificial intelligence (AI) is reshaping the agricultural landscape, offering new opportunities to convert complex data into actionable insights that support plant science, improve production efficiency, and advance sustainability goals. This talk will explore how data-driven approaches—from machine learning and remote sensing to optimization modeling—are being used to guide agricultural decisions across scales and sectors.

Through case studies spanning postharvest phenotyping, hyperspectral imaging for quality assessment, and yield forecasting in sweet potatoes, the talk will demonstrate how AI tools are being applied to real-world agricultural challenges. It will also examine the integration of satellite and ground-based environmental data into predictive models that inform agronomic management and biomass logistics for renewable energy systems.

Emphasizing responsible innovation, this presentation will highlight the importance of stakeholder engagement, interpretability, and co-designed solutions. Attendees will gain insight into how artificial intelligence can bridge the gap between data and decision-making in plant biology, agricultural engineering, and sustainable system design.

6 - Quantifying apple diversity for a future of sustainable agriculture

Zoë Migicovsky¹

¹Acadia University

Computational tools that enable genomic analyses and high-throughput phenotyping are advancing our ability to quantify and use plant genetic diversity in crop improvement. Many perennial fruit crops, including apple (*Malus × domestica* Borkh.), are clonally propagated and have a lengthy juvenile phase before producing fruit. As a result, breeding new cultivars is time-consuming and expensive with the same cultivars being used repeatedly in breeding. Making use of broader plant genetic diversity in fruit breeding is essential to supporting a future of sustainable agriculture.

Canada's Apple Biodiversity Collection, one of the most diverse collections of apples in the world, contains over 1,000 unique accessions. Over the past decade, we have used this resource to perform genome-wide association studies (GWAS) across dozens of traits, with the number of genetic markers increasing from 8,000 to over 21 million. This work has enabled the discovery of predictive genetic markers, including one in a NAC18.1 transcription factor associated with fruit ripening which is now used in genomics-assisted breeding.

To complement our increasing genomic resources, we've implemented high-throughput phenotyping approaches. We quantified 18 fruit colour phenotypes using over 12,000 images of apples from Canada's Apple Biodiversity Collection and performed GWAS using these traits. We also applied a pseudo-landmarking morphometric approach to measure apple shape and size, revealing that these traits are independent which suggests that they can likely be targeted separately using genomics-assisted breeding. Recently, we extended this approach more broadly to quantify leaf shape variation across 18 wild apple species.

In summary, our work applies computational tools including both genomics and phenomics to improve the accuracy of apple breeding, enabling the use of broader genetic diversity and supporting the long-term sustainability of perennial fruit agriculture.

PLENARY SESSION III

7 - Creative Assessment: students can write an exam and have fun, too!

Jennifer McDonald¹

¹Saint Mary's University

This January I had a meeting with a student thinking of taking my course, and I asked them what hesitations they had about their ability to succeed in my course. Their response: “I’ve heard from my friends that you make students do weird things for grades.” In this plenary I’ll talk about all of the strange and unusual assignments and tests I’ve given my students over the years: smoothie competitions, board games, creative writing assignments, nature journals, creating Frankenplants, collaborative exams, and even theoretical cyborg robot building for Comic Con costume contest purposes, as well as the feedback I’ve had from students about these unusual assignments, tests, and exams. Students express a decreased sense of anxiety and stress over writing exams, and many say they feel they are better able to convey the information they’ve learned in the course in these unusual course assignments. What few of them realize is that these exercises help them synthesize course material across course topics, apply course material in unique ways, and prepare them for careers in science disciplines by refining skills in writing, problem-solving, and science communication.

8 - The terminological thicket: helping students untangle the language of plant biology

Gillian Gass¹, Lara Gibson¹, Jennifer Van Dommelen¹

¹Department of Biology, Dalhousie University

Learning the language of plant biology requires students to acquire many novel terms to describe structures, processes, and names of groups and organisms. How can those of us teaching plant biology help our students work through this challenge in a way that helps them grow their plant vocabulary while at the same time establishing a secure foundation of plant biology knowledge? In this session, we’ll consider the importance of accessing prior knowledge, the role and value of rote learning, and tools students can use to create links between terms, processes, structures, and taxa. We’ll focus on common areas of difficulty for students learning about plants in first- and second-year biology classes, presenting research-informed strategies that we use in our own work.

9 - Destressing Exams

Barbara Ehlt¹

¹University of Victoria

Mental health-related disabilities affect a significant proportion of Canadian youth aged 15-24—19% of females and 9% of males—according to a 2024 Statistics Canada report. Among university students, academic stress is a key contributor to anxiety, especially during exam periods. This raises an important question: how can we effectively reduce exam-related stress in undergraduate students?

Therapy animals, particularly dogs, have become increasingly common in educational post-secondary institutions. These animals offer immediate emotional support, reduce feelings of homesickness, and contribute positively to student well-being. At the University of Victoria, certified therapy dogs and their handlers visit campus regularly during academic terms, providing comfort and connection. However, the calming effects of therapy dogs are often temporary and may not persist during high-stakes periods like exams.

To investigate whether therapy dog interactions just prior to exams has a beneficial impact, I conducted a study involving volunteer participants from two large-enrollment undergraduate courses. After obtaining human and animal ethics approval, students self-reported their stress levels before exams—with and without therapy dog interactions (control group). The findings reveal that students who interacted with therapy dogs reported lower anxiety levels and a greater sense of readiness to write their exams, compared to those who did not (control group). Recognizing that therapy dog access is limited, I also examine other accessible and scalable strategies

to support students during exams. These include the benefit of so-called cheat sheets, balanced assessment design (formative vs. summative), and innovative approaches such as two-stage exams. Together, these approaches can contribute to a more supportive academic environment that addresses mental health challenges and enhances student success.

Tuesday, August 12th, 2025

PLENARY SESSION IV

10 - Latitude, Light & O₂: Phytoplankton from Coast to Coast to Coast

Douglas Campbell¹, Mireille Savoie¹, Sylwia Sliwiska-Wilczewska², Maximilian Berthold³, Julie Nadeau⁴, Laurel Genge⁵, Naaman Omar⁶, Amanda Cockshutt⁷

¹Mount Allison University, ²Gdansk University, ³Umweltbundesamt, Germany, ⁴McGill University, ⁵Organigram Inc., ⁶University of New Brunswick, ⁷St. Frances Xavier University

Our phytoplankton ecophysiology group implemented integrated high-throughput culturing, bio-optics, physiology, bioinformatics and field work, to study phytoplankton responses to matrices of light, oxygen & temperature. The light-driven oxygenation of the biosphere by phytoplankters transformed global biogeochemistry, yet low-oxygen zones persist in water columns, and are indeed currently re-expanding in area. The cost/benefit trade-offs between light capture for photosynthesis, and the metabolic burden of assembling and maintaining the complexes catalyzing photosynthesis, vary across taxa interacting with irradiance, spectral shifts and oxygen levels. Surprisingly, some oxygenic picocyanobacteria thrive under low oxygen, which lowers their maintenance costs, allowing them to (potentially) exploit higher light, if low-oxygen zones extend towards the surface. In coastal habitats mixing and sinking carry phytoplankters through steep, centimeter scale environmental gradients, favouring physiological generalists. In contrast, clear offshore waters present gradients spanning many meters of depth, allowing for evolution of specialists tuned to specific conditions. Benthic microphytobenthos use motility to cope with steep millimeter scale bio-physical gradients. Irradiance, light spectral quality and oxygen levels are key, interacting elements of these niche gradients. Currently warming waters intensify depth stratifications, which retains cells in layers, while opening potential poleward extensions of temperature ranges. Yet photoperiods, particularly at the poles, will remain imposed by the annual cycle, and by depth attenuation of light. Photoperiod requirements will constrain some specialist strains from exploiting expanding temperature niches. Polar species survive prolonged darkness and sustained cold, while re-launching growth under extremely low light to generate spring blooms. We are finding that low water temperatures are critical to extreme photosynthesis under extreme low light, by suppressing wasteful dissipations of photons. Warming polar waters may disrupt this low-light survival mechanism in polar species.

11 - Canadian kelp forests in a changing ocean: consequences of loss and our narrative for the future

Karen Filbee-Dexter¹

¹University of Western Australia

Canada hosts the largest coastline in the world, and large expanses of it are dominated by kelp forests. These ecosystems are the second most vulnerable marine ecosystem to climate change and are being reshaped by humans across their global distribution. I will explore three general patterns of change in Canadian kelp forests: predicted expansion in some polar regions, overgrazing in temperate areas, and collapse to turfs and tropicalized reefs at warm range edges. Losses of kelp forests have important consequences for key ecological functions including supporting biodiversity, provision of fisheries, and carbon and nutrient cycling. Restoration and marine protected areas have been proposed as key solutions required to rebuild kelp forests and their associated biodiversity. Yet there is unfounded optimism around this 'fix it' narrative, as restoration and no-take areas often do not address core drivers of loss and degradation. In fact, these actions are rarely kelp targeted or scalable to match current rates of kelp decline. Currently there is intense discussion around leveraging kelp carbon credits for restoration and the strong need to verify their role in carbon drawdown and sequestration.

This debate is a distraction. Even if certified carbon credits come about, carbon credit revenue will not cover the cost of kelp forest restoration, and may mask the consequences of ongoing kelp forest decline on carbon dioxide drawdown. I argue that although small-scale efforts to restore kelp forests may be meaningful in some areas, a more compelling narrative is that these ecosystems and their associated functions are often irreplaceable, putting emphasis on conservation and preventing further loss as the only viable path forward.

AWARD SESSION II

CARL DOUGLAS AWARD

12 - Investigating the role of Rosaceae subgroup IV CDPKs in plant immunity

Márcia Gonçalves Dias^{1,2}, Beatriz Appezzato-da-Glória², Jacqueline Monaghan¹

¹Queen's University, ²University of São Paulo

Plant diseases pose significant challenges to biodiversity conservation and food production. However, as plants have evolved several mechanisms to cope with environmental and biotic stressors, it should be possible to boost plant resilience through genetic engineering. Calcium-dependent protein kinases (CDPKs) are well-established regulators of abiotic and biotic stress in the model Brassicaceae *Arabidopsis thaliana*, however their function in the Rosaceae - a globally important horticultural plant family that includes raspberries, strawberries, and ornamental plants - remains poorly understood. In this study, we identified 14 CDPK proteins in black raspberry (*Rubus occidentalis*) and 12 in strawberry (*Fragaria vesca*) based on predicted orthology to the *A. thaliana* CDPK family. Focusing on group IV CDPKs, particularly CPK28 due to its role in plant immunity, we identified single orthologs in these species, herein named RoCPK28 and FvCPK28. Transient expression of these proteins in *Nicotiana benthamiana* revealed their localization to the plasma membrane and showed that their overexpression dampened the immune-triggered oxidative burst. Our ongoing work aims to assess the functional role of the Rosaceae CPK28 orthologs through genetic complementation and to characterize their immune phenotypes in response to pathogen challenges. We expect these findings to contribute to a better understanding of CPK28 function conservation in Rosaceae and to provide insights into leveraging these kinases for improving crop disease resistance.

THE GOLD MEDAL AWARD

13 - Pollen acceptance or rejection? Intersecting signaling pathways in the stigma regulate Brassicaceae reproduction.

Daphne Goring¹

¹Dept Cell and Systems Biology, University of Toronto

The regulation of sexual reproduction in the Brassicaceae begins immediately after pollination of the stigma at the top of the female pistil. The stigma selectively recognizes compatible pollen and provides water to the desiccated pollen for hydration. A pollen tube then emerges and penetrates the stigmatic surface to begin its journey down the reproductive tract to deliver sperm cells to an ovule for fertilization. The Brassicaceae stigma can also have reproductive barriers to recognize and actively reject self-incompatible pollen or interspecies-incompatible pollen by suppressing compatible pollen responses. These three pollen recognition systems rely on pollen signalling peptides that are perceived by stigma receptor kinases which then regulate cellular responses in the stigma to promote or reject pollen. Working with *Brassica* and *Arabidopsis* species, the Goring research group has identified a number of novel players that function in the stigma for these pollen recognition systems and have investigated their cellular functions to discover the mechanisms by which pollen is accepted or rejected, and uncovered intersecting points between these pathways. Ultimately, these pollen recognition systems and barriers in the stigma are critical for promoting compatible pollen-pistil interactions as they affect the success of the downstream post-fertilization events of gamete fusion and seed development. As well, by elucidating the cellular regulation of these early post-pollination interactions, this research can impact strategies for improving agricultural crop seed production.

ABSTRACTS

CONCURRENT SESSIONS

Sunday, August 10th, 2025

CONCURRENT SESSION A1

A1-14 - From Arabidopsis to Canola: CPK28 Homologs in Pathogen Resistance

Virginia Miguel¹, Edel Perez-Lopez², Mark Belmonte³, Jacqueline Monaghan¹

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Plants constantly balance growth and defence, activating immune responses only when necessary to avoid compromising development. Enhancing disease resistance in economically important oilseed crops, such as canola (*Brassica napus*), without compromising growth remains a significant challenge. In *Arabidopsis thaliana*, the calcium-dependent protein kinase CPK28 negatively regulates immune signalling by modulating BIK1 stability. Specific mutations in AtCPK28 enhance immunity without adverse effects on growth, thereby positioning this kinase as a promising target for crop improvement through precision editing.

To translate these findings into canola, we employed comparative genomics to identify four *B. napus* genes with high sequence similarity to AtCPK28. In vitro kinase assays confirmed conserved autophosphorylation activity, and confocal microscopy revealed plasma membrane localization. To assess functional conservation, we performed complementation assays in the *Atcpk28-1* mutant background. Likewise, using the native *AtCPK28* promoter to express mutated versions of these candidate homologs, we are assessing their potential to enhance immunity without affecting growth, consistent with previous findings for AtCPK28. Preliminary results indicate that the *BnaCPK28* homologs successfully complement the *cpk28-1* mutant phenotypes, suggesting functional conservation of these genes across species.

We are also generating knockout lines for the four *BnaCPK28* homologs to determine their roles in immune signalling and development in canola. Altogether, these results lay the groundwork for applying precision editing of these loci in canola, offering a path toward elite lines with durable disease resistance and minimal growth trade-offs.

A1-15 - Investigating the role of CPK4-mediated phosphorylation of the Arabidopsis E3 ubiquitin ligase ATL6 in immune signaling

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Protein kinases control various cellular processes by transferring a phosphoryl group from ATP onto specific Ser/Thr or Tyr residues of substrate proteins. Identification of protein kinase substrates is a critical goal for establishing signaling cascades controlled by protein phosphorylation. Calcium-dependent protein kinases (CPKs) possess both Ca²⁺ sensing and enzymatic activities, allowing them to directly translate Ca²⁺ signals into protein phosphorylation events. In the model plant *Arabidopsis thaliana*, CPK4 participates in several pathways such as abscisic acid signaling, immune signaling, reactive oxygen species production, and central metabolism. The current study is centered on our recent discovery that the ubiquitin E3 ligase *TOXICOS EN LEVADURA 6* (ATL6) is a putative CPK4 target. Previous work has shown that ATL6 polyubiquitinates CPK28,

leading to its transient degradation and subsequent activation of immune signaling. Phosphosite mapping using mass spectrometry revealed that CPK4 efficiently phosphorylates recombinant ATL6 at multiple Ser and Thr residues *in vitro*. Our research aims to assess the impact that CPK4-mediated phosphorylation exerts on ATL6 activity and biological functions. The knowledge of how plants sense and acclimate to stress can be utilized in biotechnology to enhance crop resistance to various stressors.

A1-16 - AIL7 transcription factor contributes to defense against clubroot disease

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Clubroot, caused by *Plasmodiophora brassicae*, is a major soilborne disease of canola (*Brassica napus*) and other Brassicaceae crops, leading to significant yield losses worldwide. As chemical control options are limited for clubroot, resistant cultivars are essential for disease management. Identifying novel genes involved in clubroot resistance is therefore critical. AINTEGUMENTA-LIKE 7 (AIL7) is a member of the APETALA2/ETHYLENE RESPONSE FACTOR (AP2/ERF) transcription factor family, with some members linked to plant defense. Seed-specific *AIL7* overexpression lines showed altered expression of defense-related genes, many of which overlap with genes responsive to *P. brassicae* infection in resistant Brassica varieties. This study investigates the clubroot response of constitutive *AIL7* overexpression (OE) and knockout (KO) *Arabidopsis* lines. Following inoculation with *P. brassicae*, both OE and KO lines displayed reduced clubroot symptoms and lower resting spore loads compared to the wild type, indicating enhanced disease tolerance. Hormone analysis revealed that *AIL7*-KO lines had elevated salicylic acid (SA) and jasmonic acid (JA) levels even without inoculation, suggesting a primed defense condition. *AIL7*-OE lines showed a similar but weaker response, where only JA levels were significantly enhanced without inoculation. Gene expression analysis further confirmed these findings. In addition, *AIL7*-OE lines showed differential expression of *UGT76B1* and *SnRK1* post-infection, indicating potential roles in alternative resistance pathways. Overall, AIL7 appears to modulate clubroot tolerance by influencing phytohormone levels and related defense gene expression at least in part. These findings highlight AIL7 as a promising candidate for developing clubroot-resistant canola through breeding strategies.

A1-17 - Identification of cyclic nucleotide-gated channels in chitin-triggered immune responses in *Arabidopsis thaliana*

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Calcium (Ca²⁺) signaling plays a central role in almost all aspects of plant responses, including immunity. Following the recognition of microbes, the transient increase in cytosolic Ca²⁺ concentration triggers a number of immune responses, including reactive oxygen species (ROS) production and transcriptional reprogramming. Plant cyclic nucleotide-gated channels (CNGCs) are a family of Ca²⁺-permeable channels that contribute to Ca²⁺ signaling. In *Arabidopsis thaliana*, several members of the CNGC family have been implicated in immune responses. Here, we identified two CNGCs with previously uncharacterized roles in immunity that are transcriptionally upregulated upon infection by the fungal pathogen *Botrytis cinerea*. To investigate their immune function, we used single and double knockout mutants of these CNGCs and examined their response to chitin—a component of fungal cell walls and a known elicitor of plant immunity. We observed that the double mutant exhibited reduced chitin-induced Ca²⁺ influx compared to wild-type plants. This reduction was accompanied by diminished ROS production upon chitin recognition. To further elucidate downstream effects, we performed a genome-wide transcriptional analysis (RNA-seq) on chitin-treated plants and found that genes differentially regulated in the double mutant are enriched for Gene Ontology terms associated with defense responses and oxidative stress. Notably, the expression of multiple ROS-scavenging genes was altered in the double mutant, consistent with its impaired ROS production. Together, our findings identify two CNGCs as

important mediators of Ca²⁺ signaling upon chitin recognition, modulating ROS production and the activation of defense genes during fungal pathogen-related plant immunity.

A1-18 - Does Overexpressed BnNAC19 in *Brassica napus* Enhance Plant Resistance Against *Verticillium longisporum*

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Canola (*Brassica napus*) is one of the most significant cash crops in Canada, contributing approximately \$30 billion to agricultural revenue. *Verticillium longisporum* is an emerging soilborne fungal pathogen in Canada that causes Verticillium stripe disease in canola, resulting in significant yield losses. Verticillium stripe disease results in the formation of microsclerotia in the stalks, causing epidermal peeling and host plant decomposition, allowing microsclerotia to re-enter the soil where they lie dormant until the next growth season. There are currently no known resistant varieties, biocontrols or fungicides to help control *V. longisporum* infection. Previously, we have identified that the overexpression of a transcription factor *BnNAC19* (NAM, ATAF, and CUC) can enhance canola resistance against another important fungal disease, blackleg. This study aimed to investigate whether the overexpressed *BnNAC19* transgenic canola line also exhibits enhanced resistance against *V. longisporum*. Two-week-old seedlings of Westar and *OEBnNAC19* transgenic lines were inoculated with *V. longisporum* isolate and monitored over a 120-day growth period. Seedling phenotypic evaluations were conducted at 1, 3, 7, and 14 days post inoculation (dpi), and adult plants were rated at 120 dpi. Plant roots and leaves were sampled at 7 and 14 dpi, respectively, for gene expression analysis. Our initial results indicated that overexpression of *BnNAC19* in canola resulted in greater stunting, necrosis, and yellowing of the cotyledons and leaves in seedlings. Gene expression analysis showed an upregulation of pathogenesis-related gene 1 (*PR1*) and a downregulation of *PR2* in overexpression lines. This suggests that *BnNAC19* may act as a negative regulator for Verticillium stripe disease resistance. To validate these findings, we will further assess the expression of several pathogenesis-related (*PR*) genes and measure antioxidant activity in the overexpression of *BnNAC19* transgenic plants, aiming to characterize the role of *BnNAC19* in response to *V. longisporum*.

A1-19 - Uncovering new players in Arabidopsis kinase-mediated signaling during abiotic stress

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Receptor kinases in plants transduce signals from the outside of the cell inward. Inside the cell, Receptor-Like Cytoplasmic Kinases (RLCKs) are subsequently activated by receptor kinase phosphorylation and serve to (in)activate various downstream processes. Several recent studies have implicated the RLCK-VII subfamily in playing key roles in biotic stress response and indeed mutants of RLCK-VIIs are often impaired in their ability to respond to biotic challenge. Yet, many of the 46 Arabidopsis RLCK-VIIs (and most RLCKs in general) have no known cellular signaling role. Given that receptor kinases are known to transmit signals during abiotic stress responses, the present study seeks to uncover previously unknown abiotic stress roles for RLCK-VIIs by screening a collection of RLCK-VII mutants with abiotic challenges.

The screen was conducted using 9 lines of multi-gene RLCK-VII knockout mutants, covering 33/46 Arabidopsis RLCK-VIIs, subjected to various abiotic stress assays. In this presentation, I will discuss the newly generated data and how it advances our understanding of RLCKs in plant kinase signaling. I will also discuss planned follow-up studies on RLCK-VIIs that display altered stress responses. This work exploits Arabidopsis mutants and molecular biology techniques to better characterize the role of specific RLCK pathways in specific abiotic stress responses.

The molecular responses to abiotic stresses have been studied extensively in plants, however, the signalling pathways between the plant perceiving the stress and mounting a response are not well understood. This work serves to uncover new players in these pathways which will advance our understanding of the plant stress response and provide valuable information to inform the study of stress-tolerant crops.

CONCURRENT SESSION A2

A2-20 - Ammonium-responsive repressor NRI1 orchestrates nitrogen use strategies in *Chlamydomonas reinhardtii*

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In *Chlamydomonas reinhardtii*, the addition of ammonium to nitrate-fed cultures rapidly suppresses NIT2 (an Arabidopsis NLP7 homolog)-dependent transcription of nitrate assimilation genes, supporting preferential use of ammonium. However, the molecular basis for this ammonium repression remains unclear. We recently identified a bHLH transcription factor, NRI1 (an Arabidopsis BIM homolog), as a global repressor of alternative nitrogen assimilation pathways⁽¹⁾. Its activity, modulated by ammonium availability, offers a compelling mechanism for ammonium repression. To assess NRI1's role, we identified 107 nitrate-specific genes, of which 102 were suppressed within two hours of ammonium addition—defined as ammonium repression targets (ARTs). Transcriptome analyses revealed that over 80% of ARTs showed NRI1-dependent downregulation, indicating a central role for NRI1. The remaining 10 genes required nitrate for expression even in *nri1* mutants, classifying them as bona fide NIT2-dependent targets. Notably, these NIT2-dependent genes were no longer repressed by ammonium in *nri1* mutants, suggesting antagonism between NRI1 and NIT2. To explore how ammonium regulates NRI1, we imaged YFP-tagged NRI1 *in vivo*. Under ammonium-fed conditions, NRI1 localized to the nucleus. Upon nitrogen removal, NRI1 rapidly exited the nucleus and formed puncta, suggesting degradation. In nitrate-fed cells, NRI1 accumulated in chloroplasts, correlating with strong expression of its target genes. Introduction of ammonium restored nuclear localization. In conclusion, nitrate-induced gene expression in *Chlamydomonas* involves both NRI1- and NIT2-dependent regulations. Nuclear retention of NRI1 under ammonium-rich conditions enables repression of secondary N assimilation pathways, while its exclusion during nitrogen starvation permits rapid gene induction, providing a dynamic mechanism for nitrogen source preference.

References

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A2-21 - Genotypic variability and physiological determinants of nitrogen-use efficiency in bread wheat near isogenic lines

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Nitrogen (N) is critical nutrient for maximizing grain yield (GY) and grain protein concentration (GPC) in wheat. Understanding the physiological and genetic basis of nitrogen use efficiency (NUE) is essential for improving nitrogen uptake and its utilization across diverse wheat genotypes. We hypothesized that genotypic variation among near isogenic lines (NILs) differing in GY and GPC would enable the identification of alleles associated with NUE and related physiological traits, providing insights for physiological breeding. A panel of 85 NILs, developed from an elite spring wheat breeding population, was evaluated across three field sites over three years (nine site-years) for agronomic and physiological traits linked to nitrogen accumulation in wheat. The NUE was determined as the product of nitrogen uptake efficiency (NUpE) and nitrogen utilization efficiency (NuTE). Significant genotypic variation was observed for GY (570-1551 g/plot), GPC (14.4 -15.8%), falling numbers (318 - 648 secs), gluten strength index (71 - 124), normalized difference vegetation index at grain fill (0.50 - 0.58), and harvest index (41.8 - 46.6%). NUE ranged from 18.4% to 24.31% among the NILs, with NILs B1018-LN04C25, B1018-LN04C64, and B1018-LN04C65 exhibiting the highest NUE and GY across test environments. In contrast, NILs B1018-LN04C75, B1018-LN04C32, and B1018-LN04C08 consistently showed lower NUE. Exome sequencing analysis of 30 selected lines revealed QTLs/SNPs associated with key traits: seven QTLs for GY (chr. 1A, 2B, 3B, 4A, 6A & 6B), two QTLs for GPC (chr. 6B & 7B), four QTLs for NUE (chr. 2B, 5B, 7B & an unknown chromosome), and six QTLs for N remobilization (chr.2B, 3B, 6B, 7B, 7D and an

unknown chromosome). Overall, these findings highlight substantial genotypic variation in NUE and its components and demonstrate the potential of using high-impact SNPs and physiological traits for selecting wheat germplasm with improved GY, GPC, and NUE.

A2-22 - Nitrogen form substitution identifies nitrogen use efficiency management pathways in maize

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Plants rely on nitrogen (N) for their growth, development, and metabolic functions. However, the regulatory mechanisms modulating N assimilates allocation under varying N forms is unclear. This study examines N metabolism and spatial distribution in maize seedlings subjected to four N treatments (T1 to T4): T1, 1 mM NO_3^- (sole NO_3^-); T2, substitution of 1 mM NO_3^- with 1 mM NH_4^+ (N form substitution, NFS); T3, 1 mM NH_4^+ (sole NH_4^+); and T4, 0.5 mM NH_4NO_3 (mixed N supply). The NFS treatment induced significant physiological and molecular adaptations, such as enhanced growth and total biomass under fluctuating N conditions. NFS-treated plants exhibited improved photosynthesis, increased protein and amino acid synthesis, and increased NO_3^- and NH_4^+ accumulation. Activities of key N metabolism enzymes, such as nitrate reductase (NR), nitrite reductase (NiR), glutamine synthetase (GS), and glutamate synthase (GOGAT), were significantly upregulated, supporting efficient assimilation of both NO_3^- and NH_4^+ . Molecular analysis revealed transcriptional reprogramming under NFS, marked by the upregulation of genes related to NH_4^+ metabolism (*ZmGS1* and *ZmGOGAT1*) and transport (*ZmAMT2.1* and *ZmAMT3.1*). Furthermore, spatial and diurnal analyses revealed dynamic N partitioning and adaptive regulation, with NFS-treated plants maintaining consistently higher NO_3^- and NH_4^+ levels in leaves, roots, sheaths, and developing ears. These findings highlight the robust plasticity of maize N metabolism under NFS conditions and provide valuable insights into optimizing N use efficiency (NUE) for sustainable crop production. Future studies will focus on exploring these adaptive mechanisms across different maize genotypes and under field conditions to improve NUE and productivity in varying N environments.

A2-23 - Examining the role of a RING-type E3 in jasmonic acid-mediated regulation of iron deficiency response in *Arabidopsis thaliana*

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Iron (Fe) is a vital micronutrient for plant growth, yet its bioavailability is limited in alkaline soils, leading to a deficiency stress. In *Arabidopsis thaliana*, the iron deficiency response (IDR) is activated under low iron to increase uptake, and it involves solubilizing immobile iron via rhizosphere acidification by AHA2 (PROTON ATPASE 2), reducing the mobile Fe^{3+} to Fe^{2+} by FRO2 (FERRIC REDUCTION OXIDASE 2), and transporting Fe^{2+} into the root epidermal cell by IRT1 (IRON-REGULATED TRANSPORTER 1). This process is tightly regulated by a suite of transcription factors, including FIT1 (IRON-DEFICIENCY INDUCED TRANSCRIPTION FACTOR 1), which promotes the expression of the *AHA2-FRO2-IRT1* uptake complex. However, the stress hormone jasmonic acid (JA) suppresses the IDR, although the mechanism involved remains unclear. Another layer of modulation involves post-translational regulation via the ubiquitin-proteasome system (UPS), where ubiquitin ligases (E3s) selectively target components of the IDR for degradation. We have identified a novel RING-type E3, NRE17 (Nutrient-induced RING E3 17), as a potential regulator of iron uptake. Preliminary data suggest that NRE17 positively regulates the IDR. Interestingly, JA regulates *NRE17* expression depending on the iron status of the plant. Notably, JA suppresses *NRE17* expression under iron-deficient conditions, suggesting that JA regulates the IDR by reducing *NRE17* abundance. This study explores the role of *NRE17* in the IDR and its potential role in connecting JA signalling and tolerance to iron deficiency stress.

A2-24 - How cells know when nitrogen is scarce: A divergent glutamine synthetase essential for nitrogen sensing in algae.

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Nitrogen (N) fertilizers have significantly increased crop yields, yet poor plant N use efficiency—due to ammonium toxicity and nitrate leaching—poses major environmental and sustainability challenges. Despite the centrality of N to plant growth, little is known about how cells monitor and regulate intracellular N flow. To investigate cellular mechanisms underlying N sensing, we performed a forward genetic screen in the unicellular model alga *Chlamydomonas reinhardtii*, targeting defects in *NIT1* transcriptional regulation upon N starvation. This screen yielded two contrasting mutant classes: *nitrogen starvation insensitive 1 (nsi1)* and *nitrogen repletion insensitive 1 (nri1)*[1]. Five independent *nsi1* alleles failed to induce most N starvation-responsive genes, while ribosome and chlorophyll recycling remained active. Notably, *nsi1* cultures showed impaired growth on organic N, suggesting that NSI1 is essential for alternative N utilization. NSI1 encodes a divergent type-II glutamine synthetase (GS) conserved in chlorophytes. Given the pivotal role of GS in inorganic N assimilation, we hypothesize that NSI1 acts as a sensor of N flow into the organic pool. To investigate this, we complemented *nsi1* with a fluorescently tagged NSI1 and found the protein consistently localized to a single cortical site between the plasma membrane and chloroplast, regardless of N availability. This stable localization implies a localization-sensitive role in sensing N flow. We further propose that NSI1 responds to N status via conformational changes, potentially through dynamic oligomerization, a feature currently under investigation using *in vitro* and *in silico* approaches. These findings offer new insights into metabolic N sensing and may guide strategies for engineering crops with improved nitrogen resilience and use efficiency.

A2-25 - Role and control of the γ -aminobutyrate (GABA) shunt during acclimation of *Arabidopsis thaliana* to nutritional phosphorus starvation

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Inorganic phosphate (**Pi**) is a crucial plant macronutrient, but its limited soil availability necessitates the overuse of unsustainable Pi fertilizers in agriculture. However, plants have evolved various adaptations that improve their Pi acquisition and use efficiency when growing in Pi-deficient (**-Pi**) soils. These include enhanced expression of *Pi-starvation inducible* genes, altered post-translational modifications of many proteins, and modified flux through various metabolic pathways. One such pathway is the γ -aminobutyrate (**GABA**) shunt, a bypass of the Krebs' cycle implicated in many stress responses. The first committed step is catalyzed by glutamate decarboxylase (**GAD**), a Ca^{2+} /calmodulin-activated enzyme that produces GABA. Our previous research showed that: (i) the GAD isozyme, AtGAD1, becomes hyperphosphorylated *in vivo* following Pi-resupply to -Pi *Arabidopsis* cell cultures (Mehta *et al.* 2021 Plant J.), and (ii) growth and GABA accumulation of -Pi *atgad1* T-DNA mutants was markedly attenuated relative to wild-type (**WT**) plants (Benidickson *et al.* 2023 New Phytol.). We are currently comparing the kinetic properties of recombinant phosphomimetic and phosphoablative AtGAD1 mutants to test the hypothesis that AtGAD1 is post-translationally activated by dephosphorylation to promote controlled GABA accumulation during Pi starvation. We are also assessing the phenotype of *Arabidopsis* GABA-transaminase (*gaba-t*) mutants to assess the relative importance of GABA accumulation versus its metabolism via the GABA shunt during Pi deprivation. Initial results indicate that -Pi *gaba-t* mutants show enhanced growth and GABA accumulation relative to WT plants, supporting a beneficial adaptive role for GABA itself. However, excessive GABA is known to cause developmental trade-offs (such as reduced fertility and seed yield), emphasizing the need for tight regulation of GABA metabolism. Together, our findings highlight the importance of precise control of GABA synthesis and turnover in plant Pi starvation responses, and inform future strategies to engineer crops with improved Pi-use efficiency, reducing agriculture's overreliance on non-renewable and polluting fertilizers.

CONCURRENT SESSION A3

A3-24 - Investigating the dynamics of strigolactone receptors across the rhizosphere

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Strigolactones (SLs) are a class of phytohormone known for regulating shoot branching and root architecture in plants. In addition to regulating processes in plants SLs are known to be exuded into the rhizosphere affecting the other organisms in the area. One such interaction is with *Striga hermonthica*, an obligate root hemiparasite known for parasitizing and devastating crop plants. *S. hermonthica* perceives SL through the HTL (Hyposensitive To Light) family of alpha/beta hydrolases triggering its germination through a signalling cascade. Another effect of SL in the rhizosphere is its perception by arbuscular mycorrhizal (AM) fungi. AM fungi form symbiotic relationships with 70% of all land plants by forming mycorrhizae to exchange nutrients. Despite the ubiquitous nature of this relationship no receptor for SL has been identified in AM fungi, let alone fungi in general. Recent work in the lab has identified a strigolactone receptor in *Saccharomyces cerevisiae*, the high-affinity phosphate transporter Pho84. Using molecular docking we were able to identify a potential binding pocket for SLs on Pho84. Using the known binding position of SL on ShHTL7, the SL receptor with the highest affinity from *S. hermonthica*, and the predicted binding pocket of SL on Pho84 we performed molecular dynamics simulations on both receptors with SL. Simulations indicated SL caused a significant decrease in stability of ShHTL7, possibly promoting its interaction with its signalling partners. Meanwhile SL seemed to slightly impede the function of Pho84, preventing conformational changes needed to transport phosphate. Together these results demonstrate the effect of SLs across the rhizosphere are very diverse based on the perceiving organism.

A3-25 - CRY2-SMAX1 Crosstalk Illuminates a New Layer of Germination Control in Arabidopsis

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Karrikins (KARs) are smoke-derived compounds that promote seed germination by activating the α/β -hydrolase receptor KAI2 and the F-box protein MAX2, which together target the repressor SMAX1 for degradation. This mechanism allows seeds to bypass traditional light-dependent germination cues, offering an adaptive advantage in post-fire environments. Although the core components of KAR signalling are well characterised, the environmental regulation of SMAX1 remains poorly understood. My research investigates the role of SMAX1 as a key repressor of germination in the dark. I discovered that SMAX1 accumulates in the dark and is destabilised upon light exposure, suggesting that light functions as an additional regulatory input independent of KAR signalling. To probe this further, I am investigating the role of the blue light photoreceptor CRY2, which, like *smx1*, promotes dark germination when mutated. While red light signalling and its influence on germination are well established, the role of blue light in regulating SMAX1 stability and KAR-induced germination remain largely unexplored. So far, I have identified a physical interaction between SMAX1 and CRY2 in both yeast and *in planta*. This suggests a potential point of integration between light perception and KAR signalling in plants. Given that CRY2 forms light-induced photobodies with its interaction partners, I am investigating whether SMAX1 localises to similar subcellular structures. To test this, I am using yeast as a heterologous system to express CRY2 and SMAX1, allowing me to monitor light-dependent photobody formation. Together, these studies aim to uncover a novel point of crosstalk between the blue light and KAR signalling pathways, providing a new insight into how plants integrate environmental signals to fine-tune germination responses

A3-26 - Identifying a novel plant hormone for the orphan receptor, AtHTL/KAI2.

