

9th ANNUAL

BIOLOGY GRADUATE RESEARCH FORUM



PRESENTED BY THE DEPARTMENT OF BIOLOGY

THURSDAY OCTOBER 18TH – FRIDAY OCTOBER 19TH 2018

WESTERN UNIVERSITY

Biology Graduate Research Forum 2018 Agenda

Thursday October 18th

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|--------------|---|
| 5:00–5:30 PM | Pre-Lecture Reception – P&AB Atrium |
| 5:30–5:45 PM | Opening Remarks – P&AB 148 |
| 5:45–7:00 PM | David Laudenbach Memorial Lecture – P&AB 148 |
| 7:00–8:00 PM | Post-Lecture Reception – Grad Club |

Friday October 19th

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|-------------------|--|
| 8:00–8:30 AM | Registration/Poster Setup/Load Standard Talks – IGAB Atrium |
| 8:30–9:30 AM | Standard Talks
Concurrent Sessions – UCC 54A/UCC 54B |
| 9:30–10:00 AM | Coffee Break – UCC 54 |
| 10:00–11:00 AM | Standard Talks
Concurrent Sessions – UCC 54A/UCC 54B |
| 11:00 AM–12:00 PM | Poster Judging and Viewing – IGAB Atrium |
| 12:00–1:00 PM | Lunch and Poster Viewing – IGAB Atrium |
| 1:00–2:00 PM | Career Panel – IGAB Atrium |
| 2:00–3:00 PM | Coffee Break/Load Lightning Talks – IGAB Atrium |
| 3:00–4:00 PM | Lightning Talks
Concurrent Sessions – AHB 1B02/AHB 1B04 |
| 4:00–5:00 PM | Awards & Closing Remarks – AHB 1B04 |
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Biology Graduate Research Forum 2018

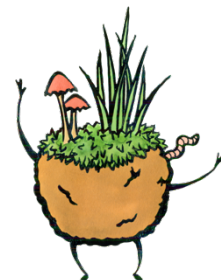
Schedule of Talks

UCC 54A Standard Talks – Concurrent Session A

8:30–8:45 AM	Aida Parvizi
8:45–9:00 AM	Andrea Boyer
9:00–9:15 AM	Claire Bottini
9:15–9:30 AM	Caitlyn Lyons

9:30–10:00 AM Coffee Break – UCC 54

10:00–10:15 AM	Matthew Meehan
10:15–10:30 AM	Carlie Muir
10:30–10:45 AM	Anna Chernyshova
10:45–11:00 AM	Olivia Colling



UCC 54B Standard Talks – Concurrent Session B

8:30–8:45 AM	Bradley Bork
8:45–9:00 AM	Emily Clayton
9:00–9:15 AM	Xi Zhang

9:15–9:30 AM

Danielle Spice

9:30–10:00 AM

Coffee Break – UCC 54

10:00–10:15 AM

Alex Kozlov

10:15–10:30 AM

Alannah Mattice

10:30–10:45 AM

Kelsey Pannunzio

10:45–11:00 AM

Wes Robinson



AHB 1B02

Lightning Talks – Concurrent session A

3:00–3:08 PM

Zoran Culo

3:08–3:16 PM

Ryley Yost

3:16–3:24 PM

Joshua Isaacson

3:24–3:32 PM

Jordan Kustec

3:32–3:40 PM

Hailie Pavanel

AHB 1B04

Lightning Talks – Concurrent session B

3:00–3:08 PM

Bonnie Alberry



3:08–3:16 PM

Jennifer Blythe

3:16–3:24 PM

Muhammad Tahir

3:24–3:32 PM

Alberto Torrez

3:32–3:40 PM

Vonica Flear

3:40–3:48 PM

Asad Lone



Oral Presentations – Standard Talks

Can spermatophore isotope composition indicate natal origin of males in the migratory true armyworm moth (*Mythimna unipuncta*)

Aida Parvizi, MSc Student

Many insect species migrate in response to habitat deterioration and we typically have little understanding of their geographic origin. A proof of principle study confirmed that deuterium (natural stable isotope of hydrogen, $\delta^2\text{H}$) levels in the wings of adults, could be used to assign the probability of origin of the migratory agricultural pest *Mythimna unipuncta*. Immigrant females may mate up to 7 times during their life. We asked if the deuterium values of the chitinous spermatophores could provide information on the natal origin of her mates. While wing deuterium values are determined uniquely by larval food sources, those of spermatophores could also be affected by nectar sources exploited by adults. Comparison of spermatophores from males reared on either control or deuterium spiked diets (both as larvae and adults) confirmed significant differences in $\delta^2\text{H}$ are detectable. Furthermore, mating control males with spiked females and vice versa, showed that the female contributes about 10% to the hydrogen content of a spermatophore. We mated field collected males with lab colony females and compared the $\delta^2\text{H}$ profile of the spermatophore with the male's wing $\delta^2\text{H}$ value. In most cases the two values were similar but in some there were significant differences. The results suggest that spermatophore $\delta^2\text{H}$ value could be a reliable source in the study on migration and nutrient allocation. However, as adult feeding can influence spermatophore $\delta^2\text{H}$ value, we are currently conducting additional experiments to determine how sustained migratory flight affects the turnover rate of deuterium obtained from larval and adult diets.

Differential seasonal effects of temperature on nocturnal migratory restlessness in white-throated sparrows

Andrea Boyer, PhD Student

Weather-related cues can influence the timing of a migratory flight along stopover sites, including temperature, and wind speed and direction. Weather conditions differ across seasons for many songbirds, thus cues in autumn influence birds differently than the same cues in spring. Food availability can also influence the timing of migration. It is difficult to determine which weather cue has a significant impact on the migratory behaviour of birds in the wild, thus we experimentally tested the specific impact of three temperature treatments and two food treatments on migratory restlessness behaviour in captive white-throated sparrows (*Zonotrichia albicollis*) during autumn and spring. Birds were randomly placed on either restricted or unlimited food diets. We manipulated temperature each night to one of three conditions: 4 °C, 14 °C or 24 °C to represent cold, average, and warm nights, respectively. Birds were video recorded each night using infrared illuminators and migratory restlessness was tracked using Noldus

EthoVision. Results are still pending, but we predict that birds will have opposite responses to temperature manipulations between seasons. In autumn, the amount of migratory restlessness exhibited should increase with decreasing temperature as birds will be motivated to reach wintering grounds with higher food availability. Birds receiving ad libitum food should exhibit less migratory restlessness, even during colder temperatures. In spring, birds should increase their migratory restlessness with warmer temperatures as they are encouraged to reach their breeding grounds. Birds receiving a restricted amount of food should exhibit an increased rate of migratory restlessness, even during warmer temperatures.

