Conference Booklet for Attendees

Schedule Information

Directions, Maps and Parking

Abstracts

Participant List

ORGANIZING COMMITTEE

Danielle Way (Chair), Mark Bernards, Norman Hüner, Susanne Kohalmi

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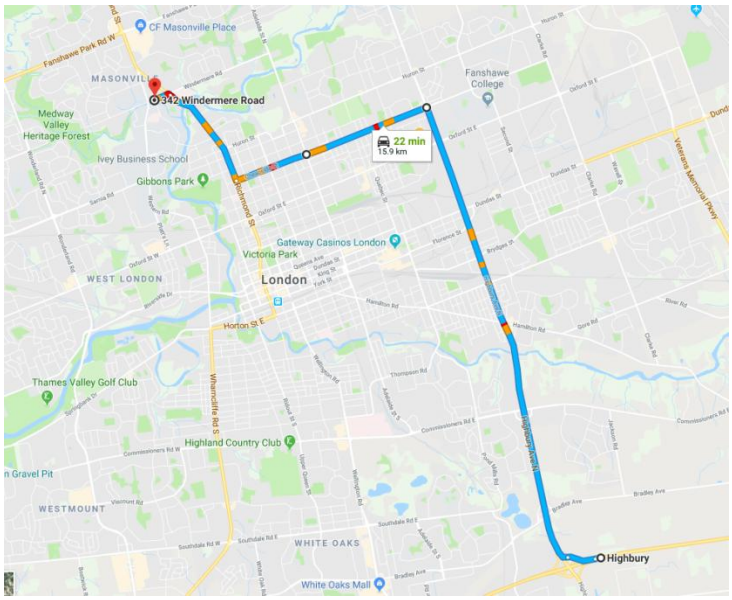
DRIVING INSTRUCTIONS and MAPS



One of the fastest ways to reach Western from the 401 (avoiding downtown area):

1. Take the Highbury exit and go North on Highbury

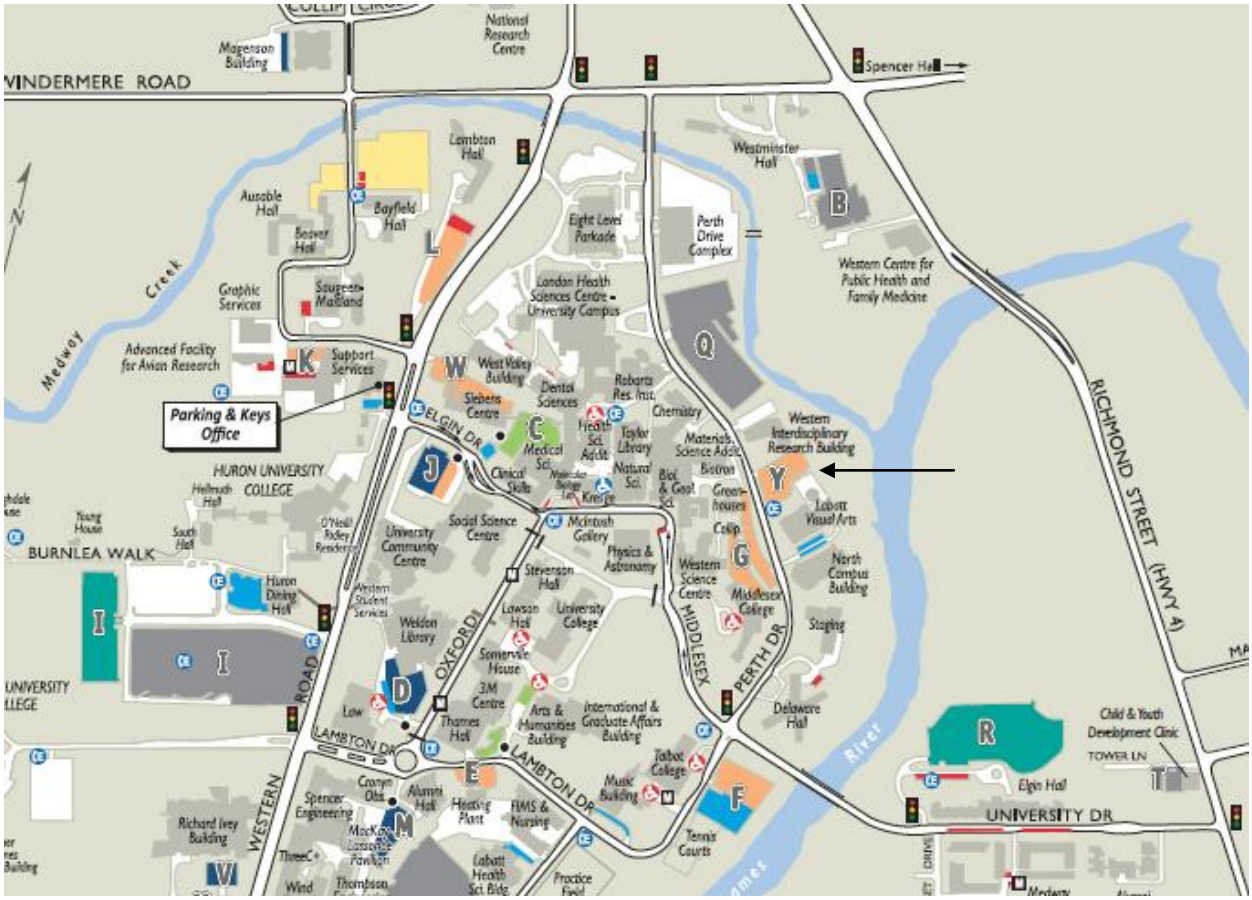
2. Turn left onto Cheapside

3. Turn right onto Richmond

4. Go PAST the University main gate as the bridge over the Thames is CLOSED

5. Turn left onto Windermere

6. Turn left onto Perth Drive



Please park in the Y lot between the Interdisciplinary Research and Visual Arts Building, marked by the arrow. The North Campus Building (NCB) is on the other side of the Visual Arts Building.

SCHEDULE

Saturday, November 24

7:30-9:00 **Registration, poster setup,** NCB atrium

9:00-9:20 **Welcoming remarks and by-law vote, Room NCB 113**

9:20-10:10 **PLENARY LECTURE I: Room NCB 113**
Arunika Gunawardena, Dalhousie University
Lace plant: An emerging model system to study developmental programmed cell death

10:10-10:30 **Coffee Break,** NCB atrium

10:30-12:00 **CONCURRENT SESSIONS (Rooms NCB 113, NCB 114 and NCB 117)**

ABIOTIC STRESS: Room NCB 113

10:30-10:45 **AS1** Lauren Erland
Direct visualization of location and uptake of melatonin and serotonin in living tissues and their redistribution in plants in response to thermal stress

10:45-11:00 **AS2** Nathieli Beltran Schiavi
Assessing the role of the Arabidopsis ubiquitin ligase XBAT31 in Fe deficiency stress response

11:00-11:15 **AS3** Nicolas Dimopoulos
Compositional changes of the grape berry (Vitis vinifera L.) cuticle during fruit development in response to water deficit stress

11:15-11:30 **AS4** Biruk A Feyissa
SPL13 and WD40-1 regulate DFR to enhance anthocyanin biosynthesis for stress tolerance in Medicago sativa

11:30-11:45 **AS5** Eliana Vonapartis
Transcriptional regulation of XERICO modulates abiotic stress response in Arabidopsis thaliana

11:45-12:00 **AS6** Marina Cvetkovska
The enigmatic loss of light-independent chlorophyll biosynthesis from an Antarctic green alga in a light-limited environment

BIOCHEMISTRY: Room NCB 114

10:30-10:45 **B1** Artyom Gritsunov
Investigation of quinate metabolism in plants

10:45-11:00 **B2** Michael Kanaris
Evolutionary insights into the role of shikimate kinase-like 1 in chloroplast development

11:00-11:15 **B3** Trish Tully
Characterization of CYP86A genes and their role aliphatic suberin deposition in soybean roots

11:15-11:30 **B4** Lisa Amyot
Assessment of glucosinolate content in Camelina sativa and its wild relatives

11:30-11:45 **B5** Kristen Van Gelder
Plant polyprenols and their role in thylakoid membrane dynamics

11:45-12:00 **B6** Nishat Islam
Pinto bean (Phaseolus vulgaris L.) orthologue of Arabidopsis TT8 regulates pro-anthocyanidin genes and seed coat darkening

DEVELOPMENT, ENZYMES AND MOLECULAR BIOLOGY: NCB 117

10:30-10:45 **DEB1** Deka Mohamed
Investigating the role of XERICO in stomatal development and cell wall function

10:45-11:00 **DEB2** Carina Carianopol
The role of Arabidopsis ABI5 binding protein 2 (AFP2) in HR-like programmed cell death

11:00-11:15 **DEB3** Purva Karia
The mitochondrial tail-anchored protein TTM1 mediates ABA-induced senescence

11:15-11:30 **DEB4** Mark Minow
Stability and change in Zea mays small RNA transcriptomes provide exciting insights into the formation of new epigenetic states

11:30-11:45 **DEB5** Alberto Torrez
Identifying interacting proteins with BdHD1 in Brachypodium distachyon

11:45-12:00 **DEB6** Emily Clayton
Piggybacking AtADT5 into the nucleus

12:00-13:00 **Lunch,** NCB atrium

13:00-14:00 **Poster session and Sponsor Exhibitions,** NCB atrium

14:00-15:15 **CONCURRENT SESSIONS (Rooms NCB 113, NCB 114 and NCB 117)**

CLIMATE CHANGE AND ENVIRONMENTAL PHYSIOLOGY: Room NCB 113

14:00-14:15 **CE1** Petra D'Odorico
Predicting the seasonal course of functional traits for white spruce populations using a high-throughput UAV-based phenotyping approach

14:15-14:30 **CE2** Vera Velasco
Variation in photosynthesis and water status in response to a simulated heatwave in seedlings of coastal and interior Douglas-fir

14:30-14:45 **CE3** Mirindi Eric Dusenge
Tropical forests in a warming world

14:45-15:00 **CE4** Jordan Demone
New breeding techniques for greenhouse gas (GHG) mitigation: plants may express nitrous oxide reductase

BIOTIC STRESS AND EDUCATION: Room NCB 114

14:00-14:15 **BSE1** Angela Fufeng
Biofilm formation by Pseudomonas syringae pv. tomato is correlated with bacterial success and is reduced in Arabidopsis plants undergoing PAMP-triggered immunity

14:15-14:30 **BSE2** Bradley Laflamme
The effector-triggered immunity landscape of a host-pathogen interaction and its implications for broad-spectrum disease resistance

14:30-14:45 **BSE3** Winfield Yim
Identification of bacterial and fungal endophytes to induce broad-spectrum immunity in Arabidopsis thaliana

14:45-15:00 **BSE4** Emilie Widemann
A strategy toward the identification of Arabidopsis phytochemicals efficient against the two-spotted spider mite pest

15:00-15:15 **BSE5** Allison McDonald
Science communication through online writing

REPRODUCTION: Room NCB 117

14:00-14:15 **R1** Victoria Lesy
Understanding the molecular mechanism behind ScFT2-OSCAR, a Synthetic anti-florigen

14:15-14:30 **R2** Benjamin Tremblay
Effects of transgenerational spermidine-restoration of fertility in 5'-methylthioadenosine nucleosidase deficient plants

14:30-14:45 **R3** Jian Wu
Regulation of reproductive development and seed abortion in Arabidopsis

14:45-15:00 **R4** Stuart Macgregor
Secretion in the Arabidopsis thaliana compatible pollen response

15:00-15:15 **R5** Hyun Kyung Lee
The search for receptor kinases that regulate compatible pollen responses in the Brassicaceae stigma

15:15-15:35 **Coffee Break,** NCB atrium

15:35-16:25 **PLENARY LECTURE II: Room NCB 113**
Keiko Yoshioka, University of Toronto
Cyclic nucleotide-gated channels and Ca 2+ signaling - Roles in immunity, cell death and beyond - Insights from genetic screens and structural analyses

16:25-17:00 **Closing remarks and awards,** NCB atrium

PLENARY LECTURES

PLI Arunika **Gunawardena**

Dalhousie University; Arunika.Gunawardena@Dal.Ca

Lace plant: An emerging model system to study developmental programmed cell death

Programmed cell death (PCD) plays a major role in plant development and defense. One fascinating example of developmentally regulated PCD is perforation formation in lace plant leaves (*Aponogeton madagascariensis*). In the lace plant, perforations form at highly predictable locations and specific developmental stages of leaves that are accessible for observation and experimental manipulations. PCD begins at the center of each areole in window stage leaves, develops outwards, and stops 4-5 cell layers from the veins. These peripheral cells do not undergo PCD and are therefore referred to as NPCD (non-PCD) cells. The presence of visible changes within an areole provides a gradient of cell death for analysis that parallels chronological changes. In addition, events in PCD and NPCD cells are readily comparable. Lace plant has outstanding potential as a model system to study developmentally-regulated PCD in plants due to: the accessibility and predictability of perforation formation, the ability to perform live cell imaging of the thin and semi-transparent leaves, and the availability of an established sterile culture system for propagation. In my talk, I will be discussing the road map to the development of the lace plant as a versatile model system.

PLII Keiko **Yoshioka**

University of Toronto, Cell & Systems Biology; keiko.yoshioka@utoronto.ca

Cyclic nucleotide-gated channels and Ca 2+ signaling - Roles in immunity, cell death and beyond - Insights from genetic screens and structural analyses

Calcium ions (Ca 2+) are universal second messengers that control a wide variety of responses to abiotic and biotic stresses (heat, pathogen infection, etc.), as well as developmental cues (reproduction, circadian clock etc.) in plants. Upon perception of stimuli, transient changes in cytosolic Ca 2+ levels are rapidly induced. Stimuli-specific Ca 2+ signals are generated by the combined action of channels, transporters, and pumps. Cyclic nucleotide-gated channels (CNGCs), one of the largest cation channel families in plants, are involved in Ca 2+ signaling in a wide range of responses, such immunity, gravitropism, and thermo-tolerance. To understand the biological role and regulation of plant CNGCs, we have studied Arabidopsis CNGCs, mainly CNGC2, 4, 11 and 12, which are involved in immunity and programmed cell death (PCD). Our results with CNGC12 indicated that it can be subject to complex regulation by the Ca 2+ sensor protein calmodulin (CaM) to induce PCD. We found multiple CaM-binding domains in CNGC12 and one of them, an IQ type domain regulates channel positively, challenging the previously proposed competitive ligand model. Currently we are working on the visualization of CNGC-mediated Ca 2+ signals using the genetically encoded Ca 2+ sensors, GCaMP and Chameleon, and found an unexpected role of CNGC2 in auxin signaling.