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Germination of angiosperms is a highly regulated process coordinated by environmental and phytohormone cues. Light, gibberellins (GA), and abscisic acid (ABA) are regarded as the main regulators of germination. However, karrikins (KARs), a smoke-derived compound, promotes germination in Arabidopsis and is perceived by the Arabidopsis thaliana HYPOSENSITIVE TO LIGHT/KARRIKIN INSENSITIVE 2 (AtHTL/KAI2) receptor. A loss-of-function mutant in *HTL/KAI2* exhibits opposite phenotypes of KARs-treated seedlings indicating that Arabidopsis produces an unknown endogenous KAI2-ligand (KL). While the HTL/KAI2 signalling pathway is known to regulate germination and other developmental processes, the biosynthetic pathway of KL remains elusive. Since mutants in this pathway have not been isolated through genetic screening of embryonic mutations, we propose KL synthesis occurs in the maternal tissue, the seed coat. In my presentation, I will present a novel genetic screen to identify mutants in the maternal and embryonic genome. I will describe my characterized mutants which phenocopy loss-of-function *htl/kai2* mutants. These mutants should help identify KL and elucidate the biosynthetic pathway of this novel hormone.

A3-27 - Silver nitrate inhibits ABA2 xanthoxin dehydrogenase, an ABA biosynthetic enzyme

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Silver nitrate (AgNO₃) is applied in agriculture to control plant growth and development. These physiological effects are attributed to the ability of silver ions to modulate plant hormone pathways. In particular, application of AgNO₃ is known to inhibit ethylene signaling. Our present work demonstrates that ABA biosynthesis is also inhibited by application of AgNO₃. Silver nitrate was identified as an inhibitor of the biosynthetic enzyme ABA2 in a chemical screen. AgNO₃ application blocked stress-inducible ABA biosynthesis in a concentration dependent manner. This reduction in ABA accumulation was independent of the effect of silver ions on ethylene signaling. AgNO₃ treatment disrupted the oligomeric state of the enzyme possibly through modulation of redox status. ABA2 orthologs showed differential sensitivity to AgNO₃ correlating with the proportion of cysteine residues present in each enzyme. Overall, our work highlights that AgNO₃ alters plant growth by affecting multiple hormone pathways, including ABA biosynthesis in addition to ethylene signaling.

A3-28 - Small Molecule Uptake Through Fungal Transporters: New Perspectives on Symbiosis and Antifungal Action

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Most vascular plants establish mutualistic relationships with arbuscular mycorrhizal (AM) fungi, which enhance the plant's uptake of water and nutrients from the soil. Among the nutrients provided by AM fungi is phosphate (Pi), which is typically in forms the plant cannot access directly. A key signal used by plants to initiate this symbiosis is strigolactone (SL), which is secreted in response to Pi deficiency. We showed that SL acts as a signal that fungi detect by internalizing their high affinity Pi transporter, Pho84, from the plasma membrane, ultimately decreasing fungal Pi uptake. By characterizing this process, we identified genes involved in Pho84 internalization and trafficking through the endolysosomal system. We discovered that the fungicide, chlorothalonil, also acts as an inducer of Pho84 internalization, suggesting that a hidden pocket on the Pho84 surface can recognize other small molecules. This suggests that major facilitator superfamily (MFS) transporters like Pho84 may serve as conduits for small molecule entry into cells. To discover additional internalizers, we performed transcriptomic analysis on yeast treated with a different SL stereoisomer, KL. This revealed upregulation of genes involved in inositol biosynthesis, echoing the Pho84 response. We found that *Itr1*, the main inositol transporter upregulated during inositol starvation, also internalizes upon KL treatment. Furthermore, specific azole antifungals—such as tebuconazole and ketoconazole—internalized *Itr1*, while

others like clotrimazole did not. These findings suggest a selective mechanism by which specific antifungals enter yeast cells via transporter internalization. This transporter-mediated internalization of small molecules, including fungicides and plant hormones, offers a novel strategy for antifungal development and improving plant-fungal symbioses to support crop health and sustainability.

A3-29 - Regulation of jasmonate biosynthesis in pepper: Temporal expression of lipoxygenases in response to wounding

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¹McGill

Globally, pathogens and chewing insect pests are responsible for ~16% and ~18% of crop yield losses, respectively, and this is projected to increase with climate change. In plants, the jasmonate phytohormone signaling pathway is primarily responsible for resistance against these pathogens and chewing insect herbivores. In response to wounding, an enzyme (lipoxygenase; LOX) in the jasmonate pathway that is constitutively present, is altered through dephosphorylation to make it more active, which initiates high jasmonate production within the first few seconds leading to defense responses. This initial burst is followed by the gene expression of other LOXs that will sustain the levels of jasmonate and, therefore, resistance. We believe that the LOX involved in the initial jasmonate burst has the amino acid serine at the phosphosite position which allows it to be regulated by phosphorylation while LOXs involved in the sustained response have an alanine at this position which means the enzyme cannot be phosphorylated and must be regulated in another way. In pepper (*Capsicum annuum*), there are two genes encoding a LOX with a serine at the phosphosite (LOX2 and LOX8), and two genes encoding a LOX with an alanine at this position (LOX6 and LOX7). This study aims to investigate the differential transcriptional regulation of these genes over time in response to wound stress. This will be achieved through a time-course wounding experiment, followed by differential expression analysis of RNA-sequencing data. Given the importance of the jasmonate signaling pathway in plant resistance to agricultural pests, understanding its regulation is of key importance and may lead to novel strategies needed to enhance crop productivity to ensure food security.

CONCURRENT SESSION A4

A4-25 - Overcoming the Challenges for Editing the Highly Polyploid Sugarcane Genome

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Sugarcane provides 80% of the world's table sugar and 40% of the global biofuel. However, its highly polyploid genome ($2n = 100-120$; $x=10-12$) has made crop improvement via traditional breeding and genome editing challenging. When considering targeted mutagenesis, co-editing of all copies/alleles is often desired to obtain a loss of function phenotype. While RNA guided nucleases show great promise for this approach, protocol optimization is required to increase the efficiency of multiallelic co-editing. To determine the number of co-edited allelic variants, deep sequencing methods such as Illumina NGS are typically deployed. To simplify mutant characterization in highly polyploid sugarcane and reduce the cost we compared non-NGS based methods including capillary electrophoresis (CE), Cas9 RNP assays, and high-resolution melt analysis (HRMA).

CRISPR-mediated targeted mutagenesis was deployed for optimizing canopy architecture for improved light capture, which has great potential for elevating biomass yield. *LIGULELESS1* (*LG1*) is a gene involved in leaf ligule and auricle development. Events ranging in loss of function of 3 to all of the 40 copies/alleles of the *LG1* gene corresponded to tunable leaf-angle phenotypes that became more upright as co-editing frequency increased. The extent of co-editing in the different lines and the associated leaf angle phenotype remained constant in vegetative progenies. Data from randomized and replicated greenhouse and field trials will be presented.

A4-26 - A simple method to efficiently generate structural variation in plants

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Phenotypic variation is essential for the selection of new traits of interest. Structural variants, consisting of deletions, duplications, inversions, and translocations, have greater potential for phenotypic consequences than single nucleotide variants. Pan-genome studies have highlighted the importance of structural variation in the evolution and selection of novel traits. Here, we describe a simple method to induce structural variation in plants. We demonstrate that a short period of growth on the topoisomerase II inhibitor etoposide induces heritable structural variation and altered phenotypes in *Arabidopsis thaliana* at high frequency. Using long-read sequencing and genetic analyses, we identified deletions and inversions underlying semi-dominant and recessive phenotypes. This method requires minimal resources, is potentially applicable to any plant species, and can replace irradiation as a source of induced large-effect structural variation.

A4-27 - Optimization of Solid Silica Nanoparticles for Transformation of Protoplasts and Microspores in Canola (*Brassica napus*)

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Nanoparticles (NPs) are increasingly being explored as promising tools for plant genetic engineering, offering potential advantages over conventional methods such as *Agrobacterium*-mediated transformation or biolistic. Among them, silica nanoparticles (SiNPs) stand out for their biocompatibility, ease of synthesis, and surface functionalization potential.

This study focuses on optimizing solid SiNPs for the transformation of protoplasts and microspores *Brassica napus* cv. Westar. SiNPs (~15 nm) were characterized using dynamic light scattering and functionalized with polyethylene glycol (PEG) to enhance cellular uptake. Fluorescein isothiocyanate labeling enabled tracking of SiNP internalization. A plasmid carrying a DsRed reporter and a kanamycin resistance gene was used to assess transformation efficiency. Cell viability, growth, and regeneration were evaluated using fluorescein diacetate staining and resazurin viability assays. Fluorescence microscopy was employed to monitor SiNP internalization and localization.

We optimized key parameters including DNA and SiNP concentrations, DNA/NP ratios, and incubation time/temperature. An SiNP concentration of 80 ng/μL yielded optimal results. Gel retardation assays confirmed effective DNA:NP complexation at ratios of 1:4 (unmodified) and 1:5 (PEGylated). Transformation efficiency improved significantly with PEG-functionalized SiNPs, particularly in microspores, where regeneration rates were enhanced. Additionally, 10 μg of plasmid DNA successfully transformed 1×10^6 protoplasts per sample. Our results highlight the potential of PEGylated solid SiNPs as efficient and minimally invasive vectors for plant transformation. This platform could offer a valuable alternative for transforming recalcitrant species and contribute to the development of nanomaterial-assisted strategies in agricultural biotechnology.

A4-28 - CRISPR Makes Its Oat Debut: A breakthrough in oat genomics

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Oat (*Avena sativa* L.), a globally important cereal valued for its nutritional profile and gluten-free nature, has remained genetically challenging due to its large (12.5 Gb), complex, and highly repetitive genome. This has impeded the use of gene-editing tools in oat, in contrast to other cereals. We report the **first-ever successful**

CRISPR-Cas9-based gene-editing in oat, targeting three agronomically important genes: *Thaumatococcus*-like protein 8 (TLP8), *Vernalization 3* (VRN3), and *VRN3D*. We achieved efficient targeted mutations, including deletions and insertions, with gene-editing efficiencies of up to 50%. Mutations in *VRN3/VRN3D*, key regulators of flowering time and yield traits, revealed striking phenotypes. Triple *vrn3* knockout mutants (*aaccdd*) exhibited prolonged vegetative growth, significantly thicker stems, and increased cell proliferation, while *vrn3D*-edited lines showed impaired flag leaf development. This study not only demonstrates a robust, high-efficiency CRISPR-Cas9 platform for oat genome editing but also establishes a foundation for functional genomics and precision breeding in this emerging crop for healthy lives.

A4-29 - Functional Characterization of Thaumatin-like Protein 8 (TLP8) Using the CRISPR-Cas9 System

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Oat (*Avena sativa*) is an important industrial cereal in Canada, notable for its substantial (1-3, 1-4)- β -D-glucan (β -glucan) content, which is beneficial to human health. Thaumatin-like protein 8 (TLP8) has been shown to bind to β -glucan, and studies in barley have associated increased TLP8 expression with reduced β -glucan content. However, the precise role of TLP8 in β -glucan metabolism is not fully understood. We hypothesize that TLP8 may play a role in β -glucan turnover, thereby affecting β -glucan content and early seedling development. To investigate this, we employed CRISPR-Cas9 genome editing. Guide RNAs targeting *TLP8* were designed and delivered into oat tissues via particle bombardment. Out of 100 bombarded calli, 39 regenerated plants were obtained, with 21 confirmed as transgenic, resulting in a transformation efficiency of 21%. Next-generation sequencing revealed that 41% of the transgenic lines carried edits at the *TLP8* target site, including 1-bp, 4-bp, and 12-bp deletions near the PAM sequence. Lines harboring a 4-bp deletion in the *TLP8-C* allele were advanced to the next generation, and homozygous mutants were isolated. These knockout lines displayed altered early seedling development, with significantly shorter roots and shoots ($p < 0.001$), suggesting a role for TLP8 in plant development. These findings indicate that TLP8 may exhibit β -glucanase activity and contribute to cell wall remodeling and carbon mobilization. Ongoing studies with these homozygous CRISPR mutants aim to further elucidate the role of TLP8 in β -glucan biosynthesis and its broader impact on oat development and nutritional quality.

A4-30 - CRISPR-Cas9 editing of de novo DNA methyltransferase alters development and cold responses in *Brachypodium distachyon*

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DNA cytosine methylation (5mC) is a key epigenetic mechanism influencing gene regulation and genome stability in plants and mammals. Its patterns result from dynamic processes of *de novo* methylation, maintenance methylation, and demethylation. In plants, *de novo* 5mC establishment occurs via the RNA-directed DNA methylation (RdDM) pathway, where non-coding RNAs direct the DNA methyltransferase DOMAINS REARRANGED METHYLTRANSFERASE 2 (DRM2) to specific DNA sequences. This regulation appears crucial for development and stress responses, particularly in complex genomes like wheat and other cereal crops. However, genome-wide studies on these crops are complicated by their size, leading to most insights coming from the model dicot, *Arabidopsis thaliana*. To better understand 5mC regulation in cereals, we utilized the undomesticated model cereal, *Brachypodium distachyon*, and CRISPR-Cas9 to mutate *BdDRM2*, a homologue of the *de novo* DNA methyltransferase. We hypothesized *BdDRM2* mutation would affect development, stress response, and patterns of 5mC and gene expression. To assess development, we measured flowering time in our edited lines (*Bddrm2*) and found a significant delay compared to wild type. To evaluate impact of *BdDRM2* mutation on 5mC patterns, gene expression and stress response, we performed whole genome bisulfite sequencing and RNA sequencing on wild type and *Bddrm2* under control and 24-hour cold stress conditions. We found decreased global 5mC levels and identified numerous differentially methylated regions and expressed genes in both conditions. These findings support conserved DRM2 function in *B. distachyon*, linking it to developmental regulation and cold stress responses. Importantly, this work

identifies potential epigenetic targets for cereal crops, which may have been lost through domestication. These insights could be leveraged for crop breeding efforts to enhance resilience. As climate change intensifies environmental stressors, understanding the interplay between epigenetic and transcriptional changes will be crucial for developing cereal varieties that can adapt to shifting climates and ensure food security.

CONCURRENT SESSION B1

B1-31 - Harnessing Plant Evolutionary Diversity to Decode *Pseudomonas* Virulence

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Since the dawn of plant evolution > 500 million years ago, pathogenic microbes have impacted plant health and survival. While much of our knowledge is centred on evolutionarily young flowering plants, emerging research in non-flowering lineages has provided new insight into the origin and evolution of plant immunity and pathogen virulence. We recently employed a comparative evolutionary approach to understand how the bacterial pathogen *Pseudomonas syringae* infects distantly related plants. By screening a wide diversity of *P. syringae* pathovars, we identified isolates causing significant disease in the liverwort *Marchantia polymorpha*, the fern *Ceratopteris richardii*, and the flowering plant *Nicotiana benthamiana*. Broad host *P. syringae* belonging to the phylogroup 2 clade of the *Psy* species complex were particularly virulent in non-flowering plants, relying on a combination of type-III secreted effector proteins and the cyclic lipopeptide phytotoxin syringomycin. Importantly, ectopic toxin application was sufficient to activate host tissue necrosis and to promote bacterial growth of toxin-deficient *P. syringae* in liverwort thalli - revealing syringomycin as a key virulence factor for broad host virulence in plants. To further complement this approach, we isolated natural *Pseudomonas* pathogens of diseased liverworts in the wild. Sequencing and comparative genomics identified all pathogenic isolates as *P. viridiflava*, with subsequent experiments demonstrating that the conserved type-III effectors AvrE and HopM1 are critical for virulence in *Marchantia* liverworts as well as the model flowering plant *N. benthamiana*. Together, these works highlight that broad host range pathogens successfully manipulate distantly related land plants. Looking forward, our research aims to identify conserved and/or lineage-specific resistance mechanisms that protect liverworts from bacterial infection and transfer them into flowering plants to bolster crop health.

B1-32 - Swiss Needle Cast and the Foliar Mycobiome of Coastal Douglas-fir (*Pseudotsuga menziesii*)

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The causal agent of Swiss Needle Cast (SNC) in Douglas-fir, *Nothophaeocryptopus gaeumannii*, begins its life cycle as a benign foliar endophyte, causing no immediate visual harm. Approximately one year post-infection, pseudothecia form within leaf stomata, blocking gas exchange. This occlusion leads to limited respiration and premature leaf senescence, with low endemic conditions resulting in a minor decrease in needle lifespan (three to four years) compared to four to five years for healthy trees. However, during outbreaks, trees may lose all but the current year's needles, significantly impacting growth and productivity.

Since the mid-1990s, SNC outbreaks have surged in Oregon and have spread northward, posing a heightened risk to coastal British Columbia's Douglas-fir forests due to favorable climate factors, such as mild winters and moist springs. Our study assessed variations in disease severity within a British Columbia breeding population, quantifying tolerance and resistance across families. We found moderate heritability for stomatal occlusion and low heritability for needle loss, suggesting additional influencing factors.

To explore these factors, we evaluated the endomycobiome—the fungal endophyte communities—associated with SNC. Utilizing culture-based techniques, we identified over 30 fungal species from 19 families in more than fifty individual trees across twelve families. ITS amplicon meta-barcoding via Illumina sequencing revealed 22 to 88 fungal species per sample, with *N. gaeumannii* being the most abundant. Only nine taxa represented over 1% of OTUs, indicating a predominance of rare species. Significant community composition differences

were observed among families, and certain fungal species showed a negative correlation with *N. gaeumannii*. These findings suggest that fungal endophyte communities may modulate SNC severity.

B1-33 - Shooting the Messenger: The Importance of Small RNAs in Plant Immunity

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Understanding the fundamental workings of plant immunity is important for improving agricultural efficiency and reducing food insecurity around the world. Plants employ a two-tiered immune response. Pattern-triggered immunity (PTI) is the first line of defense in plant immunity, and leads to a relatively mild immune response. Effector-triggered immunity (ETI) is a second and more severe immune response and results in localized cell death. Previously, these pathways were thought to be independent from one another, but recent research has shown that PTI and ETI are interlinked and can mutually reinforce each other. However, the mechanisms behind this mutual reinforcement remain unclear. Previous research has shown the importance of miRNAs in plant immunity, which modulate degradation and translation of mRNAs based on sequence complementarity. We investigated if post-transcriptional gene silencing by miRNAs could serve as an avenue of crosstalk between the two immune pathways. miR825-5p is known to suppress intracellular receptors involved in ETI, helping to prevent activation of immunity in the absence of elicitors. We demonstrate using RNAseq analyses that loss of PTI sensitivity results in altered miRNA expression, including substantial downregulation of miR825-5p, suggesting that disruption of PTI signaling leads to upregulation of ETI receptors. These results further implicate miRNAs as an avenue of crosstalk between the two layers of plant immunity. Further research will investigate the role of immune-correlated miRNAs by assessing how efficiently they reduce accumulation of their targets. Understanding the role miRNAs play in plant immunity will allow us to further elucidate how plants respond to pathogens and to understand how the two layers of plant immunity interact to mount an effective anti-pathogen response.

B1-34 - A clubroot pathogen pseudokinase induces tolerance to the disease in Arabidopsis and canola

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Plasmodiophora brassicae, the causative agent of clubroot disease, significantly impacts *Brassica* crops by inducing gall formation on roots, leading to substantial yield losses. Despite its agricultural importance, the molecular mechanisms underlying *P. brassicae* pathogenicity remain poorly understood. In this study, we identified and characterized a *P. brassicae* effector, PbPK1, which plays a crucial role in modulating host plant development. PbPK1, a pseudokinase with a non-canonical active site, is highly expressed during the secondary infection stage of *P. brassicae*. Overexpression of *PbPK1* in *Arabidopsis thaliana* resulted in delayed flowering and enhanced disease tolerance, with infected plants reaching maturity, unlike controls. Transcriptomic analyses of *PbPK1* overexpression lines demonstrated differential expression of genes associated with hypoxia response and immunity, suggesting that PbPK1 manipulates the host response to disease development to extend the plant's lifespan, thereby facilitating the completion of the pathogen's life cycle. Notably, after phenotyping five of the more than 40 *Brassica napus* (canola) lines overexpressing *PbPK1* generated in the lab, we found that these exhibited phenotypes similar to those observed in *Arabidopsis*. Protein interaction assays, including pull-down/mass spectrometry, yeast two-hybrid, and BiFC, are underway to identify the PbPK1 target in the host and determine the mechanism induced by this effector.

B1-35 - Pinpointing candidate genes in quantitative trait loci (QTL) associated with resistance to bacterial brown spot (BBS) by exploiting the molecular interactions between adzuki bean (*Vigna angularis*) and *Pseudomonas syringae* pv. *syringae*.

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Adzuki bean was dubbed “the Ontario money-maker crop of 2023”. Bacterial brown spot caused by *Pseudomonas syringae* pv. *syringae* van Hall, can cause up to 40 % yield loss in the adzuki bean crop. Genetic resistance is the most effective measure in managing this disease, however, there are no resistant commercial adzuki varieties. Elucidating the genetic basis for resistance to bacterial brown spot will accelerate the breeding of commercial resistant cultivars. QTL regions are often large, comprising 100s of candidate genes and it can be challenging to pinpoint the specific gene(s) responsible for the trait of interest. In this study, a QTL of approximately 6 Mbp associated with BBS resistance was identified on a linkage map constructed from 2,911 filtered SNPs from 105 recombinant inbred lines from a cross between Erimo and a Chinese variety (CV). One hundred and eighty candidate genes were identified within the QTL. Comparative genomics and pathogenicity screening on Erimo and CV were carried out using *P. syringae* pv. *syringae* (Pss27, obtained from AAFC Harrow) *P. syringae* pv. *aceris* (Me2 and Me3, isolated in this study) and *P. syringae* pv. *dysoxyl* (Li6, isolated in this study). Erimo showed more resistance ($p < 0.0001$). Unique virulence factors (VF) were identified in these strains. Pss27 was the most aggressive strain in adzuki and possessed 10 unique VFs (avrRpm1, hopAB1, hopAF1, hopAP1, hopH1, hopL1, sypA, ahll and ahIR). Two of the unique VFs (hopAB1 and hopL1) had a total of seven cognate target genes within the QTL. Of all the target genes, only ethylene-responsive transcription factor (ERF024) showed polymorphisms between Erimo and CV. There was an indel in CV and two extra copies of ERF024 in tandem in Erimo. ERF024 was located 20 kbp from the QTL peak marker, S02_48822524, and also was the closest candidate gene peak marker.

B1-36 - Regulation and virulence function of *Fusarium graminearum* secondary metabolites during Arabidopsis seedling infection

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Fusarium graminearum is a fungal pathogen of cereals that reduces grain quality due to the deposition of secondary metabolites during infection. Secondary metabolite production is temporally regulated, with specific gene clusters involved in virulence factor production being induced during host colonization in response to unknown plant host factors. Previous research identified the Arabidopsis gene *RbohD* as promoting production of the DON mycotoxin and promoting susceptibility to the pathogen, however its effect on secondary metabolite production is unclear. We used transcriptomic profiling to identify 712 fungal genes whose expression is dependent on *RbohD*, including biosynthetic gene clusters associated with the production of butenolide, fusaoctaxin A, fungal decalin-containing diterpenoid pyrones (FDDP), and DON. To determine if the *RbohD*-dependent mycotoxins contribute to the virulence of *F. graminearum*, we infected Arabidopsis with *Fusarium* mutant strains lacking production of butenolide, fusaoctaxin A, FDDP and DON. Preliminary results reveal that mutants lacking in fusaoctaxin A, FDDP, and DON production exhibit significantly reduced virulence compared to the wild-type strain indicating these secondary metabolites could be virulence factors during *F. graminearum* infection in Arabidopsis. Together, this suggests that host-derived factors promote virulence of the pathogen by activating secondary metabolite production.

CONCURRENT SESSION B2

B2-33 - Shrub ecophysiological responses to metal stress in degraded peatlands

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Peatlands in the Sudbury region of Ontario, Canada, have experienced severe ecological degradation due to over a century of metal and sulfur pollution from smelting activities. While recent restoration efforts aim to re-establish ecosystem function, the recovery potential of dominant shrub species under persistent copper and nickel metal stress remains uncertain. To address this, we integrate data from a suite of studies examining plant ecophysiological responses across a well-characterized metal contamination gradient. These include field surveys, manipulative field experiments, and controlled plant growth experiments. Across these efforts, we measured leaf-level gas exchange and key functional traits (specific leaf area, leaf dry matter content, elemental composition) for dominant ericaceous shrubs (e.g., *Chamaedaphne calyculata*, *Kalmia angustifolia*, *Rhododendron groenlandicum*). Preliminary results reveal reduced stomatal conductance and altered trait expression at high-contamination sites, suggesting stress-induced limitations on carbon assimilation and conservative leaf economic strategies. Our findings help clarify species-specific tolerance thresholds and inform restoration strategies that account for physiological constraints under long-term metal stress. This work highlights the value of combining field and experimental approaches to assess plant function in contaminated environments.

B2-34 - Plant-derived methane: What do we know about it?

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Anthropogenic release of carbon dioxide, methane, nitrous oxide and other greenhouse gases into the atmosphere contributes to the enhanced greenhouse effect. Methane is a potent greenhouse gas, contributing to global warming. Its current atmospheric concentration is over 1900 parts per billion. Methane has both natural and anthropogenic sources and can be generated under both anaerobic and aerobic conditions. It has been shown that all living organisms can produce methane. For the first time, aerobically-produced methane in plants was reported in 2006. Since then, many researchers have tried to determine the precursors of methane in plants and the factors that influence its emissions. As suggested, plant-derived methane may originate from pectin, lignin, epicuticular wax, dimethyl sulfoxide, methionine and other amino acids. Methane emissions from plants can be affected by different environmental stressors. Since its discovery, we were also interested in exploring the source of plant-derived methane and the factors that lead to increased or decreased methane emissions. We used multiple plant species and environmental factors to investigate this phenomenon. We have shown that pectin, lignin, wax and amino acids can serve as precursors of plant-derived methane, which can be increased by higher temperatures, enhanced ultraviolet-B radiation, water deficits, higher blue light, lower red/far-red light ratio, and higher ethylene level, but decreased by elevated carbon dioxide. We also demonstrated that methane emission was higher in stems than in leaves and roots as well as in the upper parts of the plants than in the middle and lower parts. Methane emissions decreased with plant age and varied among plant species and varieties. Recently, it was suggested that all living organisms may have a common mechanism of methane synthesis through the interaction of reactive oxygen species, iron and methyl donors. However, more studies are required to understand the mechanism of methane production in plants.

B2-35 - Warmer Autumns Delay the Development of Photoprotective Sustained Non-Photochemical Quenching (NPQ) in White Spruce

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Climate change, with shifts like rising autumn temperatures and unchanged photoperiods, may disrupt cold hardiness and low-temperature acclimation in conifers. This disruption could affect autumn photosynthesis downregulation, extending its influence into winter. In conifers, low-temperature acclimation and the downregulation of photosynthesis involve shifts in two components of photoprotective non-photochemical quenching (NPQ): a dynamic component that diminishes by the end of the growing season, and a sustained component that becomes more pronounced. Our study explores the effect of autumn warming on the development of sustained NPQ in white spruce (*Picea glauca*) using a Temperature Free-Air Controlled Enhancement (T-FACE) system in a field experiment, raising leaf temperatures by 5°C above ambient in heated plots. Warming delayed the downregulation of photosystem II efficiency and dynamic NPQ, while postponing the onset of sustained NPQ. This was evidenced by transient drops in maximum quantum efficiency of photosystem II (F_v/F_m), retention of xanthophyll cycle pigments including zeaxanthin, which dissipates excess energy safely as heat. Immunoblot analysis revealed that unheated seedlings showed earlier reductions in key thylakoid proteins: D1 (a core PSII reaction center protein), PsaD (part of the PSI reaction center), and PsbS (linked to energy-dependent quenching). These changes were consistent with a more rapid transition to sustained NPQ. In contrast, heated seedlings retained these proteins longer, indicating delayed restructuring of the photosynthetic apparatus in response to cooling temperatures. Our findings demonstrate that autumn warming delays the seasonal development of sustained NPQ and the associated downregulation of photosynthetic machinery. This disruption suggests that warm autumn conditions may interfere with the cold hardening process in conifers by slowing the protective adjustments typically triggered by low temperatures, potentially compromising their readiness for winter stress.

B2-36 - Intraspecific Variation in *Populus balsamifera* Indicates Divergent Drought and Elevated [CO₂] Responses Among Ecotypes

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Climate change, driven by rising atmospheric carbon dioxide concentrations ([CO₂]) and increasing drought frequency, is altering plant physiological function and the forest ecosystem. Understanding intraspecific variation in drought responses, particularly under elevated [CO₂], is critical for improving predictions of forest ecosystem responses and management practices that enhance resilience to future climate conditions.

We investigated the morpho-physiological responses of three *Populus balsamifera* ecotype seedlings (Thunder Bay (TB), Peace River (PR), and Zama City (ZM)), originating from regions in Canada with contrasting precipitation regimes, to factorial treatments of ambient and elevated [CO₂] (400 vs. 1000 $\mu\text{mol mol}^{-1}$) under well-watered and moderate drought conditions.

Significant ecotypic variation was observed in gas exchange, growth, and biomass allocation traits. Although all ecotypes reduced stomatal conductance (g_s) and transpiration rate (E) under drought, PR and ZM, ecotypes from drier regions, consistently exhibited higher g_s and E across treatments than TB, the ecotype from the wettest origin. The intermediate-origin PR exhibited the greatest increase in maximum electron transport rate (J_{max}) and net photosynthesis (P_n) under elevated [CO₂], along with tall growth and moderate root investment, indicating a balanced strategy supporting both productivity and drought tolerance. ZM, in contrast, displayed drought-adaptive morphological traits such as reduced height, lower total biomass, and increased root-to-shoot ratio, contributing to enhanced P_n under elevated [CO₂]. TB exhibited a more conservative physiological strategy, with low g_s , E , intercellular to ambient CO₂ concentration ratio, and J_{max} , resulting in limited P_n response to elevated [CO₂]. Morphological traits showed stronger ecotypic differentiation than physiological traits, with ZM's drought adaptation reflected primarily in biomass allocation.

These findings demonstrate that *P. balsamifera* ecotypes express distinct trait combinations shaped by their source origin, influencing their performance under interacting [CO₂] and moisture regimes.

B2-37 - Improving heat stress tolerance in canola (*Brassica napus* L.) by regulating diacylglycerol acyltransferase activity

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Canola (*Brassica napus* L.) is a globally important oilseed crop and one of the principal cash crops cultivated in Canada, contributing approximately \$43.7 billion annually. However, as a cool-season crop, canola is particularly susceptible to abiotic stresses such as heat, which can reduce yield and compromise its economic value by lowering seed oil content and quality. Triacylglycerol (TAG), the main compound of storage oil, is synthesized primarily through the Kennedy pathway in seeds, in which diacylglycerol acyltransferase (DGAT) catalyzes the final and committed step. Previous studies have shown that *DGAT* expression responds to various abiotic stresses, and the *DGAT1* mutant exhibit increased sensitivity under stress conditions. In our previous work, we generated *B. napus* *DGAT1* (BnDGAT1) variants through directed evolution, some of which exhibited enhanced enzyme activity and promoted TAG accumulation in yeast and tobacco leaves. To evaluate its potential function in plant heat stress response, we introduced both native and high-performance BnDGAT1 variants into *B. napus* cv. DH12075 and the transgenic lines were subjected to both long- and short-term heat stress treatments to assess their performance. Preliminary results showed that one transgenic line accumulated 15.9% more seed oil under short-term heat stress than the wild type, and its oil content was similar to that of the wild type under normal conditions. These findings highlight the potential of enhancing stress tolerance and maintaining seed oil content in canola through regulation of *DGAT1* activity under heat stress.

B2-38 - Microtopography and soil moisture gradients drive variation in turgor loss point, xylem conductivity, and growth in white spruce seedlings

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Although drought impacts on tree physiology are well studied, the effects of excess soil moisture and waterlogging remain less understood especially in conifers. Prolonged soil saturation can induce root hypoxia, limiting water transport, reducing growth, and compromising drought resilience. Yet, the extent to which microtopography-driven environmental heterogeneity influences intraspecific variation in hydraulic traits remains poorly characterized.

We assessed how microsite conditions affect the growth and physiology of white spruce (*Picea glauca*) seedlings in a field experiment involving 42 individuals from 14 genotypes planted across a natural topographic gradient. Daily soil moisture was monitored using 13 sensors throughout the 2023 growing season. For each seedling, we measured tree height, turgor loss point (Ψ_{TLP}) and xylem traits including tracheid diameter and specific hydraulic conductivity (K_h). We hypothesized that seedlings growing in chronically wet, low-elevation microsites would exhibit reduced height, less negative Ψ_{TLP} and lower hydraulic efficiency compared to those in drier upland zones. We also tested for genotype-by-environment interactions, expecting differential trait responses depending on soil moisture regimes.

Our findings underscore the role of microtopographic soil moisture gradients in shaping physiological trait expression and growth performance. These insights are critical for informing reforestation and assisted migration strategies under variable moisture regimes. Our study highlights the importance of accounting for fine-scale environmental variation and genotypic sensitivity in climate-resilient forest management.

CONCURRENT SESSION B3

B3-39 - Phenotypic Variation in a Batch Culture of *Chlamydomonas reinhardtii* with Different Light Tolerances

Gaganpreet Kaur Gill¹, Dion Durnford¹

¹UNB

Phenotypic variation is expected in natural microbial populations of genetically diverse populations. However, with an isogenic strain of *Chlamydomonas reinhardtii*, you would predict this variation to be minimal within well mixed, batched cultures. However, using Percoll density gradients, we were able to isolated two distinct populations (Top, Bottom) of *Chlamydomonas reinhardtii* from two-day old batch cultures based on buoyant density. The least dense cells were at the top of the gradient and made up 86% of the culture, while the denser cells collected at the bottom of the gradient and were a smaller proportion of the population (14%). The cells in the Top fraction were, on average, larger with more protein per cell, had greater short-term growth rate, and an elevated rate of respiration. The top population also had a greater chlorophyll content, greater photosynthetic maxima, and better light use efficiency. Nevertheless, the bottom population had a greater capacity for dissipating excess light energy, was more tolerant of high-light stress, and had a greater resistance to accumulation of singlet oxygen, suggesting there was activation of light-stress pathways in this population. We propose these population variants were part of a stochastic mechanism that generated phenotypic variation in the population. This may be part of a “bet-hedging” strategy, where distinct gene expression profiles in a population gives a selective advantage to a portion of the population that would optimize survival chances under abrupt changes environmental conditions, such as light intensity.

B3-40 - Do Ginsenosides Alter the Pathogenicity of *Ilyonectria*?

Anka Colo¹, Mark Bernards¹

¹Western University

American ginseng, *Panax quinquefolius* L., is an economically valuable crop used in Traditional Chinese Medicine; however, yield of ginseng is negatively impacted by ginseng replant disease (GRD). GRD is characterized by a severe root rot, primarily caused by the fungus *Ilyonectria mors-panacis* (*Imp*) (formerly *Cylindrocarpon destructans*), in ginseng planted in a former-ginseng garden. While *Imp* is typically present in ginseng garden soils during the first cultivation of a ginseng crop, *Imp*-associated root rot is more extensive during subsequent crop plantings. Furthermore, the bioactive ginsenoside compounds produced by ginseng accumulate in ginseng garden soils during cultivation and are fungitoxic toward some soil-borne fungi, while growth stimulants of others, including *Imp*. Presently, it is not clear if (1) prior exposure to ginsenosides enhances *Ilyonectria* virulence, (2) different *Ilyonectria* isolates can metabolize ginsenosides equally effectively, and (3) there is a relationship between *Ilyonectria* virulence and the metabolism of ginsenosides. Twelve *Ilyonectria* isolates, differing in geographic and host origins, were screened for pathogenicity on one-year-old ginseng seedlings. Disease progression was monitored non-invasively for 28-days using chlorophyll fluorescence imaging, with final disease severity scores indicating five low-virulence and seven high-virulence isolates. To test the effect of ginsenoside exposure, isolates from both virulence groups that had been cultivated on media with and without ginsenosides were used to inoculate two-year-old ginseng roots and lesion sizes were measured after 16-days. Two low-virulence isolates exhibited significantly increased virulence following ginsenoside exposure, whereas high-virulence isolates remained unaffected. Finally, to determine whether *Ilyonectria* isolates can metabolize ginsenosides equally effectively, *Ilyonectria* isolates were grown in liquid minimal media supplemented with ginsenosides. After 7-days, ginsenosides were quantified using liquid chromatography-mass spectrometry (LC-MS). No correlation was present between virulence and the ability to metabolize ginsenosides. Understanding whether exposure to ginsenosides enhances virulence will further our understanding of *Ilyonectria* and its implications in GRD.