Evaluation of mercury and stress exposure impacts on songbirds' moult

Claire Bottini, PhD Student

In their natural habitat, wildlife faces multiple stressors (e.g., predation, food unpredictability, climate change) in addition to pollutants burden. While growing evidence suggests that stress can amplify the impact of pollutants, toxicant exposure might alter the ability to cope with stressful situations. Such co-occurring sources of pressures brings high risks for birds as both stress and mercury exposure can alter their endocrine system, disrupting their thyroid hormones levels, which may affect their feather moult, and thus impacting their thermoregulation, migration capacity, and fitness. This project aims to determine the effects of concurrent stress and mercury exposure on adult birds' moult. Song sparrows (*Melospiza melodia*) were captured and assigned to one of four environmentally relevant treatments: control, stressed only, mercury exposed only, and both stress with mercury exposure. The exposure lasted 3 months with an additional 3 months of post-exposure monitoring where the feather moult of each bird was scored every week. The preliminary results of this study show a tendency for the birds exposed to mercury to moult sooner than the non-exposed individuals, while stress doesn't seem to affect moult. This impact on moult may be due to an action of mercury on thyroid hormones. Advanced moult may benefit the birds by allowing depuration of mercury into feather tissue. However, thyroid hormones also regulate numerous other functions such as metabolism, neuronal function, and migratory behaviour. Thus, even at low doses, mercury exposure may induce trade-offs between the timing of moult and other physiological functions.

Plant community shifts in Boreal peatlands after two years of passive warming

Caitlyn Lyons, MSc Student

Boreal peatlands are important global carbon sequesters due to the accumulation of peat (partially decomposed plant material). Therefore, the aboveground plants are important determinants for carbon storage potential. Boreal peatlands are mainly dominated by three different plant functional groups: Sphagnum mosses, Carex sedges, and shrubs. Sphagnum mosses comprise a vast majority of peat, meaning Sphagnum spp. contribute the most to carbon storage potential of Boreal peatlands. I am

performing a field experiment in northern Ontario looking at the effects of passive warming on peatland plant communities in two different types of peatlands, a Sphagnum dominated and Carex dominated peatland. Under climate warming, a shift from Sphagnum mosses to Carex sedges in peatlands is expected. I have two field seasons of plant community composition data collected using the point intercept method under passive warming and control treatments. I have also measured leaf area index (LAI) as an indicator of aboveground biomass. After only one growing season there were detectable changes in LAI under passive warming where the Carex dominated peatland demonstrated larger LAI in the warming treatments. In the Carex dominated peatland there was also a significant shift in plant communities under passive warming, demonstrating an increase in shrub dominance. In the Sphagnum dominated peatland there are trends depicting seasonal shifts in plant communities. Climate warming has the potential to impact the plant community composition at these two Boreal peatland sites and this could shift these sites from carbon sinks to carbon sources, exacerbating climate warming effects.

Warming rewires trophic linkages in belowground forest communities

Matthew Meehan, PhD Student

IPCC predicts temperatures to increase 2-8C globally by 2100, shifting environments into new stable states, and correspondingly changing food web structure. Warming has been predominantly studied in aquatic and aboveground food webs, while soil food webs remain understudied. Yet, they perform several ecological processes, including decomposition, and the cycling of nutrients, in addition to being functionally diverse. We studied the effect of warming on soil predators (Mesostigmata), prey (Collembola, Prostigmata and Oribatida) and basal resources (moisture, proxy for microbial communities), to determine how warming affects faunal composition and soil trophic dynamics. Soil samples were collected from warmed (+4C above ambient temperature) and ambient soil in 2015-2017, in the boreal forest in Lac Simoncouche, Quebec. Once collected, prey and predator groups were extracted, counted, and identified to morphospecies (excluding Prostigmata). We examined the effect of warming on community composition of soil fauna using PERMANOVA. In addition, we analyzed changes in trophic dynamics under warming using a structural equation model (SEM), with three trophic levels: resource (moisture), consumer (prey groups) and predator (arthropod-feeding Mesostigmata). We found that warming altered the community composition of Collembola alone. However, soil food webs were rewired, as basal resources were uncoupled from prey groups, and potentially, prey switching occurred amongst predators under warming. Although faunal composition did not change, trophic links were desynchronized, representing a new stable state. The implications of this are unknown; however, changes in ecosystem-level processes should be expected.



Elevated rearing temperature alters cardiac function and thermal performance in juvenile Atlantic salmon

Carlie Muir, PhD Student

Elevated water temperatures impose increased oxygen demands on the cardiorespiratory system of salmonids, and these demands are primarily met via increased heart rates. When temperature exceeds the upper limit of a fish's thermal window, it results in cardiac failure, and ultimately, death. This leaves salmon very vulnerable to high environmental temperatures. Previous work suggests that phenotypic plasticity in cardiac development helps shape thermal performance in fish, such that the heart develops to function optimally at temperatures reflecting those experienced during embryogenesis. In this study, Atlantic salmon were raised from fertilization in present-day (+0°C) or projected future (+4°C) temperature conditions. At the parr stage, we measured maximum heart rate and atrioventricular blood flow velocity at increasing temperatures using a Doppler flow velocity system. We found that higher temperatures during development led to higher optimal temperatures, as calculated by both heart rate and cardiac output. In addition, higher developmental temperatures led to higher maximum heart rates. Increased developmental temperatures were also associated with higher temperatures at which cardiac arrhythmias began, signifying the upper thermal limit for cardiac function. These results support the hypothesis that the thermal environment experienced during development can alter thermal performance later in life, possibly through plasticity in cardiac development.

Molecular signatures of kin selection: Are sterile caste-associated genes nearly neutral?

Anna Chernyshova, MSc Student

In termite societies, sexual kings and queens are highly specialized for reproduction, while more-or-less sterile workers and soldiers perform non-reproductive roles that are associated with colony growth, colony maintenance and defence. Workers and soldiers can therefore be considered reproductively altruistic because they labour to help produce large numbers of non-descendent kin (i.e., siblings, half-siblings, etc.) at the expense of their own direct fitness. The evolution of altruism, in any taxon, is therefore intriguing because in theory it requires 'genes for altruism' to evolve indirectly via selection on reproducing relatives, who carry, but do not express these genes. Therefore, the notion of indirect selection is fundamental to our understanding of social evolution, yet surprisingly, we know little about how real genes might respond to this type of selection within living populations. The 'nearly neutral' hypothesis predicts that genes indirectly selected for subfertility may experience relaxed adaptive molecular evolution, relative to genes directly selected for reproduction. If so, I expect that the ratio of non-synonymous to synonymous substitutions will tend towards a neutral value of '1', relative to loci under direct selection for which this ratio will deviate from neutrality in either a purifying or positive direction. Here, I exploit the newly available RNA sequence dataset from eastern subterranean termite to test key predictions of the nearly neutral hypothesis.