SHORT PRESENTATIONS

ABIOTIC STRESS

AS1 Lauren A E **Erland** [1], Adam Yasunaga [2], Isaac T S Li [2], Susan J Murch [2] and Praveen K Saxena [1]
1 Gosling Research Institute for Plant Preservation, Department of Plant Agriculture, University of Guelph, Guelph, ON; 2 Chemistry, University of British Columbia Okanagan, Kelowna, BC; lerland@uoguelph.ca

Direct visualization of location and uptake of melatonin and serotonin in living tissues and their redistribution in plants in response to thermal stress

Melatonin and serotonin are important phytochemicals enabling plants to redirect growth in response to environmental stresses. Despite much research on their biosynthetic routes, localization of their biosynthetic enzymes and recent identification of a phytomelatonin receptor, localization of the molecules themselves has to date not been possible. Elucidation of their locations in living tissues can provide an effective tool to facilitate indoleamine research across systems including both plants and animals. In this study, we employed a novel technique, quantum dot nanoparticles, to directly visualize melatonin and serotonin in axenic roots. Melatonin was absorbed through epidermal cells, travelled laterally, and accumulated in endodermal and rapidly dividing pericycle cells. Serotonin was absorbed by cells proximal to the crown with rapid polar movement toward the root tip. Thermal stress disrupted localization and dispersed melatonin and serotonin across cells. These data demonstrate the natural movement of melatonin and serotonin in roots directing cell growth and suggest that plants have a mechanism to disperse the indoleamines throughout tissues as antioxidants in response to environmental stresses.

AS2 Nathieli **Beltran Schiavi**, Sophia Stone
Dalhousie University; nathielischiavi@gmail.com

Assessing the role of the Arabidopsis ubiquitin ligase XBAT31 in Fe deficiency stress response

Iron (Fe) is an essential micronutrient for plants. Its deficiency can negatively impact nutritional quality, growth and limit crop productivity. Plants must sense Fe deprivation and be capable of maintaining Fe-concentration within a specific range. A number of ubiquitin ligases (E3s) have been shown to regulate the abundance of proteins involved in iron homeostasis. Substrate selecting E3s are important components of the essential ubiquitin-proteasome system, which regulate the abundance of numerous cellular proteins. Here we demonstrate a role for Arabidopsis RING-type E3 XBAT31.1 in plant tolerance of low Fe growth conditions. Under Fe-deficiency condition, XBAT31.1 expression is induced, and xbat31-1 seedlings have elevated transcript levels of the Fe-utilization genes FIT, FRO2 and AHA2. Unexpectedly, transcript and protein levels of the metal transporter IRT1 were significantly lower compared to wild type, which correlated with reduced accumulation of iron, manganese and cobalt in shoots. Despite the low iron content, xbat31-1 seedlings showed significantly longer primary roots, greater fresh weight and higher chlorophyll content when grown under Fe-deficiency condition. Our results suggest that XBAT31.1 indirectly regulates the abundance of IRT via possibly regulating the abundance of a transcriptional repressor. Further analysis is required to fully understand the role of XBAT31.1 in iron homeostasis.

AS3 Nicolas **Dimopoulos**1,3, Darren C.J. Wong1, Tegan Haslam2, Changzheng Song1 Rodrigo Lopez Gutierrez1, Ljerka Kunst2, Simone Diego Castellarin1

1. Wine Research Centre, Faculty of Land and Food Systems, University of British Columbia, Vancouver, BC V6T 1Z4, Canada; 2. Department of Botany, Faculty of Science, University of British Columbia, Vancouver, BC V6T 1Z4, Canada; 3. Department of Biology, Graduate Department of Cell & Systems Biology, University of Toronto, Mississauga, ON L5L 1C6, Canada; nicolas.dimopoulos@mail.utoronto.ca

Compositional changes of the grape berry (Vitis vinifera L.) cuticle during fruit development in response to water deficit stress

The grapevine (Vitis vinifera) berry cuticle protects the fruit from water loss, which may in turn affect the final berry size at ripening. Water deficit (WD) is a common stress in vineyards, but little is known about how the berry cuticle is modified in response to this stress. We hypothesized that under severe WD stress, the cuticular aliphatic wax biosynthetic pathway of developing grape berries would be upregulated, resulting in an increased wax load in the fruit's cuticle and a decreased transpiration rate through the berry cuticle. Candidate wax-related genes were identified by phylogenetic and transcriptomic analyses, which also showed that a number these genes are significantly upregulated in response to WD stress. We then conducted a greenhouse experiment in 2016 to test the impact of WD on cuticular wax composition (GC-MS), expression of candidate biosynthetic genes (RT-qPCR), and water transpiration (drying experiment) in Merlot grapes. A significant increase in aliphatic wax load and a decrease in the ratio of triterpenoids:aliphatic wax was observed under WD stress. This was due to upregulation of many genes of the aliphatic wax biosynthetic pathway. Berry transpiration rate was not significantly affected by WD; however, a marginally significant reduced rate was observed in WD berries.

AS4 Biruk A. **Feyissa**1,2; Muhammad Arshad1; Susanne E. Kohalmi2; Abdelali Hannoufa1,2

1. Agriculture and Agri-Food Canada, 1391 Sandford Street, London, Ontario, N5V 4T3, Canada; 2. Department of Biology, University of Western Ontario, 1151 Richmond Street, London, Ontario, N6A4B7, Canada; bfeyissa@uwo.ca

SPL13 and WD40-1 regulate DFR to enhance anthocyanin biosynthesis for stress tolerance in Medicago sativa

Developing Medicago sativa (alfalfa) cultivars that can withstand drought is critical for the crop's sustainable production. To mitigate drought stress, plants employ anthocyanins and other polyphenols to scavenge reactive oxygen species. DIHYDROFLAVONOL-4-REDUCTASE (DFR) is the first committed step in anthocyanin biosynthesis, and whose expression is regulated by several transcription factors. Here, we investigated the role of miR156 in regulating SPL13 and WD40-1 transcription factors to fine-tune DFR expression, enhance anthocyanin biosynthesis and improve drought tolerance. In this study, we used alfalfa genotypes with different levels of miR156 expression, SPL13RNAi genotypes, as well as plants with altered WD40-1 expression. We found that moderate levels of miR156 increased DFR expression by silencing SPL13 and activating WD40-1. Physiological, metabolite and transcript analysis showed that alfalfa plants with moderately enhanced levels of miR156 and WD40-1, and reduced SPL13 had an improved tolerance to drought due to enhanced accumulation of anthocyanins and other stress mitigating metabolites.

AS5 Eliana **Vonapartis**[1], Deka Mohamed[1], Mornisha Panchalingam[2], Sonia Gazzarrini[1,2]
[1] Dept. of Cell and Systems Biology, University of Toronto; [2] Dept. of Biological Sciences, University of Toronto.; eliana.vonapartis@mail.utoronto.ca

Transcriptional regulation of XERICO modulates abiotic stress response in Arabidopsis thaliana

Plants continually face a multitude of stresses that can severely compromise their ability to survive through the growing season. To mitigate the effects of a harsh environment, plants have evolved various developmental and physiological strategies through which they can effectively balance growth and defense against stressors. Arabidopsis XERICO (XER) is a stress-responsive putative RING E3 ubiquitin ligase that increases intracellular abscisic acid levels and promotes drought tolerance. To elucidate the molecular mechanism of XER induction by stress and better understand its role in regulating growth and stress responses, we first analyzed its expression pattern and determined its subcellular localization. Subsequently, we conducted a yeast one-hybrid screen and isolated six potential XER upstream regulators. Our results demonstrate that while some act as positive and others as negative regulators, one of them is particularly important for repressing XER expression during osmotic stress. Lastly, we show that Arabidopsis plants with altered XER and XER RING-inactive levels exhibit different degrees of sensitivity to drought, and this may be linked to XER's role in modulating stomatal density. Taken together, we propose that XER expression is tightly regulated following initial stress perception to control stomatal density, ultimately ensuring survival during unfavourable growth conditions such as drought.

AS6 Marina **Cvetkovska**(1), Shane Orgnero(2), Norman P. A. Hüner(2), David Roy Smith(2)
1.Department of Biology, University of Ottawa, Ottawa, ON, Canada K1N 6N5; 2. Department of Biology and the Biotron Centre for Experimental Climate Change Research, University of Western Ontario, London, ON, Canada N6A 3K7; mcvetkov@uottawa.ca

The enigmatic loss of light-independent chlorophyll biosynthesis from an Antarctic green alga in a light-limited environment

In green algae, the penultimate step in the chlorophyll biosynthesis pathway is catalyzed by two nonhomologous enzymes: a light-dependent (POR) and a light-independent protochlorophyllide oxidases (LIPOR). POR is nuclear-encoded and is only active when its pigment substrate absorbs light. In contrast, LIPOR is chloroplast-encoded and facilitates chlorophyll synthesis in the dark. Here we report the first confirmed case of LIPOR loss in a green alga from the chlorophycean lineage. Chlamydomonas sp. UWO241 is an extremophile isolated 17 meters below the ice surface of the Antarctic Lake Bonney. In addition to continuous cold (~5°C), UWO241 is exposed to perpetual low irradiance (<50 µmol photon m-2 s-1) and extreme photoperiods (24h darkness during the polar winter). We screened the nuclear and organellar genomes of UWO241 for genes involved in chlorophyll biosynthesis, but our extensive analysis failed to locate the genes encoding for LIPOR. This is particularly surprising given the extreme light-limitations in Lake Bonney. We identified homologs of all other chlorophyll biosynthesis enzymes and show that several of these genes are duplicated. We suggest that UWO241 has lost the ability to carry out light-independent chlorophyll biosynthesis and is solely dependent on POR. We discuss the evolutionary explanation for 'jettisoning' LIPOR and the shade-adapted physiology of UWO241.

BIOCHEMISTRY

B1 Artyom **Gritsunov**, James Peek, Dinesh Christendat
University of Toronto, Cell and Systems Biology; artyom.gritsunov@mail.utoronto.ca
Investigation of quinate metabolism in plants
Quinate is a plant metabolite that accumulates in green tissues. It provides astringent taste and serves as feeding deterrent. Additionally quinate is used for the biosynthesis of chlorogenic acids, which are antifungal agents, antioxidants and serve as UV-light protectants. Recently, we uncovered and characterized a family of both anabolic and catabolic enzymes involved in plant quinate metabolism [1]. Now we are working on in vivo confirmation of quinate metabolism and physiological properties associated with quinate production. We reintroduced evolutionary lost QDHs back into A. thaliana plants and are preparing to investigate the physiological significance of plant quinate metabolism.1. Gritsunov, A., Peek, J., Diaz Caballero, J., Guttman, D. & Christendat, D. Structural and biochemical approaches uncover multiple evolutionary trajectories of plant quinate dehydrogenases. Plant J, doi:10.1111/tpj.13989 (2018)

B2 Michael **Kanaris**, Dinesh Christendat
University of Toronto, Department of Cell and Systems Biology; michael.kanaris@mail.utoronto.ca
Evolutionary insights into the role of shikimate kinase-like 1 in chloroplast development
Chloroplast biology represents one of the most intensely studied topics within plant research that seeks to understand the complex processes related to photosynthesis. Shikimate kinase-like 1 (SKL1), a gene homolog of the well-studied shikimate kinase involved in the shikimate pathway, has been implicated in chloroplast biogenesis. Arabidopsis thaliana skl1 T-DNA insertional mutant (skl1-8) lacks developed chloroplasts and displays an albino phenotype. We are investigating the functional evolution of SKL1 through comparative mutational and biochemical analyses of a number plant species including A. thaliana, Physcomitrella patens, and Marchantia polymorpha, some of which include the earliest ancestral plants that contain an SKL1 homolog. Results from analyses thus far have shown that SKL1 has maintained its ancestral function by complementing skl1-8 mutants with P. patens SKL1. The absence of SKL1 in organisms predating land plants, including green algae, make it an attractive candidate to study plant evolution. The major goal of this research seeks to expand our current knowledge on the topic of chloroplast biogenesis to understand how SKL1 participates in this complex process, and to provide knowledge on the evolution of SKL1 as a novel functional protein based on the divergence from shikimate kinase.

B3 Trish. L.A. **Tully**, Pooja Kaushik, Jessica O'Connor, Mark A. Bernards
Department of Biology, The University of Western Ontario, London, ON, Canada, N6A 5B7; ttully@uwo.ca

Characterization of CYP86A genes and their role aliphatic suberin deposition in soybean roots

Soybean is a highly important crop plant; however, despite this high level of global cultivation, soybean yield is still threatened by both biotic and abiotic sources of stress. Suberin deposition has been shown to play a role in response to this stress. Particularly, resistance against P. sojae has been positively correlated with suberin deposition in soybean. The monomer with the highest abundance in soybean aliphatic suberin is 18-hydroxy-oleic acid, which is predicted to be synthesized by putative CYP86A enzymes in soybean. There are two root specific putative CYP86A genes in soybean; CYP86A37 and CYP86A38. To explore the physiological role of these two genes, an RNAi-knockdown was used. Following knockdown, root samples with reduced gene expression also showed a reduction in suberin deposition; most notable was the reduction in 18-hydroxy-oleic acid in relation to reduced CYP86A38 expression. Based on this correlative evidence, it is likely that CYP86A37 and CYP86A38 function as fatty acid ω-hydroxylases. As the expression of these two genes impacts the composition of the suberin polymer, it will be important to further explore the developmental regulation of these two genes to gain insight into the regulation of suberin deposition in soybean.

B4 Lisa **Amyot**, Tim McDowell, Justin Renaud, Abdelali Hannoufa
London Research and Development Centre, Agriculture and Agri-Food Canada, London, ON, Canada; Lisa.Amyot@canada.ca

Assessment of glucosinolate content in Camelina sativa and its wild relatives

The focus of our study was to compare the glucosinolate composition of Camelina sativa and its wild relatives. LC-MS/MS analysis of 20 Camelina accessions, from 5 different species, revealed that Camelina spp. separated into distinct chemotaxonomic groups. Three major glucosinolates were identified in our study, namely, 9-methylsulfinylonyl glucosinolate (GS9), 10-methylsulfinylonyl glucosinolate (GS10), and 11-methylsulfinylonyl glucosinolate (GS11). While differences were noted in total glucosinolate levels, our results showed that there were species-specific patterns for GS9 and GS11. HPLC analysis of 167 samples revealed that glucosinolate content ranged between 8.0 and 21.7 µmol/g seed FW, with the lowest levels observed in C. microcarpa cytotypes 4x and 6x. Our results show that wild Camelina spp. have distinct metabolomes and based on the variations in glucosinolate content, some could be incorporated into breeding programs for C. sativa improvement.

B5 Kristen **Van Gelder**, Lilia Virta, Kevin Rea, Kenna Whitnell, Tariq Akhtar
Department of Molecular and Cellular Biology, University of Guelph, Guelph, ON N1G 2W1; kvangeld@uoguelph.ca

Plant polyprenols and their role in thylakoid membrane dynamics

Despite the widespread occurrence of medium-chain polyprenols across the plant kingdom, their physiological function remains poorly understood. These lipophilic compounds, ranging in size from 45-60 carbons, are synthesized by a class of enzymes known as cis-prenyltransferases (CPTs) which are encoded by small CPT gene families. In Solanum lycoperscium, we identified a CPT, SlCPT5, which synthesizes medium-chain polyprenols in the plastid. Assays with recombinant SlCPT5 produced in E. coli demonstrated that SlCPT5 synthesizes polyprenols of 45-60 carbons with a variety of trans-prenyl diphosphate precursors. Further structural characterization of these polyprenols from tomato leaf tissue via H1-NMR showed that in vivo, SlCPT5 uses geranylgeranyl diphosphate as its bona fide substrate. Subcellular fractionation studies together with in vivo localization of SlCPT5 fluorescent protein fusions demonstrated that SlCPT5 resides in the chloroplast stroma whereas its enzymatic products are found in the thylakoid and envelope membranes. Chloroplast ultrastructure between RNA interference (RNAi)-mediated knockdown of SlCPT5 tomato plants, which are deficient in polyprenols, and wildtype plants indicated a decrease in grana stacks and an increase in stromal thylakoids. In polyprenol deficient leaves, photosynthetic electron transport rates and CO2 assimilation rates were both reduced. Together, these results demonstrate a role for polyprenols in mediating thylakoid membrane dynamics.