B3-41 - Role of Reactive Oxygen Species in the Acetate-dependent aging of *Chlamydomonas reinhardtii*.

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¹University of New Brunswick, Fredericton

Aging mechanisms are well-studied in multicellular organisms, but the factors regulating aging and longevity in microalgae remain largely underexplored. The unicellular alga *Chlamydomonas reinhardtii* serves as an ideal model for aging studies due to its nutritional flexibility and ease of culture. Under favorable conditions, *Chlamydomonas* can achieve immortality, but nutrient limitations induce conditional senescence. We have shown that higher acetate concentrations accelerated senescence and reduce longevity of *Chlamydomonas* in batch culture. This study investigated the role of reactive oxygen species (ROS) in acetate-dependent aging. We hypothesized that elevated acetate enhances metabolic activity and ROS production, causing cellular damage and reducing lifespan. The ROS hydrogen peroxide levels were measured using DCFH-DA fluorescence and were significantly higher in high-acetate cultures, correlating with reduced longevity. Treatment with ROS quenchers like catalase and 2,2-dipyridyl partially restored lifespan under high-acetate conditions, further supporting the role of ROS in calorie-dependent aging. Interestingly, cells in high-acetate conditions exhibited increased resistance to external ROS-inducing agents such as Rose Bengal, hydrogen peroxide, and methyl viologen, suggesting the induction of antioxidant defense pathways. Transcriptomic analysis also shows the upregulation of several antioxidant related genes in high acetate growing cells, showing the activation of ROS detoxification pathways. These findings suggest that ROS play a central role in the regulation of lifespan under high-calorie conditions, providing insights into conserved mechanisms of aging across biological systems.

B3-42 - Linking Metabolomic Variation to Larval Performance and Breeding Potential in Conifer Resistance

Mingming Cui¹

¹Laval University

Invasive insect pests like *Lymantria dispar asiatica* pose a growing threat to forest ecosystems. Understanding host susceptibility is critical for resistance-based management. We investigated the metabolomic and genetic basis of host defense in ten North American conifer species through larval performance assays and metabolite profiling. Feeding trials with two *L. dispar asiatica* strains revealed significant variation in larval weight gain, development, and survival across host species. *Pseudotsuga menziesii* (Douglas-fir) supported higher larval growth, while Norway spruce *Picea abies* consistently suppressed development. Using targeted and untargeted metabolomics, we identified 34 terpenoids and 108 phenolic compounds in host foliage. Phenolic profiles differentiated host species more clearly, while terpenoid composition correlated more strongly with larval physiology and performance. Further analysis of adult Douglas-fir from a breeding program revealed high foliar terpene variability and moderate to high heritability ($h^2 > 0.5$) for key compounds, such as aromadendrene and α -pinene. Positive genetic correlations among these terpenes suggest the feasibility of indirect selection strategies. Our results highlight the value of integrating larval bioassays, chemical ecology, and quantitative genetics to assess host resistance. The observed metabolite-larval performance link, combined with heritable variation in defensive compounds, provides a framework for developing resistant tree genotypes through targeted breeding.

B3-43 - Antioxidant Enzyme Responses and Gene Regulation in Early Interaction between *Brassica napus*-*Verticillium longisporum*

Ayomi Thilakarathne¹, Fei Liu², Zhongwei Zou¹

¹Wilfrid Laurier University, ²Henan University

Verticillium stripe disease, caused by the soil-borne fungal pathogen *Verticillium longisporum*, is a major concern that significantly reduces canola (*Brassica napus*) productivity. Current control measures are often costly, inconsistent, and further limited by the lack of well-characterized resistance in the host plant, making

effective management difficult. This study investigates the impact of *V. longisporum* on the activity of major antioxidant enzymes and changes in gene expression upon the pathogen progression at the young seedling stage. Phenotypic assessments revealed that susceptible Westar seedlings displayed partial cotyledon necrosis, while Quinta seedlings exhibited complete cotyledon death. However, most Westar plants were infected at the adult stage and exhibited severe symptoms. Interestingly, a higher amount of fungal DNA was detected in Quinta roots and hypocotyls at 7 and 14 days post-inoculation (dpi), while lower amounts were detected in Westar. Superoxide dismutase activity increased in leaves at 7 dpi and declined thereafter, whereas catalase activity showed the opposite trend. Glutathione reductase activity was consistently higher in Westar than in Quinta in both roots and leaves, while Glutathione-S transferase activity was comparatively higher in roots and leaves at 14 dpi in both *B. napus* lines. Notably, no glutathione peroxidase activity was detected in leaves. Differential gene expression analysis revealed six upregulated genes in the roots and leaves of Quinta at 7 and 14 dpi, while no upregulated genes were detected in Westar at the same time points. These findings provide valuable insights into pathogen progression, the role of antioxidant enzymes, and genotype-specific transcriptional responses during infection. This knowledge will aid in developing genetically improved *B. napus* lines with enhanced resistance and contribute to creating more effective and targeted disease management strategies.

B3-44 - Molecular Insights into Herbicide Resistance in Wild Oat: Toward Early Detection and Better Management

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¹Canadian Food Inspection Agency, ²Carleton University, ³Algonquin College, ⁴Agriculture and Agri-Food Canada

Herbicide resistance in wild oat (*Avena fatua*) poses a significant challenge to agriculture in the prairie provinces of Canada, especially cereal crops like wheat and barley. Annual economic losses attributed to wild oats are estimated to reach as high as \$500 million, representing the combined impact across all affected agricultural sectors in these regions. Currently, resistance is diagnosed through laboratory testing of farmer-submitted seeds, where dose-response curves are generated to determine resistance levels. This process can take up to six months, limiting the ability to implement timely in-season management. Our current research aims to develop molecular diagnostic tools for early detection of herbicide resistance in wild oat populations. Preliminary findings from dose-response experiments revealed that 10 wild oat populations, collected from Alberta, Saskatchewan, and Manitoba, exhibited different levels of resistance to various herbicides, including both group 1 and group 2 herbicides, which target the acetyl-coenzyme A carboxylase (ACCase) and the acetolactate synthase (ALS) genes. Dose-response data indicated that specific populations showed resistance primarily to group 1 herbicides, with notable resistance observed in populations exhibiting known mutations at positions 2088 (Cys to Arg) and 2078 (Asp to Gly) in the ACCase gene. Nanopore sequencing confirmed these mutations and provided allele-specific insights, revealing the allelic frequency of these mutations. The hexaploid nature of wild oats complicates herbicide resistance, emphasizing the necessity for advanced molecular diagnostics to manage this issue effectively. We are currently extending this analysis pipeline to examine resistance to group 2 herbicides to mutational analysis of the same populations at the ALS gene.

CONCURRENT SESSION B4

B4-45 - Single-cell genomics highlights the molecular impacts of *Indeterminate1* on *Zea mays* vasculature development and the floral transition

Mark Allan Alexander Minow¹, Zuhua Ali², Ankush Sangra¹, Lewis Lukens³, Joseph Colasanti², Robert J. Schmitz¹

¹University of Georgia, ²Summerlee Science Complex, University of Guelph, Molecular and Cellular Biology, ³Crop Science Building, University of Guelph, Plant Agriculture

Zea mays (maize) flowering is a critical determinant of yield, so there is value in understanding the genetics controlling the reproductive transition. *Indeterminate1* (*Id1*), a zinc-finger transcription factor, is a master

regulator of maize flowering. *Id1* is expressed in immature, developing leaves and is thought to regulate mature leaf production of a florigen, a mobile signal that moves to the shoot apical meristem to initiate flowering. However, the molecular mechanisms through which *Id1* regulates the maize floral transition remain poorly understood. To better characterize *Id1* actions, we performed single-cell assay for transposase accessible chromatin and single nucleus RNA sequencing (scATAC-seq and snRNA-seq) comparing *Id1*⁺ and *Id1*⁻ developing maize leaves. *Id1* expression was enriched in all cell-types of the developing vasculature, except for the xylem-associated vasculature parenchyma. We revealed *Id1*⁻ impacted chromatin accessibility and expression in many cell-types of the developing leaf, with pronounced effects in the phloem and vasculature parenchyma. Exploiting known ID1 DNA binding motifs, we identified regions of the genome with chromatin accessibility or expression changes consistent with direct ID1-binding. The ID1 binding motif was more prevalent near differentially expressed genes than near regions with altered chromatin accessibility, suggesting *id1*⁻ chromatin changes predominantly reflect secondary, indirect effects. Moreover, ZmDhr β -glucosidases, which were highlighted via previous bulk expression profiling, were amongst the expression changes linked to putative direct ID1 actions. These β -glucosidases also had vasculature enriched expression, again highlighting possible vasculature effects. Investigating putative targets further, we generated CRISPR/CAS9 edits for these β -glucosidases. Five independent edits of one β -glucosidase produced dominant pleiotropic phenotypes, highlighting their importance in many aspects of plant development. Our findings provide the cellular context for how *Id1* influences flowering and begins the molecular characterization of *Id1* downstream targets.

B4-46 - Effects of Ascorbic Acid Seed Priming on Germination Performance of Seven Broccoli Cultivars

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¹Dalhousie University

Seed priming with ascorbic acid (AsA) has emerged as a promising strategy to improve seed germination and early seedling growth. This study evaluated the effects of AsA seed priming on the germination of seven broccoli (*Brassica oleracea* var. *italica*) cultivars: Belstar, Covina, Patron, Luna, Green Magic, Imperial, and Gypsy. Seeds were preconditioned with AsA solutions at 0, 1, 5, 10, 20, and 40 mg/L for 24 hours and compared to unprimed control. The experiment followed a completely randomized design with five replications per treatment. AsA treatments significantly ($p < 0.05$) influenced most germination traits. The 10 mg/L treatment yielded the highest Fresh Weight (FW), Dry Weight (DW), Seedling Length (SL), Germination Index (GI), Vitality Index (VI), Seed Vigor Index (SVI) 1 and SVI 2 with average increases ranging from 45-215%, 75-166%, 53-186%, 9-47%, 62-350%, 65-214%, and 66-135%, respectively, across most cultivars when compared to the control. Mean Germination Time (MGT) was significantly affected ($p < 0.05$) in all cultivars except Luna and Patron. Germination Vigor (GV) and Final Germination Rate (FGR) were significantly affected ($p < 0.05$) in across treatments in all cultivars except Green Magic, Imperial, and Patron, with FGR showing no significance in Gypsy. While cultivar-specific responses varied slightly, AsA preconditioning consistently enhanced germination traits with statistically significant improvements. These results support the potential of AsA priming to enhance seedling performance in broccoli.

B4-47 - The Yellow Submarine: Discovering Suberization in the Silique and Its Functional Role Enabling Endocarp a Degradation

Justin Nichol¹, Marcus Samuel¹

¹University of Calgary

During reproduction in angiosperms, the precise, synchronized molecular dialogue established between the maturing seed and the encasing fruit is crucial for successful fruit/seed maturation and the ensuing fruit dehiscence and seed dispersal. Despite our understanding of the seed maturation program in plants, knowledge of how this synchrony is maintained is still lacking. In siliques of Brassicaceae, secondary cell wall deposition and degradation of cell wall material in selected layers of the valve are central for dehiscence to occur. Investigation into lignification of these layers revealed a previously unidentified suberin lamellae-like deposition within lignified cells of the end layer. Experiments using RNA sequencing and transcriptional fusions confirmed expression of known suberin biosynthetic genes in silique tissue; GC-MS analysis further verified the

presence of suberin monomers. Using a combination of lignin and suberin-defective (*nst1/3*) and suberin-defective (*cor413im1*) mutants, I have been able to show that ligno-suberization occurs in the *ena* L3-cells (first three cells of *ena* connected to replum) results in blocking the hydraulic conductivity to the rest of the *ena* cells, causing this layer to collapse, allowing coating of the inside of the valve chamber with the polarized waxy cuticular material from the collapsed *ena* layer. I propose that these layers form a three-pronged barrier—comprising lignin, suberin and cutin, —that protects the locule from moisture loss, pathogens, and desiccation.

B4-48 - Free fatty acid biosynthesis precursors are involved in pollen-stigma interactions in Brassica

Kumar Abhinandan¹, Hongtao Qin², Marcus Samuel¹, Xingguo Lan²

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Self-incompatibility (SI) is a complex molecular mechanism in flowering plants that prevents self-fertilization and promotes outcrossing. We conducted metabolome analysis of ornamental kale (*Brassica oleracea* var. *acephala*) pistils following SI and compatible pollination (CP), revealing significant alterations in lipid metabolism, particularly the accumulation of free fatty acid (FFA) metabolites during CP. Treatment of stigmas with acetyl-CoA and malonyl-CoA, key precursors in fatty acid synthesis, broke down SI and enhanced CP. Conversely, inhibiting acetyl-CoA carboxylase (ACCase), the rate-limiting enzyme in *de novo* fatty acid synthesis, significantly reduced compatible pollen attachment and tube growth, highlighting the critical role of fatty acid metabolism in mediating pollination success. We identified a novel interaction between the FERONIA (BoFER) receptor kinase and the biotin carboxyl carrier protein 1 (BoBCCP1), a subunit of the ACCase complex. Suppressing the expression of *BoBCCP1* in the stigma reduced CP response, suggesting that the FER-BCCP1 module may play a crucial role in regulating fatty acid biosynthesis and determining the outcome of pollen-stigma interactions. Our findings provide new insights into the identification of key metabolic pathways and signaling modules controlling pollen-stigma interactions, and offer a valuable resource for the targeted improvement of *Brassica* crop breeding.

B4-49 - Investigating the Receptor-Like Kinase, RKF1, and its Interactors in Arabidopsis thaliana Pollen-Pistil Interactions.

Stephen Bordeleau¹, Daphne Goring¹

¹University of Toronto

Plant reproduction is a complex, multistep process following pollination and ultimately leads to pollen tubes growing into ovules for fertilization and the initiation of seed development. The dialogue between a pollen and the pistil is essential for successfully mediating this process. Recently, members of the Leucine-Rich Repeat Malectin Receptor-Like Kinase (LRR-MAL RLKs) subfamily, have been identified as new players in the early stages of this dialogue in the pistil. Specifically, the Receptor-Kinase in Flowers 1 (RKF1) gene cluster was found to be required in the stigma to promote compatible pollen hydration, as knocking out this gene cluster resulted in decreased wild-type pollen hydration. The RKF1 cluster also participates with other RLKs to support pollen tube growth in the reproductive tract. Using the cytosolic kinase domain of RKF1 as bait, potential binding partners were identified through a yeast-2-hybrid screen of an *Arabidopsis* flower cDNA library (Hybrigenics Services). Here, we worked to confirm these interactions and identify their roles in these post-pollination processes. Top candidates identified from this screen belong to the Ethylene Response Factor VII (ERF-VII) family of transcription factors, specifically the Related to Apetala (RAP) 2.12 and 2.3 members. Additional candidates from the Hybrigenics screen were also tested for potential interactions with RKF1. Furthermore, phenotypic analyses of T-DNA knockout lines were performed to examine how wildtype pollen performed in hydration and pollen tube growth assays in the absence of these ERF-VII factors and related regulators in the pistil. Overall, these results point to the involvement of these novel interactors in an undescribed pathway regulating pollen tube growth in the pistil.

B4-50 - Investigating Arabidopsis stigmatic factors in the regulation of pollen hydration

Raymond Liu¹, Paula Chadic¹, Daphne Goring¹

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Fertilization in *Arabidopsis thaliana* is dependent on multiple factors that regulate successful pollen-pistil interactions, and this starts in the stigma which facilitates pollen hydration and pollen tube entry into the stigma. As well, the pollen-stigma interaction is widely regarded as the initial barrier to fertilization for unwanted pollen grains. Several stigma regulators of pollen hydration have been identified, including receptor kinases belonging to the CrRLK1L (FER, ANJ), LRR-MAL RK (RKF1 gene cluster) and SD-RLK (SD-RKK28) subfamilies. When these genes were knocked out, pollen hydration assays uncovered mild hydration defects following pollination with wildtype Col-0 pollen. On the pollen side, knockout mutations for the PCP-B signaling peptides also resulted in mild pollen hydration defects. Finally, PIP aquaporins were recently implicated in the stigma to contribute to pollen hydration. For all these mutants, the pollen grains were able to hydrate sufficiently to germinate and produce pollen tubes. In this research, we are testing combinations of mutants for these stigma factors to determine if stronger pollen hydration defects occur. We also identified the MscS-like (MSL) mechanosensitive ion channel, MSL7, as another stigma factor for pollen hydration and have tested *msl7* mutants in combinations with the other stigma factors. Newly generated combination mutants are being pollinated with both Col-0 and PCP-B mutant pollen to determine their impact on pollen hydration. Additional analyses using aniline blue staining are being conducted to determine if decreased pollen germination and pollen tube growth occurs with these mutants. Taken together, this research will help elucidate the relationship between several factors within the stigma seen to contribute to the regulation of pollen hydration during plant sexual reproduction.

Monday, August 11th, 2025

CONCURRENT SESSION C1

C1-51 - Exploring the Potential of *Cannabis sativa* and *Chelidonium majus* Endophytes in Promoting Growth and Abiotic Stress Tolerance in Yellow Beans

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¹McGill University, ²BioSun Solutions Inc

Abiotic stresses significantly affect crop yield and quality, prompting growing interest in the use of beneficial endophytic bacteria as sustainable alternatives to chemical fertilizers in agriculture. This study investigates endophytes isolated from *Cannabis sativa* and *Chelidonium majus* for their potential to enhance abiotic stress tolerance in yellow beans (*Phaseolus vulgaris* L.). Isolates were screened for key plant growth-promoting traits, including siderophore production, extracellular polymeric substances (EPS), and proline accumulation. Their tolerance to high temperatures, drought, and salinity was also evaluated. Among the isolates, *Bacillus sp.* (B2) and *Pseudomonas sp.* (P2) demonstrated superior tolerance to heat and salinity. These isolates also exhibited high levels of proline and superoxide dismutase (SOD) activity, with P2 producing the highest amount of siderophores. When applied to drought-stressed yellow bean plants, B2 and P2 positively regulated physiological parameters like proline, malondialdehyde, photosynthetic rate, and stomatal conductance. These improvements led to yield increases of 62.5% and 37.5%, respectively, compared to the drought-stressed water control. Our findings highlight the potential of these endophytes to improve crop resilience and promote growth, offering innovative solutions for sustainable agriculture.

C1-52 - Harnessing bacterial synthetic communities to alleviate drought stress in alfalfa and red clover

Taylor Austin¹, Md. Maniruzzaman Sikder^{2,3}, Shamsul Islam Shipar⁴, Raphael Ofoe², Lord Abbey², Rhea Lumactud²

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As Canada strives to meet its 2030 emission and carbon reduction targets, sustainable agricultural practices are increasingly vital. Drought, intensified by climate change, poses a major threat to global agriculture and rural livelihoods. This study examines the potential of bacterial synthetic communities (SynComs) as a sustainable approach to mitigating drought stress in forage plants under greenhouse conditions. The SynComs-comprising *Bacillus* sp. G3, *Bacillus* sp. G12, *Acinetobacter* sp. G13, and *Plantibacter flavus* strains 251 and 259 were isolated from stressed environments and selected for their potential to enhance plant stress tolerance. We assessed how SynCom inoculation influences plant responses under moderate and severe drought conditions at two-time points: immediately following drought and post-recovery. By inoculating forage plants with SynComs, we aim to observe changes in plant stress responses and identify patterns in microbial community interactions. This study includes higher trophic-level organisms, such as nematodes, which are highly sensitive to soil moisture levels due to their reliance on water films in soil for mobility and survival. The impact of drought on nematodes and their subsequent effect on bacteria, archaea, and fungi is largely unknown. Microscopic and molecular techniques will be employed to identify nematode communities. Soil samples will be analyzed for physicochemical properties, including soil organic matter turnover, nutrient cycling, microbial diversity, and community structure. The expected outcomes include identifying key microbial interactions that enhance plant growth and stress tolerance providing a foundation for sustainable agricultural practices that mitigate the impacts of climate change. By elucidating the ecological roles of SynComs, the findings will provide a scientific foundation for nature-based solutions that promote soil health and mitigate the impacts of climate-induced drought

C1-53 - Double Trouble in Canola: Understanding the interaction between *Verticillium longisporum* and *Leptosphaeria maculans*

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¹Wilfrid Laurier University, ²University of Manitoba

Brassica napus (canola) is one of the most commercially important oilseed crops in the world. Canada accounts for the world's largest canola production. Increasing stresses from fungal diseases are threatening the yield and quality of canola in Canada. Blackleg, majorly caused by the fungus *Leptosphaeria maculans*, is a well-known disease of canola, resulting in significant yield and economic losses in Canada. Verticillium stripe caused by the soil-borne fungus *Verticillium longisporum* has recently emerged as a potential threat to domestic canola production. Yield losses of up to 50% can be caused by this disease, which is a matter of concern given the susceptibility of most commercial Canadian canola varieties. In the field, Verticillium stripe disease symptoms can be misdiagnosed with those of blackleg, and the two diseases may co-occur in canola. Although the resistance of canola to blackleg is well studied, how this resistance might be affected by Verticillium stripe remains unclear. This project aims to understand the interaction between *L. maculans* and *V. longisporum* in canola through transcriptomic and genetic analysis. Canola varieties carrying *Rlm 1*, *Rlm 2*, *Rlm 3*, *Rlm 4*, *Rlm 7*, *Rlm 9*, *LepR1* and *LepR2* genes were inoculated with both *L. maculans* and *V. longisporum* under greenhouse conditions to screen a variety showing a significant change in resistance against both diseases. Canola variety 01-23-2-1, which exhibited a significant breakdown of the *Rlm7* gene, was selected for genome-wide transcriptome analysis. In addition, the disease severities of both blackleg and Verticillium stripe of these canola varieties were evaluated using phenotypic data. The overall outcome of this study will help to identify genes with strong breeding potential, which can be utilized to enhance canola resistance against *V. longisporum* and *L. maculans*.

C1-54 - The Role Ubiquitin Ligase, XBAT35, in Plant Response to Combination of Abiotic and Biotic Stresses

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Plants are continually exposed to various abiotic and/or biotic stresses that impact growth, development and yield. Plants response to individual stresses is well studied, however less is known about responses to combination of stresses: biotic-biotic, abiotic-abiotic, and abiotic-biotic. My research focus is on abiotic-biotic stress combinations occurring consecutively, specifically exposure to drought stress with a short period of recovery followed by bacterial pathogen infection. Abiotic stress has been shown to enhance pathogen defense, however knowledge of the molecular mechanism underlying drought induced resistance (DIR) is very limited. Plants utilize various molecular mechanisms to facilitate responses to environmental stresses including Ubiquitin Proteasome System (UPS), which regulates the abundance of numerous cellular proteins. The role of UPS is to target substrate proteins for degradation by 26S-proteasome via ubiquitination, the attachment of ubiquitin molecules to selected protein. Targeted protein degradation is governed by a large family of ubiquitin ligases (E3-enzymes), which selects the substrate and mediates ubiquitination. Of interest is the RING-type E3 XBAT35, which has two isoforms: nuclear localized XBAT35.1 and the smaller Golgi-localized XBAT35.2. Role of XBAT35.2 in negatively regulating responses to abiotic stresses such as drought and high salinity, while promoting defense against bacterial pathogen has been demonstrated. However, the role of XBAT35.1 in stress tolerance is unknown. The aim of the study is to explore the potential role for XBAT35 in DIR. Expression analysis shows that *XBAT35.1* and *XBAT35.2* are upregulated when the plant is infected with *Pseudomonas syringae* pv. *tomato* DC3000. Surprisingly, *XBAT35* transcript levels didn't change following pathogen infection of drought treated plants. Compared to pathogen infected plants, transcript levels of *PR1* (Pathogenesis-Related-Gene-1) are significantly higher in drought treated-pathogen infected plants indicating DIR, except for *XBAT35.2* over-expressing transgenic-plants as *PR1* transcript levels were substantially low. Further analysis is required to define XBAT35's role in the DIR phenomenon.

C1-55 - Effect of abiotic stress on Verticillium stripe disease in Canola (*Brassica napus* L.)

Shruti Kashyap Kashyap¹, Sean Walkowiak², Harmeet Chawla¹, Claudio Stasolla¹

¹University of Manitoba, ²Canadian Grain Commission

Canola (*Brassica napus* L.) is an economically relevant crop worldwide, yet its productivity is increasingly threatened by *Verticillium longisporum*, the causal agent of Verticillium stripe disease. This study investigates the effects of different water regimes: field capacity (FC), 60% FC and 30% FC on disease progression in *Brassica napus* variety 'Westar'. Plants at the two-leaf stage of development were inoculated with 500,000 spores/mL of *V. longisporum*. Following inoculation, water regimes were imposed and monitored using an ML3 Theta Probe soil moisture sensor. Disease progression in root, hypocotyl, and leaf was evaluated by visual scoring and RT-PCR quantification of fungal DNA. Relative to FC, disease severity was consistently lower under 30% FC, while no differences were observed for plants grown under 60% FC. Six days after imposition of the water regimes, fungal DNA in 30% FC plants was 0.6063% (of total plant DNA) in roots, 0.0025% in hypocotyls, and 0.1077% in the leaves. Respective values under FC conditions were 0.9460%, 0.0079% and 0.5334%. Comparative anatomical studies revealed that the imposition of 30% FC led to increased cell wall thickening in the root endodermal layers, accompanied by more pronounced accumulation of lignin and suberin. These structural modifications were then visualized using Phloroglucinol-HCl for lignin and Fluorol Yellow 088 for suberin. In the hypocotyl, 30% FC induced the precocious formation of a well-developed periderm, which was also rich in suberin. These reinforced apoplastic barriers were likely to delay infection of *Verticillium longisporum*, which was detected using Wheat Germ Agglutinin staining. Reduced staining intensity in 30% FC plants further suggested restricted pathogen colonization compared to plants grown under FC. Collectively, these findings suggest that severe drought induces formation of structural barriers limiting colonization of *V. longisporum*.

C1-56 - *Bacillus toyonensis* and *Pseudomonas koreensis* Cell-Free Supernatants: A Novel Approach to Enhance Soybean Growth under Saline Conditions

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Enhancing crop resilience to salinity stress is crucial for sustainable agriculture, and plant growth-promoting bacteria (PGPB) offer a promising biological solution. This study investigates the effects of cell-free supernatants (CFS) from *Bacillus toyonensis* (KJ812449) and *Pseudomonas koreensis* (AF468452) on soybean (*Glycine max* L.) seed germination and early plant development under a range of salinity stress levels. To ensure bacterial viability, their growth rate and salinity tolerance were assessed using a Cytation™ 5 imaging reader under various salinity levels (0 - 1000 mM NaCl). Two experiments were conducted: a laboratory germination test and a greenhouse study. The germination test exposed soybean seeds to three salinity levels (75, 100, and 125 mM NaCl) and five CFS concentrations (0, 1:100, 1:250, 1:500, and 1:1000 dilution), with germination rates recorded every 6 h up to 48 h. The greenhouse experiment evaluated early plant growth variables, including plant height, chlorophyll content, biomass accumulation, and root morphology using the same CFS treatments. The results showed that salinity stress impaired soybean germination and growth, but CFS application significantly improved these variables, with higher concentrations (1:100 and 1:250) offering greater benefits. Statistical analysis confirmed a strong correlation between CFS treatment and enhanced plant resilience to saline conditions. These findings suggest that *Bacillus* and *Pseudomonas* CFSs can enhance soybean growth under saline conditions, offering a sustainable strategy for improving crop resilience. Future studies will validate these findings in field trials and explore broader agricultural applications

CONCURRENT SESSION C2

C2-57 - Aldehyde dehydrogenases control the carbonylation of photosynthesis-related proteins in *Arabidopsis thaliana*

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Improving crop tolerance to stress conditions requires a deep understanding of plant responses to environmental stress. Protein carbonylation is a key oxidative modification involved in plant stress responses. To decipher how protein carbonylation impairs plant tolerance to oxidative stress, we profiled the carbonylated proteome of *Arabidopsis thaliana* leaves treated with the phytohormone abscisic acid (ABA), which is produced in response to a variety of stresses and controls the expression of stress-responsive genes in plants. The carbonylated proteomes were obtained by affinity capture and subjected to liquid chromatography coupled with tandem mass spectrometry. We identified 180 carbonylated proteins. Of these, 26 proteins became carbonylated upon ABA treatment, whereas 163 proteins initially carbonylated in untreated samples were no longer detected in the ABA-treated samples. This indicated a dynamic control of protein carbonylation within the plant cells. To obtain further insight into how protein carbonylation could be controlled in vivo, we compared the carbonylated proteome between the wild-type plant and mutants of stress-responsive aldehyde dehydrogenase enzymes. This comparison revealed a surprising susceptibility of the plant photosystems to carbonylation and the protective role of the aldehyde dehydrogenases in the cytosol and chloroplasts.

C2-58 - Teasing out the mechanism behind the elevated CO₂ attenuation of the jasmonate burst in C₃ plants.

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Recent studies have shown that plant susceptibility to chewing herbivores and necrotrophic pathogens may increase under elevated carbon dioxide (eCO₂). Wound-induced levels of jasmonates, important

phytohormones involved in signaling pathways leading to plant resistance, are attenuated in the C3 plant *Arabidopsis thaliana* grown at eCO₂. Previous work in the Bede lab found that wound-induced levels of jasmonic acid (JA) and jasmonyl-isoleucine (JA-Ile) were lower in plants grown at eCO₂ while levels of the precursor to these compounds, cis-(+)-12-oxo-phytodienoic acid (OPDA), increased in mechanically damaged plants and were not affected by CO₂ concentrations. My research objective is to understand why wound-induced JA and JA-Ile levels but not OPDA levels are affected by eCO₂. A protein that may have an important role in limiting wound-induced JA production at eCO₂ is cyclophilin20-3 (CYP20-3). CYP20-3 is an OPDA-binding protein that is regulated through redox-dependent conformational changes. We hypothesize that in wounded *A. thaliana* plants grown at eCO₂, a decrease in the oxidation state of CYP20-3, relative to wounded plants grown at ambient CO₂ (aCO₂), will lead to increased binding of OPDA to CYP20-3 and may prevent flux into downstream JA-Ile biosynthesis. Wildtype and *cyp20-3* knockout *A. thaliana* were grown at aCO₂ or eCO₂ and mechanically wounded. Wound-induced phytohormone levels and gene expression were measured by quantitative reverse-transcription PCR and ultrahigh performance liquid chromatography, respectively. We hypothesize that wound-induced JA-Ile production and JA-Ile-responsive gene expression at eCO₂ will be restored to the levels observed at aCO₂ in the *cyp20-3* mutant.

C2-59 - A Thaumatin-like Protein Bridges Photosystem II and Brassinosteroid Pathways for Guard Cell-Mediated Drought Adaptation

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Drought stress triggers complex molecular responses in guard cells (GCs) to balance water conservation with photosynthetic demands. Here, we characterize AT4G36010 (hereafter TLP10), a thaumatin-like protein, as a novel integrator of drought signalling in *Arabidopsis* GCs. Using T-DNA knockout (KO) and GC-specific (*KAT1* promoter-driven) overexpression (OE) lines, we demonstrate that TLP10 mutants exhibit cooler leaf temperatures, elevated stomatal conductance, and impaired abscisic acid (ABA)-induced stomatal closure under drought. Conversely, preliminary data from OE lines show enhanced survival rates and accelerated recovery in re-watering assays.

Yeast two-hybrid (Y2H) screening and *in vitro* validation identified a few high-confidence putative TLP10 interactors: PSBO1 and PSBO2 (core components of the photosystem II oxygen-evolving complex) and GRF1 (a 14-3-3 protein that regulates brassinosteroid signalling). Network analyses and gene ontology enrichment revealed that TLP10 might have a dual role in bridging photosynthetic repair and metabolic regulation pathways.

Our working model proposes that drought-induced DREB2A stabilization activates TLP10 to modulate GC function via two pathways:

- 1) PSBO1/PSBO2 interaction reduces PSII repair efficiency, altering ATP/NADPH balance and amplifying ROS signalling;
- 2) GRF1 interaction perturbs BRI1/BZR1-mediated brassinosteroid signalling, disrupting BAM1-dependent starch degradation and osmotic adjustment.

These synergistic mechanisms may promote stomatal closure and enhance drought tolerance. This study establishes TLP10 as a possible key gene that integrates photosynthetic machinery and hormonal signalling in GC-mediated drought adaptation, providing future targets for improving crop water-use efficiency.

C2-60 - Investigation into the roles of calmodulin-binding transcription activators (CAMTAs) in the Arabidopsis heat stress response

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Global warming is imposing stress on natural ecosystems and commercial crops. In plants, heat stress negatively impacts key physiological processes such as photosynthesis, respiration, and reproduction. Plants respond to elevated temperatures through a process termed the heat stress response (HSR). The cellular mechanisms underlying the HSR are not yet fully understood but involve calcium (Ca²⁺) signal transduction

and changes in gene expression. Plants have many Ca²⁺ sensor proteins that decode Ca²⁺ signals during growth and stress responses. Calmodulin (CaM) is the canonical Ca²⁺ sensor in eukaryotes, but plants also have evolved many calmodulin-like proteins (CMLs), some of which function in abiotic stress signaling. Among the downstream targets of CaM/CMLs are calmodulin-binding transcription factors (CAMTAs), a family of transcription factors that regulate stress-responsive genes in plants. CAMTAs have been implicated in response to cold, drought, pathogen defense, and other environmental stresses. However, their roles during heat stress have not been studied. Here, using a combination of genetics and physiological assays, I test the hypothesis that CAMTAs are important regulatory proteins during heat stress. In heat shock phenotyping assays, *camta* mutants exhibited distinct phenotypes compared to wildtype (WT) plants. RT-qPCR analysis revealed that several heat responsive genes are misregulated in *camta* knockout mutants, indicating their potential role as regulators of the HSR in *Arabidopsis*. Understanding how CAMTAs influence the HSR will provide insight into plant stress regulatory mechanisms and should help inform engineering strategies to enhance crop tolerance in our warming climate.

C2-61 - Overexpression of Canola *PLA1ly3* Altered Chloroplast Lipid Homeostasis and Reduced Thermotolerance in Canola

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Canola (*Brassica napus*) is Canada's most important oilseed crop, which is highly vulnerable to heat stress, making the enhancement of thermotolerance critical under climate change. In *Arabidopsis thaliana*, phospholipase A1ly3 (*PLA1ly3*) catalyzes the hydrolysis of a principal chloroplast membrane lipid, monogalactosyldiacylglycerol, releasing free fatty acids that may subsequently serve as precursors for jasmonic acid (JA) biosynthesis. JA is a key phytohormone involved in regulating stress responses, including thermotolerance in *Arabidopsis*. Building on this functional knowledge, we investigated the role of the canola *PLA1ly3* homolog (*Bna.PLA1ly3*) in heat stress responses by generating overexpression (OE) and CRISPR/Cas9 knockout (KO) lines. OE lines exhibited pronounced heat-sensitive phenotypes at the vegetative stage, including necrotic lesions on leaves, reduced aboveground biomass, and decreased chlorophyll content, whereas KO and wild-type plants showed no such phenotypes. Lipid profiling revealed a substantial reduction in monogalactosyldiacylglycerol and digalactosyldiacylglycerol levels in OE plants, suggesting that elevated *Bna.PLA1ly3* expression compromises chloroplast membrane lipid homeostasis. JA biosynthesis genes were constitutively upregulated in OE lines under normal conditions and further induced by heat stress, yet OE plants remained heat sensitive. Moreover, inhibiting JA biosynthesis further exacerbated the stress symptoms, indicating that while JA is necessary for thermotolerance, upregulation of its biosynthetic genes alone cannot rescue OE-induced heat sensitivity. Together, our findings demonstrate the OE of *Bna.PLA1ly3* causes heat-sensitive phenotypes and disrupts chloroplast membrane lipid homeostasis, along with increased expression of JA biosynthesis-related genes. However, this elevated gene expression is insufficient to maintain thermotolerance, highlighting the need for balanced *Bna.PLA1ly3* expression to preserve chloroplast membrane integrity and enable effective JA-mediated heat stress responses.