Differential vulnerability to window collision mortality among songbird species

Olivia Colling, MSc Student

It is estimated that billions of birds worldwide die each year from window collisions. Birds appear to behave as if windows are invisible, suggesting they cannot distinguish between glass and open space. The objective of my M.Sc. research is to determine if some bird species are more vulnerable to dying from window collisions than others. Using a combination of citizen science and bird monitoring data from the Fatal Light Awareness Program (FLAP) and regional bird banding stations, I will determine if the mortality seen in downtown Toronto is simply a function of species abundance or if it is unequal among species. In addition to among species differences in mortality, I will compare individuals within species to address if age is a factor in collision mortality vulnerability. To determine this, I will use the FLAP 2017 and 2018 bird collections to identify individuals by species and age. This information will be used to compare age ratios.

MT1-MMP mediated cell signalling in MCF-7 cells: more than just an enzyme

Bradley Bork, MSc Student

Membrane-Type 1 Matrix Metalloproteinase (MT1-MMP) is a multifunctional, membrane-bound protease involved in cell movement. Because it can degrade the extracellular matrix (ECM) and activate secreted matrix metalloproteinases (MMPs), unregulated expression of MT1-MMP has been implicated with increased cellular migration and invasion in cancerous tissue. Apart from its proteolytic activity, recent studies have shown that signalling pathways facilitated by MT1-MMP may have an equally important role in regulating cell migration, invasion, and viability. However, little is known about which proteins MT1-MMP binds to trigger such signalling pathways, especially in terms of cytoplasmic interactions. MCF-7 breast cancer cells that stably overexpress MT1-MMP or express MT1-MMP with a mutated cytoplasmic domain have been used to determine the signal transduction capabilities of MT1-MMP protein complexes. Cells that express MT1-MMP lacking its cytoplasmic domain have reduced expression of TGFB1, but not its receptor TGFB1R. The interaction between TGFB1 and MT1-MMP has been well-studied - TGF β 1 is released from its latent binding proteins via extracellular cleavage by MT1-MMP. However, it appears that the non-proteolytic cytoplasmic domain of MT1-MMP is also involved in TGF β regulation. MT1-MMP cytoplasmic binding partners potentially involved in this regulation will be isolated and identified using co-immunoprecipitation and mass spectrometry. Equally important signalling events involving MT1-MMP and its signal transduction ability may change how we perceive cell migration and invasion.

Is piggybacking required for AtADT5 nuclear localization?

Emily Clayton, PhD Student

In *Arabidopsis thaliana*, a family of six enzymes called AROGENATE DEHYDRATASES (ADTs) catalyze the last step of Phe biosynthesis, and all six *AtADT*s localize to the chloroplast. Interestingly, *AtADT5* is the only *AtADT* that also localizes to the nucleus. However, no nuclear localization signals (NLSs) were identified in the *AtADT5* sequence. Recently, we discovered an interactor specific to *AtADT5* that has two putative NLS sequences. We believe this interactor “piggybacks” *AtADT5* into the nucleus. I am interested in determining the sequences responsible for the specificity of this interaction. *AtADT4* is the most closely related to *AtADT5*, sharing 90% sequence identity, and yet it does not enter the nucleus. Comparisons of *AtADT4* and *AtADT5* sequences revealed a unique motif in the C-terminal ACT domain of *AtADT5*. To test if this motif is responsible for the interaction of the candidate interactor, yeast-two-hybrid and BiFC analyses will be performed using a series of domain swapped constructs with or without the *AtADT5* ACT domain. In addition, constructs containing targeted mutations of this motif will be generated for both *AtADT4* and *AtADT5* and tested for interaction with the candidate interactor. As plant ADT expression is often regulated by stress, understanding the mechanism of *AtADT5* nuclear localization will help to elucidate its role in the nucleus as a possible transcriptional regulator. This work will provide insight on how seemingly inconsequential differences in sequence can have striking effects on protein localization and function.

Sequencing and assembling the nuclear genome of the Antarctic psychrophilic green alga *Chlamydomonas* sp. UWO241: unravelling the evolution of cold adaptation

Xi Zhang, PhD Student

The Antarctic harbours a variety of algae that can withstand extreme cold but falter at warmer temperatures (psychrophiles), including the unicellular green alga *Chlamydomonas* sp. UWO241. Little is known, however, about the origins and evolution of psychrophilic algae, and their nuclear genomes remain largely unexplored. For my PhD, I propose to sequence and assemble the entire nuclear DNA of UWO241 using a next-generation sequencing (NGS) approach, and then employ these data to better understand the evolution of photopsychrophily. DNA sequencing technologies have undergone tremendous advancements in recent years, but assembling, annotating, and analyzing a nuclear genome is still a huge undertaking, especially for small laboratory groups, partly because many eukaryotic genomes are repeat rich and contain thousands of genes and introns. To characterize the UWO241 genome I will, firstly, develop an assembly pipeline for processing high-throughput DNA sequencing reads into genomic contigs. These contigs, alongside RNA-sequencing data, will then be fed into an annotation pipeline, which I will design based on the most up-to-date eukaryotic bioinformatics gene-profiling software. Computational analyses will be carried out on an in-house computer, which I have constructed for the Smith Lab, as well as on Western’s supercomputing network SHARCNET. Lastly, I will perform a wide range comparative

genomic analyses of the UWO241 genome with those of other model green algae, including *Chlamydomonas reinhardtii*. Preliminary data already suggest that the UWO241 genome is exceptional in many ways—including its size (>230 Mb) and gene copy number—and at least some of these features appear to have a fundamental role in surviving in the extreme cold.

CRISPR/Cas9 knockout of *Sufu* attenuates neuronal differentiation of P19 embryonal carcinoma cells

Danielle Spice, PhD Student

Hedgehog (Hh) signaling is vital in many stage of neural development. A major negative regulator of Hh signaling is Suppressor of Fused (SUFU), which acts to sequester the full length Gli transcription factor in the cytoplasm or facilitates its conversion to a repressive form. The P19 embryonal carcinoma cell line is a model of hind-brain neuronal differentiation and the involvement of Hh signaling, in particular the role of SUFU in this process has yet to be explored. We hypothesize that SUFU is required for P19 neuronal differentiation to occur, and that genetically ablating it will result in a loss of differentiation potential. Throughout normal, retinoic acid (RA) induced, differentiation of P19 cells the abundance of SUFU mRNA levels show modest changes, although protein abundance does not change. We used CRISPR/Cas9 to genetically ablate the *Sufu* gene. Ablation of *Sufu* attenuated neuronal differentiation, without changing the pluripotency or proliferation of untreated *Sufu*^{-/-} cells. This result is likely due to Hh signaling being inhibited during days 4-10 of RA induced differentiation, as seen by increased Gli repressor over those days. Together these results show that although active Hh signaling is required in the early stages of differentiation, its major negative regulator SUFU is required in order for proper neuronal differentiation to occur.