B6 Nishat S. **Islam**1,2, Frédéric Marsolais1,2, Sangeeta Dhaubhadel1,2
1 London Research and Development Centre, Agriculture and Agri-Food Canada, London ON, Canada N5V 4T3; 2 Department of Biology, University of Western Ontario, London ON, Canada N6A 5B7; nislam33@uwo.ca

Pinto bean (Phaseolus vulgaris L.) orthologue of Arabidopsis TT8 regulates proanthocyanidin genes and seed coat darkening

Postharvest darkening of seed coat in pinto bean (Phaseolus vulgaris L.) is an undesirable trait that affects its market value. Darkening is more rapid in the adapted cultivars like CDC Pintium than the newly developed slow darkening cultivar 1533-15. A single gene, SLOW DARKENING (Sd), is responsible for the slow darkening in pinto beans and the trait co-segregates with two simple sequence repeat (SSR) markers. The objective of this research is to identify and characterize the Sd gene to understand the slow darkening mechanism in pinto bean seed coat. A search for Sd within the linkage distance from the SSR markers has identified a basic helix-loop-helix (bHLH) transcription factor gene, PvbHLH333 as a candidate gene. PvbHLH333 from CDC Pintium and 1533-15 has shown sequence and functional variations. The ability of PvbHLH333 from 1533-15 to rescue the tt8 mutant phenotype in Arabidopsis suggests that PvbHLH333-1533-15 is the orthologue of TT8, which regulates proanthocyanidin biosynthesis in Arabidopsis.

DEVELOPMENT, ENZYMES AND MOLECULAR BIOLOGY

DEB1 Deka **Mohamed**, Eliana Vonapartis, Carina Carianopol, Sonia Gazzarrini

University of Toronto, Cell and Systems Biology (CSB), University of Toronto Scarborough, Biological Sciences; deka.mohamed@mail.utoronto.ca

Investigating the role of XERICO in stomatal development and cell wall function

Plants are sessile organisms that rely on the ability to perceive and respond to changing environmental conditions to survive. As a result, they have the ability to alter their development in response to a host of environmental stresses. Stomata, which are epidermal pores that control gas and water exchange, are pivotal for ensuring plant survival and growth under stress. Consequently, stomatal aperture, density and patterning are tightly regulated to ensure optimal stomatal density and spacing. XERICO (XER), a small putative RING E3 ligase with a single N-terminal transmembrane domain involved in ABA homeostasis, promotes drought tolerance when overexpressed. To better understand XER role in stress response, I performed detailed developmental and genetic analyses. My data show that two xer mutant alleles exhibit an increase in stomata density and clustering. I hypothesized that XER promotes drought tolerance by regulating stomatal development and distribution. Furthermore, a putative interactor of XER was identified through a Y2H screen using a library of ABA regulated proteins. This interactor is a part of a family of Golgi-localized glycosyltransferases implicated in cell wall modification. I have re-confirmed this interaction in planta and will discuss how this interaction may function to regulate stomatal development to ultimately enhance abiotic stress tolerance.

DEB2 Carina **Carianopol**, Sonia Gazzarrini

Department of Cell and Systems Biology, University of TorontoDepartment of Biological Sciences, University of Toronto Scarborough Campus; carina.carianopol@mail.utoronto.ca

The role of Arabidopsis ABI5 binding protein 2 (AFP2) in HR-like programmed cell death

Plants are continuously exposed to changing environmental stresses. They have thus evolved complex signaling pathways to govern responses to a variety of stimuli. For example, salicylic acid (SA) has a well-known role in biotic stress response. A response governed by SA is the development of programmed cell death (PCD) at the site of infection to prevent systemic infection and promote plant survival. Abscisic acid (ABA) is an important hormone during abiotic stress response. ABA has been implicated in modulation of plant defense response by positively regulating stomatal immunity, but also antagonistically interacting with SA to promote post-invasive immunity in some cases. Recently, a family of negative regulators of ABI5 has been identified and includes four ABI Five Binding Proteins (AFPs). AFP2 negatively regulates the transcription factor ABI5, which is involved in core ABA signaling pathway. Our data shows that overexpression of Arabidopsis AFP2 in Nicotiana bethamiana induces PCD and may be involved in both immunity and ABA signaling. Deletion studies indicate that select domains of AFP2 are required for the induction of PCD, while others for its subcellular localization. Interestingly, Y2H screens identified AFP2 as an interactor of the SnRK1 kinase complex. SnRK1's potential role in modulating AFP2-induced PCD will be further discussed.

DEB3 Purva **Karia**1, Wolfgang Moeder1, Kazuo Ebine2, Takashi Ueda2, Keiko Yoshioka1
1. Department of Cell &Systems Biology, University of Toronto, Toronto, Ontario; 2. National Institute for Basic Biology. Nishigonaka 38, Myodaiji, Okazaki 444-8585 Aichi, Japan; purva.karia@mail.utoronto.ca

The mitochondrial tail-anchored protein TTM1 mediates ABA-induced senescence

Tail-anchored (TA) proteins are characterized by the presence of a C-terminal transmembrane (TM) domain. TA proteins are integrated into organellar membranes by their TM domain and they lack a signal peptide. TA proteins perform diverse functions, including apoptosis and vesicular trafficking. Arabidopsis possess over 150 TA proteins. TTM1 and 2 of the Triphosphate Tunnel Metalloenzymes (TTMs) superfamily are TA proteins based on the presence of C-terminal TM domain. Confocal microscopy unveiled their subcellular localization at the mitochondrial outer membrane. Our earlier work revealed the involvement of TTM1 and 2 in different forms of programmed cell death; natural senescence and immunity, respectively. ttm1 knockout mutants exhibit delayed dark- and ABA-induced senescence whereas transiently overexpressed TTM1 displayed senescence-like cell death in the dark in Nicotiana benthamiana, indicating TTM1 is a positive regulator of senescence. Published phospho-proteomic data suggest increased TTM1 phosphorylation upon ABA treatment. Furthermore, a phospho-dead version of TTM1 failed to complement the ttm1 delayed senescence phenotype, suggesting that phosphorylation is crucial to fine tune senescence. We have identified a potential kinase that phosphorylates TTM1. Together, these data suggest that phosphorylation of the mitochondrial TA protein TTM1 is crucial for regulating senescence in Arabidopsis.

DEB4 Mark A.A. **Minow**1, Luis Avila Bolivar2, Vincenzo Rossi3, Lewis Lukens1, Joseph Colasanti1
1 University of Guelph; 2 U C Davis; 3 Unita di Ricerca per la Maiscoltura CREA; mminow@uoguelph.ca

Stability and change in Zea mays small RNA transcriptomes provide exciting insights into the formation of new epigenetic states

Epigenetic diversity within a species can affect numerous plant phenotypes. Small RNA (sRNA) is an important epigenetic regulator of genes and transposons in plants. Diversity in sRNA expression patterns exists within a species, but it is poorly understood how this diversity arises. To provide a window into the dynamics of maize sRNA patterning, the sRNA and mRNA transcriptomes were examined in Zea mays recombinant inbred lines (RILs) and near isogenic lines (NILs). Widescale stability in sRNA expression patterns was observed; that is, progeny retain most of the sRNA expression levels that they inherit from their parents. However, deviation of expression patterns from the parental levels also occurs. These expression level changes show a high similarity between related RILs that is inconsistent with the expected rates predicted through the RIL co-inheritance of independent trans regulators. Comparison of NIL sRNA transcriptomes revealed a larger change in sRNA expression patterns compared to the RIL profiles. This disparity in sRNA expression stability indicate that genetic differences between RILs and NILs modulate the magnitude of change in sRNA expression levels. The patterns of sRNA expression change found here may provide translational insights into sRNA silencing in other plants and possibly metazoans.

DEB5 Alberto **Torrez**, Lining Tian, Hugh Henry
University, Agriculture and Agri-Food Canada; atorrez@uwo.ca

Identifying interacting proteins with BdHD1 in Brachypodium distachyon

Stress-responsive genes can be mediated by epigenetic changes. Current evidence has indicated the involvement of histone deacetylases (HDACs) in plant stress responses. HDACs facilitate the removal of acetyl groups from the histone tails, causing gene repression. HDACs can interact with transcription factors to form repressor complexes, thus leading to the repression of target genes. In Arabidopsis thaliana, HDAC1 (HDA19) interacts with different transcription factors to regulate gene expression in response to stresses. HDAC research has been mainly conducted within dicots. There is limited HDAC research within monocots. Brachypodium distachyon is used as a model plant to investigate questions unique to monocot crops. BdHD1, belongs to the HDAC/RPD3 class, has been identified in B. distachyon. This research is investigating potential protein-protein interactions with BdHD1 with several candidate transcription factors, namely HOS15, WRKY24 and MYB22. The interactions between BdHD1 and the candidate proteins are under investigation by using yeast two-hybrid (Y2H) assays and bimolecular fluorescence (BiFC) assays . Based on my Y2H assay results, BdHD1 strongly interacts with WRKY24, as well as with MYB22. But there was no sign of interaction between HOS15 and BdHD1. The interactions between BdHD1 and WRKY24, MYB22 will be confirmed via BiFC assays.

DEB6 Emily J. **Clayton**, Sara Abolhassani Rad, Susanne E. Kohalmi
Western University, Department of Biology; eclayto3@uwo.ca

Piggybacking AtADT5 into the nucleus

Arabidopsis thaliana, a family of six enzymes called AROGENATE DEHYDRATASES (ADTs) catalyze the last step of phenylalanine (Phe) biosynthesis, and all localize to the chloroplast. In addition, ADT5 localizes to the nucleus, a localization that is not shared by any other AtADT. Interestingly, no nuclear localization signals (NLSs) were identified in the AtADT5 sequence. Recently, an ADT5 specific interactor was identified that has two putative NLS sequences. We believe this interaction allows the “piggybacking” of ADT5 into the nucleus and I am interested in determining the required sequences. ADT4 is the most closely related to ADT5 (90% identity), but there is a unique motif in the C-terminal ACT domain of ADT5. To test if this motif is responsible for the interaction with the candidate interactor, yeast-two-hybrid and Bimolecular fluorescence complementation (BiFC) analyses will be performed using a series of domain swapped proteins. In addition, proteins with specific amino acid replacements in this motif will be generated for both ADT4 and ADT5 and also tested for interaction. This work will provide insight on protein evolution between related proteins, and how seemingly inconsequential differences in sequence can have large effects on enzyme localization and function.

CLIMATE CHANGE AND ENVIRONMENTAL PHYSIOLOGY

CE1 Petra **D'Odorico**1, Ariana Besik1, Nathalie Isabel2, Ingo Ensminger1
1 Department of Biology, University of Toronto Mississauga, Ontario, Canada; 2 Laurentian Forestry Centre, Canadian Forest Service, Quebec, Canada; petra.dodorico@utoronto.ca

Predicting the seasonal course of functional traits for white spruce populations using a high-throughput UAV-based phenotyping approach

Plant breeding programs rely on phenotyping to identify and select desired plant traits, such as those linked with stress resistance and longer growing season. Traditional plant phenotyping approaches involve a high amount of time and human resources and often result in low temporal and spatial sampling. We present a UAV-based imaging phenotyping approach employing spectral vegetation indices to predict functional traits and phenology of a white spruce population. A common garden experiment with seedlings of 2000 different white spruce genotypes was sampled on nearly weekly basis over the 2017/2018 growing season. In addition to UAV-based multispectral imaging, ground validation included fluorescence measurements and needle sampling for pigment content analysis. We developed and validated a model for the prediction of functional traits, such as maximum potential quantum efficiency of photosystem II, over the growing season and estimated key phenological transition dates for the entire population. This work paves the way to an unprecedented ability to rapidly screen the performance of individual seedlings and genotypes in large tree populations to use in support of tree breeding programs.

CE2 Vera ME **Velasco**, Tomoyuki Sen, Devin Noordermeer, Sepideh Torabi, Ingo Ensminger
University of Toronto; vera.velasco@utoronto.ca

Variation in photosynthesis and water status in response to a simulated heatwave in seedlings of coastal and interior Douglas-fir

Abiotic stresses due to climate change causes a mismatch between locally-adapted species and their environment leading to increased mortality. Understanding how conifer will respond to future climatic conditions can help us secure the resilience of boreal forest ecology. In this study, we aimed to assess intraspecific variation of photosynthesis and water status in response to four weeks of extreme heat, drought and heatwave (combined heat and drought) in seedlings of four Douglas-fir provenances from broad climate gradients. We observed a 20% and 40% reduction in maximum quantum efficiency of PSII (Fv/Fm) of Douglas-fir needles exposed to four weeks of extreme heat and drought, respectively. However, four weeks of heatwave was most detrimental and resulted in a 75% reduction in Fv/Fm. We also found lower photosynthetic rates and stomatal conductance on all provenances, suggesting inhibition of CO2 assimilation in Douglas-fir due to heatwave. Interestingly, statistically significant reduction in respiration due to heatwave was only observed in provenances adapted to warm climate. Our study also showed that Douglas-fir traits associated with water status, such as relative water content, water potential and transpiration, were negatively affected by heatwave. Notably, more pronounced inhibition on water status-related traits was observed in provenances originally from wet environments. These findings suggest that intraspecific variation exists among Douglas-fir provenances and in their response to co-occurring extreme drought and heat stress.

CE3 Mirindi E. **Dusenge**1,*, Maria Wittemann2, Elisée N. Bahati3, Myriam Mujawamariya3, Etienne Zibera3, Donat Nsabimana3, Göran Wallin2, Johan Uddling2 and Danielle Way1,4
1 Biology Department, Western University, London, Ontario, Canada; 2 Department of Biological and Environmental Sciences, University of Gothenburg, Gothenburg, Sweden; 3 Biology Department, University of Rwanda, Rwanda; 4 Nicholas School of the Environment, Duke University, Durham, USA; mdusenge@uwo.ca

Tropical forests in a warming world

Tropical forests play a significant role in carbon sequestration, and help slow the rate of climate change by dampening the rise of atmospheric CO2. However, their response to climate warming is uncertain, hindering accurate projections of future climate change. We grew seedlings of two tropical montane trees, an early and a late successional species, along an altitudinal gradient (high, medium and low altitude) in Rwanda. We measured temperature responses of photosynthesis and respiration. Harungana montana, the early successional species, shifted its photosynthetic thermal optimum to higher temperatures, despite exhibiting decreased net photosynthesis at lower altitudes, whereas Syzygium guineense, the late successional species, had similar thermal responses of net photosynthesis at all elevations. Photosynthetic biochemical parameters (i.e. Vcmax, maximum carboxylation capacity and Jmax, maximum rate of electron transport) declined from high to low elevation in both species. Stomatal conductance responded similarly to temperature from high to low elevation-grown seedlings. Dark respiration thermally acclimated in both species, reducing CO2 losses in species grown at lower altitudes. Our findings show that tropical species might have a limited ability to acclimate to warming, and suggest that warming might reduce net CO2 uptake in tropical species.