C2-62 - Developing drought-tolerant and compact canopy architecture in wheat by manipulating the brassinosteroid pathway

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Wheat contributes \$27 billion in wages to the Canadian economy annually. But its yield is constantly threatened by various abiotic and biotic stresses. An annual loss of 7-10 % in grain yield (equivalent to \$55-60 million) has occurred due to drought stress over the last four decades. The situation has been exaggerated due to rapid global warming and drought conditions during critical stages of wheat development, leading to severe yield losses. Wheat yield need to double to feed the global population by 2050.

Brassinosteroids (BRs) are a class of plant steroids that function as hormones with essential roles in plant growth and development. In cereals, BRs regulate numerous important traits, including leaf angle, grain size, final plant height and senescence. An upright canopy architecture can improve cereal grain yields under high

planting densities, but broad reductions in BR activity often have negative impacts on fertility and grain size. Spatially restricted modification of BR metabolism through targeting specific genes could therefore provide an erect canopy while minimising negative pleiotropic effects. We targeted three BR pathway genes, i.e., BRI1, DWF1, and DWF4, using forward and reverse genetics approaches, and demonstrated their role in regulating canopy architecture in wheat.

Abscisic acid (ABA) hypersensitivity and brassinosteroids (BR) suppression have been reported to impart drought tolerance to crops. Higher ABA levels cause stomatal closure. This is the basis of our current chemiscreen for isolating M2 individuals from elite wheat breeding lines such as AAC Taber, AAC LeRoy, and AAC HY2090. Some of the top-performing mutants are currently being tested under controlled and field environments (across three prairie locations) for their drought tolerance compared to controls. We are currently characterizing a potential causal gene by reverse genetics approaches. This will be followed by primer design and integration with commercial CPS and CWRS wheat cultivars through breeding.

CONCURRENT SESSION C3

C3-63 - Unlocking the enzyme network driving Amaryllidaceae alkaloids biosynthesis

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Amaryllidaceae alkaloids (AAs) are a treasure trove of bioactive metabolites with immense pharmaceutical potential, yet their biosynthetic roadmap remains largely uncharted. At the heart of their formation lies the condensation of tyramine and 3,4-dihydroxybenzaldehyde—catalyzed by norbelladine synthase (NBS) and/or noroxomaritidine/norcraftsodine reductase (NR)—to yield norbelladine, a crucial precursor. Further modifications, including O-, N-methylation, and regioselective C-C couplings by cytochrome P450 enzymes, sculpt the structural diversity of these potent compounds, including lycorine, galanthamine, and crinine. In this study, we uncover key biosynthetic players from *Amaryllidaceae*, shedding light on a long-standing biochemical mystery. Transient expression in *Nicotiana benthamiana* reveals that NBS and NR11 efficiently generate norbelladine, which is subsequently methylated by N4'-OMT *in planta*. Strikingly, co-expression of NBS, NR11, N4'-OMT, and CYP96T1 results in a metabolic cascade yielding lycorine as the major product, along with haemanthamine, crinine/vittatine, and norgalanthamine. Even more remarkably, we identify CYP96T1 and CYP96T2 as monocot enzymes capable of catalyzing all three regioselective phenol couplings—unlocking a new level of complexity in AA biosynthesis. Beyond elucidating key enzymatic steps, this work paves the way for metabolic engineering strategies to sustainably produce high-value alkaloids. With growing interest in plant-derived pharmaceuticals, our findings provide an unprecedented opportunity to harness nature's chemical diversity for drug discovery and biotechnological innovation.

C3-64 - Dissecting the function of aroenate dehydratases from *Arabidopsis thaliana*

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Aroenate dehydratases (ADTs) catalyze the last step in phenylalanine (Phe) biosynthesis. In plants, the resulting Phe is a precursor for protein synthesis and many aromatic compounds. In *Arabidopsis thaliana* ADTs are encoded by a small family of closely related proteins consisting of six members. All six ADTs (ADT1-ADT6) primarily use aroenate as a substrate, but two of them (ADT1 and ADT2) can also convert prephenate into phenylpyruvate and can therefore also be classified as prephenate dehydratases (PDTs). Furthermore, ADT2 and ADT5 have been described as moonlighting proteins required in chloroplast division and for transcriptional regulation, respectively. To perform their various functions ADTs can act as homo- and heterodimers, they can form larger protein complexes involving non-ADT proteins, they have been found to associate with different subcellular localizations (chloroplast, cytosol, nucleus) and they can be modified by posttranslational modifications. To understand how small changes in sequence can differentially mediate these many aspects of ADT functions, we have taken a multipronged approach. We introduce amino acid substitutions at specific positions within ADTs and determine if such changes affect ADT dimer formation, subcellular locations, substrate specificity. Specifically we are targeting amino acids (mostly serines and threonines) which are

predicted to be phosphorylated and replace them with amino acids which are phosphomimics or phosphonulls to determine if any of the functions are regulated by phosphorylation. Using ADTs as a model system, it is our goal to elucidate how small changes in amino acid sequence can define and potentially fine-tune and regulate different aspects of each family member.

C3-65 - Discovery and Characterization of Coenzyme A Ligases Involved in Phytocannabinoid and Bibenzyl Biosynthesis in *Radula complanata*

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The Bryophyte *Radula complanata* is a non-vascular plant and a unique source of medicinal metabolites known as bibenzyls. These compounds likely serve as chemical defenses against environmental stressors, herbivory, and microbial pathogens. Among these, perrottetinene stands out due to its structural similarity to the psychoactive cannabinoid tetrahydrocannabinol (THC), differing only by a phenyl substitution and a chiral center. PET acts as a partial agonist of the cannabinoid 1 (CB1) receptor and has been shown to elicit reduced inflammatory responses compared to THC. To elucidate the biosynthetic pathway of PET, we focused on coenzyme A ligases (CoALs), which activate acyl donors to initiate bibenzyl-forming polyketide biosynthesis. In this study, we biochemically characterized several CoALs from *R. complanata* and confirmed their involvement in PET and related bibenzyl biosynthesis. Notably, RcCoAL1 displayed broad substrate promiscuity, highlighting its potential as a versatile biocatalyst for generating structurally diverse phytocannabinoids and bibenzyl-based pharmaceuticals.

C3-66 - Flavonol peroxidation in senescing arugula leaves

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The high perishability of the vegetable arugula is due to its postharvest senescence. In other Brassicaceae vegetables, leaf senescence is associated with the loss of kaempferol glycosides which culminates in the accumulation of the aglycone kaempferol. The lack of a sustained accumulation of kaempferol in senescing leaves is likely due to peroxidase activity. Kaempferol peroxidation occurs in the model Brassicaceae plant *Arabidopsis* although the precise enzyme(s) involved in this process and the nature of the peroxidation product remain unknown. The main aim of this study was to establish in-depth biochemical and proteomic evidence for a plant kaempferol peroxidase. A high-performance liquid chromatography-diode array detection (HPLC-DAD) assay was developed for the investigation of kaempferol peroxidase activity in arugula leaves. Kaempferol peroxidase activity was induced in postharvest leaves of arugula during low temperature storage. The HPLC-DAD assay revealed that kaempferol is oxidized to a benzofuranone-like product by cell-free leaf extracts in the presence of hydrogen peroxide. We are using nuclear magnetic resonance spectroscopy to confirm the molecular identity of the kaempferol peroxidase product. Finally, a three-step column chromatography procedure was used to purify a kaempferol-utilizing peroxidase from freshly harvested arugula leaves. The monomeric nature of the peroxidase was confirmed by size exclusion chromatography coupled with mass spectrometry-based proteomics. *In silico* analysis of the peptides generated from the trypsin digestion of the peroxidase revealed a high homology to plant peroxidase III family proteins. The analysis of the kinetic properties of the arugula leaf peroxidase will determine whether it is specific for flavonols. The information generated from this study will improve our understanding of the role of flavonol peroxidases in the postharvest senescence of Brassicaceae vegetables.

C3-67 - Screening of novel genes associated with seed oil production

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The growing global population led to an increasing demand for vegetable oil, and triacylglycerol (TAG) is the major seed oil component. Although advances in plant lipid biology in recent years have substantially enriched our knowledge of TAG biosynthesis and regulation, gaps still exist in many aspects of TAG production in seeds. Studies with multi-omics and genome-wide association analysis over the past years have predicted some novel genes of interest. However, many of these genes have not been verified independently, and the underlying mechanisms are largely unknown. In this study, we selected a set of candidate genes potentially involved in TAG biosynthesis from published molecular breeding papers and their associated datasets for further evaluation. We initially tested these genes in a yeast expression system but did not get conclusive results. Then we generated constitutively overexpressing (OE) Arabidopsis lines, respectively, and obtained the loss-of-function mutant lines (SALK lines). Seed oil analysis identified 5 genes that could significantly change seed oil contents. Subsequently, a *MYB-like* gene and an *alpha/beta hydrolase-like* gene were selected for further analysis. The *MYB-like* gene did not significantly alter total seed oil content in OE or SALK lines but significantly changed fatty acid composition and seed areas. The OE of the *alpha/beta hydrolase-like* gene resulted in a 9% reduction in seed oil content, and both OE and SALK lines showed significant changes in fatty acid composition and reduced seed areas. The results could serve as a good foundation for the full characterization of the biological functions and biotechnological values of the genes.

C3-68 - Mechanistic insights into the toxicity of naphthenic acid fraction compounds: Toward plant-based solutions for oil sands process water remediation

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Bitumen extraction from the Athabasca oil sands is a water-intensive process. It is estimated that more than 400 billion litres of oil sands process affected water (OSPW) are stored in all existing tailings ponds in the region. OSPW contains multiple constituents, with particular concern surrounding the organic compounds known as naphthenic acid fraction compounds (NAFCs) due to their potential acute and chronic toxicity to aquatic and terrestrial life. Plant-based remediation strategies offer a low-energy input solution to complement existing and emerging OSPW treatments, and can include constructed wetland treatment systems (CWTS). One outstanding question for most NAFC bioremediation technologies is: how do biochemical transformations of NAFCs by plants or microorganisms reduce their acute and chronic toxicity? This is still not completely understood, in part because the mechanisms underlying NAFC toxicity are not well characterized. It is assumed that NAFCs disrupt plasma membranes and act as narcosis/anesthetic agents, but there is little direct evidence to support this. Our research addresses this gap using in vitro liposome assays, which demonstrate that model NAFCs act as narcosis agents. Furthermore, the relative binding affinities of NAFCs to a model lipid membrane correlated well with their toxicity across multiple organisms—a hallmark of narcosis agents. Importantly, plant-derived metabolites of model NAFCs exhibited reduced membrane disruption and lower toxicity. These findings support the potential of plant-based remediation to transform NAFCs into less harmful byproducts. Future research will focus on characterizing these plant metabolites to confirm their reduced chronic toxicity and to identify which membrane-associated NAFC classes persist in CWTS-treated OSPW. This work will enhance our understanding of NAFC toxicity mechanisms and the role of plant-based remediation in OSPW remediation.

CONCURRENT SESSION C4

C4-69 - Developing new tools to quantify plant endomembrane dynamics

Emily Larson¹

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The plant endomembrane system is composed of complex intercellular compartments that deliver molecular cargo in and out of the cell. Practically everything a cell does requires the endomembrane system, from cell division and expansion to differentiation and functional specialisation. The aim of my research is to expand our knowledge on endomembrane compartment identity and regulation and learn more about how to improve plant

growth, especially under changing environmental conditions. However, we have limited understanding of how the identity of endomembrane compartments is maintained and how the different compartments that make up the endomembrane system are coordinated to sort molecular cargo for delivery. To begin to address these questions, I have developed fluorescence assisted vesicle sorting (FAVS) methods to quantify endomembrane compartment populations and dynamics, with preliminary results indicating that fluorescent protein-fusion constructs can be used to count and sort specific vesicle populations. I also express these fluorescent proteins in different *Arabidopsis* vesicle trafficking mutant lines to gain a holistic view of how trafficking patterns in and out of the cell are coordinated. Lastly, I am using classical genetics to discover new interacting partners associated with vesicle trafficking pathways that support polarised cell growth. These projects will build a foundation for a new lab group that will focus on plant endomembrane biology.

C4-70 - Characterizing the targeting mechanism of OEP6, a novel chloroplast outer membrane protein

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The chloroplast is a membrane-bound organelle responsible for photosynthesis in all plants and contains a diverse proteome of approximately 3,000 total proteins necessary for biogenesis and function. Approximately 95% of these proteins are nuclear-encoded, synthesized in the cytosol and post-translationally targeted to the chloroplast through the TOC and TIC translocon complexes at the outer and inner membranes, respectively. The chloroplast outer membrane proteome encompasses approximately 5% of the total proteome and is less well-characterized. Outer envelope proteins are targeted and inserted specifically into the outer membrane but have not been found to use a single conserved mechanism, leaving many unclassified and novel in terms of the targeting pathway they use. Of the roughly 138 OEPs that have been identified in *A. thaliana*, my research examines Outer Envelope Protein 6 (OEP6), previously presumed as a tail-anchored protein with little known evidence surrounding its targeting or insertion mechanism. A series of eGFP-tagged OEP6 constructs with deletions and point mutations were created to investigate the role of the predicted alpha-helical transmembrane domain (TMD) and positively charged flanking residues in its targeting to the chloroplast outer membrane. These constructs were introduced into onion epidermal cells and *A. thaliana* protoplasts via biolistic bombardment and PEG-mediated transfection where their preliminary localization was monitored using epifluorescent and confocal microscopy. To validate the confocal imaging data, cellular fractionation was performed and using anti-GFP antibodies, Western blotting was used to detect cytosolic versus chloroplast localized proteins. The results indicate OEP6 utilizes a novel targeting mechanism, with positively charged residues flanking the TMD acting as targeting determinants responsible for both chloroplast and mitochondrial targeting. The work contributes to the chloroplast outer membrane targeting mechanisms thus also advancing the overall knowledge of chloroplast protein trafficking pathways.

C4-71 - Protein interaction analyses for plant molecular chaperone HSP90C via Turbo

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Nascent polypeptides must fold into their correct three-dimensional structures before they can perform their function properly. Molecular chaperones, many of which belong to heat shock proteins (HSPs), help prevent misfolding and promote the correct folding of other proteins also known as clients. HSPs are found in animals, fungi, and plants and are known to mediate and stabilize a variety of signal transduction proteins such as protein kinases and transcription factors (TFs) to name a few. There are seven HSP90 family proteins in *Arabidopsis* and we will survey the physical interactors of the plastid-localized HSP90C. At the molecular level, HSP90C has been shown to assist in plastid protein import and thylakoid protein transport, playing an essential role in plant growth and development. However, only a very small set of clients have been identified so far. In this study, we propose to study and screen for the physical interactors of the HSP90C proteins via Turbo which captures weak and transient protein interactors and complexes. We have already generated HSP90Cpro::HSP90C-TurboID plant lines, and the transgenic protein was expressed very well and exhibited a characteristic biotinylation activity *in vivo*. Interestingly, a large proportion of the transgenic line showed albino

and/or variegated phenotypes. The possible mechanism of this phenotype was also investigated. In the future, the dynamic change of HSP90C interactome under different stresses will be mapped to decode the protein-protein interactions and the molecular basis of HSP90C-associated signalling.

C4-72 - What goes up must come down: modulation of the microtubule protein clasp is required for control of cell proliferation

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To survive, all organisms must modulate their growth in response to environmental conditions. In plants, cell proliferation is localized to the meristem and regulated by crosstalk between signalling pathways to optimise cell division rates, and thereby growth, in response to diverse environmental conditions. TOR kinase is an established evolutionarily conserved master regulator of cell proliferation in response to cellular energy cues, yet how TOR kinase activity is translated into altered cell proliferation rate is unclear. This work demonstrates that the microtubule associated protein CLASP is required for sustaining cell proliferation downstream of TOR. *CLASP* loss of function plants display reduced sensitivity to TOR inhibition at both the root growth and meristem level. We show that CLASP expression is increased in response to TOR inhibition, however this regulation requires a functional brassinosteroid responsive element within the CLASP promoter. This promoter element is required for modulation of cell proliferation in response to both brassinosteroid and TOR signalling demonstrating the essential nature of CLASP in enacting changes downstream of both pathways. Finally, we show that TOR mediated control of auxin, by sustaining PIN2 at the plasma membrane, is dependent on modulating CLASP levels, providing a mechanism by which altered TOR signalling is manifested as decreased meristem activity. These findings demonstrate that CLASP is a crucial regulator of meristem activity in response to a common BR-TOR signalling pathway and furthers our understanding of how diverse signalling pathways cooperate to optimise cell proliferation.

C4-73 - Unexpectedly Diverse - Anatomical Variations in the Rosette Leaves of *Arabidopsis thaliana*

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Leaves are essential plant organs that perform key physiological processes. *Arabidopsis thaliana*, a widely studied model organism, has provided valuable insights into plant development. Many studies on *Arabidopsis* leaf structure and function assume uniformity across all rosette leaves, often leading to selection of leaves arbitrarily for anatomical and physiological analyses. This assumption overlooks an important developmental phenomenon known as heteroblasty, which refers to the progressive changes in leaf structure as a plant matures.

Previous research on heteroblasty in *A. thaliana* has primarily focused on genetic pathways regulating the transition from juvenile to adult leaves, and changes in morphological traits such as leaf shape, trichome density, and hydathode distribution. While these studies advance our understanding of leaf development, the internal anatomical differences between the different leaves remain unexplored. If the internal anatomy changes with plant growth, this could have profound implications for plant function, particularly in areas like photosynthetic efficiency, gas exchange regulation, and water use.

This study aims to address this knowledge gap by systematically characterizing the anatomical variations among the rosette leaves of *A. thaliana*. Using 3D confocal laser scanning microscopy and electron microscopy, we investigate variations in mesophyll organization, epidermal cell size, and cell wall thickness between leaves formed at different stages of rosette development. Our findings reveal that leaf anatomy is not uniform within the rosette, challenging current practices in leaf selection for physiological analyses. The cross-sectional area of palisade mesophyll cells in paradermal view displays a distinct pattern of decline in area from the first to the third leaf within each whorl of three. Variations in cell volume were also observed in the mesophyll cells of the different leaves. Characterizing these differences is critical for refining experimental approaches in plant physiology and may have broader implications for our understanding of leaf function in other plant species.

C4-74 - Formation of a stable tubular ER network requires localized phosphatidylcholine synthesis in Arabidopsis

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Phosphatidylcholine (PC) is a major component of phospholipid to constitute bilayers of the endoplasmic reticulum (ER) membrane. Membrane fusion of different ER tubules driven by RHD3 in Arabidopsis functions to form and maintain the interconnected ER network, whilst synthesis and rearrangement of membrane lipid is required to form and remodel the ER during continuous membrane fusion. However, it is still unclear how the role of PC affects the ER formation and remodeling, especially in the process of membrane fusion mediated by RHD3. Through forward genetics, a genetic enhancer of *rhb3-1* named *ren10* was identified. In this study, the *REN10* gene was cloned by map-based cloning, which encodes phosphorylcholine cytidylyltransferase (CCT1) which is the rate-limiting enzyme in the biosynthesis of PC. The deletion or mutation of *CCT1* leads to short root hairs and aggregated ER and unsmooth ER tubules. And CCT1 is partially localized in the ER and is involved in the regulation of ER fusion. The membrane attachment of CCT1 in ER is reduced in the absence of RHD3 although there is no physical interaction between CCT1 and RHD3. The localized PC synthesis in the ER is reduced in *ren10* mutant. We conclude that the localized PC synthesis is required to form and maintain a tubular ER network.

CONCURRENT SESSION C5

C5-75 - Comparative transcriptomics of divergent wheat cultivars reveals epigenetic regulation in microspore embryogenesis

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Microspore embryogenesis refers to the process of immature male gametophytes forming embryo-like structures (ELs), which subsequently develop into haploid plantlets and then into doubled haploid (DH) plants through chromosome doubling. DHs achieve complete homozygosity in a single generation, thereby accelerating breeding programs by significantly reducing time and cost. DH technology complements marker-assisted breeding or genomic selection by enabling effective selection at the haploid or homozygous stage. Due to its economic and genetic advantages, DH technology is widely used in breeding programs. As climate change intensifies biotic and abiotic stresses, the use of DH technology becomes increasingly vital for developing resilient crop varieties. Bread wheat is one of the most important cereal crops worldwide. However, most wheat cultivars exhibit strong genotype-dependent recalcitrance to microspore embryogenesis. In this study, we explored the molecular basis of this variation using two bread wheat cultivars: Nanda, which displayed high embryogenic efficiency, and Sadash, which showed limited responsiveness. Comparative transcriptomic analysis across multiple time points during the culture period revealed dynamic changes in gene expression associated with microspore embryogenesis. Notably, we identified distinct biological processes enriched in Nanda, particularly those related to epigenetic regulation, including nucleosome assembly, chromatin remodeling, and organization. Genomic regions enriched with differentially expressed genes (DEGs) indicated possible structural variation between the two genotypes that may underlie their divergent embryogenic responses. Among the potential biomarkers identified through machine learning, a gene from the histone deacetylase family stood out and was located within a DEG-enriched region. Furthermore, analysis of homoeolog expression bias, a key feature in polyploid wheat, revealed a consistently higher suppression of B-subgenome homoeologs in Sadash, 65% of which overlapped with the DEG list from genotype comparisons. These findings highlight a significant role of epigenetic regulation and subgenome-specific expression dynamics in controlling microspore embryogenesis in bread wheat.

C5-76 - Proteomic analysis of compound leaves of two pea (*Pisum sativum* L.) cultivars reveals accumulation of regulatory clues for differential reproductive development in two phenotypes

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Field pea (*Pisum sativum* L.) is a highly valuable legume crop cultivated for both food and feed purposes. Its high protein content has increased its appeal as a sustainable, plant-based protein source. As a nitrogen-fixing crop, pea contributes to reduced greenhouse gas emissions and serves as an effective option in commercial crop rotations, particularly across the Canadian Prairies. To support continued improvements in crop productivity, a deeper understanding of leaf developmental biology is essential. In pea, leaves not only perform photosynthetic carbon assimilation but also provide mechanical support through tendrils. This study was conducted on two contrasting leafed and semi-leafless pea cultivars, which have either residual or well-developed tendrils. A comprehensive overview of the total and nuclear proteomes across different leaf parts identified approximately 7,800 total proteins and 8,600 nuclear proteins. Major changes in protein reprogramming were observed between young and mature tendrils, petiole, and leaflet tissue-type comparisons in both total and nuclear proteome studies. Among the total and nuclear proteomes about 78% and 37% proteins, respectively were found differentially abundant. In addition to the role of various transcription factors in tendril development, FLOWERING LOCUS Ta1 (FTa1) and FRIGIDA-like (FRL) proteins showed cultivar-specific accumulation differences. These proteins are known to be involved in the regulation of flowering time, and their accumulation patterns were consistent with observed phenotypic differences in flowering timing and density between the two cultivars, which supports their functional relevance in pea reproductive development.

C5-77 - Multi-Omics-Driven Genetic Improvement of Pea (*Pisum sativum* L.) Seed Quality

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Pea (*Pisum sativum* L., $2n = 2x = 14$) is a cool-season legume crop containing approximately 20-35% protein. To improve the nutritional quality and diversify the techno-functional properties of pea, it is essential to understand the genetic mechanism underlying seed quality traits. We employed multi-omics approaches, including transcriptomics, proteomics and metabolomics, to elucidate the complex regulatory networks controlling pea seed development and composition in ten diverse *Pisum* genotypes across five stages. Transcriptomics results reveal dynamic changes in gene expression associated with key developmental processes and gene co-expression networks regulating biosynthesis and deposition of seed storage compounds (protein and starch). Specific gene modules were identified for wild, landrace and cultivated *Pisum* genotypes. Global proteome profiling complemented these findings, identifying protein group type and abundance changes across developmental stages for wild and cultivated *Pisum* genotypes and protein content traits. In addition, metabolome profiling was conducted to identify the biochemical processes essential for seed development.

A panel of >300 diverse pea lines were grown in three environments and the harvested seeds were analyzed for total protein, starch, and amylose content. Genome-wide association study (GWAS) identified significant SNP-trait association for starch and amylose content on 2LG1, 3LG5 and 4LG4, and total protein content on 1LG6, 3LG5, 4LG4 and 5LG3. PCR Allele Competitive Extension (PACE) markers were developed for these

traits to facilitate rapid marker-assisted selection, accelerating breeding peas with improved quality traits. Our study also revealed that pea flour with varied amylose content, originating from diverse genotypes, displayed different techno-functional properties that can be potentially tailored for various applications. Global proteome profiling of 200 diverse pea lines identified 5,679 protein groups; significant differences were observed for major storage proteins, such as globulins and albumins and 25 allergens. Most of these allergens (60%) were 7S proteins, with varying quantities across wild, landrace and cultivated lines.

C5-78 - Identification of genetic loci for culm breaking strength in spring wheat (*Triticum aestivum* L.)

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Lodging, the displacement of plants from their upright positions, is a major constraint to spring wheat (*Triticum aestivum* L.) production which negatively impacts harvest efficiency, and grain yield and quality. Lodging is a quantitative trait regulated by many factors including plant morphological characteristics such as plant height and culm strength. Although reducing plant height has been the primary strategy for managing lodging, basal culm breaking strength is a critical trait that controls lodging as plants with enhanced mechanical strength exhibit superior standability and lodging resistance, while those with lower strength are more susceptible. This study investigated culm breaking strength quantitatively through a genome-wide association study using a diverse population of spring wheat genotypes in multiple environments. The mapping panel exhibited a wide range of breaking strength of the culm basal internode, ranging from 8 to 22 Newton. Our data also revealed a high heritability (0.87) of culm breaking strength, and significant effect of genotype-by-environment (GxE) interaction on the trait. Genotyping of the panel using the 90K iSelect BeadChip Array resulted in 18,611 filtered single nucleotide polymorphism (SNP) markers, which were used to determine their association with culm breaking strength. Our analysis identified multiple SNPs and quantitative trait loci (QTL) significantly associated with breaking strength on chromosomes 1B, 2A, 3A, 3B, 3D, 6B, and 7A. These QTL explained 3.6 to 49.0% of the variance in breaking strength; with QBs_UMb_3D explaining the least (3.6%) and QBs_UMb_2A explaining the greatest (49.0%). The findings of this study may support marker assisted selection for improving culm strength and lodging resistance in spring wheat.

C5-79 - From Infection to Insight: De Novo Assembly of the Sunflower Transcriptome Under Fungal Stress

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When you enter your kitchen, whether it is sunflower seeds that you snack on or sunflower oil that you use for cooking, you can find some products derived from sunflowers. However, despite the significant economic value that sunflowers offer, sunflower cultivation is threatened by fungal pests that can severely damage its harvest. To lay down the foundation for the development of pathogen-resistant or pathogen-resilient sunflower genotype, this research aims to utilize RNA sequencing, *de novo* transcriptome assembly, and differential gene expression analysis to investigate the molecular basis of sunflower response to fungal pathogens. More specifically, the research focuses on elucidating the role of isoprenoids, or terpenoids, in sunflower's response to infection by *Rhizopus oryzae*, *Botrytis cinerea* and *Sclerotinia sclerotiorum*. We have assembled a *de novo* transcriptome from healthy and infected plants and carried out differential gene expression analyses. Results indicated that the expression of many of the isoprenoid biosynthetic genes is altered by fungal infection. In addition, our transcriptome contains several previously uncharacterized terpene synthase transcripts. We are currently working on cloning and biochemical characterization of novel mono- and sesquiterpene synthase genes.

C5-80 - Radicle proteomics of soybean and green pea, in response to lipo-chitooligosaccharide and Thuricin 17

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Soybean (*Glycine max*) and pea (*Pisum sativum*) are leguminous plants that form nitrogen-fixing root nodules through symbiosis with *Bradyrhizobium* (soybean) and *Rhizobium leguminosarum* (pea), based on precise chemical signaling between plants and rhizobia. Two important molecular signals involved in this interaction in soybean are lipo-chitooligosaccharides (LCOs) and Thuricin 17 (Th17). The LCOs are highly specific and structurally tailored to the host, ensuring proper partner recognition, while Th17 can work synergistically with LCOs, priming the plant for improved symbiotic performance and resilience.

The germination responses of soybean and pea to two concentrations of LCOs (10^{-6} and 10^{-8} M) and Th17 (10^{-9} and 10^{-11} M) were studied and the radicles at 72 h post germination were used to extract total proteins for LC-MS/MS based proteome studies. Soybean germination was not significantly affected by these signals under optimal conditions. However, pea in Th17 showed consistently significant patterns of germination and more so at the lower concentration of 10^{-11} M. Radicle-localized proteins orchestrate the earliest responses to rhizobia in soybean and pea. While both species share core symbiotic pathways, differences in nodule types (determinate in soybean vs. indeterminate in pea) influence the spatial and temporal dynamics of protein expression. While, some core proteins were found to be common, many proteins were detected in concentration specific patterns. For example, apyrases in legumes, which plays diverse roles in nodulation, nodule senescence, stress responses, and nutrient uptake and transport by modulating extracellular nucleotide signaling pathways, was found at the lower concentrations of Th17. Understanding these proteins is crucial for enhancing biological nitrogen fixation and legume productivity. Together, LCOs prime the legume for symbiosis, while Th17 boosts plant vigor and nodulation, making them powerful tools for sustainable agriculture.

CONCURRENT SESSION D1

D1-81 - Enhancing disease resistance in potato through heterologous expression of multiple host defense peptides.

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Host defense peptides (HDPs) are essential components of the innate defense system of many organisms, including plants. These evolutionarily conserved small molecules have potent antimicrobial activities against a broad spectrum of pathogenic microorganisms, killing invading fungi and bacteria directly or by modulating host immune responses. The aim of the present study was to evaluate the antimicrobial activity and cytotoxicity of selected HDPs, and to test whether the heterologous expression of these peptides in potato has the potential to enhance host resistance against major potato pathogens. Five HDPs were assessed in this study: Shepherdin 1, from shepherd's purse (*Capsella bursa-pastoris*); SM-985, from Mexican teosinte (*Zea mays* ssp. *mexicana*); BnPRP1, from rapeseed (*Brassica napus*); P4650, from pumpkin (*Cucurbita maxima*); and Ib-AMP1Q, from spotted snapweed (*Impatiens balsamina*). Chemically synthesized HDPs were tested in vitro, alone and in combination with each other, against potato pathogens *Fusarium sambucinum*, *F. culmorum*, *F. solani* (the causative agents of dry rot and Fusarium wilt diseases), *Alternaria solani* (early blight) and *Pectobacterium carotovorum* (soft rot). Shepherdin 1, SM-985, P4650, and Ib-AMP1Q inhibited pathogen growth when used as single peptides. Strong synergistic interactions were observed for combinations of two or three HDPs, lowering the minimum inhibitory concentrations (MICs) by up to 83%. No cytotoxicity in mammalian cells was detected. Plant-optimized nucleotide sequences of genes encoding these HDPs were introduced into the genome of *Solanum tuberosum* L. cv. Desiree, via *Agrobacterium*-mediated transformation. The expression of HDP genes in transformed plants was regulated by the *PmBiPPPro1* promoter of the luminal binding protein (BiP) from Douglas-fir. Over 130 transgenic potato lines expressing single HDPs, or their combinations (gene pyramiding), were regenerated on the selective medium. In disease resistance bioassays, the lines expressing two or three HDPs significantly delayed the onset of disease symptoms, although the level of host resistance was often pathogen specific.

D1-82 - Interference of fungal nucleases protects crop plants from disease using next generation RNA fungicides

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Spray Induced Gene Silencing (SIGS) is a practical, non-transgenic approach that applies the principles of RNA interference (RNAi) to crop protection. In this approach, double-stranded (ds) RNA molecules with sequence homology to an essential fungal gene are topically applied to plant tissues - mimicking that of conventional foliar fungicide applications. Following the successful uptake of these molecules by the target fungus, gene silencing occurs resulting in reduced pathogen viability. We have previously shown that SIGS is capable of protecting canola from the necrotrophic fungal pathogen *Sclerotinia sclerotiorum* - the causative agent of white mold. While advances have been made using long dsRNA molecules, there remains opportunity to improve molecule durability and target gene knockdown. We investigated the use of a novel double-stranded RNA structure called paperclip (pc). Paperclip RNAs enter *Sclerotinia* via an alternative uptake mechanism to those used by conventional dsRNAs and are more resistant to nuclease digestion. The co-silencing of the *Sclerotinia* Mitochondrial Ribosomal Protein L3 (dsRNA nuclease) along with Chitin Synthase VI led to markedly reduced disease severity and target transcript abundance relative to the interference of a single target gene. Similarly, a reduction in disease symptoms was also observed following foliar applications of pcRNAs targeting essential *Sclerotinia* genes; with significant plant protection when pcRNAs targeting MRPL3 were applied as a stack. Together, our work shows utility of next generation RNA fungicides in plant protection against devastating fungal pathogens.

D1-83 - Exploring the Phenotypic Impacts of the Soybean Endophytic Microbiome Using Shotgun Proteomics

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Plant adaptation to environmental stress involves cellular, molecular, and biochemical processes. Some stress-mitigating strategies leverage microbes associated with the plant known as the plant microbiome or phytomicrobiome. This is the case for soybean, a major agricultural crop that releases secondary metabolites into the soil-root environment to attract beneficial bacteria. Upon colonizing host tissues, endophytic bacteria establish symbiotic relationships with a plant. The root endophytes of soybean plants form a selectively beneficial and diverse community whose direct effects on the plant and indirect effects mediated by inhibitory effects of pathogens are poorly characterized. We investigated three microbial strains isolated from the soybean root microbiome and characterized their direct plant growth-stimulating effects as well as their biocontrol activity against *Fusarium oxysporum*. The strains *Bacillus*-HT1 and *Bacillus*-HT2 increased soybean seed germination, whereas *Bacillus*-HT3 significantly increased plant leaf area. Shotgun proteomics of soybean leaves revealed the modulation of expression levels of several proteins associated with plant growth and stress tolerance, through multiple signalling pathways. The expression of several proteins involved in major metabolic pathways, such as carbon metabolism, photosynthesis, respiration, and oxidative stress pathways, was upregulated. These include glutathione transferase, cysteine proteinase, glutathione peroxidase, lipoxygenase, glutathione reductase and inositol-1-monophosphatase and stromal heat shock-related protein. Insights into the changes in protein profile will allow us to characterize the growth-enhancing metabolic pathways activated in soybean by associated *Bacillus* species and allow us to deduce how resilience to stress is established.

D1-84 - Use of a rapid screen to identify formulations that enhance plant resistance to viral and bacterial infections

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Viral and bacterial infections substantially diminish crop yield and quality resulting in reduced agricultural productivity. The use of antimicrobial formulations may mitigate the adverse effects of plant disease, however two key challenges must be addressed: 1. Development of rapid screens to identify resistance-inducing formulations and 2. Identifying effective antimicrobial formulations with minimal impact on human and environmental health. A rapid 7-day screen was developed using *Agrobacterium*-mediated delivery of Tobacco Rattle Virus expressing GFP (TRV-GFP) into *Nicotiana benthamiana* to identify magnesium chlorophyllin (Mg-chl) formulations that enhance plant viral resistance. Subjective scales based on TRV-GFP fluorescence in inoculated and systemic leaves were created and validated. Treatment with Mg-chl formulations resulted in significant reductions in TRV-GFP levels in *N. benthamiana* leaves compared to mock-treated plants, without causing phytotoxic effects. Mg-chl treatment of leaves led to H₂O₂ accumulation as expected given that Mg-chl is activated by light to produce reactive oxygen species. Expression of salicylic acid (SA) and jasmonic acid (JA) defense pathway genes (*Nb PR1*, *Nb MYC2*) was also observed in response to Mg-chl treatments, suggesting that Mg-chl treatment initiates SA- and JA-mediated defenses. Hydrothermal carbonization process water has the potential to stimulate disease resistance and promote plant growth. During hydrothermal carbonization of agricultural food waste, process water containing beneficial plant organic compounds is produced. To determine if hydrothermal carbonization process water stimulates disease resistance, the Agro-TRV-GFP/*N. benthamiana* rapid screen will be used followed by lab testing in cucumber, an important Ontario greenhouse crop, using the bacterial pathogen, *Pseudomonas syringae* pv *lachrymans*, and cucumber viruses. Given that lab research does not always transfer to genotypes commercial settings due to differences in environmental conditions, production systems and plant, resistance-enhancing Mg-chl and process water treatments will be assessed in cucumber plants grown under simulated commercial greenhouse conditions in the new McMaster greenhouse.