Lactate-mediated metabolic reprogramming in human dermal fibroblast cells

Alex Kozlov, PhD Student

Increasing evidence implicates the cellular metabolic state in the establishment of pluripotency. During the reprogramming of somatic cells to induced pluripotent stem cells (iPSCs), a 'metabolic switch' occurs whereby cell metabolism changes from oxidative phosphorylation (OXPHOS) to glycolysis. Small molecule activation of OXPHOS or glycolysis before or after the metabolic switch respectively enhances reprogramming efficiency. However, chemical-induced alterations in metabolism may compromise iPSC integrity. We hypothesized that culturing human dermal fibroblasts (hDFs) in various sole metabolite fuel sources would promote a shift in metabolism from OXPHOS towards a glycolytic or bivalent state. We discovered that culturing hDFs in media containing glucose as a fuel source promoted increased phosphorylation of the mitochondrial enzyme PDH; indicative of increased glycolysis. In contrast, fibroblasts cultured in either lactate or pyruvate-media exhibited decreased phosphorylation of PDH, reflective of elevated OXPHOS. Indeed, cells grown in pyruvate-media displayed high maximal respiration, spare respiratory capacity and low glycolytic reserve,

indicative of oxidative metabolism. Conversely, cells cultured in lactate-media exhibited no spare respiratory capacity and high glycolytic reserve, suggesting lactate-treatment induces bivalent metabolism. Interestingly, fibroblasts first cultured in lactate-media, then switched to glucose-media became more glycolytic than fibroblasts solely cultured in glucose-media. Our data implies that lactate-treatment primes hDFs to switch from OXPHOS to glycolysis. We are currently examining if lactate-induced stabilization of the glycolytic transcription factor HIF-1 contributes to this metabolic switch. Our findings indicate that sole source fuel treatment alters the metabolic state of somatic cells, which may make them more amenable to reprogramming for iPSC generation.

Genetic basis of hybrid sterility in *Drosophila pseudoobscura* and *D. persimilis*

Alannah Mattice, MSc Student

Hybrid sterility is a reproductive barrier that can prevent gene flow between species. The genetic basis of this common reproductive barrier, however, remains poorly understood. *Drosophila pseudoobscura* and *D. persimilis* are able to form sterile male and fertile female hybrids. These two species also produce heteromorphic sperm: sperm which can vary in shape and size within one ejaculate. F1 hybrids of these species produce needle-eye shaped sperm that appears to have failed to separate during spermatogenesis. This sterile needle-eye phenotype can be tracked during an introgression analysis between these species. Using ten generations of backcrossing, I will be selecting for the sterile needle-eye phenotype in order to genetically map regions of the genome linked to hybrid sterility. The presence of the needle-eye sterility phenotype in some sperm morphs but not others will allow me to simultaneously identify genomic regions affecting sperm heteromorphism and sterility. Whole genome sequencing will be performed on males of the tenth-generation backcross for specific sperm sterility phenotypes and their fertile brothers. Loci of interest for sterility will be identified as genomic introgressions present in sterile males but not their fertile brothers. Loci for sperm heteromorphism will be identified by comparing males that have sterility in only one sperm morph to those that have sterility in only the other morph. Identifying candidate loci for sterility will help elucidate the mechanisms behind hybrid sterility. Of even greater interest, this study may be the first to identify candidate loci for the production of different sperm morphs.

Identification and characterization of the *Arogenate dehydratase* gene family in soybean

Kelsey Pannunzio, MSc Student

Soybean (*Glycine max*) is one of the most valuable crops worldwide. Employed in animal and human food processing as well as a wide variety of industrial applications, its importance cannot be understated. However, many diseases and pests threaten global yields, with billions of dollars in crop losses annually. Isoflavonoids are legume-specific metabolites of the phenylpropanoid pathway. They are released as part of the

plant-stress response and also promote the attraction of rhizobial symbionts through root nodulation. As such, research targeted towards isoflavonoid biosynthesis in soybean is of paramount interest.

Previously an isoflavonoid biosynthesis metabolon was discovered using isoflavone synthase (IFS) as a bait protein in a Co-IP experiment. In this metabolon, soluble enzymes in the pathway are anchored to the endoplasmic reticulum (ER) by isoflavone synthase (IFS), the final step in isoflavonoid synthesis. One of the potential IFS-interacting partners pulled-down in the Co-IP assay was aroenate dehydratase (ADT). ADT catalyzes the conversion of aroenate into phenylalanine - the precursor to the vast array of phenylpropanoid metabolites. However, ADTs are predicted to localize within the chloroplasts, making them unlikely IFS-interacting partners. In the current study, a genome-wide analysis of soybean ADTs (*GmADTs*) was performed, and 8 *GmADTs* were identified. The *GmADTs* localize to the chloroplasts and show unique expression profiles, both tissue-specific and in response to stress. Furthermore, the ADT-IFS interaction is confirmed to be occurring in-vivo, via bimolecular fluorescence complementation. These findings suggest a novel role for ADTs in isoflavonoid biosynthesis and highlight the importance of these legume-specific metabolites.

Characterization of social spacing behaviour through genetic mutation of *Drosophila* neuroligin 3

Wes Robinson, MSc Student

Fruit flies exhibit simple social behaviours when brought in proximity to other individuals by certain environmental factors such as presence of food. Social space is one of those behaviours. A preferred spacing distance can be quantified in *Drosophila* using social space assays. Determination of spacing could be attributed to many factors such as social experience, genetics, or differential signalling of neural circuitry. So far, it has been determined that the mushroom bodies, a sensory integration center in fly brains, are important for social space. In addition, neuroligin 3 (NLG3), a post-synaptic protein that regulates transmission at the synapse, affects fly social spacing. However, we do not know how these different parts of the neural circuitry are potentially integrated. We want to map the relevant neural circuitry associated with NLG3 in the fly brain to better understand the factors affecting social spacing behaviour. Using genetic and molecular techniques we are looking to identify specific brain regions rich in nlg3 protein and have assessed how different mutations of nlg3 can differentially affect social spacing. Lastly, we will quantify the behaviour of a knockdown in NLG3 specifically within the identified brain regions containing high protein levels which may be facilitating the decision making of the fly. Our results indicate that NLG3 expression is required for standard social spacing behaviour in *Drosophila*.

Oral Presentations – Lightning Talks

Determination of digestive tract pH in the two-spotted spider mite *Tetranychus urticae* (Koch)

Zoran Culo, MSc Student

Spider mites are phytophagous arachnids and major pests to botanical industries such as farming and greenhouses. *Tetranychus urticae* is a generalist spider mite species with a host range of over 1,000 species of plants. Such a vast host range suggests *T. urticae* is capable of successful herbivory while resisting the effects defensive compounds ingested from target plants. The digestive tract of *T. urticae* is the principal interface for nutrient acquisition and detoxification of ingested defensive compounds. Understanding the pH environment within the digestive tract is key to understanding which processes for detoxification and digestion occur there, as pH is a key condition responsible for optimal performance of enzymes. The objective of my project is to determine the pH environment of the digestive tract by using multiple pH-sensitive dyes (PSDs) with unique transition ranges. I fed the PSDs to newly-molted adult female *T. urticae* mites, each with a single PSD. The PSDs changed color depending on the pH inside the mite's digestive tract. The color of the PSDs indicated if the pH of the alimentary tract was higher, lower or equal to their transition range values. I was able to determine the lumen pH of the posterior midgut to be between 6.8 and 8.0. The determined pH range will thus contribute to determination of potential digestive processes within the posterior midgut. My next steps are to fulfill the rest of my objective by using PSDs on the remaining areas within the *T. urticae* digestive tract.