CE4 Jordan J. **Demone**, Shen Wan, Maryam Nourimand, Asbjörn Erik Hansen, Qing-yao Shu, Illimar Altosaar
Department of Biochemistry, Microbiology, and Immunology, University of Ottawa, Ottawa, ON K1H 8M5, Canada National Key Laboratory of Rice Biology, Institute of Crop Sciences, Zhejiang University, Hangzhou 310058, China; jdemone@uottawa.ca

New breeding techniques for greenhouse gas (GHG) mitigation: Plants may express nitrous oxide reductase

Nitrous oxide (N2O) is a potent greenhouse gas (GHG). Although it comprises only 0.03% of total GHGs produced, N2O makes a marked contribution to global warming. Much of the N2O in the atmosphere issues from incomplete bacterial denitrification processes acting on high levels of nitrogen (N) in the soil due to fertilizer usage. Using less fertilizer is the obvious solution for denitrification mitigation, but there is a significant drawback (especially where not enough N is available for the crop via N deposition, irrigation water, mineral soil N, or mineralization of organic matter): some crops require high-N fertilizer to produce the yields necessary to help feed the world's increasing population. Alternatives for denitrification have considerable caveats. The long -standing promise of genetic modification for N fixation may be expanded now to enhance dissimilatory denitrification via genetic engineering. Biotechnology may solve what is thought to be a pivotal environmental challenge of the 21st century, reducing GHGs. Current approaches towards N2O mitigation are examined here, revealing an innovative solution for producing staple crops that can 'crack' N2O. The transfer of the bacterial nitrous oxide reductase gene (nosZ) into plants may herald the development of plants that express the nitrous oxide reductase enzyme (N2OR).

BIOTIC STRESS AND EDUCATION

BSE1 A.B. **Fufeng**, N.W. Xiao, G.M. Nunn, A. Halim, H.M. Philip, R.K. Cameron
Department of Biology, McMaster University; fufengab@mcmaster.ca
Biofilm formation by Pseudomonas syringae pv. tomato is correlated with bacterial success and is reduced in Arabidopsis plants undergoing PAMP-triggered immunity
Plants possess multiple SA signaling pathways for defense against microbial pathogens. However, during Age-Related Resistance (ARR) in Arabidopsis, SA acts as an anti-microbial and anti-biofilm agent against Pseudomonas syringae pv. tomato (Pst) in the plant intercellular space. Given that little is known about the effect of other defense responses on Pst biofilm formation, we investigated whether PAMP Triggered Immunity (PTI) acts to limit bacterial biofilm formation in an SA-dependent manner. PTI was induced with flg22 in wild-type Col-0, fls2, bak1-3 (PTI mutants) and sid2-2 (SA biosynthesis mutant) In vivo bacterial biofilm-like aggregate formation was monitored using Pst DC3000 PDSK-GFPuv and epifluorescence microscopy. Pst aggregate occurrence and size were positively correlated with bacterial success in fls2, bak1-3 and sid2-2, while fewer and smaller bacterial aggregates were observed in Col-0 undergoing PTI. To determine if Pst aggregates are actually biofilms, in vivo bacterial aggregate formation was monitored using alginate deficient Pst-GFP unable to form biofilms. Alginate deficient Pst-GFP bacterial numbers were greatly reduced in PTI-responding plants and wild-type Pst-GFP-like aggregates were not observed. This study provides evidence that SA not only contributes to defense signaling, but also acts in the intercellular space to limit growth and bacterial biofilm formation during PTI.

BSE2 Bradley **Laflamme**, Marcus M. Dillon, Alex Martel, Renan N.D. Almeida, Darrell Desveaux, David S. Guttman
University of Toronto; bradley.laflamme@mail.utoronto.ca
The effector-triggered immunity landscape of a host-pathogen interaction and its implications for broad-spectrum disease resistance
All plant species, including crops of major agricultural importance, are constantly interfacing with diverse microbial communities, including the phytopathogenic bacterium Pseudomonas syringae. While a few of these interactions will lead to disease in any given plant species, most would-be pathogens are thwarted by the robust broad-spectrum resistance exhibited by any individual plant species against unadapted pathogens. Further decoding the mechanisms through which plants establish and maintain broad-spectrum resistance is imperative for developing a more robust theoretical framework for engineering durable resistance in plant species. Using a genomics approach, our labs have surveyed hundreds of pathogenic P. syringae isolates and assembled a P. syringae virulence protein repertoire, which we have used to identify novel immune elicitors in the model plant Arabidopsis thaliana. A subset of these proteins is found in the majority of P. syringae isolates, suggesting that they play a major role in general resistance against this pathogen in Arabidopsis. Preliminary experiments in tomato show that our P. syringae virulence protein repertoire can be readily translated to an important crop species, potentially for the development of novel disease control strategies.

BSE3 W. **Yim**1, N. Morales3, Y. Arocha-Rosete4, W. Moeder1, J. Scott3, P. Wang1,2, K. Yoshioka1,2

1 Dept. of Cell and Systems Biology, University of Toronto; 2Center for the Analysis of Genome Evolution and Function (CAGEF), University of Toronto; 3 Div. of Occupational and Environmental Health, University of Toronto; 4 Sporometrics Inc.; winfield.yim@mail.utoronto.ca

Identification of bacterial and fungal endophytes to induce broad-spectrum immunity in Arabidopsis thaliana

Root-colonizing microorganisms have a significant influence on plant growth, development and immunity. Some of these non-pathogenic microorganisms can trigger an immune response called induced systemic resistance (ISR). This response elicits a priming phenomenon that allows plants to exhibit a broad-spectrum resistance against subsequent pathogen invasion in the foliar tissues. In this study, to identify novel endophytes that can induce ISR to protect plants against diseases, we screened our endophytic collection isolated from coconut plants utilizing the model system, Arabidopsis thaliana. So far, 100 endophytes have been screened for their ability to confer disease resistance against both a necrotrophic fungal pathogen, Botrytis cinerea, and a bacterial pathogen, Pseudomonas syringae pv. tomato (Pst DC3000). Through a root-dipping application of the endophytes, four bacterial endophyte strains were found to reduce disease severity significantly. These isolates belong to Orchobactrum sp., Bacillus subtilis, Bacillus sp. and Bacillus pumilus. Currently, we are analyzing the molecular mechanisms behind the induction of this observed disease resistance through gene expression analyses of JA-responsive genes PDF1.2 and VSP2.

BSE4 Emilie **Widemann**, Brendan Walshe-Roussel, Vladimir Zhurov, Julien Le Roy, Repon Saha, Mark Bernards, Vojislava Grbic

Western University, Department of Biology; ewidema4@uwo.ca

A strategy toward the identification of Arabidopsis phytochemicals efficient against the two-spotted spider mite pest

The two-spotted spider mite (TSSM) is a major pest affecting Canadian crops. This animal feeds on more than 1100 species of flora across the world, but its ravages depend on the host plant. For example, it proliferates much better on bean than on Arabidopsis thaliana. This natural resistance of Arabidopsis to TSSM is particularly due to the jasmonate hormonal pathway that triggers the production of defense compounds including the indole glucosinolates. The CYP79B2 and CYP79B3 enzymes, which assure the initiating step for the indole glucosinolate biosynthesis, are required to limit TSSM performance. However, the CYP72B2/CYP79B3 product is at the center of a complex metabolic network involving also other pathways than indole glucosinolates, which itself includes multiple reaction cascades leading to a plethora of derivatives. Our objective is to identify which compounds are efficient to inhibit TSSM performance. To dissect the metabolic network producing the toxic or repellent compounds, we performed a genetic and metabolomic screen. This led us to select key molecules that are now being evaluated for their effectiveness against TSSM. The findings of our research will be a step forward for identifying new phytochemicals that could be tested on crops to prevent TSSM propagation.

BSE5 Allison E**. McDonald**

Department of Biology, Wilfrid Laurier University; amcdonald@wlu.ca

Science communication through online writing

The ability of plant biologists to communicate their results and expertise to a wide variety of audiences is of increasing importance in many career paths. In particular, research dissemination efforts, knowledge mobilization plans, community outreach, and the training and mentoring of highly qualified personnel are metrics used to evaluate grant applications. One way to meet these criteria is to engage in science communication through social media platforms (e.g. blogging and Twitter) and by writing articles for online publications read by the public. Often as biologists, we are not trained explicitly in the skills required for science communication, or are told that such activities are a waste of time. I will make the argument that science communication efforts can be advantageous for your career and offer some examples of what is possible.

REPRODUCTION

R1 Victoria **Lesy**1, Mark Minow1, Carla Coelho1,2, Zhenhua Xu1, Zach Leblanc1, Steven Rothstein1, Antonio Chalfun Junior2, Joseph Colasanti1

1 Department of Molecular and Cellular Biology, University of Guelph, Guelph, ON, Canada; 2 Setor de Fisiologia Vegetal, Departamento de Biologia, Universidade Federal de Lavras, Lavras, Brazil; vlesy@uoguelph.ca

Understanding the molecular mechanism behind ScFT2- OSCAR, a synthetic anti-florigen

Flowering time is a tightly regulated process that is essential for seed production and proliferation in most higher plants. The vegetative to reproductive transition is controlled by diverse cues acting through distinct genetic pathways that converge at mobile floral integrators called florigens. These conserved phosphatidylethanolamine-binding proteins (PEBPs) are synthesized in leaves and migrate to the shoot apex to form a floral activation complex (FAC), which prompts reproductive growth. In sugarcane (Saccharum officinarum), several florigen gene candidates have been identified, such as ScFT2. Overexpression of this gene in Arabidopsis disrupts the floral transition and causes a dramatic change in shoot architecture. Later analysis revealed that extra amino acids were inadvertently added to ScFT2, creating a synthetic variant, called ScFT2-OSCAR. This research aims to characterize the mechanism behind this novel protein to determine how ScFT2-OSCAR overexpression causes an extreme vegetative phenotype. Interaction assays will be conducted to determine if ScFT2-OSCAR interferes with FAC formation by competition with endogenous PEBPs, and to identify new entities involved in floral regulation. Furthermore, the extra amino acids will be added to other PEBPs to determine whether they similarly alter shoot architecture. ScFT2-OSCAR may be transformed into other species to fine-tune flowering for agriculture.

R2 Benjamin **Tremblay**1, Markus Wirtz2, Ruediger Hell2, Barbara Moffatt1
1 Department of Biology, University of Waterloo, Waterloo, Canada; 2 Centre for Organismal Studies, Heidelberg University, Heidelberg, Germany; b2trembl@uwaterloo.ca

Effects of transgenerational spermidine-restoration of fertility in 5'- methylthioadenosine nucleosidase deficient plants

5'-Methylthioadenosine nucleosidase (MTN) is the first enzyme in the methionine salvage pathway. This enzyme is encoded by two genes, MTN1 (AT4G38800) and MTN2 (AT4G34840) in Arabidopsis. MTN deficiency in the double mutant mtn1-1mtn2-1 results in an accumulation of its substrate, 5'-methylthioadenosine (MTA). These double mutants have a complex phenotype which includes male and female sterility. This is believed to be due in part to possible feedback- inhibition of MTA-producing enzymes, which include the nicotianamine synthase, 1-aminocyclopropane-1-carboxylic acid synthase (produces the precursor to ethylene), and polyamine synthases. By feeding the polyamine spermidine to mtn1-1mtn2-1 double mutant seedlings for a brief time, partial fertility can be restored on random branches. Fertility is expanded to the whole plant in subsequent generations. We sought to elucidate the mechanism of transgenerational restoration of fertility by comparing the changes in key metabolites and transcriptomes between generations. Our results suggest the fertility may be linked with the recovery of cysteine, S-adenosylmethionine, and S-adenosylhomocysteine metabolite levels. These analyses also implicate changes circadian rhythm, stress response, and DNA methylation are relevant to fertility restoration. Current work is aimed at measuring related metabolites and analyzing the DNA methylome of spermidine-restored plants.

R3 Jian **Wu**, Sonia Gazzarrini
University of Toronto Scarborough, Department of Biological Sciences and University of Toronto, Department of Cell and Systems Biology; gazzarrini@utsc.utoronto.ca

Regulation of reproductive development and seed abortion in Arabidopsis

Reproduction is one of the most important developmental phases in all organisms and it is strictly regulated in plants. FUSCA3 is a plant-specific, B3-domain transcription factor (TF), which plays a crucial role in seed development and maturation. Previously, we showed that the fus3 loss-of-function mutant and overexpression lines cause seed and silique abortion, respectively, suggesting that low levels of FUS3 are also required for reproductive development. To further investigate the regulation of FUS3 expression during reproduction, a yeast one-hybrid screen was performed and BASIC PENTACYSTEINE (BPC) TFs were found to bind the FUS3 promoter. BPCs are expressed during both reproductive and seed development, and loss-of-function bpc double mutants induce severe seed abortion. We show that BPCs form homo- and hetero-dimers and recruit the Polycomb complex PRC2 to repress FUSCA3 expression during reproductive development. Indeed, the fus3-3 loss-of-function mutant is able to partially rescue bpc defects in vegetative and reproductive development, suggesting they are partially due to FUS3 overexpression. Accordingly, bpc loss-of-function mutants misexpress FUS3 in reproductive organs. Altogether, these findings show that BPCs controls FUS3 levels during reproduction reproduction to allow embryo and seed development and that FUS3 levels are tightly controlled in reproductive organs.

R4 Stuart **Macgregor**, Daphne Goring
University of Toronto; s.macgregor@mail.utoronto.ca

Secretion in the Arabidopsis thaliana compatible pollen response

The acceptance of compatible pollen in the Brassicaceae is tightly regulated through interactions between the pollen and the pistil. Secretion in the stigmatic papillae is proposed to be key to this interaction to provide resources to the pollen for hydration and germination. The objective of this research is to investigate components of the Arabidopsis thaliana secretory pathway machinery for their requirement in compatible pollen acceptance. Fluorescently-tagged markers that identify different compartments in the endomembrane system are being used examined to gain a fuller understanding of the secretory activity that occurs following compatible pollinations. In addition, the requirement of SNARE complex subunits, which are implicated in vesicle fusion and cargo release, is also being investigated through loss-of-function mutants. We have found that combinations of SNARE knockout mutants lead to a reduction in compatible pollen hydration, indicating its role in the compatible pollen acceptance pathway. Higher order SNARE knockout mutants will be used in the future to further characterize the complex's role in compatible pollen acceptance. Together with fluorescently-tagged endomembrane marker lines, this research will provide a better understanding of the stigmatic papilla's secretory system, and how this system is employed in the acceptance of compatible pollen.

R5 Hyun Kyung **Lee**, Daphne R. Goring
Department of Cell and Systems Biology. University of Toronto, ON, Canada; hyunkyung.lee@mail.utoronto.ca

The search for receptor kinases that regulate compatible pollen responses in the Brassicaceae stigma

Plant have developed ways to recognize pollen grains as providing nutrients to the wrong mating partner can be disadvantageous. The Brassicaceae family have dry stigmas, which means that they lack stigmatic surface secretions for automatic pollen acceptance. The procedures that allow pollen grains to become metabolically active is tightly regulated processes and require delivery of cellular components from the stigma. Cellular processes in the stigma that facilitate pollen acceptance are well documented, however, the initial upstream signalling components are yet to be elucidated. This project aims to identify receptor kinases that can recognize signals from the pollen and initiate basal compatibility pathway. To address this goal, the yeast two-hybrid system was employed to screen a library of receptor kinases with the previously identified stigma proteins in the Goring lab as baits. Putative kinase interactors were found to cluster to two distinct receptor kinase sub-groups and loss-of-function T-DNA lines and/or CRISPR deletion lines were then isolated for these receptor kinases for further investigations. Preliminary results gathered from these receptor kinase mutants show mild compatible pollen response defects, and further investigations are currently in progress to better understand the involvement of these receptor complexes in facilitating pollen acceptance and hydration.