CONCURRENT SESSION D2

D2-85 - Selection has acted upon regulatory alleles to improve hexaploid bread wheat.

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Plant breeding, the science of artificial selection, has generated remarkable organismal changes. In cereals, grain yields and the yield stabilities of new cultivars are far greater than those of old cultivars, enabling modern food security. Regulatory alleles that affect gene transcript abundances may have been selected. A high proportion of crop species are also recent polyploids. Allelic variants act in a genomic background with duplicated sub-genomes, and selection may favor certain types of regulatory alleles potentiated by this duplication. In this work, we identified regulatory polymorphisms between two hexaploid bread wheat (2n=6x=42, AABBDD) cultivars by sequencing the transcriptomes of 154 lines derived from a cultivar grown beginning in the 1800s, Red Fife, and a modern cultivar, Stettler. Both cultivars are adapted to the same geographic region, and Stettler is a product of decades of intense selection for grain yield and grain quality. Selection within historical and more recent breeding populations acted upon regulatory loci. Chromosomal domains where selection was weak accumulated regulatory alleles, and regulatory loci target genes were enriched for specific functions. In recent, modern wheat breeding, breeders selected hotspot alleles, and pleiotropic regulatory alleles with many gene targets. Modern cultivar alleles at hotspot regulatory loci both corresponded to known selection targets and led to improved field crop performance. Breeders also selected alleles driving the co-expression of homoeologous genes. Modern cultivar regulatory alleles had consistent effects on homeolog transcript abundances. These alleles also improved field crop performance. By enhancing the favorability of large effect regulatory alleles and expanding the range of gene transcript abundances, genome duplication contributed breeding progress.

D2-86 - Genomic Insights into Aroma Using Whole Genome Sequencing in Apple (*Malus domestica*)

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Aroma significantly influences the quality and consumer preference of apples (*Malus domestica*), one of Canada's most economically valuable fresh fruit crops. However, due to the complexity of aroma, traditional breeding approaches face considerable challenges in effectively selecting for desirable aroma traits. Genomics-assisted breeding enables the selection of apple seedlings at an early stage of development, before the production of fruit, for traits of interest. This approach can be applied to select for genetic markers predictive of aroma-related volatile organic compounds (VOCs), potentially accelerating the breeding of cultivars with improved flavour.

We performed whole genome sequencing (WGS) for 515 unique apple accessions from Canada's Apple Biodiversity Collection, one of the world's most genetically diverse apple collections. We integrated the resulting 20 million single-nucleotide polymorphisms (SNPs) with quantitative measurements of more than 100 VOCs obtained via gas chromatography-mass spectrometry (GC-MS) to perform genome-wide association studies (GWAS). We uncovered significant SNPs and identified novel genomic regions and candidate genes influencing apple aroma production.

These findings not only enhance our understanding of the genetic basis of apple aroma but also provide highly predictive SNP markers that could be used for genomics-assisted breeding, facilitating the development of apple cultivars with superior and diverse aroma profiles.

D2-87 - Weeding Out the Weeds: Genomic tools for improving identification of noxious plant species

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Noxious weed seeds can affect our trade when mixed with commodity seed imports and exports, or our crops when they remain in the soil bank, thereby impacting agriculture. Identification of noxious plants is the most effective strategy to preventing their proliferation. However, due to the morphological similarities between noxious seeds, commodities, and closely related species, phenotypic identification is challenging and time intensive, when having to check thousands of seeds for a target contaminant or having to grow weed seeds to validate their identity. Here we discuss two alternative identification tools to increase accuracy and efficiency - DNA metabarcoding and DNA megabarcoding. DNA metabarcoding allows for the identification of multiple taxa in parallel via high throughput sequencing. It relies on the amplification of short, standardized regions of the genome with enough variation to enable unambiguous species identification. Using jointed goatgrass to pilot our methodology we found that ITS2 and rbcL can distinguish this target weed from wheat and nine of its close relatives. The limit of detection of this technology is being tested by sequencing three dilutions in triplicate of wheat seeds spiked with jointed goatgrass. DNA megabarcoding on the other hand involves sequencing sizeable, medium sized barcodes (up to 10kb) that encompass multiple gene regions in one fragment with long-range sequencing technologies (LST) like Oxford Nanopore Technologies (ONT). We tested this technology by obtaining PCR amplicons (~5kb) for eight *Amaranthus* species, with multiple accessions for most, and sequencing them with ONT's Minlon. These sequences were compared with Illumina generated chloroplast assemblies. Sequence alignments showed 100% species resolution. Overall, our preliminary tests showed that megabarcoding with ONT is another effective identification tool despite the higher error rate in homopolymeric regions. We expect to apply these methodologies to other target weeds including wild mustard, wild oats, and cleavers for more robust testing.

D2-88 - Conservation Challenges and Opportunities for Native Apples (*Malus* spp.) in Canada

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Canada is home to two native apple species—*Malus coronaria* (L.) Mill. in the east and *Malus fusca* (Raf.) C.K.Schneid. in the west—each facing unique conservation challenges. *Malus coronaria* is restricted to a small region of southwestern Ontario and would likely qualify for protection if assessed, given its limited Canadian range and vulnerability to climate change. *Malus fusca*, while more widespread along the Pacific Northwest coast, holds cultural significance for Indigenous Peoples and occurs predominantly in ancestral orchard landscapes that require equitable conservation approaches.

This national review synthesizes ecological, evolutionary, cultural, and policy perspectives to identify conservation gaps and opportunities for *Malus* in Canada. We incorporate species distribution modeling (SDM) to assess recent and projected climatic suitability (environmental niche) under climate change scenarios. Results suggest *M. coronaria* is likely to lose a significant portion of its suitable habitat (across its U.S.-Canada range), with most remaining suitability confined to Canada, while *M. fusca* appears more climatically stable, showing a slight northward and inland shift across its range.

By combining predictive modeling with interdisciplinary insight, we highlight the need for coordinated *Malus* conservation in Canada that reflects both ecological priorities and Indigenous stewardship. This work contributes to national efforts to conserve crop wild relatives and culturally important plant species under climate change. To stimulate adaptation planning, our 20 Calls to Action provide direction for future *Malus* conservation efforts.

CONCURRENT SESSION D3

D3-89 - Metabolic Engineering of Terpene Metabolism in Lavender Glandular Trichomes

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The arial parts of lavender plants are covered with glandular trichomes (GT) or oil glands, which produce and accumulate large quantities of mono- and sesquiterpenes as the main constituents of an economically important essential oil. These trichomes represent excellent bio-factories for the mass production of valuable terpenes through metabolic engineering. We are engineering terpene metabolism in lavender GTs in order to enhance production of certain monoterpenes through enhancing the expression of desired biosynthetic genes using GT-specific promoters, and knocking out the expression undesired genes using CRISPR and antisense technologies. In this presentation, the results of studies aimed at the overexpression of the Bornyl Diphosphate Synthase (BPPS; responsible for the production of monoterpenes camphor and borneol), as well as Green Fluorescent Protein (GFP) and β -glucuronidase (GUS) reporter genes in *Lavandula latifolia* GTs will be discussed. I will also discuss preliminary results elucidating the role of micro RNA molecules in the regulation of isoprenoid metabolism in lavender.

D3-90 - Functions and Phosphorylation-Dependent Regulation of the CCT1 Enzyme in Phosphatidylcholine Biosynthesis and Root Development in Arabidopsis

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Phosphatidylcholine (PC) is a membrane lipid with essential roles in cellular processes and stress responses. Cytidine triphosphate:phosphocholine cytidylyltransferase 1 (CCT1) catalyzes a key step in the *de novo*

biosynthesis of PC, but its functions and regulation are yet to be well elucidated. This study explored the physiological functions and phosphorylation-dependent regulation of Arabidopsis CCT1 (AthCCT1) in PC biosynthesis and root development. We first generated *Atcct1* knockdown/*Atcct2* knockout mutant lines. These lines exhibited significantly decreased PC levels under normal growth conditions and impaired root growth under osmotic stress, which could be rescued by *AthCCT1* overexpression, indicating the functions of CCT1 in the processes. Subsequently, since AthCCT1 is phosphorylated at Serine 187 (S187) by the serine/threonine kinase sucrose nonfermenting 1-related protein kinase 1, we investigated how this phosphorylation affects its biochemical and biological functions. *In vitro* analyses revealed that the S187D phosphomimetic protein mutant of AthCCT1 exhibited reduced lipid-induced conformational changes and lower enzymatic activity than the native protein. Consistently, overexpression of *AthCCT1-S187D* did not rescue stress-induced short root phenotypes in the *Atcct1* knockdown/*Atcct2* knockout lines. In summary, our results demonstrate that AthCCT1 regulates PC biosynthesis under normal conditions and root development under osmotic stress, with its phosphorylation at S187 playing an important role in modulating its enzymatic activity and physiological functions.

D3-91 - Deciphering the role of CYP96T enzymes in the biosynthesis of galanthamine-type Amaryllidaceae Alkaloids from *Hippeastrum papilio*

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Amaryllidaceae alkaloids (AA) are well-known specialized metabolites with versatile antitumor, antiviral, and anti-acetylcholinesterase activities. Among the more than 650 reported AAs, galanthamine has gained particular interest due to its widespread use in treating mild symptoms of Alzheimer's disease. While the biosynthetic pathway of galanthamine has been partially elucidated, key steps remain unresolved—particularly the cyclization of the central intermediate, 4'-O-methylnorbelladine (4'OMN), which leads to the formation of distinct AA scaffolds, including galanthamine-, lycorine-, and crinine-type of alkaloids. These cyclization reactions are catalyzed by cytochrome p450 enzymes (CYP) with distinct regioselectivities, including *para-ortho'*, *para-para'*, and *ortho-para'*. In this study, we identified specific CYP96T enzymes capable of converting the 4OMN into nornarwedine, the precursor scaffold of galanthamine-type AA, via a *para-ortho'* C-C phenolic coupling. To determine whether additional CYP96T isoforms could catalyze such steps, we performed co-expression network analysis and correlated metabolite abundance with gene expression patterns in *Hippeastrum papillio*, a species known for its high galanthamine production. The results suggest the involvement of several CYP96T candidate enzymes in the cyclization of 4OMN. Phylogenetic analysis revealed that these CYP96T candidates belong to the oxidoreductase family and possess a different oxygen-binding motif, A(Q/G)X(NTQ), compared to the superfamily CYP86. Structural modeling and docking simulations were conducted to identify key residues within the substrates-binding sites, providing insights for engineering enzymes increased specificity and catalytic efficiency. The functional activities of the identified CYP96T candidates will be verified through heterologous expression in yeast. This study provides new insights into the enzymatic mechanism underlying galanthamine biosynthesis and lays the foundation for targeted metabolic engineering strategies to enhance its production through synthetic biology approaches.

D3-92 - A Novel Model of the PAL-Mediated Salicylic Acid Biosynthesis Pathway: A Functional Rediscovery of Two Hypersensitivity-Related Genes

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Salicylic acid (SA) is a critical hormone for plant pathogen defense, but its biosynthesis in plants is not completely understood. Two biosynthetic routes to SA are known: the isochorismate synthase (ICS) and the phenylalanine ammonia-lyase (PAL) pathway. Arabidopsis mostly uses the recently elucidated ICS pathway, while most other species, including poplar and tobacco, rely on the PAL pathway. The 2-hydroxylase, which was originally proposed to catalyze the biosynthesis of SA from benzoic acid, has not been identified or characterized in any plant, however. We first generated CRISPR-knockout poplars of a recently characterized benzaldehyde synthase. These plants show reduced benzenoid as well as SA concentrations, yet have

strongly increased benzoic acid levels. The coordinated loss of benzyl benzoate, benzyl salicylate and SA in these transgenics suggests an alternative SA biosynthesis pathway based on benzenoids. This led us to identify a benzyl salicylate-specific carboxylesterase, which we characterized using recombinant enzyme assays. Additionally, virus-induced gene silencing (VIGS) of this gene in *Nicotiana benthamiana* led to reduced hydrolysis of benzyl salicylate to SA in vivo. Based on these data, we propose a new biosynthesis model and provide evidence that benzoyl-CoA is esterified to benzyl benzoate and converted to benzyl salicylate, from which SA is released. We also identified a pathogen-induced gene as a putative benzyl benzoate 2-hydroxylase. Suppression of this gene in *N. benthamiana* reduced the conversion of benzyl benzoate to SA. Phylogenetic analyses indicated that Brassicaceae genomes do not contain the orthologs of either gene, whereas they are present in most other plants. This is consistent with the primary importance of the ICS pathway in Arabidopsis. Our work directly advances our understanding of SA biosynthesis and identifies missing steps in the PAL pathway.

CONCURRENT SESSION D4

D4-93 - Cell-Tissue Specific Expression Does Not Encode RLCK Signalling specificity

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Receptor kinase signalling facilitates a multitude of physiological and biochemical changes in plants in response to environmental and endogenous signals. Receptor kinases (RKs) are proteins located at the cell membrane that perceive ligands to initiate signalling cascades essential for various biological pathways including plant growth, development, and immunity. Though many RKs have been identified, the molecular mechanisms downstream of receptor activation are not fully understood. Receptor-like cytoplasmic kinases (RLCKs) are key components of downstream RK signalling. RLCKs associate with receptor complexes to execute downstream signal transduction via substrate phosphorylation. Despite sequence similarity, analogous mechanisms of activation, and substrate interaction, individual RLCKs can exhibit distinct functionalities and involvement in specific RK pathways. For instance, BIK1 and SGN1 are RLCKs present within the same subfamily, the RLCK-VII/PBL family, however, play central roles in immunity and casparian strip development respectively. Our research focuses on understanding the specificity within RLCK-mediated signalling. By generating promoter-swapped RLCK constructs, we demonstrated that signaling specificity is not solely dictated by RLCK expression patterns. Notably, SGN1 failed to rescue an immune phenotype even when driven by the BIK1 promoter. These findings contribute to our understanding of the molecular signaling networks that regulate diverse physiological and developmental processes in plants.

D4-94 - Biostimulant Potential of Endophytic Bacteria from *Cannabis sativa* and *Chelidonium majus* as Sustainable Alternatives to Agrochemicals

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Endophytic bacteria isolated from *Cannabis sativa* and *Chelidonium majus* were previously screened for their biostimulant and growth-promoting potential using assays for indole production, organic acid production, and nutrient solubilization. Whole genome sequencing identified several isolates as belonging to *Bacillus spp.* and *Pseudomonas spp.* This study evaluates the efficacy of two strains, *Bacillus sp.* (B1) and *Pseudomonas sp.* (P2), in enhancing the growth and productivity of yellow beans (*Phaseolus vulgaris*) under both greenhouse and field conditions. Both strains promoted plant growth, enhanced nutrient uptake efficiency, and improved stress resilience, while also contributing to soil microbiome health by promoting the recruitment of beneficial species as revealed by metagenomics studies. Greenhouse trials revealed significant improvements in morphological health traits and yield in B1-treated plants ($P < 0.05$). Physiological evaluations indicated improvements in photosynthesis, CO₂ fixation, and transpiration efficiency. Root assessments indicated enhanced nitrogen fixation through increased nodulation. Field trials demonstrated consistent yield

improvements across multiple crops, surpassing control groups. These findings position B1 and P2 as promising biostimulants that reduce dependency on chemical inputs, offering an ecologically safe and cost-effective approach to sustainable agriculture.

D4-95 - Genetic Screening with Markers can Assist Backcrossing to Introduce a Novel Trait into Cranberry Dry Bean (*Phaseolus vulgaris*) Varieties

Sajida noor¹, Peter K. Pauls¹, Qijian Song²

¹University of Guelph, ²USDA-ARS

Post-harvest seed coat darkening is a detrimental trait that affects the appearance, taste, and cooking time of several market classes of dry beans, including cranberry beans. The current research was designed to introduce the non-darkening trait, controlled by a homozygous recessive condition for the *j* gene on chromosome 10 in *P. vulgaris*, into high-yielding, regular-darkening cranberry bean varieties, OAC Navabi and OAC Firestripe. The non-darkening variety Wit-rood was crossed with OAC Navabi and OAC Firestripe to introduce the recessive *j* allele. The resulting F₁s were backcrossed with the elite parents to establish non-darkening OAC Navabi and OAC Firestripe backcross (BC) breeding populations. The BC seedlings were screened with a Kompetitive Allele Specific PCR (KASP) marker to identify *Jj* individuals that were used for additional rounds of backcrossing with the elite parents. The recovery of recurrent parent background genomes was assessed by screening genomic DNA from KASP-selected individuals with ~1,000 SNPs with a BARCBean_12K bead chip. Molecular assisted selection of individuals with the non-darkening *j* allele and the recurrent parent alleles for the background markers can facilitate the production of non-darkening cranberry beans that have not lost the quality traits of the recurrent parents.

D4-96 - Ethylene-mediated sexual plasticity as an example of plant developmental plasticity in *Cannabis sativa*

Adrian S Monthony¹, Julien Roy¹, Olivia Carlson², Susan J Murch², Davoud Torkamaneh¹

¹Université Laval, ²University of British Columbia

Cannabis sativa L. is dioecious with an XX/XY chromosome system. Typically, its floral sexual phenotype is concordant with genetic sex. However, under certain conditions, the plant develops sex organs opposite to its genetic sex, a phenomenon known as sexual plasticity. This developmental plasticity can result in male flower production, pollination and ultimately yield losses for commercial producers, who rely on unpollinated female flowers for cannabinoid production. Despite the consequences of this phenomenon, its genetic basis remains underexplored, leaving control of cannabis sexual expression out of the hands of producers and breeders.

Ethylene, and its inhibitors are known to induce sexual plasticity, but the role of ethylene-related genes (ERGs) in controlling these shifts is unclear. We employed a multi-omic approach to investigate the phenomic, genomic, transcriptomic, and metabolomic basis of sexual plasticity. Phenotypic analysis of three *C. sativa* genotypes demonstrated consistent male-to-female transitions induced by ethylene and female-to-male transitions triggered by ethylene inhibitors. Whole-genome sequencing identified ERGs with minimal genetic variation and no direct functional mutations. Transcriptomic data revealed a temporal and sex-dependent pattern of expression in candidate ERGs, indicating that the initiation and maintenance of sexual plasticity are regulated by distinct ERGs over time and across sexes. Metabolomic profiling quantified ethylene precursors, validating gene expression patterns and supporting a biochemical model of sexual plasticity.

This research offers a detailed model of how ethylene and its related genes regulate sex determination and plasticity in *C. sativa*, with potential implications for understanding similar processes in other dioecious plant species. Our findings enhance understanding of cannabis sex expression and offer a foundation for targeted breeding strategies to develop sex-stable varieties, with broader implications for other dioecious plants.

ABSTRACTS

POSTER SESSIONS

Sunday, August 10th, 2025

POSTER SESSION I

P1-97 - Interrogating *Zea mays* F1 chromatin accessibility inheritance to uncover genetic trans-regulatory relationships

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Crop heterosis is the widespread over-dominant inheritance of many yield-associated traits that follows crossing certain genetically distant inbreds. Heterosis magnitude varies and is genetically determined. However, despite being a cornerstone of *Zea mays* (maize) productivity, the genetics influencing strong heterosis remain unclear. F1 plants retain the transcription factors (TF) binding sequences, or *cis*-regulatory elements, exactly as found in the inbred state, albeit at a halved dose. Rather it is the transcriptional *trans*-regulatory environment, including TF variation, that differs in the heterotic F1. The utilization of *cis*-regulatory elements requires accessible, nucleosome-free DNA for most TF binding. Therefore, to determine how chromatin accessibility varies in F1 maize, we measured seedling chromatin accessibility from three inbreds (Oh43, B73 and Ki3) and their F1 progeny via Assay for Transposase Accessible Chromatin sequencing (ATAC-seq). Using a concatenated reference approach, we comprehensively interrogated their accessible chromatin regions (ACRs), uncovering haplotype shared, unique and duplicated ACRs. By comparing chromatin accessibility between parents and offspring we categorized the mode of inheritance (additive, dominant, under/over-dominant) for ACRs genome-wide. Although, chromatin accessibility was largely inherited additively, dominant and over-dominant ACR inheritance was most common in the F1 exhibiting strong heterosis. Finally, exploiting F1 *cis*-regulatory conservation and allele-specific chromatin accessibility, we uncovered ACRs with the hallmarks of differential F1 *trans*-regulation - these differentially *trans*-regulated ACRs were enriched for over-dominant or dominant chromatin accessibility inheritance. Moreover, we discovered haplotype bias in the F1 *trans*-regulation that co-occurs with strong agronomic heterosis, providing mechanistic insights into the genetic architecture behind strong hybrid vigour. Future work aims at using a tissue and environmentally matched diversity panel to genetically map the specific *trans*-regulators underpinning this heterosis, which will help inform the breeding and engineering of good heterosis into novel inbred backgrounds.

P1-98 - Bio-Analytic Resource for Plant Biology

Asher Pasha¹, Nicholas Provart¹

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The BAR is a widely-used, open-access plant biology 'omics database resource. The BAR was designated as a Global Core Biodata Resource by the Global Biodata Coalition in 2023. The BAR consists of 100s of databases, including those for gene expression levels, protein-protein interactions, and protein structures. The data in the BAR databases are provided to researchers via modern web applications such as eFP Browsers, ePlants, ThaleMine and a BAR API. The BAR's GAIA tool can search genes, BAR tools, more than ten thousand genetic models from the literature, and undertake BLAST searches. Feedback from researchers has helped develop and improve the BAR apps. The BAR will work as part of recently-awarded 'Centre for Sustainable Plant Innovation and Resilience through International Teamwork' effort to develop a database for plant single cell data. The BAR is celebrating its 20th anniversary in 2025.

P1-99 - A simple method to create structural variation in plants

Naiyara Ahsan¹, PRV Satyaki¹, Aysha Hussain¹

¹Cell and Systems Biology, University of Toronto

From Donut Peaches to improved drought response in Maize, the introduction of genomic alterations, known as Structural Variants (SV), can yield desirable phenotypes in plants. Current methods of generating SVs such as treatment by gamma-irradiation are often inaccessible due to limited facilities. In this study, using *A. thaliana* as a model, we demonstrate that treatment with an accessible and affordable drug, etoposide, is an effective method for the induction of SVs. Additionally, etoposide mutagenesis also demonstrated a large effect size with distinct visible phenotypes, as well as heritability of mutations. Computational exploration of SVs poses challenges of their own, particularly when limited by sequencing techniques with short reads. We utilized a pipeline which detects high-confidence SV events, by filtering false positive calls arising from mapping artifacts and visually inspecting calls on a genome browser. To identify SVs too large to be detected accurately by short-read SV callers, we employed a read-depth based approach to detect deletions and duplications spanning megabases. Finally, we found that etoposide-treated plants do not exhibit a significant difference in single nucleotide variant (SNV) count or distribution compared to controls.

P1-100 - Transposons expression landscape in seed tissues

Alefiyah Bahrainwala¹, PRV Satyaki¹

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Transposable elements (TEs) or transposons are repetitive DNA sequences in the genome that can increase their copy number within the genome. TE families make up approximately 21% of the *Arabidopsis thaliana* genome, and their insertion can mutagenize genes and induce ectopic recombination. TEs are therefore highly regulated and silenced by DNA and histone methylation. In this study, we describe TE expression in different seed and gametic tissues. In particular, we hypothesize that the endosperm, whose genome is hypomethylated during development, has a high TE activity. We test this hypothesis and explore molecular players controlling TE expression in the endosperm.

P1-101 - Examining co-inheritance of reproductive and vegetative traits in hybrid grapevines

Alison Purcell¹, Zoë Migicovsky¹, Peter Cousins²

¹Acadia University, ²Gallo Winery

Grapevine (*Vitis* spp.) is one of the most economically significant and widely cultivated fruit crops globally. Most species produce lateral determinant meristems (tendrils and inflorescences), which are found opposite the leaf at the node, at two successive nodes, followed by a skip (a node lacking a tendril or an inflorescence). However, the position of the first lateral meristem and the length between nodes (internode length) can vary among individuals. These traits may influence canopy structure, reproductive potential, and overall vine performance.

In this study, we examined tendril development and other vegetative traits including shoot growth, internode length, leaf size, and chlorophyll content, across eight hybrid *Vitis* crosses. These crosses also segregated for two reproductive traits, flower sex (perfect vs pistillate) and fruit colour (black skin berries with red flesh vs all other berry colour categories) which were determined in the seedlings using molecular markers. We measured ~500 seedlings to examine how these reproductive traits influenced early vegetative development, in particular tendril quantity and distribution.

Our results offer insights into how vegetative traits may be co-inherited with reproductive traits. Ultimately, this work contributes to our understanding of vegetative traits in grapevine and can be used to help inform targeted breeding strategies for hybrid grapevine production.

P1-102 - An evaluation of genomic prediction for barley improvement

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¹University of Guelph, ²Université Laval, ³Agriculture and Agri-Food Canada

Increases in cereal grain crops' yields and yield stabilities have been critical for food security and are primarily due to the development of new varieties. The first step in generating a new variety is to cross two parents with desirable traits. Each cross generates individuals within the same family. These individuals are then repeatedly self-pollinated to generate near pure-breeding lines. Breeders estimate lines' genetic yield values from one or two plots in a single location, termed preliminary yield trials. A small number of the lines considered best are then grown in multiple locations, termed advanced yield trials. These trials' lines that are judged genetically superior to current varieties become new varieties. Genomic prediction, where a line's yield is estimated from its DNA profile, is an alternative or supplemental method to estimate genetic value. In this study, genomic predictions of a line's future performance were compared to predictions generated from its family membership and from its field-trait values. Compared to genomic prediction, family membership was an equivalent or better predictor of lines' yields. However, predictions combining both genomic data and family membership were the most accurate. Genomic predictions captured variation due to Mendelian sampling, the segregation of alleles within a family. The change in means between preliminary and advanced trial lines suggested similar yield improvements using phenotypic and genomic selection.

P1-103 - Elucidation of the UDP-glycosyltransferase(s) Involved in Phenolic Glycosylation in Trees

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Lignin is essential for plant growth and development, as it confers strength, rigidity, and hydrophobicity to plant cell walls and aids in the transport of water and nutrients through the vasculature. This vital polymer also inherently creates a significant obstacle for industrial processing of lignocellulosic biomass. Poplar trees are models for studying woody plant development. Moreover, they have been the subject of numerous gene editing initiatives aimed at modifying lignin, including several studies targeting the inclusion of non-canonical monolignols by strategically expressing exogenous genes, originating from plant and bacterial origin. Such efforts clearly demonstrate that the target metabolites can be incorporated into the cell walls, while notable amounts of their corresponding glycosides accumulated in the vacuoles—a common occurrence in angiosperms. The changes in lignin deposition are partially influenced by the extent of glycosylation and the magnitude of monolignol glycosides storage in the vacuole, a process involving one or more UDP-glycosyltransferases (UGTs). To elucidate which UGT(s) govern the glycosylation and storage of the exogenous non-canonical monolignol influencing the spatial and temporal distribution of lignin in trees, we employed PacBIO sequencing to build a haplotype-resolved assembly and annotation genome for a highly transformable hybrid poplar genotype, P39 (*P. alba* × *P. grandidentata*). Using the assembled reference genome, we performed RNA-seq to examine the expression patterns of UGTs within the developing xylem of wild-type and five different uniquely transformed genetically modified P39 poplar lines. More than 40 UGTs were significantly upregulated in the transgenic poplar lines, including 27 associated with 'anthocyanidin 3-O-glucosyltransferase activity,' which was among the top 20 significantly enriched GO terms for differentially expressed genes. As a next step, using the newly assembled genome, we plan to generate knockout lines for select candidate UGT genes to evaluate how UGT-mediated glycosylation affects vacuolar storage of phenolic glycosides, lignin structure, and plant phenotype.

P2-104 - Three-step biosynthesis of salicylic acid from benzoyl-CoA in plants

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¹UBC, ²Sichuan University

Salicylic acid (SA) is a small molecule historically recognized for its therapeutic use in humans, originally extracted from willow bark and later synthesized as aspirin—the most extensively used drug worldwide. In plants, SA plays a central role as a signaling hormone that activates immune responses against pathogen attack. While the biosynthetic pathway of SA has been elucidated in *Arabidopsis thaliana*, where it originates from chorismate, the mechanisms underlying SA production in other plant lineages remain poorly understood.

Here, we reveal a conserved SA biosynthesis pathway that operates widely across seed plants. Using *Nicotiana benthamiana* as a model system, we identified three enzymatic steps responsible for SA production, distinct from the *Arabidopsis*-based pathway. First, the enzyme BEBT catalyzes the condensation of benzyl alcohol with benzoyl-CoA to form benzyl benzoate. This compound is then hydroxylated by BBO to yield benzyl salicylate, which is subsequently cleaved by BSH to release free SA.

Genetic and biochemical evidence, including gene knockouts, complementation assays, and *in vitro* enzymatic assays, demonstrates the functional relevance of each step in the pathway. Moreover, homologous genes encoding BEBT, BBO, and BSH are present in a wide range of plant species, indicating that this pathway is evolutionarily conserved and broadly utilized in the plant kingdom.

Our findings uncover a previously unrecognized and widely distributed route for SA biosynthesis in plants, shedding new light on the evolutionary diversity of plant metabolic pathways and immune signaling mechanisms.

P2-105 - Class IV plant U-box proteins function redundantly to optimize protein accumulation of receptor-like cytoplasmic kinase BIK1

Virginia Miguel¹, Ruoqi Dou¹, Lauren Grubb¹, Mansuba Rana¹, Brandon Saltzman¹, Jacqueline Monaghan¹

¹Queen's University

Plant immune responses must be tightly regulated to avoid detrimental effects on growth and fitness. In *Arabidopsis thaliana*, the kinase BIK1 serves as a key signaling hub downstream of several immune receptors and is subject to post-translational control to prevent excessive immune activation. Previous studies have identified class IV plant U-box (PUB) E3 ligases as negative regulators of immunity. The class IV PUBs PUB25 and PUB26 are phosphorylated by the calcium-dependent protein kinase CPK28 and polyubiquitinate BIK1, targeting it for proteasomal degradation. We hypothesized that additional class IV PUBs contribute redundantly to BIK1 turnover. Using *in vitro* phosphorylation and ubiquitination assays, we demonstrate that PUB22 and PUB24 are also phosphorylated by CPK28 and can poly-ubiquitinate BIK1. In a quintuple mutant *pub22 pub23 pub24 pub25 pub26*, we observed strongly enhanced immune-triggered ROS production and increased resistance to the bacterial pathogen *Pseudomonas syringae*. Moreover, BIK1 turnover is strongly reduced in *pub22 pub23 pub24 pub25 pub26* protein extracts compared to wild-type. These findings support a model in which multiple class IV PUBs act redundantly to regulate BIK1 stability and fine-tune immune output in plants.

P2-106 - Purification and Characterization of Methionine Aminotransferase from the Microalgae *Tetraselmis*

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Dimethylsulfoniopropionate (DMSP) is a key algal metabolite enzymatically synthesized from methionine by a 4-step pathway. Algae synthesize DMSP in massive quantities and appear to use DMSP for osmoprotection and scavenging reactive oxygen species to cope with environmental stresses. After release into the ocean, bacteria use DMSP as a carbon and sulfur source, yielding dimethylsulfide (DMS) as a byproduct. Once volatilized, DMS constitutes ~50% of the global atmospheric sulfur budget and may play a role in global climate regulation. As the precursor to DMS, key DMSP biosynthesis enzymes have recently been discovered.

Isotope labelling and radiometric enzyme assay were used to study the DMSP pathway in *Ulva intestinalis* macroalgae and *Tetraselmis* microalgae. This macroalgae initiates DMSP synthesis using a L-methionine:2-oxoglutarate aminotransferase (Met AT, E.C. 2.6.1.88). The same pathway was suggested in *Tetraselmis*, but the gene(s) that encode it remain unidentified. The involvement of a Met oxidase was also proposed. With the release of several *Tetraselmis* -omic databases, the Waller group has conducted bioinformatic investigations of *Tetraselmis* microalgae. The development of high-throughput Met AT assays alongside these results, immunological and enzyme assays suggest that another type of Met AT is present in this DMSP-producing species, distinct from that in *Ulva*. To uncover the gene encoding this enzyme, we have purified the *Tetraselmis* Met AT. Progress in characterizing this enzyme using various protein biochemistry techniques will be presented.

P2-107 - Targeted Modification of Essential Oil in *Lavandula latifolia* Using CRISPR/Cas9

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Lavenders (*Lavandula*) are commercially significant crop plants cultivated globally for their multifunctional essential oils (EO), which are constituted mainly of monoterpenes. Lavender EOs used in cosmetics and alternative medicine products are negatively impacted by elevated concentrations of undesirable off-odor monoterpenes borneol and camphor. The biosynthesis of these metabolites begins with the conversion of geranyl diphosphate (GPP), to bornyl diphosphate (BPP), catalyzed by the enzyme bornyl diphosphate synthase (BPPS). Further enzymatic steps transform BPP into Borneol, which is then dehydrogenated to produce camphor. This investigation aims to determine whether the biosynthesis of borneol and camphor may be halted or substantially reduced through knocking out the *BPPS* gene. In this study, we utilized the CRISPR/Cas9 genome editing platform to knock out this gene in *Lavandula latifolia*, which produces large amounts of borneol and camphor. We identified and selected highly specific guide RNA (gRNA) molecules that target the *BPPS* gene. Plasmids carrying these gRNAs along with the Cas9 nuclease cDNA were then constructed and introduced into *L. latifolia* plants using *Agrobacterium*-mediated transformation. Transformants are currently undergoing molecular analysis, including sequencing, to confirm mutations in the *BPPS* gene. Additionally, preliminary EO analysis using gas chromatography-mass spectrometry (GC-MS) suggested a significant decrease in borneol and camphor levels in the EO of some transformants, indicating that the *BPPS* gene was successfully knocked out. These results demonstrate that the CRISPR/Cas9 technology can be used to knock out undesired genes in order to improve the quality of lavender oil.

P2-108 - Investigation into the biochemical properties, downstream targets, and physiological roles of *Arabidopsis thaliana* calmodulin-like proteins-26 and -27

Brittany Menard¹, John Filipic¹, Kyle Symonds¹, William Plaxton¹, Wayne Snedden¹

¹Queen's University

Ca²⁺ ions are ubiquitous second messengers in eukaryotes that have been implicated in most plant (a)biotic stress signaling pathways. Ca²⁺ signals are decoded by sensor proteins, which bind Ca²⁺ with a high affinity and specificity to activate downstream target effector proteins and execute the appropriate physiological response to the perceived stimulus. There are 3 main families of plant Ca²⁺ sensors, of which calmodulin (**CaM**) and CaM-like proteins (**CMLs**) are the largest. While CaM is one of the most evolutionarily conserved proteins across eukaryotes, CMLs are unique to plants and some protists. Although CMLs are predicted to be regulatory proteins capable of propagating stress-induced Ca²⁺ signals similar to CaM, many have yet to be characterized. Of the 50 CML isoforms found in *Arabidopsis thaliana*, CML26 and CML27 are close paralogs (72% sequence i.d.) predicted by transcriptomic and proteomic databases to be the most highly and broadly expressed. However, despite their high level of expression, their functions have not been reported. Thus, our objective is to assess the biochemical properties, downstream targets, and physiological roles of CML26 and 27. Recombinant CML26 and 27 was heterologously expressed in *E. coli* and purified to near homogeneity. Biochemical analysis revealed that, like CaM, CML26 and 27 bind Ca²⁺ with a high affinity and undergo a conformational change for interaction with downstream targets. Work is in progress to determine if changes in

the phosphorylation status of each CML influences their biochemistry, subcellular localization and/or interaction with putative targets. Qualitative and quantitative phenotypic analyses are also underway, comparing *Arabidopsis* wild-type plants to *cm126* and *cm127* T-DNA knockout mutants under a variety of abiotic stress conditions. Results of this research will contribute to our understanding of Ca^{2+} -induced signal transduction events, and the biological significance of reversible phosphorylation in the post-translational control of plant CMLs.