Neuroigin3 is required for a response to the social environment in *Drosophila melanogaster*

Ryley Yost, MSc Student

The social environment affects behaviour by altering molecular processes and neuronal functioning. In *Drosophila melanogaster*, social isolation affects complex behaviours including interactions with others, as well as, social spacing: the distance between flies. Social isolation is known to cause changes in gene expression, including *Drosophila* neuroigin3 (*nlg3*), an autism-associated post-synaptic cell adhesion protein. Neuroigin3 ensures proper synaptic formation, maturation, and function and is important for proper social spacing. However, the involvement of *nlg3* in response to isolation and how this alters social space is unknown. Using the social space assay, we observed that flies lacking *nlg3* did not respond to social isolation. Furthermore, Western blot analysis of NLG3 protein levels revealed that NLG3 was unchanged after isolation, indicating it is required for a response to the social environment but not solely responsible for the effect of isolation on social space. This is the first evidence of the involvement of *Drosophila* *nlg3* influencing the response to environmental stimuli. These findings begin to shed light on the genetic and environmental regulation of behaviour and the underlying molecular processes could be conserved in other organisms.

Determining the effect of mistranslating tRNAs on development, behaviour, and disease

Joshua Isaacson, PhD Student

Proteostasis is process that controls the production, maintenance, and degradation of proteins within cells. Disruptions to proteostasis, which can occur through aging or disease, can greatly affect cell and organism viability. My goal is to understand how mistranslation of transfer RNAs (tRNAs) can affect behaviour and viability, particularly when mistranslation is accompanied by latent conditions that affect proteostasis. I have integrated mutant tRNA genes that mistranslate serine at proline codons into the fruit fly, *Drosophila melanogaster*, to characterize the effect of mistranslation on behaviour, development, and lifespan. To determine if mistranslating tRNAs can exacerbate diseases that affect proteostasis, I have created strains that contain mistranslating tRNAs and a Huntingtin gene with a polyglutamine expansion (Htt-PolyQ). Preliminary assays have shown that even mild mistranslation has enhanced the severity of neurodegeneration in flies. Other neurodegenerative diseases, such as Parkinson's Disease or amyotrophic lateral sclerosis, could also be tested for interactions with mistranslating tRNAs. My work represents the first time the effect of mistranslating tRNA genes have been characterized in a multicellular organism and would help elucidate how organisms tolerate disruptions to proteostasis. It could also uncover a novel enhancer of neurodegenerative and other age-related diseases that affect a substantial fraction of the human population.

Warming homogenises springtail communities in boreal forest soils prioritising small bodied species

Jordan Kustec, MSc Student

Climate and anthropogenic change can act as ecological stressors within soil communities. Soils are understudied compared to terrestrial and aquatic systems in the context of ecological shifts and impacts on communities, despite being responsible for carbon storage and decomposition processes. Boreal forest floor soil is particularly important in the context of climate change, where greater average temperature increases are expected within this zone that covers 29% of the world's forested area. Anthropogenic inputs of nitrogen from agriculture are increasing nitrogen deposition rates in the boreal forest as well. One particular group of soil animals abundant in the boreal forest system that are vulnerable to anthropogenic change are springtails (Collembola: Hexapoda), which function as secondary decomposers in the soil system. My research objective was to assess springtail community level responses under warming and increased rates of nitrogen deposition in a three year study in boreal forest soils. Springtail communities shifted to become more homogenous under warming and shifted under nitrogen deposition. Communities body sizes decreased under warming due to an increase of small bodied individuals. The results of this study indicate that warming is affecting springtail community structure and body size, which can lead to a loss of functional diversity within soils, which can impact soil carbon and nutrient cycling.

Does heterozygosity breed mutations: The case of the outbred mice

Hailie Pavanel, MSc Student

Across genomes, the level of mutation is not a constant value. The reason why is not fully understood. It has been reported that heterozygosity correlates with increased mutagenesis in *Arabidopsis*. Further elucidation of this phenomenon would clarify why mutation frequency differs along the landscape of a genome. This study investigates the correlation between heterozygosity and deletions and duplications in two outbred mouse stocks, CD-1 (Caesarian-derived 1) and NMRI (Naval Medical Research Institute). Publicly available Mouse Diversity Genotyping Array (MDGA) data were used to detect single nucleotide polymorphisms (SNP) and copy number variants (CNV) in 99 CD-1 mice and 279 NMRI mice. SNP genotypes can be homozygous – containing the same alleles or heterozygous – containing different alleles. SNP genotypes were used to identify localized heterozygosity and CNVs were assayed as an indicator of associated mutations. The level of heterozygosity was found to be 10.7 and 6.2% and there were approximately 17 and 14 CNVs per mouse in CD-1 and NMRI mice, respectively. Clusters of heterozygosity were observed across the autosomes in both stocks. Association of clusters of heterozygous SNPs and CNVs was investigated to see if CNVs tended to co-occur in clusters of heterozygosity. 32.4% and 13.0% of chromosomes containing CNVs analyzed in CD-1 mice and NMRI mice, respectively, display proximal association between heterozygous SNPs and CNVs. CD-1 mice display more proximal than distal association, whereas NMRI mice do not. Further analysis of links between heterozygosity and mutagenesis, is relevant to better understanding the origins of genetic variation in health and disease.

FASD: an epigenetic neurodevelopmental disorder modeled in mice

Bonnie Alberry, PhD Student

Most neurodevelopmental disorders involve genes, environment and poorly understood epigenetic interactions that are multifaceted, dynamic and complex. Clarifying these interactions is critical but difficult, requiring a suitable model. We propose Fetal Alcohol Spectrum Disorder (FASD), a common neurodevelopmental disorder resulting from prenatal alcohol exposure modeled in C57BL/6J mice, is highly suited for this. Here we demonstrate that following continuous mild prenatal alcohol exposure in mice during neurodevelopment, behaviour phenotypes seen in FASD are present. Also, they have many lasting hippocampal molecular changes including genome-wide gene and microRNA expression (RNA-Seq), as well as DNA methylation (meDIP-Seq). The relationship between these features is not understood. It may account for initiation and persistence of behavioral abnormalities in FASD. Interestingly, DNA methylation alterations in response to prenatal alcohol exposure in mice have also been demonstrated in buccal swabs from children with FASD. More importantly, postnatal manipulations or treatment paradigms potentially applicable to children can be applied and tested in young mice during development. In fact, behavioural outcomes can be improved or worsened by favourable and unfavourable postnatal environments in mice

subject to prenatal ethanol exposure, in this case early life stress via maternal separation. Additionally, behavioural outcomes are reflected in newly established molecular features in response to postnatal environment. Critical genes and pathways identified may provide a foundation for development of FASD diagnosis using DNA methylation and interventions for children born with FASD. Finally, pathways and ontologies associated with these features implicate a role for epigenetic regulation of gene expression common in neurodevelopmental disorders.