POSTERS

P1 Diana **Bonea**, Sonia Gazzarrini, Rongmin Zhao

Department of Biological Sciences, University of Toronto Scarborough and Cell & Systems Biology, University of Toronto; diana.bonea@mail.utoronto.ca

Structural adaptations of the proteasome in response to oxidative, osmotic, and salt stresses in A. thaliana

Under adverse conditions, the ubiquitin-proteasome system (UPS) is crucial for protein quality control. Under this system, substrates tagged with polyubiquitin chains are targeted for hydrolysis by the 26S proteasome, which is canonically composed of multisubunit 19S regulatory (RP) and 20S core (CP) particles. Most stress-based UPS studies have focused on the substrates targeted for degradation, and the ubiquitin ligases that recruit them. However, the way in which proteasomes adapt to an increased demand for protein degradation remains unclear. In this study, we investigated the impact of abiotic stresses, chiefly oxidation, hyperosmolarity, and salinity, on proteasome complex profile and activity. Oxidative and salt stresses were found to induce a shift in the dominant proteasome complex, from doubly-capped (RP2CP) to uncapped CP alone. Furthermore, oxidative stress also seems to enhance the activity of the isolated CP, by rendering the core proteolytic chamber more accessible to substrates. Immunoprecipitation of the proteasome has revealed little change in subunit composition, suggesting that while the proteasome's basic constituents remain the same, abiotic stress may affect their assembly into higher-order complexes. We have identified several putative proteasome assembly chaperones and, via genetic analyses, we are investigating their role in structural remodeling of the proteasome during stress adaptation.

P2 Indira Q **Castillo** 1,2, Isabel Molina 2, Mark A Bernards 1

1 Department of Biology, Western University, London Ontario, Canada; 2Department of Biology, Algoma University, Sault Ste. Marie Ontario, Canada; yqueralt@uwo.ca

Functional Characterization of Arabidopsis thaliana HXXXD-motif Acyltransferases involved in suberin metabolism

Plants synthesize suberin, a lipophilic extracellular barrier that controls water and nutrient loss and protects plants against pathogen infection. In Arabidopsis thaliana, suberin is deposited in the cell walls of seed coats, endodermis, and periderm of roots. Suberin is a complex lipid polymer formed by an aliphatic polyester and an aromatic polymer, co-deposited with soluble waxes. The aliphatic polyester is composed of ?-hydroxy fatty acids, a,?-dicarboxylic acids, fatty alcohols, glycerol, and ferulate. Alkyl ferulates, alkyl coumarates and alkyl caffeates are major components of suberin-associated root waxes, which also contain alkanes, fatty acids and fatty alcohols. Two enzymes of the BAHD family of HXXXD-type acyltransferases, ASFT (AT5G41040) and FACT (AT5G63560), function as feruloyl- and caffeoyl-CoA transferases, respectively. However, the enzyme responsible for transferring coumarate remains unknown. Individual mutants of these enzymes are not affected in either alkyl coumarates or suberin-bound coumarate, but in vitro both enzymes can transfer coumaroyl-CoA to fatty alcohols, suggesting that these enzymes may have partially redundant function in suberin and wax biosynthesis. To investigate redundant functionality in vivo, we generated double mutants (asft × fact). We will report our progress in the characterization of asft x fact suberin and root-associated waxes.

P3 Cecily D. **Costain**, Michael J. Emes, Ian J. Tetlow

Department of Molecular and Cellular Biology, University of Guelph; ccostain@uoguelph.ca

The Role of Starch in the Development, Physiology, and Reproduction of Arabidopsis thaliana

Starch is an agronomically important polyglucan synthesized by plants as a means to store photoassimilates as an osmotically inactive carbon store for subsequent use in metabolism and growth. The synthesis of starch involves several enzymes working in concert, including starch branching enzyme (SBE), responsible for cleaving alpha-(1,4)-bonds on pre-existing glucan chains and reattaching them to the same or to neighbouring chains via alpha-(1,6)-linkages. SBE activity creates branch points and contributes to starch's semi-crystalline structure. Recently, SBEI and SBEIIb cloned from maize (Zea mays) endosperm were constitutively expressed in a starchless, null line of Arabidopsis, lacking endogenous SBEs. While both ZmSBEI and ZmSBEIIb were able to individually restore starch synthesis in the null line, transformants exhibited altered starch metabolism. Here, we have re-transformed the null line with ZmSBEI and ZmSBEIIb under the control of a CaMV 35S promotor and are in the process of characterizing these lines. Transgenic Arabidopsis displays an intermediate phenotype between wild-type and the null line. Transformants take longer to bolt and to produce inflorescences, indicating that substituting the activity of Arabidopsis SBEs with those of maize prolongs the transition from juvenility to adulthood.

P4 Laura D. **Cox**, Gale G. Bozzo

Department of Plant Agriculture, University of Guelph, Guelph, ON, N1G 2W1; lcox05@uoguelph.ca

Transcriptome analysis of the interaction between common bean and Xanthomonas axonopodis pv. phaseoli, the casual agent of common bacterial blight

Common bacterial blight (CBB) is a disease that affects common bean (Phaselous vulgaris) production and is caused by Xanthomonas axonopodis pv. phaseoli (Xap). Xanthomonad virulence in susceptible plants is mitigated by type III secretion system effector proteins. Overall, plant resistance to xanthomonads is afforded by immune responses encoded by plant defense genes, including those associated with hormone signaling (e.g., brassinosteroid) and secondary metabolite (e.g., phenylpropanoid) pathways. The mechanisms by which CBB-susceptible (i.e., OAC Seaforth) and CBB-resistant (i.e., OAC Rex) common bean genotypes interact with Xap are little understood. We will test the hypothesis that CBB-resistance in common bean is dependent upon the induction of a plant defense response, whereas CBB-susceptibility is associated with the absence of a defense response. Leaves of CBB-resistant and CBB-susceptible recombinant inbred lines generated from a cross between 'OAC Rex' and 'OAC Seaforth' will be inoculated with Xap and their RNAseq transcriptome profiles will be compared to those of mock-inoculated plants. RNAseq data will be mapped to the OAC Rex genome; gene ontology enrichment will establish the putative function of all differentially expressed genes. The research will be valuable for bean breeders aiming to create elite common bean germplasm that is fully resistant to CBB.

P5 André G. **Duarte**, Fred J. Longstaffe, Danielle A. Way
Western University, Department of Biology & Department of Earth Sciences; aduarte4@uwo.ca

Do I grow or do I fight? Plant growth and defense compounds at past low [CO2]

For most of the recent evolutionary history of plants, atmospheric CO_2 concentrations have been far lower than modern values. Since CO_2 availability is reduced at low CO_2conditions, photosynthesis decreases, and so does carbon available for growth and production of defense compounds against herbivory. To investigate how plants balance growth requirements and defense compound production at low CO_2, we grew *Picea mariana* at ambient (~400 ppm) and low CO_2 conditions (~180 ppm). We measured biomass, photosynthetic rates, and leaf anatomy, including resin duct morphology, which are the main sites of resin production in conifers. Total biomass in low CO_2-grown plants decreased by 73% and photosynthetic rates by 47% compared to ambient-CO_2 plants. At ambient CO_2, plants formed two resin ducts in their needles, but 28% of needles from low-CO_2 plants lacked either one or both ducts, and duct lumen area was 45% smaller. Reduction or loss of resin ducts in plants grown at low CO_2 implies that these plants invest less carbon in defense than plants grown at modern CO_2 conditions. Thus, the relationship between plants and herbivores may have changed over time as the ability of plants to chemically defend themselves was affected by changes in atmospheric CO_2 levels.

P6 Corey **Flude,** Eric Lyons
University of Guelph, Department of Plant Agriculture; cflude@uoguelph.ca

Plant growth regulators effect on survival of putting greens during thawing events in late winter

Winter survival is dependent on a plants ability to acclimate and then to remain acclimated during thaw refreeze which can occur in the late winter and early spring. The ability of plant growth regulators (PGR) to influence acclimation status and survival of creeping bentgrass and annual bluegrass throughout the winter and simulated warming events was determined using a novel pot-in-pot system that allows harvesting of plants throughout the winter. Gibberellic Acid (GA) (2mg/m2), Abscisic acid (ABA) (11 mg/m2) and Trinexapac-Ethyl (TE) (4.4 mg/m2) were applied during the fall acclimation period and acclimation status, measured by LT50, was not influenced by plant growth regulators in the months of January, February, and March for both species. In April PGR treatments resulted in a loss of acclimation status for annual bluegrass and ABA and GA applications increased acclimation status for creeping bentgrass. TE application to creeping bentgrass resulted in lower net carbon exchange rate than the control in the presence of light during a warming period, indicating retention of acclimation status during warming as photosynthetic rate was correlated with LT50 ($r = 0.59$, $p = 0.0157$). The results indicate that PGR treatments can be beneficial for retaining the acclimation status of creeping bentgrass.

P7 Joshua J.R. **Frank**1, Mamdouh Abou-Zaid1,2, Tod Ramsfield1,3, Danielle Way1,4

1. Department of Biology, Western University, London, ON; 2. Great Lakes Forestry Centre, National Resources Canada, Ottawa, ON; 3. Northern Forestry Centre, National Resources Canada, Edmonton, AL; 4. Nicholas School of the Environment, Duke University, Durham, NC; jfrank22@uwo.ca

Impacts of root-associated fungi on tree growth under elevated temperature and CO2

Growth of poplars, an economically important group of trees, has been declining due to elevated temperatures and droughts associated with climate change. Symbiotic microbes, such as root-associated fungi (RAF) may increase plant growth under climate change conditions, by increasing tree access to water and nutrients. To address this hypothesis, three RAF were isolated from poplar roots in the field. We then determined the effects of RAF inoculation on poplar growth under a range of future climate scenarios: ambient (400 ppm) or elevated CO2 (750 ppm) with either ambient temperatures, or a +4 °C or +8°C warming treatment. Inoculation with RAF did not increase plant height or total dry weight in plants grown at ambient CO2. However, inoculated trees grown under +4 ?C with elevated CO2 were taller and had greater biomass than trees from ambient CO2 treatments with either ambient or +4 °C temperatures. Non-inoculated trees also had a significant increase in biomass when grown at higher CO2 and temperature conditions, although this CO2 effect was reduced in the +8 °C treatment. Our results suggest that RAF increases tree growth under moderate warming (+4 °C) and may provide resilience to future climatic stresses.

P8 Tim **Garant** 1 , Li Wei 2 , Jordan Roberts 1 , Rebecca Kalinger 1 , Suo-Min Wang 2 , Owen Rowland 1,2

1 Department of Biology and Institute of Biochemistry, Carleton University, Ottawa, ON, Canada

2 State Key Laboratory of Grassland Agro-ecosystems, College of Pastoral Agriculture Science and Technology, Lanzhou University, Lanzhou, P. R. China; TimGarant@cmail.carleton.ca

The effects of drought and salt stress on cuticular wax composition in the leaves and stems of the extremophile Zygophyllum xanthoxylum

Drought and salinity are abiotic stresses that reduce crop growth. Xerophytes and halophytes are plants that have unique mechanisms to survive under drought and salt stress, respectively. Zygophyllum xanthoxylum is a succulent shrub that has been defined as both a xerophyte and a halophyte. All land plants deposit a waxy (hydrophobic) cuticle on their aerial organs to help prevent uncontrolled non-stomatal water loss. To determine if the cuticular wax layer of Z. xanthoxylum helps confer salt and drought tolerance, we used gas chromatography to analyze leaf and stem cuticular wax composition of Z. xanthoxylum under control conditions and under salt or drought stresses. Under control conditions, the leaf cuticular waxes of Z. xanthoxylum were comprised of very-long-chain (VLC) alkanes (86.21 %), of which 81.18 % was 31:0 alkane, VLC alcohols (9.02 %), sterols (2.83 %), VLC aldehydes (1.94 %), and wax esters (trace). Stem cuticular waxes were comprised of the same chemical classes at 79.5 %, 12.16 %, 4.99 %, 3.35 %, and trace amounts respectively. For the leaf and stem cuticular waxes, the salt stress induced a 3.7-fold and 2.8-fold increase in total VLC alkanes, respectively, and the drought stress induced a 2.2-fold increase and no change in VLC alkanes, respectively. These results imply that Z. xanthoxylum increases the amount of VLC alkanes in its cuticular waxes in response to stresses, likely to reduce water lost via the cuticle.

P9 Satinder K. **Gidda**1, Michal Pyc1, Yingqi Cai2, Michael S. Greer2, Franziska K. Kretzschmar3, Damien Seay4, Nathan Doner1, J. Joe Hull4, Till Ischebeck3, Kent D. Chapman2, John M. Dyer4, and Robert T. Mullen1

1 Department of Molecular and Cellular Biology, University of Guelph, Guelph, ON, N1G 2W1, Canada; 2 Department of Biological Sciences, Center for Plant Lipid Research, University of North Texas, Denton, TX, 76203, USA; 3 Department of Plant Biochemistry, Albrecht-von-Haller-Institute for Plant Sciences, University of Goettingen, Goettingen, Germany; 4 U.S. Department of Agriculture, Agricultural Research Service, U.S. Arid-Land Agricultural Research Center, Maricopa, AZ, 85138, USA; sgidda@uoguelph.ca

The role of LDIP in regulating lipid droplet size and neutral lipid homeostasis in plant cells

Lipid droplets (LDs) are unique organelles that contain a core of neutral lipids, such as triacylglycerols (TAGs), enclosed by a single phospholipid monolayer and decorated with a diverse array of 'coat' proteins. In plants, LDs are found in high abundance in pollen and oilseeds, and are ultimately mobilized in the latter as a carbon and energy source during post-germinative growth. While some of the proteins associated with seed LDs have been characterized, relatively little is known about LDs in other tissues. Our group has identified several new LD proteins involved in the biogenesis, maintenance, and turnover of LDs in vegetative tissues. Here we describe the identification and characterization of LDIP (Lipid Droplet-Associated Protein-Interacting Protein), which is a plant-specific protein that is ubiquitously expressed in Arabidopsis. We show that LDIP localizes to the LD surface via an amphipathic a-helix, forms associations with other known coat proteins, and influences LD size and neutral lipid homeostasis in both leaves and seeds. Efforts are underway to elucidate LDIP's role(s) in LD formation and its influence on the lipid composition of LDs. These findings contribute to our growing understanding of LD biogenesis and modulation of TAG content in both seed and non-seed tissues of plants.

P10 William **Hargreaves**1, Amidou N'Daiye2, Curtis J. Pozniak2 and Lewis Lukens1

1Department of Plant Agriculture, University of Guelph, Crop Science Building, 50 Stone Road E, Guelph, ON, Canada, N1G 2W1; 2Department of Plant Sciences and Crop Development Centre, University of Saskatchewan, 51 Campus Drive, Saskatoon, SK, Canada, S7N 5A8; whargrea@uoguelph.ca

An investigation of Canadian bread wheat diversity reveals distinct germplasms corresponding to breeding groups and reduced genetic diversity within groups due to recombination suppression

We used a Wheat 90K SNP iSelect array to genotype 388 bread wheat cultivars grown in western Canada across 15,321 markers. Genetic analyses of variety relationships are congruent with reported pedigrees, although some varieties' genetic histories differ from expected. Based on grain attributes and growth habit the three largest variety groups, roughly corresponding to market classes, are Hard Red Spring (HRS), Hard Red Winter (HRW), and Soft White Spring (SWS). Clustering array data splits the varieties into five clusters, one each for HRW and SWS, three for HRS, and a noise group that contains cultivars outlier varieties. These groups account for 29% of the allelic variation in the sample with clustered SWS varieties the most divergent group. In comparisons of HRS to HRW and HRS to SWS, frequencies of marker alleles greatly differ near the gene VRN-A1 that is partially responsible for growth habit. Expected heterozygosity and linkage disequilibrium analyses reveal very large haplotypes proximal to the centromere on chromosomes 1A, 2A, 4A, 6A, 7A, 5B, and 6B segregating in every sample cluster. Thousands of genes have behaved as single locus in wheat breeding.