P2-109 - Uncovering Aggressive Ammonium Uptake via Intracellular Nitrogen Storage in *Chlamydomonas reinhardtii*

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Bioavailable nitrogen (N) is a key limiting factor for global biomass production. In fluctuating environments, organisms must employ effective nitrogen use strategies (NUS) to exploit transient N availability. The recent discovery of intracellular guanine crystals in diverse algal species(1,2) raises intriguing questions about their roles in N storage and supply. To investigate NUS in *Chlamydomonas reinhardtii* as a model for photosynthetic eukaryotes, we compared three ubiquitous N sources—ammonium, nitrate, and urea—by analyzing biomass-to-N conversion efficiency and batch-culture growth kinetics under mixotrophic conditions. Saturated growth was achieved at 4 mM ammonium, 4 mM nitrate, and 2 mM urea, indicating equivalent molar efficiency of N utilization and suggesting that growth was limited by factors other than N availability. To assess N uptake relative to biomass production, we monitored residual N in the medium at mid-log and stationary phases. Nitrate- and urea-fed cultures exhibited near one-to-one conversion of extracellular N to biomass. In contrast, ammonium-fed cultures rapidly depleted external ammonium well before reaching maximum biomass, yet continued to grow, accumulating up to fourfold more biomass after ammonium was mostly exhausted in the medium. These findings support a model in which *C. reinhardtii* aggressively acquires ammonium during exponential growth and stores it intracellularly, enabling sustained biomass production following episodic N availability. Ongoing comparative transcriptomic analyses aim to identify N-source-dependent regulation of N metabolism and elucidate the molecular bases of intracellular N storage. This work uncovers a previously unappreciated NUS in microalgae and offers potential strategies for enhancing N resilience in crop systems.

P2-110 - Application of exogenous ergothioneine as a strategy to limit the postharvest senescence of broccoli

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Green vegetables tend to undergo senescence-related yellowing during postharvest handling. The mushroom-derived antioxidant ergothioneine limits discolouration of meat products and the postharvest browning of arugula, but its impact on other vegetables is unknown. Here, broccoli heads were dipped into one of four ergothioneine solutions (0, 20, 100, or 500 μM) prior to storage at 10 °C for up to 10 days. HPLC-DAD analysis revealed the exogenously supplied ergothioneine was distributed throughout the broccoli head, and in the greatest abundance on day 0 of storage but declined thereafter. Floret yellowing was evident in most broccoli heads by day 7 of storage but was delayed by 3 days in broccoli treated with 20 μM ergothioneine. Total chlorophyll concentrations decreased with storage, but to a lesser degree in broccoli treated with 20 μM ergothioneine. We also profiled the temporal changes of oxidative stress-related metabolites in the stored broccoli. This included H_2O_2 , ascorbate, glutathione, and pyridine dinucleotide-related metabolites. H_2O_2 increased with storage, and the greatest accumulation was in broccoli treated with 100 μM and 500 μM ergothioneine. The ergothioneine dip treatments affected the temporal fluctuations in the concentrations of ascorbate, dehydroascorbate, glutathione, glutathione disulfide, and the reduced and oxidized forms of pyridine dinucleotides. Interestingly, broccoli treated with 20 μM ergothioneine had greater ratios of ascorbate/dehydroascorbate and glutathione/glutathione disulfide and higher activities of glutathione reductase and dehydroascorbate reductase relative to the other dip treatments. Conversely, a smaller increase in ascorbate peroxidase activity was apparent in 20 μM ergothioneine-treated broccoli than the other treatments. Moreover, the temporal alterations in the pyridine dinucleotide ratios (e.g., $\text{NADPH}/\text{NADP}^+$) were also impacted

by the antioxidant treatments. Taken together, the application of 20 μM ergothioneine delayed postharvest senescence of broccoli by limiting the accumulation of the reactive oxygen species H_2O_2 and boosting the operation of the endogenous ascorbate-glutathione recycling pathway.

P2-111 - Volatile emissions, temperature, and chlorophyll fluorescence as indicators of apple maggot injury in 'Honeycrisp' apple fruit.

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¹Agriculture and Agri-Food Canada

Apple maggot (*Rhagoletis pomonella*) (Walsh) is a detrimental pest to apple fruit (*Malus x domestica* Borkh.) in commercial orchards located in eastern Canada and the northeastern and midwestern United States. Additional non-destructive indicators for maggot injury could be useful for identifying compromised apples that may have reduced quality and a shortened storage life. Changes in volatile emissions, chlorophyll fluorescence, and/or surface temperature may serve as useful early stress indicators. 'Honeycrisp' fruit collected from the experimental orchard at the Kentville Research Station was assessed using chlorophyll fluorescence, infrared thermography and profiled for volatile organic compounds and then characterized for the number of stings and for maggot development. There were no differences in the quantum yield of photosystem II or surface temperature on the green side of 'Honeycrisp' fruit. Interestingly, fruit that acquired a high number of stings had a lower quantum yield of photosystem II and a higher surface temperature. This corresponded with a greater number of larvae present on the red as opposed to the green sides of an apple. More than 50 volatile organic compounds were identified using headspace solid phase microextraction and analysis with gas chromatography x gas chromatography-time of flight-mass spectrometry. A greater number of stings on a fruit resulted in a decline in volatile organic compounds associated with apple aroma such as 1-hexanol and 1-butanol. Whereas fruit with a greater number of stings produced more stress-induced volatile organic compounds such as ethanol and ethyl-acetate. Future work will characterize differences in bi-colour apple fruit chemistry that may influence apple maggot 'side preference'.

P2-112 - Engineering plant phenolic metabolites via overexpression of microbial feruloyl-CoA synthetase in *Arabidopsis thaliana*

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In living organisms, coenzyme A (CoA) plays an important role in activating metabolites by forming a high-energy thioester bond between the pantetheine moiety of CoA and a carboxyl group of diverse metabolites. The biosynthesis of CoA thioesters requires acyl-CoA synthetases/ligases, which catalyze the ligation of CoA onto the carboxyl group of acids. In plants, many acyl-CoA ligases are involved in, but not limited to, the biosynthesis of secondary metabolites, including lignins, lignans, and flavonoids, while analogous ligases in microbes participate in catabolic pathways—implying different kinetic and substrate specificities between microbial and plant ligases. Given these differences, heterologous expression of microbial CoA ligases *in planta* may offer potential for uniquely modifying plant metabolic pathways.

In this study, we investigated the impacts of overexpressing a microbial CoA ligase in *Arabidopsis*. The *ferA* gene from *Sphingomonas paucimobilis* SYK-6, which encodes a feruloyl-CoA synthetase, was selected based on its *in vitro* activity toward plant-related phenolics in a cell-free wheat germ assay. To characterize the *in vivo* function of feruloyl-CoA synthetase, the *ferA* gene was cloned into a plant expression vector under the control of the 35S promoter and transformed into wild-type *Arabidopsis thaliana* Col-0. Methanolic extracts were subsequently analyzed using UHPLC, revealing differences in phenolic metabolite levels between transgenic lines and wild-type plants. Additionally, sinapic acid, released by alkaline hydrolysis of methanolic samples, was found to be more abundant in transgenic lines, suggesting that exogenous feruloyl-CoA synthetase may affect the native phenylpropanoid pathway. These findings highlight the potential of microbial CoA ligases as tools for engineering plant secondary metabolites, with possible implications for improving the industrial value of plant-based products.

P2-113 - Role of a gibberellin 3-oxidase gene in regulating photosynthetic traits in wheat

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Wheat is an important staple crop and plays an indispensable role in ensuring global food security. However, due to limited arable land and environmental challenges, the current yield growth rates are insufficient to meet future demand. Photosynthesis, a primary determinant of crop yield, is highly influenced by canopy leaf area index, leaf light interception and absorption, and CO₂ fixation. Gibberellin (GA), a classical phytohormone, regulates a wide range of crop morphological and physiological traits, such as tiller formation and leaf expansion that influence canopy photosynthetic capacity, and GA 3-oxidase (GA3ox) is a key regulator of GA biosynthesis in plants. This study investigated the role of *GA3ox* gene in regulating traits associated with photosynthesis in wheat using *taga3ox* mutant. Mutation in *GA3ox* resulted in 27% reduction in leaf area and a 1.7-fold increase in the number of tillers. Treatment of the *ga3ox* mutant plants with bioactive GA reversed these phenotypes to wild type (WT). Average leaf absorbance from 520-600nm (photon flux density) was 76% in mutants, followed by 74% in WT and 71% in GA-treated mutants. Compared to WT, the mutant also showed 13%, 16%, and 18% higher CO₂ assimilation rate at 800, 1600 and 2000 $\mu\text{mol m}^{-2} \text{s}^{-1}$ (ePPFD, 400-750nm), respectively. Furthermore, the maximum rates of carboxylation (V_{cmax}) and electron transport (J_{max}) values were higher (9-11% and 7-8%, respectively) in the mutant relative to WT and the GA-treated mutant, suggesting increased Rubisco activity. Overall, the findings of this study highlight the potential of *GA3ox* gene as a target for improving photosynthetic traits in wheat.

P2-114 - Investigating the roles of *Arabidopsis thaliana* calmodulin-like protein 46 during abiotic stress

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Plants rely on signaling pathways to perceive and respond to environmental stresses. Calcium (Ca²⁺) is an example of a key second messenger regulating many of these pathways. To interpret Ca²⁺ signals, calcium sensor proteins are required. Our lab's research focuses on calmodulin and calmodulin-like proteins (CMLs). While some *Arabidopsis* CMLs have been shown to act as Ca²⁺ sensors and have been implicated in stress adaptation, the functions of many remain unknown. My research investigates the role of CML46, a largely uncharacterized CML, in abiotic stress response.

Transcriptomic analysis from online databases indicates that *CML46* expression is induced under cold, salt, and UV stress conditions, suggesting a potential role in abiotic stress response. To explore its function, phenotyping experiments were conducted on *Arabidopsis cml46* knockout mutants under control, NaCl, and abscisic acid (ABA) treatments. Results reveal that *cml46* mutants exhibit accelerated seed germination and establishment compared to wild-type (WT) plants across all tested conditions. This phenotype suggests that CML46 may be playing an important role in germination that is not specific to stress conditions. Seed coat permeability analysis indicated that *cml46* mutants behave like WT plants. Assays are underway to assess whether *cml46* mutants have altered sensitivity to stratification during seed development.

CML46 is predicted to have an N-terminal transmembrane region, a property which is very uncommon among CMLs. Biochemical characterization of CML46 Δ TM (lacking the predicted transmembrane domain) showed that the protein exhibits atypical properties compared to classic Ca²⁺ sensors. CML46 Δ TM does not bind to phenyl-sepharose (hydrophobic interaction chromatography) and does not undergo an electrophoretic mobility shift in the presence of Ca²⁺, although fluorescence assays indicate some conformational changes in response to Ca²⁺. Confocal microscopy studies on CML46-GFP and CML46 Δ TM-GFP are underway to determine the protein's subcellular localization.

These findings suggest CML46 is a unique Ca²⁺-binding protein with potential roles in germination.

P2-115 - LC-HRMS based untargeted metabolomics in exploring how the metabolic profile of pea (*Pisum* sp.) varies with seed age

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The pea is one of the oldest domesticated legume crops, and it is still widely cultivated to this day. The plant is largely farmed for the edible seeds and seed pods, however it also has use as a forage crop and a green manure. The seeds are high in nutrition, containing high levels of protein as well as beneficial secondary metabolites such as phenolics. To explore how these metabolites vary as the seed ages, pea seeds from ten different cultivars at five different ages were characterized using a HPLC-MS untargeted metabolomics approach. The BioCyc database was used to identify potential biosynthetic pathways of interest, and various online and in-house databases were used for compound identification. After filtering the data, 4239 compounds with MS2 data and a potential database match were detected, and a selection of the most abundant compounds were manually evaluated to confirm the assignments provided by the software. These selected metabolites belonged to a variety of different compound classes, including flavonoids, coumarins, amino acids, and soyasaponins. Interestingly, the majority of these metabolites decreased in abundance as the pea seed aged except for soyasaponins and some others, which increased with age in all cultivars. A similar trend was observed in the biosynthetic pathways that were explored using BioCyc, with the majority of them decreasing with seed age. A detailed analysis of the varying metabolites, with a focus on flavonoids and saponins, will be presented in this work.

P2-116 - Genetic variation in induction and maintenance of seed dormancy in wheat is mediated by alteration in the balance between ABA and GA signaling

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Induction and maintenance of dormancy are physiological processes that occur during seed maturation; however, much less is known about the mechanisms regulating these events in wheat. This study showed that seed maturation in wheat is characterized by decreases in the levels of abscisic acid (ABA) and gibberellin (GA) levels irrespective of seed dormancy level and the decreases in ABA and GA levels are mediated via expression of specific ABA (*TaCYP707A1*) and GA (*TaGA3ox2*, *TaGA2ox3* and *TaGA2ox6*) metabolism genes. Consequently, the ABA to GA level ratio exhibited a decrease during seed maturation in both highly dormant and low-dormant seeds and no difference was observed in the ratio of ABA and GA levels between the two seed samples. Our data, however, revealed that variation in induction and retention of dormancy during seed maturation is closely associated with modulation of the balance between ABA and GA signaling mainly via expression of specific genes encoding ABA and GA signaling components that act as positive regulators seed response to ABA (*TaPYL5* and *TaABI5*) and GA (*TaGAMyb*). In agreement with these results, the highly dormant and low-dormant seeds exhibited substantial variation in their sensitivity to ABA and GA during their maturation. The findings of this study highlight that a shift in balance between seed sensitivity to ABA and GA independent of a shift in balance between their levels can mediate genetic variation in induction and maintenance of dormancy during wheat seed maturation.

P2-117 - CRISPR-Cas9: A Powerful Tool for Monoterpene Indole Alkaloid Biosynthesis

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Monoterpene indole alkaloids (MIAs) constitute an extensive category of natural compounds highly valued for their medicinal attributes, holding a pivotal role in pharmaceutical research. These compounds originate from the common precursor, strictosidine. The removal of the glucose moiety from strictosidine yields strictosidine aglycone, which subsequently undergoes an enzyme-catalyzed reduction, leading to the formation of

numerous diverse alkaloid intermediates. In this study, CRISPR-Cas9 gene editing technology was employed to integrate several strictosidine aglycone reductases into the genome of a yeast strain previously engineered to produce the substrate strictosidine aglycone. Successful genomic integration was confirmed by detecting the strictosidine aglycone reduction products using liquid chromatography-mass spectrometry (LC-MS). Through the genome integration of the corresponding enzymes, the modified yeast has demonstrated the capability to produce ajmalicine, geissoschizine, and rauwolscine alkaloids. These findings highlight the significant promise of utilizing CRISPR-Cas9 for the future biosynthesis of a multitude of pharmaceutical compounds.

P2-118 - Investigating Transporter Candidates for Indole Alkaloid Movement and Exploring Alkaloid Trafficking

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Monoterpenoid indole alkaloids (MIAs) are a diverse class of pharmacologically active compounds produced by plants like *Catharanthus roseus*, including clinically important drugs like vinblastine and vincristine. The biosynthesis of MIAs is remarkably complex, involving more than 28 enzymatic steps and spanning multiple cell types, such as the leaf epidermis, idioblasts, and internal phloem-associated parenchyma (IPAP) cells. These spatially distributed biosynthetic steps necessitate a sophisticated transport system for intermediates and final products, yet the molecular identities of the associated transporter proteins remain largely unknown.

This study aims to identify and characterize membrane transporters involved in the intra- and intercellular trafficking of MIAs in *C. roseus*. Using tissue-specific transcriptomic datasets, we profiled transmembrane proteins and identified candidate genes enriched in MIA-producing tissues. Candidates were prioritized based on sequence homology, expression pattern, and phylogenetic relatedness to known transporter families (ABC, MATE, NPF, and PUP). Functional validation is underway using virus-induced gene silencing (VIGS), metabolite profiling via liquid chromatography mass spectrometry (LC-MS), and gene expression analysis. Upon confirmation of candidate transporters, their subcellular localization will be assessed using fluorescent protein fusions, while transport activity will be evaluated in *Xenopus laevis* oocytes.

By elucidating the missing transporters in MIA trafficking, this research addresses key knowledge gaps in plant-specialized metabolism. Completion of the project will lay the groundwork for future metabolic engineering strategies to enhance alkaloid yields in planta or heterologous systems.

P2-119 - Mapping the Subcellular Compartmentalization of Enzymes in Monoterpene Indole Alkaloid Biosynthesis

[Hannah Tran](#)¹, Yang Qu²

¹Graduate Student, ²Principal Investigator

Monoterpene Indole Alkaloids (MIAs) constitute a large and diverse group of specialized plant metabolites known for their potent biotherapeutic properties and high drug candidacy rates. This class includes clinically significant compounds such as vinblastine and camptothecin, used in chemotherapy, and reserpine, a treatment for hypertension. MIA biosynthesis is characterized by intricate enzyme compartmentalization across various cell types and subcellular organelles, including the vacuole, nucleus, endoplasmic reticulum, and peroxisome. Recent discoveries have expanded our understanding of MIA biosynthetic enzymes, including cytochrome P450 monooxygenases, berberine bridge enzyme-like oxidases, reductases, and methyltransferases. To further investigate the subcellular localization of these enzymes, we employed fluorescent tagging combined with transient expression in *Nicotiana benthamiana*. This approach aims to provide deeper insights into MIA biosynthesis and trafficking, clarifying how compartmentalization influences metabolic flux. A better understanding of these spatial dynamics will facilitate targeted engineering strategies to enhance the production of therapeutic MIAs in heterologous systems.

P3-120 - The effect of miR408 over-expression on growth, photosynthesis, and drought resilience in alfalfa

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With the global population projected to reach 9.8 billion by 2050, and climate change intensifying adverse weather events, there is a growing need to increase agricultural productivity under environmental stress. Yield gains in many crop species have plateaued in recent years, and thus enhancing both climate change resilience and photosynthetic efficiency simultaneously presents a promising strategy to boost crop performance. MicroRNA408 (miR408) has emerged as a positive regulator of photosynthesis in various plant species, primarily through its ability to down-regulate the expression of several genes encoding copper-containing proteins. This project investigates the role of miR408 in photosynthesis, growth, and drought response in *Medicago sativa* (alfalfa), which is a critical forage crop in Canada. Using *Agrobacterium*-mediated transformation, miR408 over-expression constructs were introduced into alfalfa, and transgenic lines were confirmed through stem-loop qRT-PCR. The three highest miR408-overexpressing genotypes were selected for further analysis. Transgenic lines exhibited a consistent trend towards increased biomass and decreased stem width compared to wild-type plants; however, no significant differences in photosynthetic characteristics were observed among genotypes. Additionally, preliminary drought assays suggested a potential role for miR408 in resilience to this type of stress. To explore the underlying molecular mechanisms driving miR408 function, comparative RNA-Seq was performed using RNA derived from transgenic and wild-type leaf tissue, identifying four candidate miR408-regulated genes: Plastocyanin-like domain 4, Uclacyanin 5, Uclacyanin 6, and Copper-transporting ATPase PAA1. CRISPR/Cas9-mediated vectors were subsequently constructed and transformed into alfalfa with the aim of knocking out these targets to evaluate their possible roles in photosynthesis, plant growth, and abiotic stress response to further our understanding of these processes in alfalfa

P3-121 - Identifying a novel plant hormone for the orphan receptor, AtHTL/KAI2.

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Germination of angiosperms is a highly regulated process coordinated by environmental and phytohormone cues. Light, gibberellins (GA), and abscisic acid (ABA) are regarded as the main regulators of germination. However, karrikins (KARs), a smoke-derived compound, promotes germination in *Arabidopsis* and is perceived by the *Arabidopsis thaliana* HYPOSENSITIVE TO LIGHT/KARRIKIN INSENSITIVE 2 (AtHTL/KAI2) receptor. A loss of function HTL/KAI2 mutant exhibits opposite phenotypes of KARs-treated seedlings indicating that *Arabidopsis* produces an unknown endogenous KAI2-ligand (KL). While the HTL/KAI2 signalling pathway is known to regulate germination and other developmental processes, the biosynthetic pathway of KL remains elusive. Unable to uncover mutants in this pathway through genetic screening of embryonic mutations, I propose KL synthesis occurs in the seed coat. In my presentation, I present a unique genetic screen identifying novel mutants in the seed coat. I also characterize novel mutants that phenocopy loss of function HTL/KAI2 mutants and are potential biosynthetic mutants of KL. Therefore, my presentation sheds light on key mutants potentially involved in the synthesis of a novel plant hormone, KL.

P3-122 - Generation of alfalfa genotypes with tetra-allelic mutations in *MsWOX13-2* to enhance waterlogging resilience

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Alfalfa (*Medicago sativa* L.), also called the queen of forage crops, is an autotetraploid perennial leguminous species esteemed for its high productivity, excellent feed value, and versatility in its use as hay, pasture and silage. However, its sensitivity to soil waterlogging significantly limits productivity and leads to a decline in forage quality. We previously found that the RNAi-mediated down-regulation of the alfalfa transcription factor-encoding *WUSCHEL-RELATED HOMEBOX 13-2* (*MsWOX13-2*) led to improved waterlogging resilience compared to wild-type controls, which corresponded with alterations in redox-related characteristics. To provide further validation of these findings, we developed alfalfa genotypes carrying bi-allelic frameshift mutations in *MsWOX13-2* using CRISPR/Cas9 technology in the T₀ generation. In a similar fashion to our previous *MsWOX13-2* RNAi genotypes, these bi-allelic mutants also displayed enhanced waterlogging tolerance compared to wild-type genotypes. Subsequently, these genotypes were crossed with the waterlogging-tolerant cultivar AAC Trueman to segregate out the transgenic cassette, and a second round of intercrossing was performed to successfully obtain genotypes with tetra-allelic mutations in *MsWOX13-2*. Current efforts are focused on evaluating their waterlogging response through metabolomic profiling, as well as assessments of cell wall composition and nutritional quality under both normally-watered and waterlogged conditions. Additionally, the development of a non-transgenic breeding population with disruptions in *MsWOX13-2* is underway, which would serve as a resource for future field trials. Overall, these efforts aim to determine the role of *MsWOX13-2* in waterlogging response in alfalfa, with potential applications in enhancing crop resilience under a changing climate to support sustainable forage and beef production in Canada.

P3-123 - A chromosome-level, allele-aware genome and stress transcriptome atlas for the forage legume sainfoin (*Onobrychis viciifolia*)

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Sainfoin (*Onobrychis viciifolia* Scop.) is a perennial autotetraploid forage legume valued for its stress tolerance, nitrogen fixation, and its ability to reduce methane emissions and prevent bloat in ruminants. We present a high-quality, haplotype-resolved, chromosome-scale genome assembly for a genotype of the cultivar Mountainview, constructed using PacBio HiFi, Oxford Nanopore, Illumina, and Hi-C sequencing. The 2.36 Gb assembly spans all 28 chromosomes (7 chromosome groups × 4), with an N50 of 77 Mb and over 97% BUSCO completeness, demonstrating strong structural and genic integrity. Gene prediction with BRAKER3, guided by full-length Iso-Seq and short-read RNA-Seq evidence, identified 117,890 protein-coding genes, of which more than 95% were functionally annotated. Transposable elements comprise approximately 65% of the genome, dominated by Gypsy and Copia LTR retrotransposons. Phylogenomic and Ks analyses placed sainfoin within the Hologalegina clade of legumes, near the *Medicago* lineage, and revealed evidence of conserved whole-genome duplication events. Tissue-enriched gene sets, derived from RNA-Seq across 9 tissues, revealed distinct developmental expression programs. Transcriptome profiling under drought and waterlogging stress further uncovered both shared and stress-specific gene expression responses. miRNA-target prediction highlighted a conserved miR156-*SQUAMOSA PROMOTER-BINDING PROTEIN-LIKE* (*SPL*) regulatory module, with *SPL* genes consistently downregulated under both water stress conditions. This integrative resource enables functional and comparative genomics, trait discovery, and genome-assisted improvement of sainfoin for climate-resilient agriculture.

P3-124 - Development of transformation and gene editing platforms in the forage legume sainfoin (*Onobrychis viciifolia* Scop.)

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Sainfoin (*Onobrychis viciifolia* Scop.) is a forage legume that holds great potential due to the presence of condensed tannins (CTs) in its shoots, bringing benefits such as decreased greenhouse gas emissions from ruminants and reductions in the incidence of pasture bloat. Despite these advantages, it has largely been underutilized by livestock producers, at least in part due to a paucity of breeding progress to date. As a means of facilitating downstream efforts to improve agronomic traits in sainfoin, our aim is to establish an *Agrobacterium tumefaciens*-mediated transformation protocol and efficient gene editing platform for an established sainfoin genotype of the AAC Mountainview cultivar for which we have whole genome sequencing data. In this study, we assessed several strains of *A. tumefaciens*, including GV3101, EHA105, GVA2260, LBA4404, and AGL1, along with various explant types derived from vegetatively-propagated, greenhouse-grown plants. Stem explants were the most effective in terms of forming viable callus, especially when paired with a 1:1 ratio of 6-benzyl amino purine (BAP) and naphthalene acetic acid (NAA) in tissue culture media. However, this genotype has proven somewhat recalcitrant to shoot, and especially root, production in tissue culture. As such, various phytohormone concentrations and additives are currently being assessed in this context. Furthermore, gene editing vectors designed to target sainfoin *PHYTOENE DESATURASE* (*PDS*), which allows for the visual identification of edited genotypes through the presence of a bleached phenotype, have also been generated to evaluate the efficacy of CRISPR/Cas9, CRISPR/intronized Cas9, and openCRISPR platforms in this species. Overall, this study highlights the potential of gene editing in sainfoin, and holds promise to lessen the gap in knowledge currently facing sainfoin genetic research.

P3-125 - CRISPR-Cas9 mediated Characterization of Glycosyltransferase 8 (GT8) in Oat

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Glycosyltransferases (GTs) are a diverse group of enzymes involved in glycosylation, with key roles in maintaining plant structure, facilitating detoxification, and modulating hormone signaling. They are also central to glycan biosynthesis by assembling monosaccharide units into both linear and branched glycan structures. This implicates some GTs in the synthesis of beta-glucan, a dietary fiber known for its health-promoting properties. One of the GT family members, *AsGT8*, has been linked to two known quantitative trait loci (QTLs) associated with beta-glucan levels, making it a strong candidate for functional analysis. In this study, we are characterizing the GT34 family in oat (*Avena sativa*), with a focus on elucidating the function of a specific member, *AsGT8*, using a recently optimized CRISPR-Cas9 system for oat.

A total of twelve GT34 genes (*AsGT1-AsGT12*) were identified in oat, including seven putative orthologs of GT34 genes from rice (*Oryza sativa*) and Arabidopsis, all sharing the PFAM domain PF05637. To investigate the role of *AsGT8* in beta-glucan biosynthesis, specific guide RNAs were designed, and the CRISPR-Cas9 system was introduced into the oat cultivar 'Park.' Several transgenic lines were confirmed through hygromycin-specific PCR. T₀ plants are currently undergoing deep sequencing to assess the gene-editing events in *AsGT8*. Additionally, we are examining potential protein interactors of *AsGT8* that may modulate its function synergistically or antagonistically. Through this study, we aim to uncover novel mechanisms for beta-glucan biosynthesis in small grain cereals, with potential implications for nutritional improvement through genome editing.

P3-126 - Wheat proteins as cryopreservation agents for human pancreatic organoids

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Cryopreservation is an essential procedure for the long-term storage of mammalian cells and higher order cellular structures such as spheroids and organoids. An efficient storage procedure helps maintain an adequate supply of biological materials that can later be used for various applications such as reproductive, regenerative and transfusion medicine, transplantation and drug toxicity testing. Effective cryopreservation requires that specific protocols are established for various cell types and cellular structures to maintain their viability and functions upon thawing after ultra-low temperature storage. These protocols use cryoprotectants added to the freezing medium to minimize freeze-thaw damage. The most widely used cryoprotectant is dimethyl sulfoxide (DMSO), but this chemical is suboptimal because it decreases post-thaw viability and metabolic functions. We have previously shown that recombinant wheat proteins associated with freezing tolerance (WCS120, TaIRI-2, TaENO and TaBAS1) are at least as efficient, and are less toxic, than DMSO to cryopreserve viability and metabolic activity of insulin-secreting human cells and other cell types. In the current work, we show preliminary data that these proteins are also efficient in maintaining post-thaw viability and integrity of more complex structures such as pancreatic organoids generated from pluripotent stem cells. Studies are underway to determine if thawed organoids maintain their glucose-induced insulin secretion function. This approach using non-toxic wheat proteins as cryoprotectants should prove useful in the development of optimized protocols to maintain biobanks of pancreatic organoids that are readily available to treat type 1 diabetes patients.

P3-127 - Plant-production of protein nanoparticle-based vaccines for animal diseases.

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Novel, edible, plant-based vaccine candidates for enteric diseases of animals could serve as safer, more effective, and inexpensive alternatives to currently available vaccines. Nanoparticle-based vaccines show improved interaction with the immune system due to the larger particle size mimicking viral and bacterial pathogens thus enhancing interaction with immune cells. Protein nanoparticles are composed of individual monomers that self-assemble into stable structures and act as scaffolds for the display of antigenic peptides. Some are based on viral coat proteins such as the tobacco mosaic virus and are referred to as virus-like particles (VLPs), whereas others originate from bacteria and form smaller cage-like structures such as lumazine synthase from *Aquifex aeolicus* or ferritin from *Helicobacter pylori*. We have shown that these protein nanoparticles can be expressed and accumulate well in plants, and that they assemble into correctly sized nanoparticles. In some cases, the nanoparticles led to stabilization of the antigen and to higher accumulation levels. We have also shown that they can display antigenic epitopes from several bacterial and viral animal pathogens on their surface, and that they elicit an antigen-specific immune response in mice. These protein nanoparticles can be targeted to different subcellular compartments within plant cells to modulate glycosylation patterns in the secretory pathway, as well as to prevent glycosylation by targeting the cytosol or the chloroplast. Controlling these post-translational modifications enhances the flexibility of the plant production platform. This is especially important because many viral antigens require N-glycans for proper folding, high accumulation, or to confer protective immunity. In contrast, antigens from bacteria are not usually glycosylated and vaccines against these pathogens should avoid glycosylation.

P4-128 - Exploring the function of protein palmitoylation in ROP6-mediated auxin signaling

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The membrane-bound Rho-like GTPases from plants (ROPs) are master regulators of diverse cell signaling events, controlling various agriculturally important traits such as leaf morphogenesis, lateral root and root hair development, and responses to biotic and abiotic stresses. Despite decades of research, critical knowledge gaps remain in understanding how ROPs function at the membrane to achieve signaling specificity. Recent studies have shown that ROP6, a member of the ROP family, clusters into plasma membrane nanodomains in response to the phytohormone auxin. However, the molecular mechanisms underlying this lateral segregation remain poorly understood. To identify potential regulators of this process, we mapped the ROP6 proxitome using TurboID-based proximity labeling combined with mass spectrometry (PL-MS). Among the candidates, we focused on a palmitoyl acyltransferase (PAT), an enzyme known to catalyze the addition of saturated palmitic acid to target proteins, thereby influencing their membrane association and domain partitioning. Using biochemical and cell imaging approaches, we showed that this PAT directly interacts with ROP6, and that the interaction is enhanced by auxin. We are currently investigating whether PAT can palmitoylate ROP6 and regulate its dynamics and nano-organization at the plasma membrane. This work has the potential to reveal a novel class of regulatory enzymes within the auxin-triggered ROP6 signaling pathway, offering new strategies to modulate auxin responses and optimize plant developmental traits.

P4-129 - CRISPR-Cas9 Editing of *Nicotiana benthamiana* Architecture for Space-Efficient Protein Production in Controlled Environments

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Molecular farming leverages plants as biofactories to produce high-value biopharmaceuticals. Given its large biomass, short cultivation cycle and metabolic plasticity, transient expression host *Nicotiana benthamiana* has become a gold standard for this purpose. With the rise of *controlled-environment agriculture* (CEA), efforts are now deployed to develop vertical farming systems adapted to *N. benthamiana*. CEA systems present advantages for plant production, allowing for a year-round production calendar, a fine control of cultural conditions and an efficient recycling of water and nutrients. A challenge at this point for CEA systems is to optimize plant space use, which makes the 'compact architecture' trait an attractive target for plant improvement. Plant form is determined by the auxin/cytokinin ratio, which determines the relative importance of apical dominance and axillary bud outgrowth and, as a result, the overall phenology and spatial footprint of the plant. Using CRISPR-Cas9 gene editing, we engineered *N. benthamiana* lines to knock out the expression of CCD7 or CCD8, two key enzymes of the strigolactone biosynthetic pathway. Strigolactones promote apical dominance and inhibit bud outgrowth by a repressing effect on cytokinin synthesis. The idea was to generate CCD-depleted lines with a reduced auxin/cytokinin ratio promoting axillary growth over apical dominance. The mutant lines showed an increased cytokinin content compared to the parent line, and also presented a compact phenotype characterized by an unchanged shoot biomass but a reduced height and area footprint compatible with a higher planting density per square meter. Despite a smaller stature, these lines produced equivalent yields of recombinant protein relative to the parent line. Most interestingly, they showed reduced levels of Rubisco post-agroinfiltration, allowing for a pre-enrichment of the recombinant protein before harvesting. These attributes of the CCD-deficient lines make them promising host candidates for the design of space-efficient CEA systems adapted to the *N. benthamiana* biofactory.

P4-130 - Identification of novel components in Arabidopsis CEP peptide signalling

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C-TERMINALLY ENCODED PEPTIDES (CEPs) are small peptide hormones that regulate numerous physiological processes in plants including root growth and development, root system architecture, responses to nitrogen stress, and immunity. Previous studies have shown that when CEPs (CEP1/4) are exogenously applied to Arabidopsis, plants exhibit reduced root and seedling growth, increased resistance to infection, and strong MAPK activation. Arabidopsis CEPs are recognized by receptor kinases CEPR1 and CEPR2, while CEP4 signals through the related receptor-like kinase 7 (RLK7) as well. However, the downstream components involved in CEP signaling remain undefined. Receptor-like cytoplasmic kinases (RLCKs), are known to be involved in signal transduction during root development and involved in pattern-triggered immunity signaling, often acting downstream of other receptor kinases. This overlap in signaling outputs suggests a potential role for RLCK-VIIs in CEP signaling.

To investigate the potential roles of RLCK-VIIs, I will test Arabidopsis CEP-induced responses in 9 RLCK-VII polymutant lines, each containing multiple loss-of-function alleles within a given RLCK-VII subfamily. If RLCK-VII members are involved in CEP signaling, I anticipate RLCK-VII knockouts will disrupt the CEP pathway resulting in immune signaling disruption, less severe root/seedling growth inhibition, and reduced MAPK activation when compared to CEP treated wild type plants. I anticipate 1 or 2 subfamilies will demonstrate involvement in the pathways. I will present the results of screening the lines for involvement in CEP immunity and growth signaling; as of now the *rlck-vii-8* polymutant line demonstrates no involvement in CEP immunity signaling. Understanding the players involved in CEP signaling pathways will advance our understanding of plant development and immune responses, and may provide insights for enhanced growth and immunity in crops.

P4-131 - Cell-tissue specific expression does not encode RLCK signalling specificity

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Receptor kinase signalling facilitates a multitude of physiological and biochemical changes in plants in response to environmental and endogenous signals. Receptor kinases (RKs) are proteins located at the cell membrane that perceive ligands to initiate essential signalling cascades essential for various biological pathways including plant growth, development, and immunity. Though many RKs have been identified, the molecular mechanisms downstream of receptor activation are not fully understood. Receptor-like cytoplasmic kinases (RLCKs) are key components of downstream RK signalling. RLCKs associate with receptor complexes to execute downstream signal transduction via substrate phosphorylation. Despite sequence similarity, analogous mechanisms of activation and substrate interaction, individual RLCKs can exhibit distinct functionalities and involvement in specific RK pathways. For instance, BIK1 and SGN1 are RLCKs present within the same subfamily, the RLCK-VII/PBL family, however, play central roles in immunity and casparian strip development respectively. Our research focuses on understanding the specificity within RLCK-mediated signalling. By generating promoter-swapped RLCK constructs, we demonstrated that signaling specificity is not solely dictated by RLCK expression patterns. Notably, SGN1 failed to rescue an immune phenotype even when driven by the BIK1 promoter. These findings contribute to our understanding of the molecular signaling networks that regulate diverse physiological and developmental processes in plants.