Bacterial activity, community composition, and mercury methylation along a latitudinal sulphate deposition gradient in Ontario peatlands

Jennifer Blythe, MSc Student

Peatlands are wetlands typically dominated by sphagnum mosses, with over 40cm of organic soil. Their nutrient-poor, inundated soils are dominated by a variety of anaerobic microbes, including sulphate-reducing bacteria (SRB). The SRB are the principal methylators of inorganic mercury (Hg) in the natural environment, and are the primary reason peatlands are known "hotspots" of methylmercury (MeHg) production. The factors that control the activity, growth, and composition of SRB communities are of particular relevance to ecosystem health as MeHg is the bioaccumulating, and neurotoxic form of Hg. As a key nutrient in SRB metabolism, sulphate constitutes a well known control on SRB methylation of Hg. Recent research suggests that elevated sulphate deposition to peatlands may shift the composition of the SRB community, with potential consequences for bacterial Hg methylation in response to further sulphate inputs.

The goal of my MSc research is to investigate these proposed community shifts in response to elevated sulphate deposition, and to link community composition with MeHg production. To do this, I am performing a series of laboratory experiments in which sulphate solutions are applied to peat cores in a flow-through system. The peat cores were collected from three sites along a latitudinal sulphate gradient in Ontario. Preliminary results suggest that the bacterial community in the southern peat from Sifton Bog in London, Ontario where historic sulphate deposition has been the highest, does not exhibit an increased Hg methylation response compared to the more northern peat from White River, Ontario when exposed to the same level of sulphate. I will further investigate whether this difference in methylation response can be attributed to differences in bacterial community composition or activity between the two sites.

Study the relationship of *HD2* family genes in *Arabidopsis thaliana* against drought stress

Muhammad Tahir, PhD Student

Histone deacetylases (HDACs) are important modulators of core histone proteins. Plants contain a specific type of HDACs family called Histone Deacetylase2 (HD2) along with RPD3 and SIR2 families. HD2 family consists of HD2A, HD2B, HD2C and

HD2D genes in *Arabidopsis*. Characterization of HD2s has indicated that the members of this family play an important role in regulating gene expression in several biological processes, especially in abiotic stress response. I hypothesized that members of HD2 family interact with each other in mediating the plant response against drought stress in *Arabidopsis*. To verify this hypothesis, firstly in-vitro Yeast 2 Hybrid assay was performed to study the interactions. HD2A showed interaction with HD2C and HD2D proteins. Also, HD2C showed interaction with HD2D. None of the HD2 family members showed interaction with HD2B. These results indicate that HD2A, HD2C and HD2D proteins might act together to regulate target gene expression. Interaction results of Y2H assay will be verified by BiFC. To study the relationship of HD2 genes in *Arabidopsis*, hd2 single mutant lines have been obtained which will be used to develop hd2 double and triple mutant lines. HD2 genes have been cloned into expression vector under 35S promoter to develop HD2 over expressing lines. Gene expression analysis will be performed to analyse the impact of HD2 gene knockouts on other HD2-type gene expression in mutants against drought. Knowledge generated from this research will be useful to improve understanding about the interactional role of HD2 protein family in mediating the plant response against drought stress.

Identifying interacting proteins with histone deacetylase BdHD1 in *Brachypodium distachyon*

Alberto Torrez, MSc Student

Stress-responsive genes can be mediated by epigenetic changes. Current evidence has indicated the involvement of histone deacetylases (HDACs) in plant stress responses. Histone deacetylases 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, as there is limited HDAC research within monocots. *Brachypodium distachyon* is used as a model plant to investigate questions unique to monocot plants. BdHD1 has been identified in *B. distachyon* which belongs to the HDAC1-type HDAC. 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 investigating by using yeast two-hybrid (Y2H) assays and bimolecular fluorescence (BiFC) assays. Based on my Y2H assay results, BdHD1 showed a strong interaction with WRKY24, and 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.



On the genetical evolution of altruism: The importance of genetic responsiveness to changing social environments

Vonica Flear, MSc Student

Genes underlying altruistic behaviors such as self sacrifice and sterility are prevalent in a number of ant, bee, wasp, and termite species. These behaviors have been a point of fascination for evolutionary biologists because they pose a problem for traditional Darwinian natural selection: How could genes that cause a detriment to personal fitness evolve? Hamilton's inclusive fitness theory has illustrated how indirect selection may overcome the direct fitness costs (c) of altruistic behavior if the benefit of the behavior (b) is sufficiently large, and both interactants possess the gene identical by descent (r), i.e. if $rb > c$. As a result, reproductive altruism can only evolve via indirect selection such that genes for altruism (G) are transmitted, but not expressed, by reproducing relatives. Hamilton's work specified how G could only evolve if its expression pattern followed a conditional rule causing some kin to express it, and others not. However, Hamilton did not explore how G may evolve if its expression were insensitive to social circumstance. The aim of my study is to determine the importance of social environment in the evolution of genes for altruism and discover how stringent the conditional rule must be for G to evolve. To do this, I am developing models of varying conditional expression rules and using the Price formula to determine whether the frequency of G is increasing or decreasing in over one generation. I will further explore the stability of these conditions by introducing a penetrance level of G in each model.

Beyond amyloid: The metabolic regulator p66Shc as a therapeutic target for Alzheimer's disease

Asad Lone, PhD Student

The amyloid hypothesis has dominated drug discovery and therapeutic strategies in Alzheimer's disease (AD) for the last 20 years, regardless of several unsuccessful clinical trials. A significant population of the elderly have pronounced amyloid beta ($A\beta$) deposition within their brains, yet show no symptoms of dementia, indicating that some cells are resistant to $A\beta$ toxicity. Several studies suggest that CNS cells that are resistant to $A\beta$ toxicity display a metabolic shift from mitochondrial-dependent oxidative phosphorylation (OXPHOS) to aerobic glycolysis for their energy needs. Expression & activation of the adaptor protein p66Shc has been shown to shift the cellular metabolic state from OXPHOS to aerobic glycolysis. Hence, we propose that the expression & activation of p66Shc in neuronal and glial cells promotes both increased OXPHOS and sensitivity to $A\beta$ toxicity. Expression and activation of p66Shc repressed glycolytic enzyme expression and increased mitochondrial electron transport chain activity and ROS levels. The opposite effect was observed when endogenous p66Shc expression was knocked down. Activation of p66Shc increased sensitivity to $A\beta$ toxicity, whereas silencing p66Shc protected cells from $A\beta$ insult. Preliminary data hints to p66Shc's metabolic & protective effects being mediated through the transcription factor NRF2. Thus, expression and activation of p66Shc renders CNS cells more sensitive to $A\beta$ toxicity by promoting mitochondrial OXPHOS while repressing aerobic glycolysis, and p66Shc may represent a potential therapeutically relevant target for AD.