P11 Solmaz **Irani**, Caitlin M.A. Simopoulos, Mitchell J. R. MacLeod, Wilson W.L. Sung, Peter S. Summers, G. Brian Golding, Elizabeth A. Weretilnyk
Department of Biology, McMaster University, Hamilton, ON, L8S 4K1, Canada; iranis4@mcmaster.ca

Genome-wide transcriptional analysis of different response of two Eutrema salsugineum ecotypes to progressive drought treatment

The halophyte Eutrema salsugineum is an extremophyte and relative of Arabidopsis and Brassicaceae crops. In this study we compared the molecular response of two E. salsugineum ecotypes, originating in Yukon, Canada and Shandong, China, to a progressive water deficit. Plants that were well-watered (WW1) were subjected to a water deficit (D1), plants were then re-watered, allowed to recover (WW2) and subjected to a second water deficit (D2). Previous work shows that Yukon and Shandong plants respond similarly during D1 (eg. with respect to turgor, solute and water potentials) and that Yukon plants manage reduced water availability during D2 better than Shandong plants. We hypothesized that during D1 and/or recovery during WW2, Yukon plants are primed to improve their water deficit tolerance during D2 whereas Shandong plants are not. The fraction of transpirable soil water for D1 and D2 plants was 10% when tissue was collected for RNA. Leaf transcriptomes from plants undergoing the progressive drought stress treatment were compared. We detected >1000 differentially expressed genes (DEGs) between WW1 and D1 Yukon plants but only 60 DEGs between WW1 and D1 Shandong plants. For the second drought treatment (WW2? D2), about 2000 and 5000 DEGs were found for Yukon and Shandong plants, respectively. Co-expression network analysis suggests Yukon plants undergo re-programming of gene expression that correlates with drought, but Shandong plants do not.

P12 Emine **Kaplanoglu**, Igor Kolotilin, Rima Menassa, Cam Donly
Agriculture and Agri-Food Canada, London Research and Development Centre, London, ON; The University of Western Ontario, London, ON; emine.kaplanoglu@canada.ca

Plastid transformation of Micro-Tom tomato for in planta RNA interference to manage pest insects

Heat, humidity, and crowding in greenhouse environments are conducive to establishment of insect pests, such as the greenhouse whitefly (Trialeurodes vaporariorum). Greenhouse whitefly causes damage by feeding on sap and by vectoring plant viruses. Currently, this pest is managed by biological and chemical control; however, biological control fails to provide rapid control while chemical control is hampered by insecticide-resistance development. Therefore, alternative control strategies are needed. RNA interference (RNAi) is an emerging technology for crop protection, and modifying plant genomes to express dsRNA is one strategy to utilize RNAi for pest control. RNAi is most effective when insects consume intact dsRNA; hence, nuclear transformation of plants provides limited protection from pest damage due to the plant's intrinsic ability to process dsRNA into small interfering RNA in the cytosol. This limitation can be overcome by producing dsRNA in the prokaryotically-derived plastids of plants, which lack the RNAi processing machinery, allowing accumulation of intact dsRNA in the plastids. Therefore, as a proof of concept, we constructed a transformation vector for the tomato plastid genome, and using this vector, produced transplastomic Solanum lycopersicum (cv. Micro-Tom) plants expressing dsRNA for an essential gene of greenhouse whitefly.

P13 Sophie **Krolikowski**1, Ling Chen1, Tim McDowell1, Justin Renaud1, Sangeeta Dhaubhadel1,2, Ian Scott1,2

1 London Research and Development Centre, Agriculture and Agri-Food Canada, London ON; 2 Department of Biology, University of Western Ontario, Lindon, ON; sophie.krolikowski@canada.ca

Isoflavonoid levels in soybean (Glycine max) cultivars and associated anti-herbivore activity

Soybean aphid (Aphis glycines) and two-spotted spider mite [(TSSM) Tetranychus urticae] are damaging herbivore pests of soybean (Glycine max) in Southern Ontario. Management of these two pests includes monitoring for economic thresholds and applying insecticides when necessary. Host plant resistance is a recognized strategy in integrated pest management (IPM); therefore, it is useful to know which soybean cultivars at different maturity groups are resistant to aphids and TSSM. Isoflavonoids, a group of legume-specific compounds, are known to have a negative effect on crop pests. Based on preliminary HPLC analysis of isoflavonoid concentrations in soybean leaves, twelve cultivars were selected to screen aphid and TSSM resistance. A qualitative assessment of each plant at the end of the 4-week experiment indicated the more tolerant cultivars were OAC Lakeview and OT06 -22 to both aphids and TSSM. However, the observed resistance to aphid and mites is not completely explained by the levels of the six most common isoflavonoids. A metabolomics approach has since provided much additional information on a broader phytochemical profile allowing us to evaluate trends between low and high resistant lines. Soybean farmers will benefit from this research through improved knowledge of which soybean cultivars have higher resistance to insect and mite pests.

P14 Vincent **Lau**, Asher Pasha, Nicholas Provart

Cell & Systems Biology, University of Toronto; vincente.lau@mail.utoronto.ca

Arabidopsis Interactions Viewer 2.0: A multifaceted-integrative client-side web based application for visualizing PSICQUIC protein-protein and protein-DNA interactions

The Arabidopsis Interactions Viewer 2.0 (AIV 2.0) is a vast improvement of a former web application (AIV 1.0) which allows a network visualization of 3 million experimentally-validated and 70,000 predicted Arabidopsis protein-protein and protein-DNA interactions curated by the Bio-Analytic Resource (BAR) for Plant Biology. Additional interaction data can also be loaded from other Proteomics Standard Initiative Common QUery InterfaCe (PSICQUIC) servers such as BioGrid or IntAct. Major technical improvements include using newer, faster technology such as cytoscape.js, allowing a much larger network to be loaded. Moreover, AIV 2.0 integrates multiple facets of biology such as subcellular localization pie-chart overlays, plant gene function terms (MapMan), group-by-localization layouts and filtering by co-expression scores. Multiple export options are available such as exporting to a Cytoscape-tailored JSON and tabular format (CSV) which allows user-defined scripting. To our knowledge, this is the only interactions visualization tool for plant biologists to feature the above facets of biological knowledge. Our goal is therefore to allow researchers use this tool's multiple data layers to generate new hypotheses relevant to plant biology.

P15 Jonathan **Lee**, Dinesh Christendat
University of Toronto; jonathans.lee@mail.utoronto.ca

Investigating the functional evolution of plant shikimate kinase-like 1 (SKL1)

Chloroplasts are essential for plant development as they house the components of the photosynthetic machinery and several other metabolic processes. As such, the biogenesis of chloroplasts is one of the most important topics in plant biology. While there has been much progress in deciphering this highly complex process, many aspects are still not well understood. In particular, the roles of several genes have yet to be fully elucidated. One such example is shikimate kinase-like 1 (SKL1), an ancient gene duplicate of shikimate kinase (an enzyme of the shikimate pathway) that arose during the evolution of early land plants between 400 and 500 million years ago. T-DNA insertion mutants of Arabidopsis thaliana (skl1-8) exhibit an albino phenotype with vesiculated plastids. Part of our work is focused on investigating the functional evolution of SKL1 in land plants. This is being done through studies of the liverwort species Marchantia polymorpha, one of the most basal species of land plant. Results have shown that there are two highly similar homologs of shikimate kinase in M. polymorpha with only one showing shikimate kinase activity. Future characterization of these genes will be achieved through mutagenesis and crystallographic studies.

P16 Marwa **Louati**, Cuneyt Uçarli, Amel Salhi Hannachi, Turgut Kara Neslihan, Ian Tetlow
1 Université de Tunis El Manar. Faculté des Sciences de Tunis. Laboratoire de Génétique Moléculaire, Immunologie et Biotechnologie LR99ES12. Campus Universitaire El Manar 2092 Tunis, Tunisia; 2 Istanbul University, Faculty of Science, Department of Molecular Biology and Genetics, 34134, Istanbul, Turkey; 3 Department of Molecular and Cellular Biology, University of Guelph, Ontario, Canada, N1G 2W1; mlouati@uoguelph.ca

A mutation screening and genotyping by high-resolution melting (HRM) to detect delta-6-desaturase (D-6-D) variants in Argania spinosa

The argan tree is a multipurpose tree of great socio-economic interest. Its oil is used in food, medicine, and in cosmetics, the tree is considered a good buffer against desertification and erosion and provides a good quality wood for various purposes. It's rare to find this species outside of Morocco but hundreds of trees exist in Tunisia in different locations. Few argan genes are sequenced, and such sequenced gene is the delta-6-desaturase (D-6-D) a key enzyme to oil biosynthetic pathway. The purpose of this study is to report the use of hight resolution melting (HRM) analysis for scanning and genotyping Argania spinosa individuals which has never been reported before, to detect delta-6-desaturase (D-6-D) variants. The present study describes an HRM analysis of natural sequence variation within a small region of D-6-D gene from 60 argan trees belonging to four different sites in Tunisia. Methylated DNA and unmethylated DNA acquire different sequences after bisulphite treatment resulting in PCR products with markedly different melting profiles, the efficiency of the HRM procedure was affected by various factors, including specificity and efficiency of PCR, amplicon length and position, and DNA template quality. In addition to these factors, the use of PCR product rather than genomic DNA in HRM increased the quality of melting curves, thus affecting the accuracy and sensitivity of the assay. Using the shape of the melting curves, we could assign the differences between curves of the different trees under investigation in fact HRM classified 10 groups of variants carrying deletion mutants and single and multiple SNPs.

P17 Mimmie **Lu**, Lining Tian

London Research and Development Center, Agriculture and Agri-Food Canada, London ON N5V 4T3, Canada; mimmie.lu@canada.ca

Regeneration and Agrobacterium-mediated transformation of soybean: towards trait improvement via genome editing

Genome editing using CRISPR/Cas9 (clustered regularly interspaced short palindromic repeats/CRISPR-associated 9) has become a popular approach to induce targeted mutations for crop trait improvement. Soybean (Glycine max) is an economically important crop worldwide. In order to modify soybean genes using genome editing, it is crucial to first achieve regeneration and stable transformation. While genetic transformation is routine for many plant species, soybean transformation is still challenging. A binary vector containing reporter gene ß-glucuronidase (GUS) and selectable marker encoding phosphinothricin acetyltransferase was used for Agrobacterium-mediated transformation of two soybean cultivars: Williams 82 and Bert. Cotyledonary node explants were inoculated with Agrobacterium tumefaciens strain EHA105, and cultured on media containing herbicide glufosinate-ammonium. Shoots developed from selection were transferred to non-selective medium for root development. Plantlets recovered were positive for GUS staining analysis for both cultivars, indicating success of genetic transformation of soybean. To develop CRISPR/Cas9 genome editing technology in soybean, four constructs targeting different sites of phytoene desaturase (PDS) gene were created. Each construct contains 35S promoter driving the expression of Cas9 translationally fused to green fluorescent protein (eGFP), and Arabidopsis AtU6 promoter driving the expression of a 20-nucleotide target sequence along with single-guideRNA (sgRNA) scaffold. Transformation with the genome editing constructs is being carried out.

P18 Gregory J. **MacNeill**, Michael J.Emes. Ian J. Tetlow

Department of Molecular and Cellular Biology, College Biological Sciences, University of Guelph, Ontario, Canada; macneilg@uoguelph.ca

The role of post-translational modification in the regulation of Arabidopsis starch branching enzyme 2.2

Starch is a major component of the human diet and provides an insoluble carbon store for plants. Unlike storage starch, produced for long term or cross-generational usage, transient starch is produced and degraded in the leaves over the diurnal cycle. This short-term carbon store in chloroplasts provides a source of energy and fixed carbon while plants are not photosynthesizing. Starch biosynthesis requires the coordinated activities of multiple classes of enzymes, such as starch synthases which polymerize ADP-glucose into a-glucan chains, and starch branching enzymes (SBE) which introduce branch points to the growing glucan chain. SBEs form phosphorylation dependent complexes with other starch biosynthetic enzymes important for normal function. Arabidopsis has two SBE isoforms (SBE2.1 and SBE2.2), of which SBE2.2 accounts for most of the measurable catalytic activity. Recombinant SBE2.2 was phosphorylated by soluble chloroplast extracts on residues Ser290 and Ser301. A putative protein-protein interaction domain, conserved across all class II SBEs, has also been identified. Site-directed mutagenesis was used to alter this conserved domain, Ser290 and Ser301, and a C-terminal Cys residue predicted to be involved in redox modulation to investigate their importance in catalysis and the formation of heteromeric complexes in vitro.

P19 Boris F. **Mayer**, Jean-Benoit Charron
Department of Plant Science, Macdonald Campus of McGill University, 21111 Lakeshore Road, Ste-Anne-de-Bellevue, QC, H9X3V9; boris.mayer@mail.mcgill.ca

Developing under a changing environment: cold acclimation and vernalization in Brachypodium distachyon

With the onset of climate change, it has become increasingly relevant to understand how plants respond to changing environmental conditions. Yet, temperate plants regularly face seasonal change and to persist in these conditions, have adapted by adjusting their stress tolerance, phenology and development. Understanding how temperate plants follow seasonal cues during their development can help elucidate adaptation mechanisms in plants. Cold acclimation (CA) and vernalization (VRN) are processes that ensure persistence in temperate climates by regulating freezing tolerance and flowering time respectively. However, how these two processes are integrated into a coordinated developmental response remains poorly understood. The model grass Brachypodium distachyon has emerged as a model to study CA and VRN in temperate cereals. By identifying key seasonal cues that occur within the native range of the species, we designed a diurnal freezing treatment (DF) that combines prevailing summer-to-winter transition signals. Under DF, B. distachyon accessions of different climatic origins manifest coordinated and novel cold acclimation and vernalization responses. Altogether, our results demonstrate a direct link between CA and VRN, and that typically used constant-temperature cold treatments induce an “over-vernalized” molecular state at the expense of freezing tolerance. This work also stresses the importance of reproducing natural signals in laboratory conditions.

P20 Sarah J. **McDonald** 1 and Danielle A. Way 1,2
1 Department of Biology, University of Western Ontario, London, ON, Canada2 Nicholas School of the Environment, Duke University, Durham, NC, USA; smcdon83@uwo.ca

How will climate change affect nectar and pollen quantity and quality in an economically important crop?