P4-132 - Influence of inhibiting gibberellin biosynthesis and nitrogen fertilization on lodging related traits in spring wheat

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Lodging is a major constraint to wheat productivity, particularly under high nitrogen input conditions, where excessive vegetative growth can compromise stem integrity. This study evaluated the interactive effects of

nitrogen fertilization and plant growth regulators on lodging-related morphological traits in spring wheat. To this end, the study involved two genotypes that exhibit varying resistance to lodging, three different rates of nitrogen, and application of a PGR that inhibits gibberellin biosynthesis in plants. Elevated nitrogen levels increased plant height (5 to 12 %), plant fresh weight (27 to 33%), bending moment (36 to 41%) and second internode length (3 to 9%) in both genotypes, resulting in a higher lodging index (10 to 32%) despite the increase in the breaking strength of the second internode which varied with genotype and nitrogen level. PGR application effectively reduced lodging index (4 to 37%) through reduction of plant height (8 to 32%), plant fresh weight (13 to 32%), and bending moment (15 to 53%) as well as breaking strength (up to 25%) in both genotypes; however, the effect of PGR on all parameters studied was more pronounced in the taller cultivar. Overall, the reduction in elongation traits due to low nitrogen rates and PGR treatment contributed to lowering the lodging risk. Thus, nitrogen and PGR interactively affect lodging-associated morphological traits in wheat, with their effectiveness strongly influenced by genotype.

P5-133 - Identifying New Components in the Arabidopsis Self-Incompatibility Pathway

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Self-incompatibility (SI) is a genetic mechanism in flowering plants that prevents self-fertilization and promotes outcrossing. In the Brassicaceae family, SI is initiated by allele-specific interactions between the SP11/SCR ligand on the pollen and the S Receptor Kinase (SRK) on the stigma surface. This interaction activates a signaling cascade that ultimately results in the rejection of self-pollen. Several SI signaling components in *Brassica* SI species have been characterized: the ARC1E3 ubiquitin ligase which targets compatible pollen factors, the FERONIA (FER) receptor kinase which activates RBOHD to generate reactive oxygen species (ROS) to inhibitory levels, and the M Locus Protein Kinase (MLPK), also connected to RBOHD activation. However, some elements of the related SRK signaling cascade in *Arabidopsis* species remain unknown. This research aims to identify and characterize new regulators of the *Arabidopsis* SI response. Using yeast two-hybrid screening, novel *Arabidopsis* SRK-interacting proteins from the Receptor-Like Cytoplasmic Kinase (RLCK) family were identified. To investigate their function in the SI pathway, T-DNA insertion mutants for these RLCKs are being crossed into the transgenic *Arabidopsis thaliana* SI-Col-0 line, which expresses functional SCR, SRK and ARC1 alleles. Additionally, the roles of FER and RBOHD in Arabidopsis SI signaling will be assessed. Loss-of-function *fer* and *rbohD* mutants are being crossed into the SI-Col-0 line to test whether disrupting ROS signaling compromises the SI response. Pollen hydration assays and aniline blue staining will be used to evaluate SI defects in all these lines. Together, this research will uncover new components of the SRK-mediated SI pathway in *Arabidopsis* and provide mechanistic insights into ROS signaling in reproductive self-recognition. Findings may inform strategies to manipulate SI systems for hybrid seed production and crop improvement in Brassicaceae.

P5-134 - The down-regulation of SUPPRESSOR OF OVEREXPRESSION OF CONSTANS 1a (SOC1a) in alfalfa leads to delayed flowering, altered growth, and alterations in degradability

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Alfalfa (*Medicago sativa* L.) is the most cultivated forage legume in the world, primarily due to its high yield and nutritional value, as well as its leguminous and perennial nature. Forage quality decreases in alfalfa during the transition from vegetative to reproductive growth, largely due to cell wall lignification and decreased leaf-to-stem ratios, leading to declines in digestibility and palatability as a feed. As such, one proposed strategy to both extend the potential harvest window and improve forage quality in alfalfa is to modulate flowering time genes in order to delay flowering and potentially improve quality. SUPPRESSOR OF OVEREXPRESSION OF CONSTANS 1 (SOC1) is a floral integrator gene that is responsible for integrating floral promotion and inhibition signals from several upstream pathways, and subsequently activating downstream floral meristem identity genes to induce flowering. The down-regulation of SOC1 in other plant species results in delayed

flowering and altered stem characteristics, but no such studies have been carried out in alfalfa as of yet. Therefore, in this study, RNAi was utilized to knock down the expression of *MsSOC1a* in alfalfa as a means of examining its role in morphology, growth, and development. *MsSOC1a*-RNAi genotypes exhibited delayed flowering, as well as decreased plant height, internode length and biomass production, and increased leaf-to-stem ratio compared to wild-type controls. Furthermore, *MsSOC1a*-RNAi genotypes displayed a trend towards increased neutral detergent fibre (NDF) and acid detergent fibre (ADF) degradation under *in vitro* conditions compared to wild-type plants. These results highlight the role of *MsSOC1a* in floral promotion and stem architecture, and suggest a potential increase in forage quality in *MsSOC1a* knockdown alfalfa plants. To gain further insight into the precise role of *MsSOC1a* in alfalfa, we have also generated genotypes bearing mutations in all four *MsSOC1a* alleles using CRISPR/Cas9, which will be evaluated in the near future.

P5-135 - Linking SPL3 and VRN1: Evidence of a Potential Regulatory Interaction in Oat Flowering

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Oat (*Avena sativa* L.; 2n = 6x = 42, AACCCDD) is an important cereal crop with high nutritional value. SQUAMOSA promoter binding-like proteins (SPLs) are key transcription factors targeted by miR156, play essential roles in developmental phase transitions in plants. We previously identified 28 *AsSPL* genes in oat, classified into six phylogenetic clades, and characterized a novel *AsmiR156* family with 21 precursors. Sixteen *AsSPLs* were predicted targets of miR156 (Mehtab-Singh et al., 2024). Among these, *AsSPL3* exhibited high transcript abundance during early inflorescence (GS-54), a period marked by low *AsmiR156* expression and elevated levels of *AsVRN1*, a key floral meristem identity gene. This temporal co-expression suggested a possible regulatory interaction. Using electrophoretic mobility shift assay (EMSA), we confirmed that *AsSPL3* directly binds to *AsVRN1* promoter, providing the first molecular evidence of a link between the SPL/miR156 module and the vernalization pathway in oat. To further dissect this regulatory network, we are currently targeting *AsVRN1* and *AsSPL3* individually using CRISPR-Cas9 system. Our findings support the existence of a conserved flowering regulatory mechanism in oat and provide promising genetic targets for the acceleration of breeding programs aimed at improving oat adaptation and productivity.

P5-178 - Deducing a Genetic Network for Seed Protein Analysis in *Arabidopsis thaliana*

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Plant-based protein consumption is a fast-growing consumer trend. Globally, plant-based protein markets have expanded rapidly in the past decade to meet the growing demand for meatless protein sources. By understanding the mechanisms by which plants control the amount of protein that accumulates in their seeds, researchers may work toward developing enhanced-protein crops. The goal of this research is to bring clarity to the complex web of gene regulation that controls seed protein levels in plants using *Arabidopsis thaliana* as a model. The genes that are investigated in this study are a series of transcription factors. The first and most central is Transcription Factor 1 (TF1), a master regulator of seed protein accumulation in *Arabidopsis*. This study also investigates the interrelated effects of four additional genes hypothesized to be negative regulators of TF1. Three of these genes have further been hypothesized to act as a complex in the negative regulation of TF1. By creating a series of digenic mutant combinations of these 5 mutants, we have been able to observe their combined and interrelated effects on seed protein levels. The interdependent effects of these genes will help reveal the nature of the pathway or pathways controlling seed protein accumulation in *Arabidopsis*. This fundamental research will then be applied to equivalent knockouts in canola and chickpeas. Detangling this regulatory network may help us to produce crops, particularly canola, peas, and chickpeas with a higher seed protein content.

POSTER SESSION II

P6-136 - Investigating barley sensitivity to a lipopeptide virulence factor

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Fusarium graminearum (Fgr) is the primary causal agent of Fusarium Head Blight (FHB), a devastating fungal disease that impacts the economic value of cereal crops globally. Fgr produces a range of secondary metabolites, some of which act as virulence factors, making infected crops unsuitable for consumption and industrial use. One such newly identified metabolite, gramillin, enhances Fgr virulence in maize and barley by disrupting membrane potential, triggering host stress responses, and promoting production of fusaoctoxin. Notably, wheat is resistant to gramillin, while barley exhibits quantitative resistance. QTL analysis has mapped gramillin sensitivity in barley primarily to chromosome 5H, with phenotypic characterization linking gramillin sensitivity to necrotic lesion formation on barley leaves.

A range of gramillin sensitivities observed across barley genotypes has prompted us to investigate the genetic basis for this variation. Comparative transcriptomic analysis reveals that gramillin triggers distinct gene expression patterns in barley, unlike conserved PAMPs such as flg22. Gene ontology (GO) enrichment analysis of gramillin-insensitive barley highlights downregulation of defense-related genes and upregulation of genes associated with catalytic activity and membrane function. To further investigate the link between plant defense and gramillin sensitivity, gramillin-induced lesion assays were performed on barley leaves that were pre-treated with flg22, a known inducer of plant stress response. Preliminary results support the hypothesis that timing or amplitude of induced stress responses may influence gramillin sensitivity. In addition, preliminary biochemical analysis of gramillin degradation in barley apoplastic fluid indicates a potential role of extracellular proteolytic enzymes in conferring resistance. Overall, this research aims to identify molecular markers and traits linked to gramillin resistance and contribute to the development of FHB-resistant barley cultivars. Given that FHB causes an estimated \$1.5 billion in annual losses to the Canadian grains industry, these findings have significant agricultural and economic implications.

P6-137 - Understanding host defense against a fungal threat to North American corn production

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Tar spot is an emerging foliar disease of corn for which no genetic resistance exists in cultivated varieties. Caused by the fungus *Phyllachora maydis* and first detected in Ontario corn fields in 2020, the disease poses a growing threat to maize production. Recent work has identified a resistance-associated genetic locus on chromosome 8, highlighting five candidate genes. Two genes were selected for further investigation based on their potential roles in disease resistance: a putative UDP-xylose transporter and a leucine-rich repeat receptor-like kinase (*LRR-RLK*). While maize encodes over 248 *LRR RLKs*, only five UDP-xylose transporters are known to be expressed.

Sequencing of both genes in resistant and susceptible maize lines revealed a number of differences in their genomic sequences. Comparative analyses identified 56 SNPs in the *LRR-RLK* coding region, including three frameshift mutations, as well as eight SNPs in the UDP-xylose transporter, which may impact protein function. Quantitative PCR analysis shows elevated expression of *PR5*, a marker gene for pathogen-related pattern-triggered immunity, along with increased expression of the UDP-xylose transporter in the resistant line CML495. Additionally, reactive oxygen species (ROS) assays indicated a stronger oxidative burst in the resistant line.

Future work will focus on microscopic comparisons of cell wall differences between the resistant and susceptible lines, as well as subcellular localization of the proteins during transient expression. Understanding the genetic mechanisms underlying pathogen resistance will help inform breeding programs and provide deeper insights into plant immune responses against emerging pathogens.

P6-138 - Exploring Genetic Resistance to Sclerotinia Stem Rot in Canola Through VQ Gene Expression Profiling

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¹Wilfrid Laurier University, ²Henan University

Sclerotinia Stem Rot (SSR), caused by *Sclerotinia sclerotiorum*, poses a significant threat to canola (*Brassica napus*), which is crucial for Canada's agricultural economy. This disease can lead to drastic yield losses and adversely affect seed quality, impacting export values. Current management strategies, including fungicides and crop rotation, are inconsistent, costly, and can harm the environment. Since there is currently no evidence of Resistance (*R*) genes conferring complete qualitative resistance against *S. sclerotiorum* in canola, other genes involved in building quantitative resistance must be studied. Valine-glutamine (VQ) genes are involved in plant immune responses to biotic and abiotic stress. In this study, various canola lines were screened for enhanced SSR resistance. Quinta and 1065 exhibited the greatest susceptibility, while Surpass 400 and Jet Neuf demonstrated the strongest resistance. RNA sequencing was done to obtain gene expression data from Jet Neuf and a more susceptible line, Westar. Inoculation was done in planta via mycelial agar plugs, and samples were taken at 24 and 48 hours post inoculation (hpi). We expect to identify differentially expressed genes (DEGs), particularly among VQ genes, between the two cultivars. DEG analysis will be complemented by Gene Ontology (GO) and Kyoto Encyclopedia of Genes and Genomes (KEGG) enrichment analyses to pinpoint genes potentially involved in SSR resistance. These candidate genes can be used to develop transgenic canola lines with enhanced SSR resistance. Improving SSR resistance will help reduce disease burden for farmers and improve the profitability and sustainability of canola production.

P6-139 - Arabidopsis *LURP1* regulates salicylic acid-mediated and PAMP-triggered immunity

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Transcription factors play essential roles in plant immunity. In Arabidopsis, calmodulin-binding transcription activators (CAMTAs) negatively regulate plant immune responses by suppressing the biosynthesis of two key defense hormones, salicylic acid (SA) and N-hydroxy-pipecolic acid (NHP). This is achieved through inhibition of the master immune transcription factor *SAR Deficient 1* (*SARD1*). The *camta2/3* double mutant exhibits obvious autoimmune phenotypes, including dwarfed size and enhanced immunity, which can be partially suppressed by *sard4*, an enzyme involved in NHP precursor biosynthesis. To search for positive immune regulators downstream of CAMTAs, a forward genetic screen was performed using *camta2/3 sard4* mutant. Here, we report on a loss-of-function allele of *Late Upregulated in Response to Hyaloperonospora parasitica 1* (*LURP1*) that suppresses the autoimmune phenotypes of *camta2/3 sard4*. *LURP1* encodes a cytoplasmic protein with structural similarity to mammalian tubby domain-containing proteins. Its transcripts are strongly induced by pathogens, SA, and NHP. Chromatin immunoprecipitation (ChIP) assays revealed that *LURP1* is directly regulated by *SARD1*. Knocking out *LURP1* and its three close homologs (*LOR1*, *LOR10*, and *LOR15*) compromises basal resistance to virulent oomycete and bacterial pathogens. Furthermore, PAMP-triggered immunity (PTI) is impaired in the *lurp1* quadruple mutant. Notably, flg22- and nlp20-induced resistance and SA accumulation are significantly reduced in this mutant, indicating that *LURP1* and its homologous are essential components of PTI and serve key roles in SA-mediated defense signaling.

P6-140 - The trans-Golgi network adaptor protein 4 complex contributes to plant immunity mediated through plasma membrane immune receptors

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Receptor like proteins (RLPs) play important roles in plant immunity. Loss of the RLP SUPPRESSOR OF NPR1-1, CONSTITUTIVE 2 (SNC2) in *Arabidopsis* results in enhanced disease susceptibility, whereas the gain-of-function mutant *snc2-1D* exhibits autoimmunity including a dwarfed morphology and constitutively activated defense responses. Mutations in the transmembrane protein BIAN DA 1 (BDA1) lead to complete suppression of *snc2-1D* autoimmunity, suggesting that BDA1 is an important regulator of SNC2. Two master transcription factors, SYSTEMIC ACQUIRED RESISTANCE DEFICIENT 1 (SARD1) and CALMODULIN-BINDING PROTEIN 60g (CBP60g), are also required for the autoimmunity of *snc2-1D*. Constitutive defense responses in *snc2-1D* are attenuated by *cbp60g* single mutants, but completely suppressed by the *sard1 cbp60g* double mutant. Here, we report the identification and characterization of the ADAPTOR PROTEIN 4 (AP4) complex in SNC2-mediated immunity. By performing a suppressor screen in the *cbp60g-1 snc2-1D* background, mutations in AP4 μ , a subunit of the AP4 complex, were identified. The AP4 complex is composed of four subunits, AP4 ϵ , β , μ and σ . Knocking out genes of the other subunits in the AP4 complex consistently suppressed *cbp60g-1 snc2-1D*, suggesting that the AP4 complex is required for SNC2-mediated defense. The AP4 complex is involved in target protein sorting in the trans-Golgi network, and AP4 μ physically interacts with BDA1 *in vivo*, suggesting that the AP4 complex is likely required for the precise subcellular localization of BDA1. Furthermore, mutating *AP4M* in wild-type plants compromises both pattern- and effector-triggered immunity, indicating a broader role of the AP4 complex in plant immunity.

P6-141 - Plant microbiome responses to clubroot infection

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Clubroot is a soil-borne disease caused by *Plasmodiophora brassicae*, an obligate biotrophic pathogen that significantly impacts economically important crops such as canola and cabbage. Infected plants develop root galls that interfere with water and nutrient uptake, leading to stunted growth and, in severe cases, plant death. The deployment of resistant cultivars has been the primary management strategy; however, the emergence of virulent *P. brassicae* pathotypes necessitates the development of sustainable alternative approaches. The plant microbiome holds promising potential for biocontrol applications. Nevertheless, the mechanisms underlying the tripartite interactions among the microbiome, the plant host, and *P. brassicae* remain poorly understood and most of the studies focused on the rhizosphere and root endosphere. Moreover, the response of the microbiome associated with the aerial parts of plants (the phyllosphere) remains largely understudied, despite its crucial role in plant nutrition and defense against both abiotic and biotic stresses. This study aims to enhance our understanding of the diversity and functional roles of plant-associated microbiome during clubroot infection. By comparing microbiome responses across clubroot-resistant and susceptible ecotypes of *Arabidopsis thaliana* infected with *P. brassicae* as well as studying the temporal dynamics by profiling microbial communities at multiple infection stages, we seek to uncover species-specific microbial shifts across different plant compartments, including the rhizosphere, endosphere, and phyllosphere. Furthermore, we will isolate and identify bacterial taxa associated with resistant plants to investigate their roles in disease suppression. To this end, we are designing synthetic microbial communities composed of multiple taxa with biological control and plant growth-promoting properties, aiming to evaluate their contribution to modulating the plant's response to clubroot. We aim to evaluate the extent to which insights gained from *Arabidopsis* can be translated to economically relevant crops such as canola, thereby addressing the broader question of host-specific microbiome responses to clubroot infection.

P6-142 - Characterization of *Plasmodiophora brassicae* putative effectors by reverse genetics

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Clubroot is a devastating disease caused by *Plasmodiophora brassicae*, affecting cruciferous crops worldwide. In Canada, this disease poses significant economic and environmental challenges, particularly in canola fields. Despite its widespread impact, little information is available regarding virulence factors, such as pathogen effector proteins. In this study, we aim to investigate the role of 24 putative clubroot effectors that are highly expressed during *P. brassicae* infection and/or contain functional domains previously identified as key in manipulating plant immunity in other pathosystems. To achieve this, we will phenotype *Arabidopsis thaliana* transgenic lines that overexpress these pathogen candidate effectors during clubroot infection. Phenotypic traits such as rosette morphology, leaf, flower, root size and shape, size of galls, and disease severity will be recorded. Additionally, the subcellular localization of the putative effectors will be analyzed. We discovered candidates that could be avirulence factors, potentially triggering plant resistance or tolerance. To date, several candidate effectors have been evaluated, and the characterization of the remaining ones is ongoing. These efforts aim to provide insights into the potential roles these effectors may play in the interaction between *P. brassicae* and its host. Our goal is to create a resource for the scientific community to expand the characterization of *P. brassicae* effectors and further understand the mechanisms behind this disease.

P6-143 - Comparison of two media for the detection of botryodiplodin (a mycotoxin) produced by *Botryodiplodia theobromae*

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The mycotoxin, botryodiplodin, plays a crucial role in plant infection. Botryodiplodin is phytotoxic to plants and cytotoxic to humans and animals, affecting agricultural production. The in-culture technique has been developed to screen pathogenic *Botryodiplodia theobromae* in plant materials for their ability to produce botryodiplodin. The Czapek Dox agar medium supplemented with glycine (CDA+G)} has been used for screening fungi producing botryodiplodin, but this technique is expensive and unable to detect certain fungal isolates producing the mycotoxin. Furthermore, in order to develop a cost-effective procedure for detecting botryodiplodin produced by *B. theobromae*, the study compares the commonly used but expensive CDA+G medium with the cost-effective sucrose agar with glycine (S+G) medium for detecting botryodiplodin. The results obtained from the study revealed that the S+G agar medium detected botryodiplodin in more isolates of *B. theobromae* than the CDA+G medium. This finding suggests that utilizing S+G agar medium could significantly reduce costs for researchers while enhancing the reliability of detection methods for this pathogen. Consequently, the adoption of S+G medium may lead to more effective management strategies for crops affected by *B. theobromae*. Also, the visibility of botryodiplodin produced by each isolates of *B. theobromae* following the incubation periods were documented and categorized into four pattern as observed in the S+G agar medium. Additionally, the disease severity index (DSI) was established based on pathogenicity test of *B. theobromae* on coconut fruits. Also, the DSI in relation to botryodiplodin produced by each the isolates were established. The results indicated a significant correlation between the levels of botryodiplodin and the severity of disease symptoms observed in the coconut fruits. Further analysis revealed that isolates producing higher amounts of botryodiplodin consistently resulted in more or less severe fruit rot, highlighting the importance of this mycotoxin in the pathogenicity of *B. theobromae*.

P6-144 - Soybean Response to Fulvic Acid Under Field Conditions

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Soybean is a vital crop for food, fuel, and feed, playing a key role in the Canadian economy. Given the rising costs and environmental impact of synthetic fertilizers, organic alternatives like humic and fulvic acids are gaining interest. Fulvic acids, organic compounds formed through the decomposition of plant material, are known to improve soil quality and plant growth. However, their potential as seed treatments under field conditions and their influence on the soybean microbiome remains underexplored. This study evaluated the effects of fulvic acid seed treatments (0.8% and 2%) on soybean development and microbial diversity in field trials conducted in Carman, Manitoba. Emergence was recorded four weeks after seeding. Samples from the rhizosphere and bulk soil were collected at growth stages V3, V6, R5, and R6 for microbial DNA extraction and analysis. At R5, plants treated with 2% fulvic acid were taller with denser foliage and had 39% more nodules than control plants. Yield responses were variable: the 0.8% treatment increased yield by 6%, while the 2% treatment showed no difference compared to the control. Fungal community dynamics of the 0.8% fulvic acid treatment were analyzed at the class level using QIIME2. In bulk soil, vegetative stages (V3 and V6) showed increases in *Leotiomycetes*, *Eurotiomycetes*, and *Microbotryomycetes*, while reproductive stages (R5 and R6) showed increases in *Tremellomycetes* and *Dothideomycetes*. In the rhizosphere, vegetative stages had increases in *Agaricomycetes*, *Tremellomycetes*, and *Zoopagomycetes*, while reproductive stages showed increases in *Dothideomycetes*, *Eurotiomycetes*, and *Rhizophlyctidomycetes*.

These results suggest fulvic acid may enhance the abundance of key saprotrophs and decomposers in soil microbial communities. Future work will explore its effects on bacterial dynamics, providing further insight into fulvic acid's potential to support soybean growth.

P6-145 - AtSnRK1 plays a role in regulation of jasmonate-mediated plant resistance

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Every year, 20%-40% of global crop production is lost to pathogens and insect pests costing the global economy around \$290 billion. Jasmonates are hormones critical for plant resistance against necrotrophic pathogens and chewing herbivores. In response to wounding or mechanical damage, jasmonate levels increase within seconds, suggesting the biosynthetic pathway is constitutively present and regulated post-translationally. *Arabidopsis thaliana* AtSnRK1 is a heterotrimeric kinase complex that acts as a key metabolic sensor adjusting cellular metabolism during growth, energy signalling, and stress conditions. However, the role of AtSnRK1 in wounding has not been explored. Here, we show that AtSnRK1 positively regulates jasmonate biosynthesis. In *Atsnrk1* mutants CS26 and CS27, the jasmonate burst is muted after wounding compared to wild type plants (Col-0). Feeding assays using an important Canadian agricultural caterpillar pest, the cabbage looper, further supports the involvement of AtSnRK1 in plant resistance against chewing insects. Understanding the regulation of plant resistance will support efforts to generate pest resistant crops to mitigate agricultural losses.

P6-146 - Potential use of free-base porphyrin derivatives in agriculture: in vitro, and in vivo test against *Botrytis cinerea*

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Agriculture faces increasing challenges due to climate change, requiring innovative and sustainable solutions to ensure food security. Conventional fungicides, while widely used, pose environmental risks, toxicity concerns, and limited efficacy against resistant fungal strains. In this study, we explore the potential of ionic free-base porphyrins as light-activated antifungal agents, specifically targeting *Botrytis cinerea*, a major agricultural pathogen. These porphyrins generate singlet oxygen (¹O₂) upon light exposure, effectively inhibiting fungal growth. Our research demonstrates significant antifungal activity in vitro at concentrations of 5-30 ppm under illuminated conditions, with ionic derivatives outperforming their neutral counterparts due to their enhanced interaction with fungal spores.

To bridge laboratory findings with real-world applications, we conducted *in vivo* experiments on infected plants under controlled greenhouse conditions. Results confirm that porphyrin-treated plants exhibit reduced fungal infection rates compared to untreated controls, validating the efficacy of these photosensitizers beyond in vitro settings. The next phase of our research focuses on optimizing formulation stability, application methods, and large-scale deployment strategies in agricultural fields. By scaling up these light-activated porphyrins for field trials, we aim to establish a cost-effective, environmentally friendly alternative to conventional fungicides, paving the way for sustainable disease management in modern agriculture.

P6-147 - Characterization of Virulence Factors in *Golovinomyces ambrosiae*, the Causal Agent of Powdery Mildew in Cannabis

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The use of cannabis dates back thousands of years, for therapeutic, industrial, and ritual purposes. Native to Central Asia, this plant from the Cannabaceae family is now cultivated and utilized worldwide, particularly for its medicinal and industrial applications. However, like all plants, cannabis is susceptible to various infections caused by pathogens such as bacteria and fungi. Among these, powdery mildew caused by *Golovinomyces ambrosiae* poses a significant threat to cannabis crops. This biotrophic fungus secretes virulence factors (effectors), which are small proteins that manipulate plant immune systems, allowing the pathogen to bypass host defenses and facilitate infection.

The hypothesis is that effectors produced by *Golovinomyces ambrosiae* modulate host plant immune responses, thereby promoting infection and pathogen spread. The main objective of this study is to characterize these virulence factors, with a focus on the role of effectors in interacting with plant defense mechanisms. The first part of the research aims to initiate the functional characterization of effector mechanisms within plant cells by determining their subcellular localization. To achieve this, effectors will be transiently expressed in plant cells via agroinfiltration, and their localization will be observed using confocal microscopy.

The second part of the study will assess the impact of these effectors on specific plant defense responses, such as reactive oxygen species (ROS) production and callose deposition. To this end, *Arabidopsis thaliana*, a model plant, will be genetically transformed to stably express individual effectors. These plants will then be subjected to various stress conditions, including exposure to other pathogens and to *Golovinomyces ambrosiae* itself. ROS production under these conditions will be quantified using a luminometer, and defense gene expression will be analyzed to better understand the molecular interactions between fungal effectors and host immunity.

P6-148 - Targeting Subgroup IV CDPKs to Enhance Soybean Immunity: Insights from Arabidopsis CPK28

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Soybean (*Glycine max*) is a significant crop of agricultural importance across Canada and all over the world. Every year, soybean farmers must deal with various pests and pathogens that cause diseases that can dramatically reduce yields. Currently, the primary way to mitigate diseases is by using pesticides that are effective but environmentally-unfriendly and often quickly overcome by the evolution of pesticide resistant strains. New, genetics-based, sustainable solutions are therefore urgently needed. Calcium-dependent protein kinases (CPKs/CDPKs) play vital roles as immune signaling proteins in plants, modulated by changes in intracellular calcium levels. While many CPKs are involved in immune signal propagation, subgroup IV CPKs—including Arabidopsis CPK28—act as negative regulators of immunity in Arabidopsis as well as certain crop plants such as rice and cotton. CPK28 is a prime target for engineering enhanced disease resistance in crops, because unlike most plants with enhanced immunity that suffer autoimmune damage, *cpk28* loss-of-function mutants demonstrate heightened immunity without autoimmune side effects. Soybean subgroup IV GmCDPKs share 70%-95% amino acid sequence similarity to AtCPK28, suggesting that their functional similarity could be harnessed for increased immunity in soybean. Therefore, we undertook a functional analysis of subgroup IV GmCDPKs in soybean immunity, which is expected to provide insight into new ways to generate broad spectrum disease resistance traits.

P6-149 - Preliminary investigation of the potential role of dehydrins in clubroot resistance and drought tolerance in Arabidopsis

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Clubroot disease caused by *Plasmodiophora brassicae* Woronin results in significant economic loss in canola (*Brassica napus* L.) and other *Brassica* species. The infection leads to the formation of galls on roots, impaired water and nutrient uptake, resulting in reduced seed yield and quality. Previous omics studies from our lab identified a set of dehydrin genes exhibiting increased expression in resistant canola lines compared to susceptible lines in response to *P. brassicae* infection. Dehydrins belong to class II Late Embryogenesis Abundant (LEA) protein family known to alleviate abiotic stresses such as drought, salinity and frost along with limited evidence suggesting antimicrobial activity. Given that clubroot disease causes water-deficit stress during later stages of infection, dehydrins may contribute to tolerance through both biotic and abiotic stress response pathways. The objective of our research is to characterize the expression of different dehydrin genes of *Arabidopsis thaliana*, another susceptible host, following *P. brassicae* infection. Four *A. thaliana* dehydrin genes namely *COR47*, *AtDhn5*, *XERO1* and *RAB18*, showed significant upregulation of expression in roots and shoots following *P. brassicae* infection at multiple time points (4, 7, 14, 21 days post inoculation). The cDNAs of these, along with that of one *B.napus* dehydrin gene *ERD 14-like*, which we identified in earlier transcriptomic studies, were cloned into the pBinGlyRed3 vector under the CaMV 35S constitutive promoter and transformed into *Arabidopsis* (Col-0) via *Agrobacterium*-mediated floral dip. Transgenic seeds (T₀) were identified based on red fluorescence from the seeds and were advanced to T₃ generation. The overexpression lines are currently being evaluated for resistance to *P. brassicae* pathotype 3H and for drought tolerance. The findings from these studies will be presented to provide insights into the potential role of dehydrins in pathogen defense.

P6-150 - Tuning the Dual Identity: Phospho-Regulation of two related multi-domain receptor-like cytoplasmic kinases in *Arabidopsis thaliana*

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Phosphorylation and ubiquitination are two major post-translational modifications (PTMs) that regulate proteins. Underscoring their importance in diverse signaling pathways, plant genomes encode thousands of protein kinases and E3 ubiquitin ligases, the enzymes that catalyze these PTMs. While these enzymes are typically encoded by distinct genes, a conserved family of receptor-like cytoplasmic kinases (RLCKs), classified under the RLCK IXb group, uniquely combines a kinase domain with a plant U-box (PUB) E3 ubiquitin ligase domain. In the model plant *Arabidopsis thaliana*, the most highly and broadly expressed RLCK IXb proteins are the paralogs PUB32 and PUB33. Orthologs of AtPUB33 in potato, tomato, and *Nicotiana benthamiana* have been identified as direct targets of at least three unrelated pathogenic effectors, suggesting a critical role in either pathogen virulence or plant immunity. We will present our ongoing work to understand the biological and biochemical functions of PUB32 and PUB33 using cellular, biochemical, and genetic approaches.

P6-151 - Development and validation of approaches for the detection, propagation, and synthesis of Hop Latent Viroid: toward the development of a biotechnological tool for functional genomics in *Cannabis sativa*.

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Viruses and viroids have emerged as powerful tools in plant functional genomics, offering innovative approaches to study gene function through sequence-specific gene silencing. Despite advances in other model plants, *Cannabis sativa* still lacks an efficient and widely adopted system for gene function analysis. This study aimed to establish and validate robust methodologies for the detection, propagation, and synthetic production of Hop Latent Viroid (HLVd), laying the foundation for its use as a functional genomics tool in cannabis biotechnology. We first optimized and compared existing RT-qPCR-based protocols for the reliable detection of HLVd across various tissue types and experimental conditions. Multiple inoculation strategies were then evaluated to ensure efficient viroid transmission and systemic infection in *C. sativa*. In parallel, synthetic biology approaches were employed to produce in vitro transcribed HLVd RNA, enabling precise control over viroid quantity and facilitating reproducible delivery. To demonstrate the applicability of this system, five synthetic HLVd constructs were engineered to carry sequences targeting the *phytoene desaturase* (*PDS*) gene, a commonly used visual marker in gene silencing studies. The *PDS* gene was selected due to its well-characterized role in chlorophyll biosynthesis, its silencing leading to visible photobleaching, allowing clear phenotypic assessment. Upon inoculation, *C. sativa* plants exhibited consistent chlorophyll loss, confirming the ability of recombinant HLVd to mediate targeted gene silencing. Collectively, these findings establish a functional viroid-based platform for gene silencing in *C. sativa*, opening new research directions for the study of gene function and the development of traits related to metabolism, stress tolerance, and pathogen resistance.

P6-152 - *Arabidopsis thaliana* as a Model Fungal-Pathosystem for *Elsinoë brasiliensis*

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Elsinoë brasiliensis is a fungal pathogen that affects many crops including *Manihot esculanta* (cassava). Across the Caribbean and Latin America the fungus causes Superelongation disease in cassava characterized by necrotic lesions and rapid stem growth, resulting in substantially decreased crop yield. *E. brasiliensis* produces gibberellic acid (GA), which is suggested to be responsible for rapid elongation symptoms. The

pathogen also produces elsinochrome pigments that are photosensitized perylenequinone phytotoxins. Similar phytotoxins from other fungal pathogens act as virulence factors, however, this has not been demonstrated for *E. brasilensis*. The purpose of this study is to determine if *Arabidopsis thaliana* can be used as a model pathosystem to study plant responses to *E. brasilensis*. Utilizing an *Arabidopsis* pathosystem will allow us to determine if 1) elsinochrome toxin produced by *E. brasilensis* acts as the virulence factor and 2) if the toxin affects plant GA synthesis and/or signalling. For this study, *Arabidopsis* seedlings were treated with increasing amounts of toxin. Expression of select GA pathway, pattern-triggered and effector-triggered immunity (PTI and ETI) genes were examined at four timepoints via reverse transcription-polymerase chain reaction (RT-PCR). In addition, RNA sequencing (RNASeq) analysis was used to observe global gene expression changes. Decreased expression of certain PTI genes was observed in the RT-PCR results and increased PTI gene expression was observed in the RNASeq analysis. ETI genes were observed to increase in expression in both the RT-PCR and RNASeq analyses. This indicates that an immune response consistent with exposure to a virulence factor occurred when *Arabidopsis* was exposed to toxin. GA signalling repressor genes increased in expression across the RT-PCR and RNASeq analyses. This suggests that GA signalling suppression is involved in the *Arabidopsis* response to toxin. These results will assist efforts to develop protection strategies to reduce the impact of *E. brasilensis* on cassava crop production.

P6-153 - Healthy Crops Start with Healthy Soil: Evaluating Humaterra's Compost-Derived Bioinoculants for Soil Health and Agronomic Yield

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Soil degradation threatens global food security by reducing crop productivity and biodiversity. Sustainable agricultural practices that enhance soil health while maintaining high yields are essential to address this challenge. This study evaluates the impact of Humaterra's compost-derived bioinoculants (H-start product) on soil health and agronomic yield of wheat (*Triticum aestivum*) and canola (*Brassica napus*) under controlled greenhouse conditions. Bioinoculants, composed of beneficial microorganisms, have the potential to improve soil microbial activity, nutrient availability, and plant resilience. 200 pots were utilized for this study in which each pot contained two biological replicates grown in a mixture of soil and sand (3:1). The treatment conditions were: (1) 100% FRR, (2) 75% FRR, (3) 100% FRR + H-start, (4) 75% FRR + H-start, (5) 0% FRR + H-start. Preliminary findings suggest that bioinoculant-treated soils exhibit improved agronomic yield in canola crops. By exploring bioinoculants as an alternative to synthetic inputs, this study contributes to the development of eco-friendly soil management strategies that enhance long-term agricultural sustainability. Future research will focus on field trials to validate greenhouse findings and assess long-term soil health benefits under real-world conditions.