Posters

P1. The effect of age on the astrocyte-neuron lactate shuttle and memory

Ariel Frame, PhD Student

Aims:

Over the last decade glial cells have emerged as key regulators of memory. One way glial cells influence memory is by aiding in brain metabolism. Studies in vertebrate models such as mice, rats and chicken have revealed that astrocyte derived lactate promotes long-term memory (LTM). Specifically, astrocytes convert pyruvate to lactate during the last step in glycolysis using the enzyme lactate dehydrogenase and this metabolite is subsequently shuttled to neurons for fueling oxidative metabolism. Compromised neuro-metabolic coupling may underlie age-related memory impairment observed in most animals; a theory that has yet to be formally tested. The recent finding that glial glycolysis in the brain of fruit flies (*Drosophila melanogaster*) provides a source of lactate to ensure neuronal survival indicates that invertebrate brain glia may function similarly to the astrocyte-neuron lactate shuttle present in vertebrates. The objective in this study was to determine whether lactate metabolism in flies can influence LTM formation.

Methods:

Drosophila lactate dehydrogenase (dLdh) expression was altered in glial cells. Memory was assessed using courtship-conditioning, whereby the ability of male flies to remember exposure to unreceptive females 24 hours prior is a measure of LTM. dLdh protein level was measured using western blot analysis.

Results and Conclusions:

In wild-type flies, dLdh expression increases with age and LTM decreases. Preliminary results suggest that elevated dLdh expression is detrimental to the health of flies and reducing dLdh may increase lifespan and promote LTM. These preliminary findings suggest that elevated lactate production may actually be detrimental to central nervous system function, particularly with age.

P2. Express yourself: AROGENATE DEHYDRATASES

Jordan Van Brenk, MSc Student

Phenylalanine (Phe) is a building block for proteins and serves as a precursor to many specialized metabolites in plants, such as flavonoids (pigments and scents) and lignins (structural organic polymers). Phe is not synthesized in animals, who acquire this essential amino acid via diet. Understanding Phe synthesis and regulation is crucial for understanding the importance of Phe for both plants and animals.

In *Arabidopsis*, the AROGENATE DEHYDRATASES (ADTs) encode six enzymes (ADT1-ADT6) catalyzing the final step of Phe biosynthesis. These six ADTs have been shown to differentially channel carbon to downstream end uses. For example, lignin biosynthesis is more dependent on ADT5 than the other ADTs. Therefore, we expect all

ADT expression levels to be individually regulated in response to internal and environmental cues. To determine how their expression differs, we used microarray data from the BAR Database eFP Browser to visualize ADT expression patterns across different *Arabidopsis* tissues and developmental stages.

To best compare patterns among ADTs, we discovered that different RNA expression thresholds needed to be set as it allowed comparison of high and low values, without sacrificing resolution. This way, along with patterns among ADTs, we are also able to directly compare between any two ADTs. We found that although ADTs are almost ubiquitously expressed in tissues, their patterns of expression do, in fact, differ.

These in silico analyses can now be tested by in vivo experiments. An in vivo study using ADT promoter-reporter constructs is currently being performed to fine-tune and expand the study of ADT expression.

P3. Niche segregation among three sympatric species of swallows in Southern Ontario

Kaelyn Bumelis, MSc Student

Aerial insectivores have experienced the highest rates of declines of all avian guilds in North America but causes of these declines are unknown. The first year of life is thought to have the lowest survivorship and so a focus on this early life stage is likely to be informative. Barn Swallows (*Hirundo rustica*), Cliff Swallows (*Petrochelidon pyrrhonata*) and Tree Swallows (*Tachycineta bicolor*) are aerial insectivores which are often sympatric and are experiencing differential population declines. So, understanding ecological differences among these species may provide insights into causes of their differential population trajectories. Similar ecological species avoid competition through resource partitioning and so tend to occupy different ecological niches. These factors may benefit species differentially and be linked to population trajectories. By studying nestling diet and post-fledging movement of the three species in sympatry, I hope to gain insight into how niche segregation among these species may be contributing to differential population declines. I predict that these swallow species will avoid competition by exploiting different diets, in turn facilitated by differential space use during foraging and differences in post-fledging movements. Regardless, this information will be useful for conservation and management of these species by helping to identify potential factors contributing to differential declines operating on the breeding grounds.

P4. Long-term impacts of fungicides on soil

Farhaan Kanji, MSc Student

Azole-based compounds with antifungal properties prevent the production of ergosterol, which is an essential cell wall component of many fungi and protozoa, by inhibiting the enzyme lanosterol 14 α -demethylase. Together with the echinocandin and polyene classes, azoles comprise a major class of first-line antifungals used for treating conditions such as dermatophytosis (ringworm), tinea pedis (athlete's foot), and

candidiasis (thrush). However, unlike other antifungals used for human medical conditions, azoles are also commonly deployed for use as antifungals in agricultural and industrial settings. Thus, the purpose of this study is to determine the extent of antifungal resistance in 12 mock agricultural plots receiving 0, 0.1, or 10 mg/kg soil of clotrimazole and miconazole since 2011. It is anticipated that approximately 100 fungal isolates from the plots will be isolated by plating the soil samples on media selective for fungi and the identity of the fungal isolates will be determined using phenotypic and genomic classification methods. Overall, we anticipate that the phenotypic and genomic data gathered from the fungal isolates will shed light on the prevalence, mechanisms, and severity of the spread of fungicide resistance over time.

P5. The effect of climate change and global warming on harmful algal blooms growth

Malihe Mehizadeh Allaf, PhD Student

Climate change is a driving force for increasing harmful algal blooms in oceans. *Heterosigma akashiwo* is a golden-brown dinoflagellate with a high potential to cause catastrophic fish kills at mariculture facilities around the world. Increasing the temperature, variation in salinity, and increase of CO₂ concentration which results in ocean acidification, are the three major consequences of climate change in oceans. The effect of this three parameters on the growth of *Heterosigma akashiwo* was studied in this work, using design of experiment (DOE) approach. The benefit of using DOE approach is to investigate the effect of more than two factors on the same response simultaneously while the interaction of these factors can be estimated in a more precise manner as well. The results suggested that the optimum conditions to produce maximum growth was observed when the temperature was 25 °C, salinity of 30 and CO₂ level of 400 ppm.