Due to anthropogenic release of CO2 and other greenhouse gases, the average surface temperature of the earth is increasing. It is well established that increases in temperature and [CO2] affect the physiology of Earth’s flora, a notable consequence being decreased nutritional value of agricultural crops. While there is abundant information on the effects of climate change on human nutrition, whether pollinator food supplies are affected by these same drivers has received less attention. With recent declines in bee populations due to pathogens and pesticide use (among other factors), adequate nutrition is becoming increasingly important because it directly affects pollinator susceptibility to these factors. To investigate how pollinator food sources respond to climate change I will expose Cucumis sativa to elevated CO2 and temperature using a full factorial design. This will elucidate interactive and singular effects of these drivers on two main pollinator food sources: pollen and nectar. To determine uptake and allocation of carbon I will measure leaf carbohydrates, allometry, flower number and morphology, and photosynthetic rate. Finally, I will conduct a pollinator preference test to investigate in vivo effects of nutritional changes. Ultimately, this study will assess the viability of pollination as an ecosystem service under a changing climate.

P21 Muhammad S. **Tahir**, Jim Karagiannis, Lining Tian
University of Western Ontario,London Research and Development Centre - Agriculture and Agri-Food Canada; mtahir25@uwo.ca

Investigating the relationship of HD2 family members of histone deacetylases in response to drought stress in Arabidopsis thaliana

Three distinct families of histone deacetylases (HDACs) have been identified in plants: the RPD3/HDA1 family, the SIR2 family, and the plant-specific HD2 family which is the focus of this study. Even though the research on HD2-type HDACs has demonstrated their important role in regulating gene expression in several biological processes, especially in abiotic stress response, the relationship of HD2 family members with each other in response to environmental stresses remains largely unknown. Objective of this study is to investigate the relationship of four HD2 family members in Arabidopsis. For this, firstly in-vitro yeast two-hybrid assay was performed. HD2A showed interaction with HD2C and HD2D proteins. Also, HD2C showed interaction with HD2D. None of the HD2 family members showed interaction with HD2B. This indicate that HD2A, HD2C and HD2D proteins may be related to regulate target gene expression. Interaction results of Y2H assay will be verified by BiFC. To study whether HD2-type genes relate each other in responding to drought stress, hd2 single, double and triple mutant lines will be developed. Additionally, HD2 over expressing lines will also be created. All the genotypes will be assessed for their growth and survival under drought stress. Gene expression pattern, plant phenotypes and survival of different genotypes will reveal the relationship of HD2 members in response to drought. Knowledge generated from this research will be useful to improve our understanding of the roles of HD2-type proteins in mediating plant response toward drought stress.

P22 Jordan B **Van Brenk**, Emily J Cornelius, Susanne E Kohalmi
The University of Western Ontario, Department of Biology; jvanbre@uwo.ca

Differentiating the expression of ADTs in Arabidopsis

Phenylalanine is an essential amino acid and precursor to many specialized metabolites such as lignins (structural polymers) and flavonoids (pigments/scents) in plants. The final step of phenylalanine biosynthesis is a decarboxylation/dehydration reaction catalyzed by AROGENATE DEHYDRATASEs (ADTs). In Arabidopsis thaliana, six isoforms (ADT1-ADT6) can perform this reaction,
each of which have differing subcellular localization and expression patterns. Interestingly, individual ADTs have been shown to differentially channel carbon to downstream metabolites. For example, lignin biosynthesis is more dependent on ADT5 than other ADTs. The variation in expression patterns, combined with special roles in carbon-channelling suggest a modular control of ADT expression in response to endogenous and exogenous cues. To uncover differential ADT transcriptional regulation, ADT promoter sequences (1kb upstream of the transcriptional start site) were analyzed in silico for putative regulatory elements, finding common and unique motifs across the gene family. Additionally, mRNA expression data were compared for the six ADTs in different Arabidopsis developmental stages and tissues. By comparing each possible combination of ADT in pairs, distinct differences between their expression patterns were clear. Using these analyses as a stepping stone, in vivo experiments are underway exploring ADT transcriptional regulation in Arabidopsis grown under standard and stress growth conditions.

P23 Lilia **Virta**, Kristen van Gelder
University of Guelph; sophie.krolikowski@canada.ca
Isoflavonoid levels in soybean (Glycine max) cultivars and associated anti-herbivore activity
The occurrence of polyprenols throughout the plant kingdom is well documented, yet their functional role is not yet clear. These compounds are known to be assembled from isoprenoid precursors by enzymes referred to as cis-prenyltransferases (CPTs). In this study, RNAi-mediated knockdown of members of the tomato (Solanum lycopersicum) and Arabidopsis thaliana CPT families (SlCPT5 and AtCPT7) is shown to reduce polyprenols in leaves by ~66-70%. Subcellular fractionation studies and in vivo localization of SlCPT5 fluorescent protein fusions show that SlCPT5 resides in the chloroplast stroma, however its enzymatic products (polyprenols 45-60 carbons in length) accumulate in thylakoid and envelope membranes. Fluorescence anisotropy measurements revealed a more disordered state of the RNAi envelope membranes in tomato, as well as in thylakoid membranes of Arabidopsis. In polyprenol-deficient tomato leaves, thylakoid membranes exhibited lower phase transition temperatures and calorimetric enthalpies, indicating alterations in integral membrane protein stability. Taken together, these results suggest that polyprenols play a part in governing chloroplast membrane dynamics.

P24 Kevin **Xiong**, Ying Wang, Chris Bergin, Shelley R. Hepworth
Department of Biology, Carleton University, Ottawa, Ontario, Canada; kevinxiong@cmail.carleton.ca
Investigating the role of clade III TGA transcription factors in BLADE-ON-PETIOLE-dependent regulation of development in Arabidopsis thaliana
BLADE-ON-PETIOLE1 and 2 (BOP1/2) are members of the BTB-ankyrin protein family found in Arabidopsis thaliana. Members of this group play important roles in defense and development. BOPs play a crucial role in patterning organ boundaries. Loss-of-function bop1 bop2 mutations disrupt boundaries resulting in fused organs and altered patterning at the base of leaves and flowers, including loss of floral organ abscission. BTB-ankyrin proteins function as co-transcription factors, having a domain for activating transcription but no DNA binding domain. BOP1/2 thereby interact with TGA (TGACG-motif binding) basic leucine zipper transcription factors for recruitment to DNA. The Arabidopsis genome contains ten TGA factors, subdivided into five clades. Clade V and clade I TGAs function in BOP-dependent regulation of plant development. However, little is known about the function of clade III TGA transcription factors (TGA3 and TGA7).Through a yeast two hybrid screen, we find that BOP1/2 interact strongly with TGA3. We also show that TGA3/7 are expressed at the same locations as BOP1/2 using a GUS reporter gene assay. In addition, we show that plants overexpressing BOP2 require TGA3/7 to function. These data provide evidence that clade III TGA transcription factors interact with BOP1/2 to regulate plant development.

P25 Lingjie **Zhang**, Bona Mu, Homaira Hamidzada and Rongmin Zhao
Departments of Biological Sciences and Cell & Systems Biology, University of Toronto; bona.mu@mail.utoronto.ca

Complementation analysis of a newly designed HSP90C gene in Arabidopsis

HSP90C in Arabidopsis is a chloroplast stroma-localized HSP90 family molecular chaperone. It is required for the active chloroplast protein import and protein transport into the thylakoid. It has been previously observed that when attempting to overexpress the HSP90C gene under the CaMV 35S promoter, transgene-induced silencing of the endogenous HSP90C gene occurs very frequently, causing a variegation phenotype and making it difficult to obtain stable overexpression lines. In this study, we reconstructed the HSP90C coding sequence and generated a new HSP90C coding gene termed HSP90C' that has no region longer than 21nt that is identical to the endogenous HSP90C gene. The newly designed HSP90C' gene was expressed in Arabidopsis under CaMV 35S promoter and screening variegation phenotypes revealed no transgene-induced silencing. We also introduced HSP90C' gene into the HSP90C T-DNA insertion knockout line and showed that HSP90' under CaMV 35S promoter is able to partially complement the endogenous HSP90C gene. Our study on the newly designed HSP90C coding sequence not only confirmed that the native HSP90C is very sensitive to transgene induced silencing, but also provided a new tool for further studies of the role and mechanism of action of the chloroplast HSP90C protein.

P26 Xi **Zhang**, David Smith
Biology Department, Western University; xzha25@uwo.ca

Sequencing and assembling the nuclear genome of the Antarctic psychrophilic green alga Chlamydomonas sp. UWO241: Unravelling the cold-adaption

Antarctica harbours a variety of green algae surviving in permanently cold climate. However, little is known about how these psychrophiles can adapt in frigid conditions. To gain insight into this issue, we sequenced Antarctica psychrophilic green alga Chlamydomonas sp. UWO241 with multiple sequencing datasets. DNA sequencing technologies have been undergoing tremendous development and acting as engine to generate a wealth of genome data in the past 40 years. Although NGS makes genome sequencing handy, the followed data analysis and biological explanations are still the bottle-neck in understanding genomes. During my PhD studies, I will look into Bioinformatics approaches including genome assembly, genome annotation and comparative genomics to analyze the high throughput data of green algae, specially C. sp. UWO241. Firstly, I will process nuclear genome assembly pipeline to acquire high quality assembly. Then the genome assembly contigs will be the inputs for genome annotation. Genes essential for survival in the cold (e.g. HSPs) might be predicted. Lastly, comparative genomics of UWO241 with other model green algae will be applied to reveal how plastid-bearing psychrophiles function under harsh conditions. Thereafter, it might explain some unique traits of psychrophilic Chlamydomonas sp. UWO241 in extreme environment and shed light on the evolution and diversification of photosynthetic eukaryotes in permanently cold climate.

P27 Kurtis **Clarke**; Steven Ngo; Tomas DeFalco; Van Phan; Keiko Yoshioka
University of Toronto, Centre for the Analysis of Genome Evolution & Function; kurt.clarke@mail.utoronto.ca

A system for evaluating the roles of cyclic nucleotide gated channel proteins in cytosolic calcium changes using a fluorescent reporter

Calcium signalling regulates a wide variety of developmental, abiotic stress, and biotic stress responses. However, how plant cells create and decode stimulus specific calcium signals is still ambiguous. The creation of these calcium signals is dependent on calcium channel proteins such as the cyclic nucleotide gated ion channels (CNGCs). In Arabidopsis the CNGC family has 20 members and reverse genetics studies have revealed the influence of specific family members on pathogen and developmental responses, though the roles of many CNGC members during calcium signalling are still uncharacterized. In order to investigate the role of each CNGC member in stimulus specific calcium signalling we are generating CNGC knockout mutants that carry the fluorescence-based calcium ion sensitive reporter GCaMP3. Currently we have developed a fluorescence based plate reader analysis and time lapsed fluorescent imaging for moderate throughput screening of CNGC family members in a semi-quantitative way in response to various biotic and abiotic based stimuli.

P28 Angelica **Miraples**, W. Moeder, Keiko Yoshioka
University of Toronto, angelica.miraples@mail.utoronto.ca

Creating higher order cyclic nucleotide-gated ion channel mutants using CRISPR/Cas9

Despite the importance of Ca2+ signals, Ca2+ channels in plants are still not well understood. Cyclic nucleotide-gated channels (CNGCs), one of the largest cation channel families in plants, are involved in Ca2+ signaling. CNGCs have been shown to be involved in a diverse array of physiological processes in plants through their Ca2+ channel activity. So far, genetic approaches have not been able to reveal the specific role of most individual CNGCs probably due to redundancies in their function. We have shown roles for several clade I CNGCs in pathogen resistance, senescence and programmed cell death. However, most single mutant phenotypes are very subtle. Therefore, we decided to create higher order knockouts of the six members of CNGC clade I using a combination of T-DNA insertion lines and CRISPR/Cas9 technology. These mutants will be tested in conditions where they have been reported to be involved in, and where they show similar expression patterns. These conditions include salt and osmotic stress, heavy metal toxicity, and pathogen resistance.

P29 Devin **Noordermeer**, Vera Velasco, Ingo Ensminger
Department of Cell and Systems Biology, University of Toronto Mississauga; devin.noordermeer@utoronto.ca

Effects of warmer climate on the development of autumn cold hardiness in seedlings of coastal and interior Douglas-fir

Climate change is anticipated to pose a significant adaptational challenge to conifers due to their long generation times. Determining how warmer autumn temperatures will affect the development of winter cold hardiness is essential for improving our ability to select and breed conifers that will be best adapted to future climates. In this experiment, we subjected seedlings of four Douglas-fir provenances from contrasting British Columbia ecoregions (two coastal and two interior) to eight weeks of short photoperiod with normal autumn temperatures (historical range from 1961-1990) and warm autumn temperatures (projected range for 2085). Following this, we assessed chlorophyll fluorescence of Douglas-fir needles exposed to a range of freezing temperatures from 0 °C to -40 °C within an environmental test chamber. We intend to use the resulting FV/FM measurements to determine the temperature at which 50% of seedlings are damaged by freezing (LT50) and use this as a proxy for freezing tolerance. Preliminary data suggests that interspecific variation exists among the four Douglas-fir provenances, and that Douglas-fir from the interior provenances may be better adapted for cold hardiness in future climates.

P30 Luc A. **Ouellette**, Boris F. Mayer, Jean-Benoit Charron
Department of Plant Science, Macdonald Campus of McGill University; luc.ouellette@mail.mcgill.ca

Investigating the role of the DNA methyltransferase DOMAINS REARRANGED METHYLTRANSFERASE 2 (DRM2) in Brachypodium distachyon abiotic stress response

Abiotic stresses such as drought and salinity have negative impacts on the productivity of important cereal crops each year. Brachypodium distachyon is a model grass closely related to wheat and barley that has become an important tool for studying stress responses in such cereals. Plants respond to various stresses by altering the expression of different genes and increasing scientific evidence connects this regulation with epigenetic modifications. DNA methylation is a major epigenetic modification involved in development and stress response. Genome-wide DNA methylation patterns depend on both de novo methylation and maintenance methylation, as well as active demethylation. DOMAINS REARRANGED METHYLTRANSFERASE 2 (DRM2) is the primary de novo methyltransferase in Arabidopsis thaliana and it is involved in RNA-directed DNA methylation. An ortholog of DRM2 has been identified in B. distachyon, but its function in the context of stress response remains unclear. We report the stable transformation of four independent B. distachyon lines overexpressing BdDRM2 under the control of the maize ubiquitin 1 promoter and intron. Preliminary results show that BdDRM2 may also regulate DNA methylation in B. distachyon and play a role in its abiotic stress response. Further work will focus on determining the sites and patterns of DNA methylation involved in B. distachyon responses to abiotic stress.

P31 Marc **Possmayer** (1), Marina Cvetkovska (2), Nina Malczewski (1), Norman P.A. Hüner (1)
(1) Department of Biology and the Biotron Centre for Experimental Climate Change Research, University of Western Ontario, London, ON, Canada N6A 3K7; (2) Department of Biology, University of Ottawa, Ottawa, ON, Canada K1N 6N5; mpossma2@uwo.ca

Heat shock protein accumulation is uncoupled from heat stress in the Antarctic alga Chlamydomonas sp. UWO241

The accumulation of Heat Shock Proteins (HSPs) in response to high temperature stress has been described across all taxa, and serves to prevent heat-induced protein denaturation and aggregation. Chlamydomonas sp. UWO241 is a green alga isolated from the perennially ice-covered Antarctic Lake Bonney, which is characterized by stable low temperatures (~5°C). UWO241 is an obligate psychrophile, unable to grow >18°C. Despite its adaptation to the cold, we detected an expanded HSP gene family in the UWO241 genome, as compared to the model mesophile Chlamydomonas reinhardtii. UWO241 grown at 4-15°C constitutively expresses high transcript and protein levels of major HSPs, in relative to C. reinhardtii grown at 22-37°C. Finally, we demonstrate significant differences in how these organisms respond to temperature stress. When exposed to non-permissive temperature (42°C) HSP accumulation is rapidly induced in C. reinhardtii. In contrast, HSP levels in UWO241 do not increase upon exposure to non-permissive growth temperature (24°C). We conclude that despite its evolution at constant low temperature, UWO241 has retained the HSP gene family; however, the heat shock response in UWO241 is not dependent on HSP accumulation.