P7-154 - Connecting stomatal conductance and ionic stress in heathland shrubs

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Soils contain a variety of ions in solution, including essential plant nutrients like nitrate and potassium as well as acids, metals, and salts that can be detrimental or even toxic at high concentrations. Soils with excessive ion concentrations occur naturally, such as coastal habitats and peat bogs, while others result from anthropogenic disturbances from, for example, industrial contamination, acid rain, or over-fertilization. A plant's ability to tolerate such ionic stress may be influenced by their water-use strategy, as high ion concentrations decrease soil water potential, limiting water uptake and lowering leaf water potential and turgor pressure. Given that changes in water potential are often seen first through stomatal regulation, variation in stomatal conductance (g_s) may be a useful trait to assess species' sensitivity or tolerance to ionic stress.

Using the LI-600, our lab is assessing how variation in ionic stress from marine salts, acid deposition, fertilizers, and metal pollutants influence g_s in heathland shrubs across a range of environments in eastern Canada. Sites include coastal forests and barrens, naturally acidic wetlands, and continental peatlands subjected to decades of experimental fertilization or metal contamination from copper and nickel mining. We are finding that, in general, g_s decreases with increasing ionic stress, although responses are species specific.

Some species respond in ways that mimick drought stress, with tight stomatal regulation and closure while other species are largely unresponsive across varying soil conditions. We report the lowest g_s in saline and acid-impacted sites, followed by metal-contaminated sites. Largest g_s have been in relatively undisturbed heathlands and forests, and in peatlands receiving intermediate levels of fertilization. We continue to investigate shared responses to water and ionic stress, with implications for plant resilience under increasingly variable and extreme environments.

P7-155 - Interspecific variation in methane emissions from wind-exposed plants

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Methane is a major greenhouse gas, second only to carbon dioxide for its contribution towards global climate change. In 2006, it was discovered that plants emit methane under aerobic conditions and since then it has been established that environmental stress factors, such as high temperature and drought, increase plant-derived methane emissions, while other factors, such as elevated carbon dioxide, can reduce emissions. Wind is an environmental stressor to which plants are commonly exposed but has not been investigated for its potential role in the production of methane emissions from plants. In this study, we selected two plant species - kale (*Brassica oleracea* L. var. *sabellica*), a wind resistant plant, and radish (*Raphanus sativus* L.), a wind sensitive plant - which were grown in environment-controlled growth chambers under a temperature regime of 22/18 °C (16 h day/8 h night) for 20 days. After the initial growth period, the plants were exposed to three wind velocities (0, 6, 12 km h⁻¹) for 1 hour each day, over a period of 10 days, after which methane emissions were collected and measured. Other traits, such as plant growth, biomass accumulation, chlorophyll fluorescence, nitrogen balance index, photosynthetic pigments, and water potential were also measured to thoroughly investigate the effects of wind on tolerant and intolerant plant species. We found that wind exposure increased methane emissions from both plant species, although not significantly. However, radish plants emitted significantly more methane in comparison to kale plants. Moreover, radish plants had significantly lower nitrogen balance index, chlorophyll content and non-photochemical quenching, but higher photochemical quenching compared to kale plants. The results of this study indicate that wind increased stress in both plant species, and that radish plants were more stressed by exposure to wind than kale plants, as shown by higher methane emissions and changes to physiological traits.

P7-156 - Unlocking the Potential of Biochar to Enhance Maize (*Zea mays* L.) Growth under Cadmium Stress

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Heavy metal contamination in agricultural soils and water bodies poses a significant challenge to food security and environmental health. This study investigates the efficacy of peanut shell-derived biochar in mitigating cadmium (Cd) stress in hydroponically grown maize plants. Biochar is a novel solution for mitigating abiotic stresses and remediating contaminated environments. The biochar's various properties, including bulk density, biochar yield, organic matter, pH level, electric conductivity, were assessed to determine its suitability as a soil amendment, and SEM, FTIR, and XRD techniques were employed to investigate the physicochemical features of the biochar, which was produced by pyrolysis at 500 °C. These techniques showed the surface area, porosity, maximum carbon and average carbon content, functional groups, crystalline nature, and adsorption capacity of biochar. Four treatments were tested: control (CK), Cd stress (100µM CdCl₂), biochar alone (1% BC), and biochar with Cd stress (BC+Cd). Cd stress raised levels of reactive oxygen species (ROS) and malondialdehyde (MDA) by 110%, which are indicators of oxidative stress, while dramatically decreasing plant development, photosynthetic pigments, relative water content, total soluble protein, and sugar levels. Co-application of biochar increased the amount of chlorophyll, antioxidant enzyme activity, and biomass of roots and shoots and decreased oxidative damage by lowering ROS production. According to these results, biochar can be employed as a sustainable amendment to reduce heavy metal toxicity, promote plant growth, and boost crop stress tolerance.

P7-157 - The effect of isoprene on membrane protein composition, growth and reproduction

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Significant reductions in crop yields have been predicted under global warming, and Canadian farmers are already experiencing crop losses. Thus, understanding the molecular and cellular mechanisms that protect leaves and reproductive organs in plants from high temperature will facilitate the development of new and sustainable tools to improve the resilience of crops. Isoprene, a volatile molecule produced by some plants, can protect plants from different stresses, notably heat stress. The mechanism behind how isoprene protects plants from heat is poorly known. The objectives of the proposed master's project are: **(1)** to find whether isoprene has the ability to alter membrane protein composition in leaves, **(2)** to understand the effects of isoprene as a long-distance signaling molecule on the growth and resilience of non-emitting plants in the vicinity of an emitter, and **(3)** to investigate the effect of isoprene on the reproductive success of emitting plants, under heat stress. For the first objective, proteins will be extracted from different membranes isolated from *Arabidopsis thaliana* rosettes exposed to either air (control) or 20 ppm isoprene for 1h or 5h under ambient or heat stress, and analyzed using liquid chromatography/mass spectrometry. I hypothesize that isoprene can affect protein recycling and consequently membrane protein composition. For the second objective, non-emitting Arabidopsis plants will be placed near isoprene-emitting trees and the growth of Arabidopsis will be monitored over time along with their tolerance to viral infections. Finally, I will assess the effect of isoprene on the reproductive success of plants by monitoring inflorescence growth, pollen viability and seed production of non-emitting and isoprene emitting Arabidopsis grown under ambient and high temperature. Collectively, data from the proposed study will provide valuable information on the possible use of isoprene as a tool to mitigate the significant yield reductions predicted under future climate scenarios.

P7-158 - Assessing the Impact of *Bri1* and *Dwf4* Mutations on Drought Tolerance in Canola

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There is mounting evidence for the antagonistic relationship between brassinosteroid (BR) and Absciscic acid (ABA) signaling pathways during plant development and stress adaptation. To exploit this interaction to improve drought tolerance in canola, we have created gene-edited canola lines compromised in both the BR biosynthetic enzyme, *DWF4* (both A and C copies) and BR receptor, *BRI1* (C copy only). In this study, we have investigated the drought tolerance in unmodified and double mutant (*dwf4/bri1*) canola lines. Both mutant and wild type plants were exposed to drought stress (30% soil moisture after 6 leaf stage), and their phenotypic responses were tracked. Our initial findings indicate that *dwf4/bri1* mutant displayed improved drought tolerance traits, such as reduced wilting, better-maintained seed production, and improved seed quality. These effects may stem from BR deficiency traits, including smaller plant size, decreased water requirements, and potentially improved water use efficiency. Furthermore, suppressed BR signaling might enhance drought-responsive mechanisms, such as absciscic acid (ABA)-mediated stomatal control and activation of stress-related genes. These results suggest that targeting BR-related genes like *DWF4* and *BRI1* could be a viable strategy for enhancing drought resilience in canola.

P7-159 - Blue light-mediated regulation of canola growth and methane emissions

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Earlier studies have shown that blue light influences plant growth and aerobically-produced methane emissions from plants. It has also been well documented that blue light affects stomatal form and function. However, the effects of blue light on plant growth and stomatal density and the relationship of plant-derived methane emissions with stomatal characteristics have not been fully addressed. We therefore investigated the effect of

blue light on plant growth, physiological traits, stomatal density and methane emissions in canola (*Brassica napus* L. cv. 6056 CR). Plants were grown from seeds under a temperature regime of 22/18°C (16 h light/8 h dark), and photosynthetic photon flux density of 300 $\mu\text{mol m}^{-2} \text{s}^{-1}$. After two weeks, half of the plants were exposed to supplemental blue light (4 mW cm^{-2}). Plant growth (stem height and diameter, internode length, stem branching, leaf number, leaf area), biomass (leaf, stem, root, total), stomatal characteristics (stomata number, stomata size), physiological traits (nitrogen balance index, chlorophyll, flavonoids, effective quantum yield of photosystem II, maximum quantum yield of PSII, nonphotochemical quenching, photochemical quenching), reproductive yield (silique length, silique diameter, silique number per plant) and methane emissions were measured, using at least three biological replications. In this study, blue light influenced many plant growth and physiological traits, including a two-week delay in the initiation of reproductive phase. Supplemental blue light significantly decreased stem diameter, internode length, stem branching, plant biomass, chlorophyll content, flavonoid content, and silique size, but increased the number of stomata and the number of siliques. Blue light also caused an increase in the aerobically-produced methane emissions from plants. Overall, the results suggest that blue light influences plant growth, physiology and aerobic methane emissions, and there is a strong positive relationship between methane emission and stomatal density. These findings may have important implications for understanding the mechanism of plant-derived methane.

P7-160 - Ecophysiological responses of Atlantic heath shrubs across coastal gradients

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Coastal heathlands are unique ecosystems in Nova Scotia and are considered endangered in North America. They can harbor endemic and at-risk plant species that provide economical and ecological importance. Despite their importance, these ecosystems are at a risk of increasing salt exposure from climate change impacts such as sea level rise, intensified storms, and coastal erosion. These impacts can change species distribution patterns and filter out more salt sensitive plants. However, the potential vulnerability to saline conditions is not currently known for many heathland plants in Nova Scotia. Therefore, understanding the effects of salinity on plant diversity and function is crucial for conserving biodiversity and assessing ecological resilience in this region. To address this, I am measuring variation in heath diversity, abundance, and ecophysiological traits along coastal gradients in Polly's Cove and Duncan's Cove. I am measuring stomatal conductance and chlorophyll fluorescence, salt tolerance (leaf Na/K), and morphology (plant height, specific leaf area and thickness) in six dominant Ericaceae shrub species: *Empetrum nigrum*, *Corema conradii*, *Gaylussacia baccata*, *G. bigeloviana*, *Vaccinium angustifolium* and *V. macrocarpon*. I predict that low-growing evergreen species such as crowberries (*Empetrum*, *Corema*) with small, thick leaves will be more salt tolerant and abundant closer to the coast than deciduous shrubs like huckleberries and blueberries (*Gaylussacia*, *Vaccinium*) with larger, thinner leaves. Therefore, I expect a more stable Na/K ratio, lower stomatal conductance, and higher electron transport rate with increased PS II activity in evergreen shrubs compared to deciduous shrubs as well as in plants closer to the coast than further inland areas. This study supports the identification of salt sensitive species that may need targeted conservation efforts and habitat management to ensure their survival under future elevated salt exposure in the context of climate change.

P7-161 - Determining plant responses to metal contamination: Implications for restoration

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Peatlands are terrestrial wetlands that play a vital role in combatting climate change by acting as long-term carbon (C) sinks. Anthropogenic disturbances such as industrial contamination can negatively impact peatland plant communities leading to reductions in their C-storage capacity. One well known example of these disturbances is the copper (Cu) and nickel (Ni) mining in Sudbury, ON. Sulfuric acid and toxic levels of metal particulate deposition resulted in the loss of biodiversity and ecosystem function. Despite much success in the greening of the lakes and forests, restoration has yet to be attempted for the extensive area of peatlands in

the Sudbury region. Peatlands close to the smelters are still heavily contaminated with Cu and Ni and have lost much of their natural plant diversity. There has been complete loss of moss species in the contaminated sites, which results in a major loss to ecosystem C-storage capabilities. To effectively restore these contaminated peatlands, it is critical to first understand the biological impacts of metal contamination on peatland plants, especially the keystone *Sphagnum* mosses.

We assessed variation in photosynthesis, morphology, and leaf chemistry in three fen peatlands ranging from high disturbance to outside the contamination zone. We measured light-saturated photosynthesis, specific leaf area, specific capitula area, capitula size, and tissue P, Cu, and Ni concentrations in dominant shrub and moss species: *Chamaedaphne calyculata*, *Kalmia angustifolia*, *Sphagnum fuscum*, and *S. rubellum*.

We saw a decrease in photosynthesis and capitula size with increasing contamination in *Sphagnum rubellum*. Interestingly, we found no negative effects of metal contamination on photosynthesis in the shrubs or *Sphagnum fuscum*, despite differences in leaf metal concentrations. These results provide hopeful species recolonization potential in the heavily contaminated sites, particularly for *Sphagnum fuscum*, thus informing restoration for a healthy C storing state.

P7-162 - Effects of legacy metal pollution on *Sphagnum* recovery in contaminated peatlands

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Peatlands are the strongest terrestrial carbon (C) sink, storing approximately 25% of the global soil C stock. In northern peatlands, *Sphagnum* mosses are the keystone species that create and maintain peatland functions. If the *Sphagnum* layer is removed or disturbed, peatlands can shift from C sinks to sources contributing to climate change. One example of industrial disturbance is Sudbury, Ontario, where over a century of nickel and copper mining has contaminated the area with metal and sulphuric acid deposition, decimating vegetation including the local extinction of *Sphagnum* mosses. Despite reductions in air pollution starting in the 1970's, there is no *Sphagnum* recolonization in the most contaminated peatlands. However, recent evidence shows some localized moss recolonization where other vegetation has recovered via greening efforts, but the spatial extent and mechanisms of the moss growth is unknown. In order to better understand the potential for *Sphagnum* recolonization on metal contaminated peat, I am targeting highly contaminated peatlands to identify the extent of moss colonies and measure colony height, area, species composition, and what substrates moss are growing on. It is unlikely *Sphagnum* moss are growing directly on contaminated peat and instead a substrate acting as a buffer (such as herbaceous or woody material). I have also set up a controlled restoration growth experiment where I am testing the effects of different buffer substrates on the growth and functional responses of two *Sphagnum* species: *S. fallax* and *S. fuscum*. I am measuring moss growth, metal and nutrient content of the capitula and stem, and both gross and microscopic morphological characteristics, such as fascicle length, leaf area, and hyaline cell density. Determining the conditions that promote recolonization will be critical for restoration efforts aimed at returning *Sphagnum* and peatland C storage to Sudbury's impacted landscape.

P7-163 - In vitro and greenhouse germination and propagation of *Iva frutescens* L.

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Iva frutescens L., commonly known as marsh elder, is a shrub native to Nova Scotia salt marshes. Dense stands provide numerous ecosystem services, including wave energy attenuation, sediment stabilization, erosion control, habitat protection, pollinator support, and carbon storage. This species tolerates a range of salinities although is intolerant to flooding, restricting its distribution to narrow belts at higher elevations. This study explored *in vitro* and greenhouse germination and propagation of *I. frutescens* to develop protocols for *ex situ* conservation and site recovery strategies. Seeds were collected in Fall 2024 from Atlantic coast (Surettes Island, Johnson Cove) and Bay of Fundy (Port Williams) salt marshes. Multiple variables were tested for

greenhouse growth, including seed coat removal, cold stratification, and growth hormone application. *In vitro*, seeds were placed onto four nutrient media types using plant tissue culture to assess germination and propagation response. *In vitro* trials revealed Johnson Cove seeds had high germination rates (85%) over 21 days when cold stratified and placed on saline and growth hormone media with no seed coat. *In vitro* germination varied significantly for cold stratified seeds, presence/absence of seed coats, and media type. Greenhouse germination varied significantly by collection site and seed coat presence. This is the first study to assess and successfully use *in vitro* micropropagation on *I. frutescens*. *In vitro* and greenhouse derived plantlets remain under observation. These protocols are expected to assist with vegetative recovery of restored salt marshes and nature-based coastal management projects.

P7-164 - Enhancing Coastal Resilience: Marsh Elder (*Iva frutescens*) as a nature based solution

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Tidal wetlands provide important ecosystem services such as wave and storm surge buffering, erosion prevention, and essential habitats for many species. These habitats are dominated by grasses and forbs, whose aboveground structures reduce overland flow rates and trap sediment, mitigating the effects of coastal erosion. However, these structures die back in the winter, leaving the coastline vulnerable. Marsh Elder (*Iva frutescens*) is a native salt marsh shrub found along the eastern coast of North America. In Nova Scotia, it is the only shrub species that occurs in salt marshes and is exclusively found in the Minas Basin and Yarmouth areas. Marsh Elder has a greater salinity tolerance than other shrubs, allowing it to grow lower in the tidal frame. This shrub is potentially valuable to living shoreline and dyke foreshore projects as its woody structures are present in winter, providing year-round erosion control. Despite its potential importance, little research has been conducted on Marsh Elder. This research aims to determine Marsh Elder's ecological niche (elevation, inundation frequency, soil nutrients and characteristics, and plant associations) and identify different planting techniques to aid in the successful integration of Marsh Elder into living shorelines. This study will provide the first ecological assessment and planting experiment of Marsh Elder in Canada.

P7-165 - Hyperspectral Fluorescence Imaging Systems for Biological Component Analysis

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For the isolation of both natural and tagged fluorescence within biological samples we explore an in-lab developed fluorescence imaging system [1] and parallel factor analysis techniques [2]. Different methods for excitation were used for this work, including direct mercury lamp ultraviolet (UV) illumination and evanescent wave illumination with a series of fiber-coupled lasers in either the UV or visible light range. Using both methods, fluorescence of a given plant tissue microscopy sample was recorded between 400 and 1100 nm for each pixel using a hyperspectral camera, thus collecting an excitation emission matrix for each pixel across the sample to serve as signatures for unique components within a heterogeneous sample.

To examine potential limitations of this technique for the *in vivo* spatial and spectral isolation of fluorophores, transgenic *Arabidopsis thaliana* plants expressing one to four green fluorescent protein variants with known localization were used [3]. Specific genetic lines were selected to align the peak absorption of the fluorescent proteins with the available visible light spectrum lasers to optimize excitation under the evanescent wave illumination technique.

As an example of autofluorescent compounds with broad excitation and emission ranges between the UV and visible spectrum, we targeted cross sections of plants covering both woody and green bark tissues. Preliminary analyses targeted the phenolic biopolymer lignin and the spectrally distinct photosynthetic pigment chlorophyll [4]. Further analyses evaluated potential spectral differences in lignin originating from distinct species to assess power of the system to infer compositional differences. Specifically, angiosperm and gymnosperm species with well characterized syringyl and guaiacyl lignin monomer ratios were considered.

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P7-166 - Immunoquantification and Activity Analysis of Dimethylsulfoniopropionate Enzymes from *Ulva* sp. Subjected to Altered Salinity

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The organosulfur compound, dimethylsulfoniopropionate (DMSP), is an important osmolyte produced and utilized by many marine species, including algae and phytoplankton, and is especially important in those species inhabiting intertidal zones which encounter frequent fluctuations in salinity. Previous studies have shown that intracellular DMSP content fluctuates with abrupt changes in salinity, but the mechanism by which this is regulated is not well known. To address this, the present study aims to investigate if these changes in DMSP content in response to salinity changes are regulated at the level of the DMSP biosynthesis enzymes methionine aminotransferase (Met AT) and methylthiohydroxybutyrate S-methyltransferase in *Ulva* sp. To investigate this, protein was extracted from *Ulva* sp. tissues that had undergone salinity stress trials, then immunoquantification of these enzymes of interest was attempted and Met AT activity was analyzed. From these analyses it was determined that there is some large macromolecule present in the native *Ulva* sp. protein extract that interferes with DMSP enzyme binding to membranes used in immunoquantification. Furthermore, it was determined that there is no clear trend in Met AT activity in response to changes in salinity. Further analysis of DMSP biosynthesis enzyme activities and DMSP degradation enzymes, as well as quantification of compounds such as sugars and other metabolites, should be conducted in order to gain further insight into how DMSP is regulated as an osmoprotectant.

P7-167 - The protection of algal lipids and proteins from singlet-oxygen mediated damage by nonphotochemical quenching

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Nonphotochemical quenching (NPQ) plays a crucial role in protecting photosynthetic organisms from the damaging effects of excess light by dissipating absorbed energy as heat. This study investigates the impact of NPQ on protecting lipids and proteins from singlet-oxygen mediated damage in three strains of the green alga *Chlamydomonas*: wild type *C. reinhardtii* (WT), a high-light tolerant mutant (CC-4107), and a species isolated from a Saskatchewan water body (DJX-J). Using Fourier transform infrared (FTIR) spectromicroscopy, we examined the biochemical responses of individual living algal cells to high light and the photosensitizer rose bengal. Our results indicate that the DJX-J strain, despite its high capacity for NPQ, exhibited significant lipid and protein damage under oxidative stress, suggesting a trade-off between NPQ efficiency and cellular damage. In contrast, the CC-4107 strain, with its stable accumulation of zeaxanthin, showed enhanced protection against oxidative damage. These findings highlight the complex interplay between NPQ mechanisms and cellular protection, providing insights into the adaptive strategies of photosynthetic organisms under stress conditions.

P8-168 - Investigating the Effect of Chemistry on the Cuticular Water Barrier in Rosaceae

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The plant cuticle is a hydrophobic barrier composed of a complex mixture of wax compounds that coats all aerial surfaces, including leaves and fruit. While the cuticle plays a crucial role in reducing water loss, the relationship between its chemical composition and permeability remains poorly understood. This study investigates whether specific cuticular wax constituents correlate with water barrier effectiveness in the leaves and fruit of Rosaceae species in British Columbia, Canada. To address this, we analyze cuticular composition using gas chromatography-mass spectrometry (GC-MS) and gas chromatography-flame ionization detection (GC-FID) to characterize and quantify wax compounds. Additionally, we use scanning electron microscopy (SEM) to detect the presence of epicuticular wax crystals. Permeability is assessed through water loss measurements in intact fruit and across leaf discs. By integrating these datasets, we aim to identify relationships between cuticle composition and structure with reduced permeability. These results allow us to speculate on the molecular arrangement of wax compound classes in the cuticle and better conceptualize how water moves through this largely hydrophobic layer. Insights from this work may inform future molecular studies on enhancing drought tolerance and improving post-harvest traits, such as prolonged shelf life, in agricultural and horticultural contexts.

P8-169 - SINE-ing PHS off: a potential marker for building resistance

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Pre-harvest sprouting (PHS) refers to precocious germination of wheat grain within the grain head, occurring due to excessive rainfall. Already a major cause of yield and quality loss, the effects may become even more pronounced under climate-change scenario. Although breeding and genetic interventions have had some success, emerging evidence indicates that RNA-directed DNA methylation (RdDM) pathway mediated by Argonaute (AGO) proteins plays a crucial role in regulating seed dormancy and PHS tolerance. In the current study, the role of the TaAGO802B gene in PHS tolerance was probed, with focus on the presence of a short interspersed nuclear element (SINE) retrotransposon insertion within its 3'UTR. A panel consisting of 41 diverse wheat cultivars elucidated strong association between PHS tolerance and SINE. SINE-insertion was observed in 73.2% of the total lines and 92.6% of the PHS-tolerant cultivars. The SINE-insertion decreased global DNA methylation levels by 54.7% and TaAGO802 expression by 73.3% when compared to cultivars without the insertion. A strong positive correlation between PHS score and methylation levels was also seen. This raises the possibility that SINE-insertion has a silencing effect and might be functioning as a cis-regulatory sequence obstructing RdDM pathway. The SINE element thus contributes to dormancy regulation by modulating the AGO802B. Subsequent genotypic analysis of a DH population (Sadash × P2711) supported the hypothesis that the SINE insertion co-segregates with PHS resistance. Thus, our findings identify the SINE insertion in TaAGO802B as both a putative epigenetic regulator and a potential marker for screening wheat germplasm for developing PHS-tolerant cultivars.

P8-170 - Europium toxicity in *Lemna minor* is not related to phosphorus deficiency.

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A potential mechanism of toxicity of Rare Earth Elements, including europium, is phosphorus deficiency. This claim stems from the fact that these elements are known to form insoluble complexes with phosphate, preventing the plant from absorbing the nutrient. To investigate this claim, the extent of the europium-induced phosphorus deficiency in *Lemna minor* was studied. A chemical equilibrium modelling software determined that all the europium was phosphate-bound in Hoagland's E+ growth medium. To test the relative magnitude of

europium-induced phosphorus deficiency, four treatment groups comprised of modified Hoagland's E+ were compared: a control, one spiked with $\text{Eu}(\text{NO}_3)_3$, one with reduced KH_2PO_4 , and the final group with both $\text{Eu}(\text{NO}_3)_3$ and additional KH_2PO_4 . The frond count, surface area and chlorophyll a content were measured; plants treated with europium were 53-89% of control, whereas the phosphate-deficient plants had reductions of only 71-97% of control and adding phosphate did not counteract europium toxicity. Chlorophyll b contents did not vary among the europium and phosphate treatments and were about 66% of the control. By comparing the magnitude of responses to europium and phosphorus deficiency on frond count and surface area, I have determined that 50-60% of europium toxicity can be explained by phosphorus deficiency. To further understand the direct effects of europium on the plant, ICP-MS will be used to quantify the uptake of europium and total phosphorus content. I will also measure both alkaline and acidic phosphatase activities to quantify the magnitude of europium-induced phosphorus deficiency.

P8-171 - Allantoin-Mediated Stress Response in Arabidopsis Seedlings under Excess Potassium

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Soil salinity refers to the concentration of soluble salts in the soil, typically comprising Na^+ , K^+ , and other ions. Approximately 85% of salt-affected topsoils are classified as agriculturally unfavorable saline soils due to a mix of salts, including both sodium and potassium. However, most studies on plant salinity stress focus primarily on Na^+ , leaving the effects of excess K^+ largely underexplored. Allantoin is a heterocyclic nitrogenous compound found in plants and is an intermediate product of purine catabolism. It has been implicated in enhancing tolerance to various abiotic stresses, likely through its roles in scavenging reactive oxygen species and activating abscisic acid biosynthesis. Prior research has demonstrated that exogenous allantoin application increases *Arabidopsis* tolerance to high sodium levels and upregulates the expression of antioxidant enzyme genes in response to NaCl stress. In this study, we investigated the effects of excess K^+ and exogenous allantoin on the growth and development of *Arabidopsis thaliana* seedlings. High K^+ concentrations caused toxicity symptoms and reduced biomass, whereas allantoin-treated seedlings showed improved tolerance, including greater biomass and root length. We further analyzed the expression of stress-related and potassium transporter genes. Allantoin treatment downregulated membrane-associated basic leucine zipper (bZIP) transcription factors, which are putative ER stress sensors, and modulated the expression of several potassium transporter genes in seedlings exposed to high K^+ .

P8-172 - The role of reticulon-like B3 as a receptor for autophagy of the endoplasmic reticulum in Arabidopsis

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Plants as sessile organisms evolved many sophisticated cellular mechanisms that allow them to tolerate unfavourable conditions in their environment. One of these mechanisms is autophagy, the self-degradation of damaged cellular components for clearing out cellular waste and recycling resources in stress recovery. A common autophagy target is the endoplasmic reticulum (ER), the largest organelle known for its efficient synthesis, modification and transport of proteins. The ER is an interconnected network of membrane-bound tubules and sheets but its actual shape consistently remodels to meet the need of cell development and response to environmental challenges. Reticulons are a family of ER-localized proteins that promote membrane curvatures needed for the formation of ER tubules and edges of ER sheets. Recent studies in mammalian cells indicated that Reticulon3 also serves as a receptor for autophagy of the ER (ER-phagy) needed for proper remodeling of the ER. In *Arabidopsis*, there are 21 reticulons, reticulon-like B3 (RTNLB3) being one of the common reticulons known for providing curvature to the ER membrane. We recently found that our *Arabidopsis rtnlb3* mutant is hypersensitive to tunicamycin, an antibiotic that inhibits protein glycosylation in the ER, causing the accumulation of unfolded proteins and inducing ER stress. Based on this, we hypothesize that RTNLB3 acts as an ER-phagy receptor in *Arabidopsis* for proper remodeling of the ER in stress response. To conclude this hypothesis, we will 1) examine if *rtnlb3* is hypersensitive to carbon starvation,

a condition that induces autophagy of the ER, 2) test if ER-phagy in *rtnlb3* is defective or not in ER stress, 3) investigate if RTNLB3 interacts with autophagy-related 8 (ATG8), a key ligand that interacts with a specific receptor for cargo recruitment into autophagophores.

P8-173 - Variation in *Chamaedaphne calyculata* ecophysiology across a smelter pollution gradient in Sudbury, Ontario

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Industrial contamination alters ecosystems with negative impacts on biodiversity and ecosystem function, with sulfur (S) and some trace metals like copper (Cu) and nickel (Ni) being toxic to plants at high concentrations. Excess Cu and Ni can impact photosynthesis by structural and functional destruction of chlorophyll thylakoids, inhibiting Photosystem II (PSII) efficiency, and decreasing stomatal conductance. S compounds accumulating in plants impairs metabolism such as reduced mesophyll conductance. In Sudbury, Ontario, thousands of hectares of forests and wetlands have suffered almost complete vegetation loss from metal and sulfur dioxide pollution. Despite successful forest restoration since the 1970's, many metal-contaminated peatlands and wetlands remain species-poor with no *Sphagnum* cover and are dominated by a single shrub, *Chamaedaphne calyculata*. It remains unclear why *C. calyculata* can survive in these contaminated wetlands and if its growth could support or hinder *Sphagnum* restoration efforts. To address this gap, my research question is: How does *C. calyculata* ecophysiology vary across a gradient of increasing Cu, Ni, and S? I measured *C. calyculata* ecophysiology in contaminated and reference (low impact) peatlands and shoreline wetlands. Stomatal conductance (g_{sw}) and chlorophyll fluorescence were measured on 10 leaves per individual for 216 plants across 12 sites. *Chamaedaphne calyculata* displayed variation with peatland plants being taller with larger leaves and higher PSII quantum efficiency in the reference sites but similar g_{sw} and electron transport rate across contamination levels. Shoreline plants were shorter in the reference sites, but had similar leaf size, g_{sw} , PSII quantum efficiency, and electron transport rate across contamination levels. These results indicate that *C. calyculata* is resilient to metal contamination and responds differently in shorelines compared to peatlands, possibly due to ecosystem changes from adjacent limed lakes and forests. This study provides baseline data to inform current and future restoration efforts in Sudbury and other metal-contaminated wetlands.

P8-174 - Accumulation of allantoin in response to abiotic stress in Arabidopsis (*Arabidopsis thaliana*)

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Plants are constantly exposed to a wide variety of biotic and abiotic stresses which influence their growth and development. These stresses include fluctuations in temperature, extremes of salinity, exposure to heavy metals and other toxic substances, variation in water availability, and encounters with pathogens and pests. The impact of these stresses on agriculture is resulting in substantial crop yield losses that incur heavy economic costs on a global scale. To cope with these stresses, plants have evolved complex physiological and molecular responses regulated by various signaling and metabolic pathways. A common mechanism is the synthesis of specific metabolites that act to mitigate the effects of stress. Allantoin is a nitrogen-rich compound and an intermediate in the ureide catabolic pathway responsible for recycling nitrogen from purine bases in plants and other organisms. Some studies indicate that allantoin may also be involved in various abiotic stress responses, yet the specific mechanism by which it contributes to stress tolerance in plants has not been resolved. Absciscic acid (ABA) is one of the phytohormones involved in a variety of physiological processes in plants including stomatal closure, seed dormancy and abiotic stress tolerance. It is synthesized via de novo and conjugation pathways, with 9-cis-epoxycarotenoid dioxygenase (NCED) acting as a key enzyme in the synthesis pathway. We employed a genetic approach using Arabidopsis mutants to explore how ABA signaling interacts with allantoin accumulation during salt stress. Using mutants like *aln-3* (deficient in allantoinase), *ALNox* (overexpressing allantoinase) and several ABA mutants (*abi*, *nced3*), we ask how these responses are connected and begin to look at the physical and biochemical impacts of stress in these genotypes. This research aims to enhance our understanding of the interaction between nitrogen metabolism and stress-

responsive hormone signaling in plants, potentially guiding strategies to engineer crops with greater tolerance to environmental stressors

P8-175 - Transcriptome analysis of beneficial and inhibitory effects of nitrate on symbiotic nitrogen fixation in common bean

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Both beneficial and inhibitory effects of nitrate concentrations on symbiotic nitrogen fixation (SNF) in legumes were observed. The objective of this study was to evaluate different nitrate concentrations on SNF in common bean (*Phaseolus vulgaris*). Six bean genotypes were grown in a growth chamber under six nitrate concentrations (0-20 mM) in a split-plot design, with nitrate concentrations as the main plot factor and genotypes as the subplot factor. Low nitrate concentrations (≤ 5 mM) enhanced nodule formation and biomass, whereas concentrations above 10 mM inhibited nodulation, reducing nodule number and biomass. Total RNA from root samples of selected genotypes at three nitrate concentrations (0, 5, and 10 mM) was extracted for RNA sequencing. Analyses of differentially expressed genes (DEGs), gene ontology (GO), Kyoto Encyclopedia of Genes and Genomes (KEGG) pathways, protein-protein interactions, and transcription factors were performed. Four key pathways were identified in the root nodulation response to low (5 mM) and high (10 mM) nitrate concentrations: metabolic pathways, biosynthesis of secondary metabolites, ribosome, and carbon metabolism. Key transcription factors, primarily involved in plant stress and development, were identified, including auxin-responsive factor, LSD1-like 3, SALT TOLERANCE HOMOLOG2, and BEL1-like homeodomain 10. These findings provide molecular insights into the mechanisms regulating SNF under varying nitrate concentrations.

P8-176 - Developing new tools to quantify plant endomembrane dynamics

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The plant endomembrane system is composed of complex intercellular compartments that deliver molecular cargo in and out of the cell. The endomembrane system participates in almost all aspects of cell biology, from cell division and expansion to differentiation and functional specialisation. We aim to expand our knowledge of endomembrane compartment identity and regulation by developing new tools for measuring compartment populations, identifying vesicle cargos, and testing the cellular effects of a disordered endomembrane system. We developed a fluorescence assisted vesicle sorting (FAVS) method to quantify endomembrane compartment populations and dynamics, with preliminary results indicating that fluorescent protein-fusion constructs can be used to count and sort specific vesicle populations. We can also express these fluorescent proteins in Arabidopsis vesicle trafficking mutant lines to examine how trafficking patterns are coordinated and what molecular mediators are required for endomembrane organisation. Lastly, we are exploring how endomembrane organisation and function affect cellular biomechanics such as cell wall elasticity and cell turgidity. Understanding these complex systems could open new doors for engineering plant-based resources to produce useful biomolecules or developing plants that are better suited for changing or new growth conditions.

P9-177 - A One-Day Workshop to Generate Interest in Plant Biochemistry Amongst Pre-College Youth

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As research on improving plant growth and resilience to stress takes center stage in finding solutions to the impacts of climate change on global food production, so has the need for more plant scientists. However, a steady recruitment of plant scientists can only be achieved by more people taking on careers in science, technology, engineering and math (STEM). In Canada, only about 20% of the population aged 25 - 64 has a postsecondary certificate, diploma or degree in the STEM fields. Furthermore, less than 50% of Canadian students pursue STEM education above secondary school. This concerning disconnection about young people has been shown to be largely due to their negative view about science, the inability to draw the link between STEM education and future career paths, and inaccessibility to active and integrated learning approaches. As plant biologists, we have the capacity to reach out to pre-college youth to teach them about plants and their importance in everyday life. We have developed a fun and highly interactive workshop titled: "Antioxidants in food: Do strawberries have more antioxidants than blueberries?" for high school students. The short-term goals of the workshop were to provide: (1) a basic understanding of the importance of plant biochemistry for human health and nutrition, (2) hands-on experience on how to conduct a biochemical assay and analyze data, and (3) an opportunity to talk one on one in an informal setting with scientists, undergraduate and graduate students working in the field of plant biology and biochemistry. The long-term objective was to motivate and enhance enthusiasm in plant biology amongst precollege youth and help them realize their academic and career goals. Details of the workshop and outcomes will be discussed.

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