P6. Nitrogen source and availability alter low CO₂ effects on plant growth and N isotopic composition

André Duarte, PhD Student

Low atmospheric CO₂ conditions prevailed for most of the recent evolutionary history of plants. While low CO₂ concentrations reduce photosynthesis and therefore plant growth in comparison to modern CO₂ levels, the effect of low CO₂ on plant performance may also be affected by nitrogen availability. Plants often display a preference towards a specific form of inorganic N, although preference for either NO₃⁻ or NH₄⁺ can shift depending on environmental conditions. To investigate how different forms of inorganic N can affect plant performance at low CO₂, we grew *Elymus canadensis* fertilized with either NO₃⁻, NH₄⁺, or a mixture of both at CO₂ levels representing modern conditions (~ 400 ppm) and the Last Glacial Maximum (~ 180 ppm). Plants in each CO₂ treatment were split into three N-fertilization regimes: nitrate-only, ammonium-only, and a full mix of nitrate and ammonium. Regardless of N treatment, low CO₂ reduced plant biomass and root [C] but increased specific leaf area in comparison to ambient CO₂-grown plants. Reductions in growth were less pronounced in the nitrate in comparison to the ammonium treatment, implying that NO₃⁻ assimilation was facilitated by low CO₂ and

higher photorespiration rates. Compared to plants that were fertilized with a full mix, low CO₂ plants in the nitrate and ammonium treatments showed an up-regulation of net CO₂ assimilation rate when measured at 400 ppm CO₂, which was correlated with higher maximum electron transport rates (J_{max}); while maximum Rubisco carboxylation rates (V_{cmax}) were not affected by the CO₂ treatment. In summary, many of the parameters analyzed were affected by the interaction of growth at low CO₂ and different N-fertilization regimes. Considering that N-sources available for plant uptake varies considerably, consideration of past and present nitrogen natural variability is important for generating a more ecologically realistic approach to low CO₂ studies.

P7. Developing an IgA biobetter antibody against *E. coli* O157:H7 with enhanced stability by rational design of a bovine Fc chain

Adam Chin-Fatt, PhD Student

Immunoglobulin A (IgA)-nanobody fusions are useful *E. coli* O157:H7 therapeutics that modularly comprise two highly ordered constant domains, collectively called the fragment crystallisable (Fc), fused to an antigen binding partner known as the variable heavy chain fragment (VHH) derived from camelids. Towards the goal of improving the accumulation of the VHH-Fc fusion, we employed rational design to improve the stability of the Fc. Two rational design strategies, supercharging and disulfide bond introduction, were tested for their effects on accumulation. Since a plant platform offers the prospect of oral delivery, the mutagenized candidates were transiently expressed in leaves of *Nicotiana benthamiana* and then screened for protein accumulation, thermostability and solubility. We have identified and characterised five supercharging and one disulfide mutant that, in comparison to native, all improve accumulation and enhance thermostability as either Fc alone or when fused to a VHH that binds *E. coli* O157:H7. We also have found that pyramiding of these mutations result in a complementary increase of accumulation. A binding assay also showed that binding efficacy is retained in the engineered fusion compared to native. A neutralisation assay also showed that the engineered fusion prevents adhesion of the bacteria to mammalian gut cells similar to native. The goal of this project is to demonstrate as a proof of concept that a bovine Fc chain can be rationally designed for improved stability and be a viable strategy for improving accumulation of the therapeutic without sacrificing efficacy.

P8. Patellin 3 and 6, two members of the *Arabidopsis* Sec14-GOLD proteins, interact with both the coat protein (CP) and cylindrical inclusion protein (CI) of Turnip mosaic virus and inhibit virus accumulation

Zhaoji Dai, PhD Student

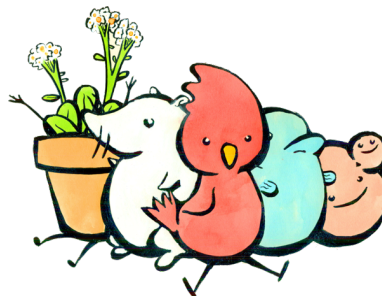
A successful viral infection results from complex interaction between an invading virus and a particular host. Molecular identification of host proteins that affect viral infection will advance knowledge of the viral infection process and assist in the development of novel antiviral strategies. I used the coat protein (CP) of Turnip mosaic virus (TuMV) as bait and conducted a yeast two-hybrid (Y2H) screening for CP-interacting *Arabidopsis* proteins. I identified several novel protein interactions that could potentially modulate TuMV infection. In this study, I focused on two of the novel CP-interacting partners,

PATL3 and PATL6, which are members of the *Arabidopsis* Patellin protein family that are implicated in membrane trafficking, cytoskeleton dynamics and lipid metabolism. The ability of PATL3 associated with CP was confirmed by bimolecular fluorescence complementation (BiFC) assay and co-immunoprecipitation (Co-IP) assay in planta. The biological significance of PATLs was assayed by TuMV infection of PATLs T-DNA insertion mutants and stable overexpression lines. Knockout of PATL3 or PATL6 or both resulted in strongly increased accumulation of TuMV genomic RNA in the inoculated *Arabidopsis* leaf. A comparative analysis of viral accumulation in protoplasts isolated from wild-type plants or the knockout mutants showed that the absence of PATL3 and PATL6 facilitates viral multiplication, whereas overexpression of PATL3 inhibits TuMV accumulation in *Arabidopsis* protoplasts. These data suggest that PATL3 and PATL6 negatively regulate TuMV infection in *Arabidopsis* plants and protoplasts. Interestingly, subcellular localization study demonstrated that PATL3, but not PATL6 was redistributed to and co-localized with virus replication complexes (VRCs) upon TuMV infection. In addition, the viral protein cylindrical inclusion protein (CI), which presents in VRCs and are critical for TuMV multiplication, interacted with PATL3 both in yeast and in planta. Therefore, I proposed that *Arabidopsis* protein PATL3 act as a host restriction factor, possibly by targeting TuMV VRCs through its interaction with CI.

P9. Dietary costs and benefits of lakeshore vs aggregate pit breeding in Bank Swallows (*Riparia riparia*)

Corrine Genier, MSc Student

Bank Swallows (*Riparia riparia*), a threatened species in Ontario, breed primarily in either banks at lakeshores or at exposed surfaces in man-made aggregate pits. Pits are suspected to be ecological traps for this species but the relative trade-offs in nesting at pits vs. natural lakeshores are poorly known. Availability of aquatic emergent insects is expected to be highest at lakeshore colonies with associated nutritional benefits including a richer Omega-3 fatty acid diet. However, Bank Swallows may experience higher mercury (Hg) exposure at lakeshores. Potential differences in dietary quality among sites may directly influence juvenile body condition, with an expected higher body condition at lakeshores. This study seeks to compare these breeding habitats to evaluate dietary differences as revealed by fatty acid, stable isotope, and mercury analyses in tissues, and DNA barcoding of fecal matter. This information will be important for management decisions related to the use of pits by this species and conservation of suitable nesting habitats.



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