P32 Beth **Szyszka-Mroz**, Marina Cvetkovska, Alexander G. Ivanov, David R. Smith, Marc Possmayer, Denis P. Maxwell, Norman P.A. Hüner

Biology Department and the Biotron Centre for Experimental Climate Change Research, University of Western Ontario, London, Canada N6A 5B7; bszyszkamroz@gmail.com

Protein kinases of the Antarctic polyextremophile, Chlamydomonas sp. UWO241, confer a distinctive thylakoid protein phosphorylation pattern associated with a remodelling of thylakoid membrane architecture and light energy distribution between photosystem I and II

Light-dependent [?-33P]ATP labelling of thylakoid membranes from the Antarctic polyextremophile, Chlamydomonas sp. UWO241, exhibited a distinct low temperature-dependent phosphorylation pattern, compared to C. reinhardtii, characterized by low levels of LHCII labelling and minimal phosphorylation of Lhcb4 and Lhcb5 as well as the PSII core polypeptides despite comparable levels of the Stt7protein kinase. The sequence and putative structure of the UWO241 Stt7 kinase domain exhibited significant alterations which we suggest predisposes it to be more active at low temperature and less sensitive to the protein kinase inhibitor staurosporine, compared to C. reinhardtii. Comparative chloroplast ultrastructure, purification of PSII and PSI pigment-protein complexes combined with digitonin fractionation of thylakoid membranes indicated that adaptation of UWO241 to its cold, high-salt environment altered its thylakoid membrane architecture and reorganized the distribution of PSI and PSII compared to C. reinhardtii. Growth under high salt/ low temperature stimulated PSI cyclic electron flow by 50% compared to growth at low salt/low temperature. The deconvolution of 77K fluorescence emission spectra of intact UWO241 cells allowed quantification of changes in energy distribution between PSII and PSI that was sensitive to the redox state of the PQ pool, to the NaCl concentrations of the growth medium and to external [Mg2+]of isolated thylakoids. We conclude that

light energy distribution in UWO241 is regulated primarily by cation-induced energy spillover which stimulates PSI cyclic electron flow rather than by the modulation of PSI antenna size by LHCII-dependent phosphorylation as a consequence of adaptation to its extreme Antarctic environment.

P33 Danielle **Williams**, Vince Qu, Vincenzo De Luca
Brock University; dw15qi@brocku.ca

***Catharanthus roseus* monoterpenoid indole alkaloid biosynthesis: from intact plants to the bioreactor**

The basic pathways involved in the assembly of monoterpene indole alkaloids (MIAs) from geraniol and tryptophan have recently been characterized at the molecular and biochemical level in the medicinal plant, *Catharanthus roseus*. Synthetic biology experiments are now making it possible for transferring these pathways to other organisms to produce drugs used for the treatment of various human diseases. The discovery of over 30 genes involved in the assembly of the dimeric anticancer MIAs, vinblastine and vincristine, is speeding up characterization of many new genes responsible for the decoration of various biologically active MIA ring structures found in nature. Recently, the solution of the multistep pathway from 19*E*-geissoschizine to tabersonine was published (PNAS 2018, 115(12):3180-3185) and previously the seven-step synthesis of the valuable aerial-specific MIA vindoline from the aspidosperma MIA tabersonine was accomplished in yeast (PNAS 2015, 112(19):6224). The aspidosperma alkaloids are the most prevalent root-specific alkaloids, and the biosynthetic pathway to form echitovenine remains unsolved. The present study used a bioinformatic approach to identify new candidate genes that may be involved in the assembly of MIAs in *C. roseus* roots that accumulate several aspidosperma-type MIAs, including minovincinine, echitovenine, lochnericine, and hörhammericine. To form these abundant derivatives, several oxidizations/substitutions of the aspidosperma backbone are required. The search for enzymes homologous to leaf-specific cytochrome P450s uncovered a novel root-specific monooxygenase required for the formation of echitovenine from vincadifformine. The biochemical characterization of this enzyme in yeast and its expression properties in *C. roseus* will be discussed.

P34 Kayla Riane **Dias**, David Kamelchuk, Barb Thomas, Nathalie Isabel, Katharina Bräutigam
Cell and Systems Biology, University of Toronto, Toronto, ON, Canada; Department of Renewable Resources, University of Alberta, Edmonton, AB, Canada; Natural Resources Canada, Canadian Forest Service, Laurentian Forestry Centre; Department of Biology, University of Toronto Mississauga, Mississauga, ON, Canada; kayla.dias@mail.utoronto.ca

Integrated analysis of stress and recovery responses in poplar

Efficient responses to environmental change are particulary important for long-lived plants such as trees. Here, we investigate how Populus balsamifera responds to two related key abiotic stresses - osmotic and ionic stress – during acute perturbation and recovery. For an integrated understanding, responses are studied at the physiological, molecular and epigenetic level. Based on preliminary screening, one genotype (AP4326) was investigated in detail during four weeks of water withholding and three weeks of NaCl exposure (two stress levels, 100 and 150 mM), followed by an extended recovery period of 50 days. All stressed individuals exhibited growth retardation and minimal stomatal conductance. While the osmotic stress treatment group showed an acclimation response and resumed normal growth rates and stomatal conductance post stress, trees exposed to severe ionic stress had

only a 50% survival rate; the moderate ionic stress treatment were slow to recover. Elemental analyses provide insights into precise soil conditions experienced under ionic stress and ongoing work focuses on molecular mechanisms, such as DNA methylation patterns and gene expression profiles, that govern physiological responses. Findings from this work will enhance our understanding plasticity and survival in trees and might be useful for informed planning of plantations or reclamation efforts.

CONFERENCE PARTICIPANTS

Last Name	First Name	Company/Org	Email
Amyot	Lisa	Agriculture and Agri-Food Canada	Lisa.Amyot@canada.ca
Ananvoranich	Sirinart	University of Windsor	anans@uwindsor.ca
Austin	Ryan	Agriculture & Agri-Food Canada	ryan.austin@agr.gc.ca
Beltran Schiavi	Nathieli	Dalhousie University	nathielischiavi@gmail.com
Bennett	Kristyn	The University of Western Ontario	kbenne44@uwo.ca
Bernards	Mark	The University of Western Ontario	bernards@uwo.ca
Boersma	Paul	The University of Western Ontario	pboersm@uwo.ca
Bonea	Diana	University of Toronto	diana.bonea@mail.utoronto.ca
Bozzo	Gale	University of Guelph	gbozzo@uoguelph.ca
Braeutigam	Katharina	University of Toronto Mississauga	katharina.braeutigam@utoronto.ca
Cameron	Robin	McMaster University	rcamero@mcmaster.ca
Carianopol	Carina	University of Toronto	carina.carianopol@mail.utoronto.ca
Castillo	Y Indira Q	The University of Western Ontario	yqueralt@uwo.ca
Chen	Ling	Agriculture and Agri-Food Canada	ling.chen@canada.ca
Clarke	Kurtis	University of Toronto	kurt.clarke@mail.utoronto.ca
Clayton	Emily	The University of Western Ontario	eclayto3@uwo.ca
Colasanti	Joseph	University of Guelph	jcolasan@uoguelph.ca
Costain	Cecily	University of Guelph	ccostain@uoguelph.ca
Cox	Laura	University of Guelph	lcox05@uoguelph.ca
Cvetkovska	Marina	The University of Western Ontario	mcvetkov@uottawa.ca
Dhaubhadel	Sangeeta	Agriculture and Agri-Food Canada	sangeeta.dhaubhadel@canada.ca
Dias	Kayla	University of Toronto	kayla.dias@mail.utoronto.ca
Dimopoulos	Nicolas	University of Toronto	nicolas.dimopoulos@mail.utoronto.ca
D'Odorico	Petra	University of Toronto Mississauga	petra.dodorico@utoronto.ca
Doner	Nathan	University of Guelph	donern@uoguelph.ca
Duarte	André	The University of Western Ontario	aduarte4@uwo.ca
Dusenge	Mirindi Eric	The University of Western Ontario	mdusenge@uwo.ca
Ensminger	Ingo	University of Toronto	ingo.ensminger@utoronto.ca
Erland	Lauren	University of Guelph	lerland@uoguelph.ca
Feyissa	Biruk A	The University of Western Ontario	bfeyissa@uwo.ca
Flude	Corey	University of Guelph	cflude@uoguelph.ca
Frank	Joshua	The University of Western Ontario	jfrank22@uwo.ca
Fufeng	Angela	McMaster University	fufengab@mcmaster.ca
Garant	Timothy	Carleton University	timgarant@cmail.carleton.ca
Gazzarrini	Sonia	University of Toronto	gazzarrini@utsc.utoronto.ca
Gidda	Satinder	University of Guelph	sgidda@uoguelph.ca
Gonzales-Vigil	Eliana	University of Toronto Scarborough	e.gonzalesvigil@utoronto.ca
Goring	Daphne	University of Toronto	d.goring@utoronto.ca
Gritsunov	Artyom	University of Toronto	artyom.gritsunov@mail.utoronto.ca
Halim	Abdul	McMaster University	halima4@mcmaster.ca
Hanly	Alexandria	The University of Western Ontario	ahanly2@uwo.ca

Hannoufa	Abdelali	Agriculture and Agri-Food Canada	Abdelali.Hannoufa@agr.gc.ca
Hargreaves	Max	University of Guelph	whargrea@uoguelph.ca
Hepworth	Shelley	Carleton University	shelley.hepworth@carleton.ca
Huner	Norm	University of Western Ontario	nhuner@uwo.ca
Irani	Solmaz	McMaster University	iranis4@mcmaster.ca
Islam	Nishat	The University of Western Ontario	nislam33@uwo.ca
Kaberi	Karina	The University of Western Ontario	kkaberi@uwo.ca
Kaplanoglu	Emine	Agriculture and Agri-Food Canada	emine.kaplanoglu@canada.ca
Kanaris	Michael	University of Toronto	michael.kanaris@mail.utoronto.ca
Karia	Purva	University of Toronto	purva.karia@mail.utoronto.ca
Kazemi	Leily	The University of Western Ontario	lkazemi@uwo.ca
Kohalmi	Susanne	The University of Western Ontario	skohalmi@uwo.ca
Krolikowski	Sophie	Agriculture & Agri-Food Canada	sophie.krolikowski@canada.ca
Laflamme	Bradley	University of Toronto	bradley.laflamme@mail.utoronto.ca
Lambert	Megan	The University of Western Ontario	mlambe9@uwo.ca
Lau	Vincent	University of Toronto	vincente.lau@mail.utoronto.ca
Lee	Jonathan	University of Toronto	jonathans.lee@mail.utoronto.ca
Lee	Hyun Kyung	University of Toronto	hyunkyung.lee@mail.utoronto.ca
Lemon	Karen	The University of Western Ontario	klemon9@uwo.ca
Lesy	Victoria	University of Guelph	vlesy@uoguelph.ca
Liu	Jun (Kevin)	The University of Western Ontario	jliu924@uwo.ca
Louati	Marwa	University of Guelph	mlouati@uoguelph.ca
Lu	Mimmie	Agriculture and Agri-Food Canada	mimmie.lu@canada.ca
Ludba	Kaitlyn	Agriculture and Agri-food Canada	kludba@uwo.ca
Macfie	Sheila	The University of Western Ontario	smacfie@uwo.ca
Macgregor	Stuart	University of Toronto	s.macgregor@mail.utoronto.ca
MacNeill	Greg	University of Guelph	macneilg@uoguelph.ca
Mayer	Boris	McGill University	boris.mayer@mail.mcgill.ca
McDonald	Allison	Wilfrid Laurier University	amcdonald@wlu.ca
McDonald	Sarah	The University of Western Ontario	smcdon83@uwo.ca
Menassa	Rima	Agriculture and Agri-Food Canada	rima.menassa@canada.ca
Minow	Mark	University of Guelph	mminow@uoguelph.ca
Miraples	Angelica	University of Toronto	angelica.miraples@mail.utoronto.ca
Moeder	Wolfgang	University of Toronto	wolfgang.moeder@utoronto.ca
Moffatt	Barb	University of Waterloo	moffatt@uwaterloo.ca
Mohamed	Deka	University of Toronto	deka.mohamed@mail.utoronto.ca
Mott	Adam	University of Toronto Scarborough	adam.mott@utoronto.ca
Mu	Bona	University of Toronto Scarborough	bona.mu@mail.utoronto.ca
Mullen	Robert	University of Guelph	rtmullen@uoguelph.ca
Nasrollahi	Vida	Agriculture and Agri-Food Canada	vnasroll@uwo.ca
Nordermeer	Devin	University of Toronto Mississauga	devin.noordermeer@utoronto.ca
Nunn	Garrett	McMaster University	nunngm@mcmaster.ca

Ouellette	Luc	McGill	luc.ouellette@mail.mcgill.ca
Poirier	Mackenzie	University of Waterloo	mcpoirier@edu.uwaterloo.ca
Possmayer	Marc	The University of Western Ontario	mpossma2@uwo.ca
Provart	Nicholas	University of Toronto	nicholas.provart@utoronto.ca
Radford	Julianne	The University of Western Ontario	jradfor8@uwo.ca
Rowland	Owen	Carleton University	owen.rowland@carleton.ca
Ryser	Peter	Laurentian University	pryser@laurentian.ca
Saha	Repon	The University of Western Ontario	rks7@uwo.ca
Summers	Peter	McMaster University	summers@mcmaster.ca
Szyszka	Beth	The University of Western Ontario	bszyszkamroz@gmail.com
Tahir	Muhammad	The University of Western Ontario	mtahir25@uwo.ca
Torrez	Alberto	The University of Western Ontario	atorrez@uwo.ca
Traa	Annika	University of Waterloo	atraa@edu.uwaterloo.ca
Tremblay	Benjamin	University of Waterloo	b2trembl@uwaterloo.ca
Tully	Trish	The University of Western Ontario	ttully@uwo.ca
Unterlander	Nicole	University of Guelph	nunterla@uoguelph.ca
Van Brenk	Jordan	The University of Western Ontario	jvanbre@uwo.ca
Van Gelder	Kristen	University of Guelph	kvangeld@uoguelph.ca
Vaziriyeganeh	Maryamsadat	University of Alberta	vaziriye@ualberta.ca
Velasco	Vera	University of Toronto	Vera.velasco@utoronto.ca
Virta	Lilia	University of Guelph	lvirta@uoguelph.ca
Vonapartis	Eliana	University of Toronto	eliana.vonapartis@mail.utoronto.ca
Way	Danielle	The University of Western Ontario	dway4@uwo.ca
Weretilnyk	Elizabeth	McMaster University	weretil@mcmaster.ca
Widemann	Emilie	The University of Western Ontario	ewidema4@uwo.ca
Williams	Danielle	Brock University	dw15qi@brocku.ca
Wu	Jian	University of Toronto	wujian.wu@mail.utoronto.ca
Xiong	Kevin	Carleton University	kevinxiong@cmail.carleton.ca
Yim	Winfield	University of Toronto	winfield.yim@mail.utoronto.ca
Zhang	Xi	The University of Western Ontario	xzha25@uwo.ca